DEVELOPMENT AND EVALUATION OF FLOW THROUGH OHMIC HEATING ASSISTED PULSED LIGHT TREATMENT SYSTEM FOR PRESERVATION OF FRUIT JUICE

by

ASHITHA G N

(2016-28-001)

DEPARTMENT OF PROCESSING AND FOOD ENGINEERING

KELAPPAJI COLLEGE OF AGRICULTURAL ENGINEERING AND TECHNOLOGY

TAVANUR, MALAPPURAM-679573

KERALA, INDIA

2020

DEVELOPMENT AND EVALUATION OF FLOW THROUGH OHMIC HEATING ASSISTED PULSED LIGHT TREATMENT SYSTEM FOR PRESERVATION OF FRUIT JUICE

by

ASHITHA G N

(2016-28-001)

THESIS

Submitted in partial fulfillment of the

requirement for the degree of

DOCTOR OF PHILOSOPHY

IN

AGRICULTURAL ENGINEERING

(Agricultural Processing and Food Engineering)

Kerala Agricultural University

DEPARTMENT OF PROCESSING AND FOOD ENGINEERING

KELAPPAJI COLLEGE OF AGRICULTURAL ENGINEERING

AND TECHNOLOGY

TAVANUR, MALAPPURAM-679573

KERALA, INDIA

2020

DECLARATION

I, hereby declare that this thesis entitled **"DEVELOPMENT AND EVALUATION OF FLOW THROUGH OHMIC HEATING ASSISTED PULSED LIGHT TREATMENT SYSTEM FOR PRESERVATION OF FRUIT JUICE"** is a *bonafide* record of research works done by me during the course of research and the thesis has not previously formed the basis for the award to me of any degree, diploma, associateship, fellowship or other similar title, of any other University or Society.

Place: Tavanur **ASHITHA G N**

Date: **(2016-28-001)**

CERTIFICATE

Certified that this thesis entitled **"DEVELOPMENT AND EVALUATION OF FLOW THROUGH OHMIC HEATING ASSISTED PULSED LIGHT TREATMENT SYSTEM FOR PRESERVATION OF FRUIT JUICE"** is a record of research work done independently by **Ms. ASHITHA G N (2016-28-001)** under my guidance and supervision and that it has not previously formed the basis for the award of any degree, diploma, fellowship or associateship to her.

Place: Tavanur,

Date:

Dr. PRINCE M V

(Major Advisor, Advisory Committee)

Professor and Head, Dept. of PFE

KCAET, Tavanur

Dedicated to

My Parents and Teachers

Acknowledgement

ACKNOWLEDGEMENT

It is with extreme contentment and reverence, that I express my sincere gratitude to my chairman ***Dr. Prince M V****, Prof. and Head of Dept. of PFE, KCAET, for having showered up on me his profound knowledge, rich experiences, valuable advice, support and encouragement throughout the course of this endeavour. It was his trust in me that fuelled my efforts towards successful completion of this work against all odds.*

I extend my sincere thanks and gratitude to ***Dr. Sathian K. K.,*** *Dean, Prof. and Head of Dept. of SWCE, KCAET, Tavanur, for the help and encouragement rendered during the course of the work. I am highly obliged to my advisory committee member,* ***Dr. Sudheer K. P.,*** *Prof. and Head, Department of Agrl. Eng., COH, Vellanikkara, whose constant advice and help has been a critical part of this work. It was extremely kind of him for having taken time off his busy schedule to do necessary corrections in my research. My sincere thanks to advisory committee member* ***Dr. Rajesh G. K.****, Asst. Prof., Dept. of PFE, KCAET, for his timely advice, suggestions, support and motivation which has helped greatly in improving quality of my work.*

Words fall short in putting across my gratitude to my advisory committee member ***Dr. Jayan P. R.,*** *Prof. and Head, Dept. of FPME, KCAET, for extending his whole hearted support and encouragement for this work. I take great pleasure in extending thanks to advisory committee member Smt.* ***Sreeja R.,*** *Asst. Prof., Dept. of PFE, KCAET, for her incessant guidance in biochemical analysis, encouragement, and support throughout my research work.*

I humbly express my wholehearted thanks to ***Dr. Santhi Mary Mathew,*** *former Dean, Retired Prof., Dept. of PFE, KCAET, for her love, care and timely advice in each stages of my research work. I also express my gratitude towards* ***Er.George Mathew****, Retired Asso. Prof., for his support, advice and motivation throughout my research.*

I would like to express my gratitude towards ***Kamen technologies****, Coimbatore, for the technical support offered for the development of Pulsed Light system. I also wish*

to express my sincere wholehearted gratitude and special thanks to ***Er. Pragadeesh, Er. Vijayan*** *and* ***Er. Aswin****, for their assistance in the design of electronic circuit systems.*

I owe my indebtness to ***Dr. Jalaja Menon****, Prof. and Head ,* ***Mr. Manoj*** *and other staffs of Cashew Research station, Madakkathara, for providing the raw materials and background support for my research. I place on record my sincere thanks to* ***Dr. Ganapathy****, Prof. and Head, Dept. of PFE, TNAU and* ***Nisha R,*** *Ph.D. Scholar, TNAU, for permitting and helping me to carry out a part of my research in TNAU laboratory. I express my gratitude to* ***MTCC****, Chandigarh, for providing bacterial culture for my research work.*

I express my sincere gratitude towards ***Dr. Riju C.****, Asst. Prof. (C), Microbiology, KCAET, for assisting and guiding me in all the microbiological aspects of my research work. I am thankful to* ***Mr. Radhakrishnan, Mrs. Geetha, Mr. Vipin, Mr. Lenin, Mr. Sudheer, Mr. Surjith, Mr. Shibu, Mr. Manoj, Mrs. Jojitha, Er. Praveena N.*** *and other non-teaching staffs of Dept. of PFE, KCAET, for their valuable help rendered during the period of study.*

I am thankful to ***Er. Namitha M. R., Er. Athira Prasad*** *and* ***Er. Aswathi M. S****., Asst. Prof. (C), KCAET, for their valuable help rendered during the period of study. I also take this chance to gratify my seniors* ***Dr.Sankalpa K. B. and Er. Pritty S. Babu*** *for their advice and support. I wish to place on record constant support provided by my batch mates* ***Dr. Dipak S Khatawker, Er. Gilsha Bhai and Er. Jyothi Narayanan*** *for their timely assistance and support. Words cannot express the gratitude towards my friends* ***Er. Anjaly C Sunny, Er. Mamatha Prabhakar, Er. Shahama and Er. Arya K T*** *for their support, constant encouragement, and motivation from the beginning to the end of my research work. I express my sincere thanks to my juniors* ***Er. Amrutha K, Sanjay P and Anjaly Palekkal*** *for the assistance and support during my thesis work.*

I'm highly indebted to my Father ***Mr. Gopi M.*** *and my mother* ***Mrs. Nishi P.*** *for supporting me throughout during this ardous pursuit. I express my ineffable gratitude to my sister* ***Mrs. Anjusha G. N.*** *and brother in law* ***Mr. Biljith*** *for their unrelenting support and encouragement.*

Last but not the least, I owe everything to ***God Almighty*** *for strengthening me to overcome all odds and kept me moving towards the successful completion of this endeavour.*

(Ashitha G N)

CONTENTS

LIST OF TABLES

LIST OF FIGURES

LIST OF PLATES

SYMBOLS AND ABBREVIATIONS

%	:	Percentage
<	:	Less than
=	:	Equal to
°C	:	Degree Celsius
µF	:	Micro Farad
µg	:	Micro gram
µs	:	Micro second
A	:	Ampere
AC	:	Alternating current
ANOVA	:	Analysis of variance
AOAC	:	Association of Official Analytical Chemistry
C	:	Capacitance of the capacitor
cfu	:	Colony forming unit
cm	:	Centimeter
CV	:	Coefficient of variation
df	:	Degrees of freedom
DNA	:	Deoxyribo Nucleic Acid
E. coli	:	*Escherichia coli*
Eq. wt	:	Equivalent weight
et al	:	And others
etc	:	Et cetera

F value	:	Fischer value
FAO	:	Food and Agricultural Organization
FDA	:	Food and Drug Administration
FSSAI	:	Food Safety and Standards Authority of India
g	:	Gram
Ha	:	Hectare
HACCP	:	Hazard Analysis Critical Control Point
HHP	:	High hydrostatic pressure
HIPL	:	High intensity pulsed light
Hz	:	Hertz
I	:	Current
i.e.	:	That is
IPL	:	Intense pulsed light
IUPAC	:	International Union of Pure and Applied Chemistry
J	:	Joule
K	:	Consistency Index
kg	:	Kilogram
kHz	:	Kilo Hertz
kV	:	Kilo Volts
L. monocytogenes	:	*Listeria monocytogenes*
L_λ	:	Absorption length
m	:	Metre

m^2	:	Square meter
Min	:	minute
mJ	:	Milli joule
ml	:	Milli litre
ml	:	Milli litre
mm	:	Milli meter
mm	:	Milli meter
MPa	:	Mega Pascal
MSS	:	Mean sum of square
MT	:	Metric tonnes
n	:	Flow behavior index
NACMCF	:	National Advisory Committee on Microbiological Criteria for foods
NEBI	:	Non-enzymatic browning index
NTU	:	Nephelometric turbidity unit
OH	:	Ohmic Heating
P value	:	Probability value
PEF	:	Pulsed electric field
PL	:	Pulsed light
PME	:	Pectin methyl esterase
Prr	:	Pulse repetition rate
PUV	:	Pulsed ultra violet
RTD	:	Resistance temperature detector

s	:	Second
Sm^{-1}	:	Siemens per meter
SS	:	Sum of Square
TPC	:	Total plate count
Tr	:	Transmittance
TSS	:	Total soluble solid
Tur	:	Turbidity
UHT	:	Ultra High Temperature
US	:	Ultra sound
UV-C	:	Ultraviolet light –C
V	:	Voltage
viz.	:	Namely

Introduction

CHAPTER I

INTRODUCTION

Fruits and vegetables are the important supplements to the human diet as it provide the essential minerals, vitamins, and fibre required for maintaining good health. The consumer's outlook towards the daily diet plans has been changed over recent years and they are more aware of the importance of inclusion of the fruits and vegetables in their daily diet. India is the second largest producer of the fruits and vegetables in the world with a production of 259 million tonnes (NHB, 2017). However, there exists a huge gap between the per capita availability of fruits and the recommended intake levels, due to huge post harvest losses. An annual wastage of 4.6-15.9% is witnessed in fruits and vegetables, owing to lack of efficient post harvest management and effective technological interventions (MoFPI, 2015). Also, only about 2% of the total production of fruits and vegetables are processed to value added products (MoFPI, 2015). Value addition of perishable commodities adopting cost effective, indigenous, state of the art and region specific technologies could resolve this issue to a considerable level.

The major share of the processed food sector includes products like juice, jam, jelly, squash, fermented beverages *etc.* Fruit juices are enjoyed by people due to its good taste and flavour apart from its usual thirst quenching ability and are consumed in fresh and processed form. Also it supply vital components to enervate the physiological condition of human beings both physically and mentally (Bates *et al.,* 2001). Seasonality of the fruit production restricts availability of fresh fruit juices throughout the year. There has been a long run for the development of suitable technologies in food industry for making minimally processed fruit juices available on consumer shelf throughout the year.

Pineapple (*Ananas comosus*) is well-known for its unique flavour and taste as well as beneficial nutritional components. It is an excellent source of calcium, potassium, and vitamin C and also contains, vitamin B6, vitamin B1, copper, and dietary fibre. Pineapple contains bromelain, an important proteolytic enzyme complex. Being rich in antioxidants and proteolitic enzymes pineapple juice has been proven to lessen the occurrence of

cardiovascular disease, inflammation and some chronic and degenerative diseases allied with oxidative damage. India is a leading producer of pineapple with a production of 1.86 million tonnes (2016-17). Pineapple is a major tropical fruit crop in Kerala and marked as the 2nd highest producer (3.10 lakh tonnes) in the country (NHB, 2017). The processing and long term storage of fresh pineapple juice is hectic without the use of chemical preservatives and thermal processing due to reduction of ascorbic and citric acid contents and colour changes. The flavour compounds of pineapple fruit is extremely sensitive to transformations taking place during thermal processing (Barros *et al.,* 2003). Hence it is essential to adopt non thermal processing technologies for the preservation of fresh fruit juices without losing its authenticity.

Cashew tree (*Anacardium occidentale* L.) is a tropical evergreen tree cultivated all over the world. It is also known as 'Gold Mine' of the wasteland as it requires less input for its production. The cashew fruit comprises of two parts, cashew nut, the true fruit and cashew apple, the pseudo fruit. The cashew is cultivated mainly for the nuts which is having a higher commercial value. While, India produces approximately 7.79 lakh tonnes of cashew nuts annually, Kerala produces 0.83 lakh tonnes and is the 4th highest producer (NHB, 2017). While 1 tonne of cashew nut is produced, about 10–15 tonnes of cashew apples are wasted in the fields after its separation from nut (Attri, 2009). It is estimated that about 77.9 million tonnes of cashew apples are produced yearly in India (NHB, 2017). Highly perishable nature of cashew apple limits post harvest handling, storage and marketing. Therefore, improved processing methods are to be derived to better utilisation of this potential fruit for increasing its commercial value.

Cashew apple juice is an excellent source of organic acids, polyphenols (prominently carotenoids, flavonoids, tannins, and anacardic acid), and vitamins. Cashew apple is also a rich source of minerals such as sodium, potassium, calcium, phosphorous, zinc, iron, copper, and magnesium. It is enriched with vitamin C content and is approximately six times than that present in orange (Azoubel *et al.* 2005). Rapid microbial spoilage due to high moisture and sugar content (55-65%) and poor acceptability due to astringent and acrid taste limits the processing of cashew apple. The

storage life of the cashew apple juice is also hampered due to astringency, sedimentation, and browning. Hence it requires special processes such as clarification, sedimentation for removal of astringency. In addition to that advanced non thermal and mild thermal processing technologies need to be developed for improving the market horizon of cashew apple juice. Only limited studies have been reported in the processing of cashew apple juice.

Consumption of fresh or unpasteurised fruit juices cause approximately 16000 to 48000 cases of illnesses in a year in United States alone (Foley *et al.,* 2002). Recently, juice industries are facing various challenges due to the pathogenic bacteria developing resistance to stress caused by process metthods. The microorganisms adapt themselves to hostile environments which were earlier detrimental to them. Thus fruit juices need to be processed to inactivate pathogenic microorganisms to improve the shelf life adopting novel technologies ensuring safety as well as quality. The NACMCF (2006) (National Advisory Committee on Microbiological Criteria for Foods) has recommended the use of *Escherichia coli* O157:H7 as a target microorganism in fruit juices and suggested a 5-log reduction in the target pathogen. Pasteurisation is an effective and the most used technology in order to reach safety requirements. It has been used for preservation of fruit juices for longer time, as they render juices safe from spoilage microorganisms and undesirable enzymatic reactions.

Thermal treatments can achieve a 5-log reduction in the number of most resistant pathogens. Hence large percentage of processed juices available in the market is thermally processed or may contain added preservatives. Thermal processing of fruit juices effectively inactivate microorganism, but on the other hand has adverse effects on the nutritional as well as sensory qualities. Thermal processing of fruit juices could result in vitamin and volatile flavour loss, non enzymatic browning and colour degradation. Therefore, in the recent years non thermal technologies have gained much of the interest as an alternative to thermal pasteurisation for better retention of product qualities and ensured safety. Different non thermal technologies like high pressure processing, ultrasound, pulsed electric field, oscillating magnetic field and ultraviolet radiation, etc.

are being employed for preservation of different food systems. Pulsed light treatment is one of such non thermal processing technologies found to be effective towards microbial destruction with retention of nutritional qualities.

The pulsed light system involves a wide band spectrum of waves ranging from 100-1100 nm, with 54% of energy attributed to UV region. Other than UV rays, it includes visible and near infrared rays. During pulsed light treatment, electrical energy cyclically stored in a capacitor gets released over a xenon gas lamp which emits an intense short pulse of light with duration of a few hundred microseconds and can be flashed many times per second. The lethal effects of pulsed light system include photo chemical, photo thermal and photo physical mechanisms. Several product parameters such as opacity of the liquid, presence of particulate materials and operating parameters like treatment time, distance of sample from the light source and volume of the sample affect the performance of pulsed light system (Krishnamurthy *et al.*, 2007).

Ohmic heating, a mild thermal processing technology provides uniform heating of foods. The food positioned between two electrodes is subjected to electrical current and acts as an electrical resistor which dissipates electrical energy. Compared to conventional heating, where heat is conducted from the outside of a hot surface, in ohmic heating, heat is generated inside the food material, creating a rapid and uniform heating minimising thermal abuse. The microbial destruction is accomplished through a mechanism of heat generated due to the resistance of food material to the electric field, and formation of pores due to charges developed in cell membrane of microorganisms called electroporation (Lee and Yoon, 1999). The effect of ohmic heating of foods are influenced by characteristics of food such as electrical conductivity and system operating parameters such as applied voltage, residence time *etc.*

Pulsed light is a rapid and effective microbial inactivation technology for solid and liquid foods. Pulsed UV light has more instantaneous energy than continuous UV light for the same total energy supplied and therefore, is more effective than continuous UV-light sources since the energy is multiplied many fold. Continuous UV radiation may result in photo degradation of fructose (fruit sugar) eventually producing free radicals

which may affect other constituents of food (Tikekar *et al.*, 2011a). Pulsed light can effectively limit oxidation reactions compared to continuous UV light, because of the short pulse duration (typically 300 ns to 1 ms) and the short half-life of excited π-bonds (1029 to 1024 s), leading to the prevention of coupling with dissolved or free oxygen (Fine and Gervais, 2004). Moreover, the pulsed UV-light provides cooling periods between pulses and hence reduces the temperature build-up.

Though pulsed light system by itself was found to posses higher penetration depth and emission power, together with relative advantages as mentioned in comparison with UV-C radiation, the in depth penetration to turbid fluid system is limited by the "shadow effect" which may limit the microbicidal effect. Therefore, pulsed light treatment could be effectively combined with other such technologies resulting in a hybrid effect in which the weaknesses of one could be the strength of the other towards microbial destruction. Ohmic heating system could be combined with pulsed light system thus producing a mild thermal process effecting efficient microbial destruction with less nutrient loss and better retention of sensory qualities. Therefore, it could be hypothesised that the ohmic heating produces electroporation effect, apart from producing a volumetric mild uniform heating. Through the combination of ohmic heating with pulsed light technology microbial destruction could be achieved by the synergic mechanisms of each technology with less energy, dosage and time.

Though studies on ohmic heating process and pulsed light treatment on preservation of fruit juices were reported separately, no detailed research were found on combination of these technologies. These studies were carried out on application of pulsed light in static chambers. However, very limited studies were found reported on flow through pulsed light systems. Besides, the raw material behaves differently with applications of ohmic heating and pulsed light, the process parameters of the combined process towards microbial destruction needs to be optimised for these raw materials under study. The present study envisages development of a flow through pulsed light system assisted with ohmic heating for preservation of fruit juices and evaluation of the developed system in retaining the quality characteristics along with microbial safety of

selected fruit juices such as pineapple and cashew apple. Such a study could lead to the production of fruit juices which are safe, nutritionally and organoleptically superior, with minimum energy for processing. This can attract remunerative price in the market, reduce the postharvest losses, improve the financial status of the growers, and thus contribute meaningfully towards the economy of the country.

Taking the above points in to consideration, an investigation was carried out to develop an ohmic heating assisted pulsed light treatment system for liquid foods with the following objectives;

- To develop a flow through ohmic heating assisted pulsed light treatment system for fruit juice preservation
- To evaluate the developed system towards preservation of pineapple and cashew apple juice leading to standardisation of the operating parameters
- To conduct characterisation and shelf life studies of optimally treated fruit juices

Review of Literature

CHAPTER II

REVIEW OF LITERATURE

This chapter deals with a comprehensive review of the research work done by various researchers on ohmic heating, pulsed light and combination treatments. A review on the characteristics and processing of the raw materials under this study has also been explained.

2.1 PINEAPPLE JUICE

Pineapple has long been one of the well known of the non-citrus subtropical and tropical fruits, mostly due to its appealing flavour and refreshing taste (Bartolome, 1995). India is one of the major pineapple producing countries in the world with a total production of 1.86 million tonnes from an area of 0.121 million ha. Kerala is the second highest pineapple producing state in India with a production of 3.10 lakh tonnes after West Bengal (NHB, 2017). This fruit can be consumed fresh or processed into a variety of products like canned pineapple slices, juice, jam, jelly, squash etc. The popularity of pineapple juice is attributed to its pleasing aroma and flavour (Rattanathanalerk *et al*., 2005).

Pineapple juice contains a wide variety of minerals, especially potassium, magnesium and calcium along with amino acids, various sugars, polyphenols, flavinoids and vitamins. It is considered as a functional drink due to its health-promoting properties and its antiaging, antiatherosclerotic, anti-inflammatory and many other health promoting qualities (Khalid *et al.*, 2016). Bromelain is a proteolytic enzyme complex prominently present in pineapple which provides pharmacological and therapeutic properties such as digestive system support, tumor growth modulation and also anti-inflammatory (Taussig and Batkin, 1988; Hale *et al.*, 2005; Chobotova *et al.*, 2010). Alternatively, phenols and vitamin C are popular for their antioxidant characters. Epidemiological studies suggest that inclusion of natural antioxidants in the daily diet lowers the risk of cardiovascular disease and prevents cancers. In addition to the valuable health components, pineapple juice is also well known for its yellowish colour and characteristic mouth feels.

Generally, the shelf life of fresh pineapple juice is restricted by enzyme and microbial activities. The pineapple juice is an abundant source of phenols, vitamins, carbohydrate, and organic acids (Bates *et al.,* 2001). Hence juice undergoes different undesirable biochemical reactions such as changes in colour, development of off flavour, nutrient degradation, microbial spoilage, etc. which leads to deterioration in quality of fruit juice (Nisperos-Carriedo *et al.,* 1990; Achinewhu and Hart, 1994; Bartolomé *et al.,* 1995).

The microbiological spoilage of pineapple juice greatly depends on initial inherent microbiota like *autochthonous* bacteria present in the mature pineapple fruits selected for juice preparation. The microorganisms such as, *Pichia fermentans*, *Pichia guilliermondii*, *Pichia membranifaciens*, *Candida stellata*, *Hanseniaspora uvarum*, *Rhodotorula spp.* along with many other bacteria or fungi causes spoilage of pineapple juice (Chanprasartsuk *et al.,*2013; Hounhouigan *et al.,*2014). In fact, fruits can be considered as the primary source of contamination of juices other than infection from processing operations (Hounhouigan *et al.,* 2014).

2.1.1 Chemical Preservation of Pineapple Juice

Pineapple juice is generally processed with chemical preservatives such as benzoic acid, sorbic acid, sodium or calcium salts and combinations thereof. The chemical preservatives showed a positive effect on the microbial growth reduction and shelf life extension of pineapple juice. But Masamba and Mandalira (2016) observed a reduction in vitamin C content in pineapple juice treated with chemical preservatives such as 0.005% sodium metabisulphite 0.05% sodium benzoate and combined use of sodium metabisulphite and benzoate at 0.005 and 0.04%, respectively, and stored at freezing (-17.3°C±0.2), room (22.4°C±1.3), and chilling (-1.2°C±0.1) temperatures.On the other hand, the juice treated with combined chemical treatments was observed to be more effective in the vitamin C stabilisation than other treatments.

Recently natural preservatives are gaining more popularity due to the adverse health effects of consuming chemical preservatives. A study of pineapple juice treated with garlic and ginger separately and the combination of both observed a reduction in

microbial count. Among the treatment, garlic alone with 2 g in 200 ml reported a lowest microbial count (Nwachukwu and Ezejiaku, 2014). Similarly pineapple juice treated with nisin and essential oils such as thyme oil and clove oil and their combinations found to be effective in reducing the microbial population of juice (Pandhare *et al.*, 2018).

2.1.2 Thermal Preservation of Pineapple Juice

Thermal processing is a globally accepted processing method for fruit juices which provides better safety and long term preservation. Even though thermal processing could offer a higher microbial inactivation, these methods create some undesirable changes in the quality of fruit juice such as changes in colour, flavour and other nutritional attribute.

Rattanathanalerk *et al.* (2005) studied the quality losses of pineapple juice during heat treatment ranging from 55 to 90°C. The changes in colour attributes 'a' and 'b' followed a first order kinetics and changes in non enzymatic reaction (5-hydroxymethylfurfural (HMF) and brown pigment formation) followed zero order kinetics. The reaction rate was strongly influenced by processing temperature. Pasteurisation at 90°C for 90 s resulted in a degradation of the nutritional attributes *viz.* vitamin C, total phenols and antioxidant in pineapple juice (Zheng and Lu, 2011).

Obeta and Ugwuanyi (1997) found that pasteurisation of pineapple juice at 80°C for 30 min treated with sodium benzoate protected the juice from the spoilage of ascospores for 64 days of storage. Slongo and Aragao (2008) observed that an increase in the ratio between degree brix and acidity in the medium of heating increased the thermal resistance of *Neosartorya fischeri* ascospores in pineapple juice.

2.1.3 Non Thermal Processing of Pineapple Juice

Non thermal processing prevented the growth of microorganism along with preserving the nutritional qualities of fruit juices. Different researchers have reported on non thermal processing technologies of pineapple juice such as UV, pulsed light, high pressure processing and microfiltration.

Pineapple juice was successfully sterilised and clarified by cross flow microfiltration coupled with enzymatic action using a polyethersulfone membrane

(Carneiro *et al.,* 2002). Laorko *et al.* (2013) in evaluated pineapple juice clarified with microfiltration using a hollow fibre module and reported the absence of microbial growth for six months of storage with retention of soluble components and photochemical properties.

Pulsed high pressure processing of pineapple juice was effective in the inactivation of *Listeria monocytogenes* and *Escherichia coli*. A significant increase in the inactivation of both bacteria especially after 5 pulses was observed in pulsed high pressure treatment (350 MPa, 20°C for 5 pulses, 60 s). The treated juices did not exhibit any injury recovery or repair of microorganisms for 3 weeks of storage (Buzrul *et al.,* 2008). Helena *et al.* (2009) observed that application of high pressure cycles was more effective in *Byssochlamys nivea* ascospores inactivation than the sustained high pressure treatments.

Mansor *et al.* (2014) reported that a 5 log reduction of *Salmonella typhimurium* TISTR 292 was obtained in UV-C treatment at 13.75 mJ/cm^2 dosages and pump frequency of 30 Hz using Dean Vortex technology. It was also found that an increase in UV-C dosage showed a decrease in microbial counts of *Salmonella typhimurium.* The repetitive ultraviolet irradiation (UV–UV) in combination with dimethyl dicarbonate (UV–DMDC–UV) resulted in a log reduction of 2.61 cfu/ml in total plate count and 4.87 log cfu/ml reduction of yeast and mould count in pineapple juice. The repetitive UV treatment alone resulted in only a 2 log cfu/ml reduction in bacterial population (Shamsudin *et al.,* 2014).

Pulsed light processing of pineapple juice showed a 6.3 log cfu/ml reduction of *Escherichia coli* with a dosage of 19.2 J/cm^2 and when the product was at 5 cm distance from the lamp. It was also reported that total phenols, acidity, vitamin C, pH and colour of the fruit juice did not show any significant difference from that of the fresh juice. The maximum inactivation was achieved in the treatment with higher number of pulses, minimum distance and lower depth (Preetha *et al.,* 2016a).

2.1.4 Combined Technologies for Pineapple Juice

The combination of two non thermal or non thermal and mild thermal technologies could be an effective method to improve the quality of fruit juice. Also, adverse effects of each technology can be reduced by the application of lower levels and synergic action of both treatments could lead to a better final product. Chew *et al.* (2014) studied the effects of combined treatment of pineapple juice with UV and mild heat. A combination of mild heat treatment of 55°C for 10 min and UV treatment at 5.61 J/cm^2 resulted in a reduction in pectin methylesterase (PME) activity along with relatively better bromelin activity and total phenolic content.

Pineapple juice qualities treated with different methods such as thermal pasteurisation (80°C for 15 min), ultrasound (US) combined with mild thermal pasteurisation (65°C for 15 min) and ultrasounds alone were studied. A higher reduction in microbial growth was observed in thermal treatment followed by combination of ultrasound and mild thermal processing and further followed by ultrasound processing alone. The combination treatment was found to be effective in the inactivation of microorganisms and pectin methylesterase with higher retention of phenols for a 60 days storage period (Lagnika *et al.*, 2017). It was reported that a combination of ultrasound treatment, citrus extract and sodium benzoate effectively reduced the microorganisms. The US treatment alone was found, not effective in reducing the microorganisms; but helped to reduce the initial contamination whereas the sodium benzoate and citrus extract eliminated the survivors and reduced the same to below detection limits (Bevilacqua *et al.*, 2015).

2.2 CASHEW APPLE JUICE

Cashew (*Anacardium Occidentale* L.) fruit is a member of the *Anacardiaceae* family. Cashew apple is a pseudo fruit or a thick receptacle of the cashew tree to which the cashew nut is attached. The global cashew production during 2016-17 was 3.97 million tons (FAO STAT, 2017). In India the total production amounts to 7.79 lakh tonnes from an area of 1.03 million ha (NHB, 2017) in case of India. Almost in all countries, nuts are harvested as a major crop while cashew apples are discarded as waste

(Rocha *et al.,* 2006). About 10-15 tons of cashew apples are obtained as a by-product for every ton of cashew nuts produced (Talasila and Shaik, 2013). A fully developed/ripened cashew apple is firm and juicy with strong exotic flavour, high sugar concentration, high astringency and low acidity (Figueiredo *et al.,* 2002).

Researchers have reported that cashew apple juice is composed of significant quantities of polyphenols (carotenoids, flavonoids, tannins and anacardic acid), vitamins and organic acids (Campos *et al.*, 2002; Cavalcante *et al.,* 2005; Azevedo and Rodrigues, 2000; Trevisan *et al.,* 2006; Carvalho *et al.,* 2007; Honorato and Rodrigues, 2010). It is also a rich source of vitamin C, which is about three to six times higher than that of orange juice and approximately ten times more than that in pineapple juice (Michodjehoun-Mestres *et al.,* 2009; Adou *et al.,* 2011). Cashew apple also contains thiamin, niacin and riboflavin in addition to the significant amount of minerals, such as magnesium, potassium, phosphorous, calcium, sodium, copper, iron and zinc (Lowor and Agyente-Badu, 2009).

About 65-80% of the juice can be recovered from the fruits depending upon maturity, variety and process of extraction. Cashew apple juice is highly perishable and deteriorates very fast. It often gets spoiled within a few hours after extraction. The quality of the juice is adversely affected by physical, chemical, biochemical and microbiological changes. These changes bring about sedimentation, browning and foul alcoholic smell. Several deteriorative reactions in the extracted juice cause degradation of ascorbic acid, development of cloudiness and off-flavour. Fruits undergo a change in colour, flavour, texture, appearance and nutritional value, all of which are related to loss of quality of the extracted juice. Thus the inhibition of microbial growth and retention of quality are two important parameters for the extension of shelf life of the cashew apple juice (Das and Arora, 2017).

2.2.1 Clarification of Cashew Apple Juice

The astringency in cashew apple juice is attributed to the presence of polyphenols, especially tannin. Tannin is a phenolic compound, which induces the astringent taste in the fruit by forming strong complexes with other macromolecules and proteins (Fontoin *et al.,* 2008). Further, polyphenols oxidises with carbohydrates and proteins to produce melanins because of the catalyses action of enzyme, polyphenol oxidase (PPO). Melanins adversely affect the nutritional qualities and sensory attributes of juice. Hence, it is highly recommended to remove the tannin content of fruit juice (Queiroz *et al*., 2011). A number of researchers have studied about different cost effective and economic methods for clarification of cashew apple juice. The processes efficacy can be evaluated with respect to the final tannin content and recovery of the clarified juice.

Jayalekshmy and John (2004) studied about the efficacy of various clarifying agents such as PVP, starch, sago and gelatin. For one kg of fruit juice 1.4 g of PVP, 4 g of starch and 5 g of gelatin were dissolved in lukewarm water, whereas the powdered sago at 2 g in 10 ml of water was boiled and then used. The cashew apple juice samples were added with each clarifier separately, with constant stirring and kept overnight for settlement. Among the four materials used for the study, Sago was found to be the most efficient clarifier, since it provided a higher juice recovery and best visual clarity. Hence sago can be considered as an economic and efficient clarifying agent for cashew apple juice. Another investigation of the astringency removal of cashew apple juice also found that sago as a clarifying agent at a concentration of 2 g/l reduced the tannins by 42.85% with a visual clarity of 94%. The same treatments combined with sterile filtration, improved the visual clarity to 96% with reduction in tannin concentration by 41.75% (Talasila *et al.,* 2012). Dedehou *et al.* (2015) reported that rice starch was more effective in clarifying cashew apple juice in short period of time. Rice starch at a concentration of 10 ml/l for 193 minutes decreased tannin content to 42.18% and visual clarity of 94.8%, but 34.2% reduction in tannins and 93.75% visual clarity was obtained for cassava starch at a concentration of 6.2 ml/l for 300 minutes.

Other than the addition of clarifying agents there are some other physical methods such as application of steam and microfiltration to remove astringency of cashew apple. The tannin content of cashew juice was reduced up to 97% in the tangential microfiltration process. But the microfiltration resulted in an alteration in the volatile profile of cashew juice due to the influence of selective properties of the membrane (Rocha-Andrade *et al.,* 2018**).** Steaming at 0.4 N/m^2 pressures for 5-15 min under boiling salt water before 15 minutes of juice extraction decreased astringency of cashew apples (Morton, 1961; Jagtian, 1980; Akinwale and Aladesua, 1999). Ten minutes of steaming prior to juice extraction was found to be helpful in retaining the quality attributes such as colour, specific gravity, pH and total soluble solids (Akinwale *et al.,* 2000). However, Inyang and Abah (1997) reported a reduction in ascorbic acid content in steamed cashew apples.

2.2.2 Thermal Treatment of Cashew Apple Juice

Costa *et al.* (2003) studied the thermal processing and preservation of cashew apple juice for 12 months. The cashew apple juice was subjected to heat treatment at 90°C for 60 seconds in a plate heat exchanger and then bottled by aseptic processing and hot fill methods. Both the filling operations were found to be effective in maintaining the quality of fruit juice for 12 months of storage without any significant changes in the physico-chemical qualities of the juice. But the juice packed through the aseptic process observed a lower viscosity, less variation in ascorbic acid content and total sugars as compared to hot fill process. Studies on the non enzymatic browning on the thermal treatment of clarified cashew apple juice at 88°C to 121°C temperatures, suggest that thermal treatments at 120°C for a lower residence time in plate heat exchanger retains high vitamin C content. A correlation between loss of vitamin C and colour formation due to browning was also observed (Damasceno *et al.,* 2008). The synergic action of thermal pasteurisation at 95°C for 7 min and chemical treatments with sorbic and benzoic acid inactivated enzymes and prevented microbial activity along with the reduction in the astringency of cashew juice (Kabuo *et al.,* 2015). The adverse effect during thermal

processing includes nutrient loss, non-enzymatic browning and formation of undesirable product like 5-hydroxymethylfurfurals (Beveridge *et al.,* 1986).

2.2.3 Chemical Preservation of Cashew Apple Juice

A majority of cashew apple juice is preserved mainly by using chemical additives. It was widely reported that chemical preservatives have a great potential to prevent microbial spoilage of foods (Gould, 1996). However, no single preservative was found to be completely effective against all the microorganisms.

Talasila *et al.* (2012) reported that the cashew apple juice processed with chemical preservatives such as sodium metabisulphate and sodium benzoate at 0.1 g/l, citric acid and sodium benzoate at 0.1 g/l each and potassium metabisulphate and sodium metabisulphate at 0.5 g/l each extended the storage life of juice up to 20 days. The vitamin C content and total sugar in the juice was observed to be stable throughout the storage. It was also observed that cashew apple treated with the citric acid and benzoic acid at 0.1 g/l each and stored at refrigerated condition (4°C) had extended shelf life to 90 days (Talasila *et al.*, 2012). Another attempt to extend the shelf life of cashew apple juice, combining three unit operations such as clarification, sterile filtration and chemical preservation resulted in prolonging the shelf life up to 3 months (Talasila *et al.*, 2011).

2.2.4 Non-thermal Treatment of Cashew Apple Juice

Non-thermal treatment of fruit juices helps to protect most of their nutritional attributes after treatment and storage, in addition to preventing growth of microorganisms and activity of enzymes. The cashew apple juice was treated with high pressure processing within the range of 250 to 400 Mpa and holding times of 3 or 7 minutes. A reduction in the growth of aerobic mesophilic bacterial count to less than the allowable detection limit for the treatments 350 MPa for 7 min and 400 Mpa for 3 or 7 min was reported. No growth of micro population and *Escherichia coli* repair action was observed for fruit juice processed with 400 Mpa for 3 min for 8 week of storage under refrigerated condition (Lavinas *et al.,* 2008).

Rodríguez *et al*. (2017) studied the indirect cold plasma technology of cashew apple juice. The experiments were conducted in a bench top plasma system with two independent variables such as N_2 plasma flow rate (10, 30 and 50 ml/min) and treatment times (5, 10 and 15 min). An increment of total phenol content, vitamin C and antioxidant activity were observed in plasma treatment with low N_2 plasma flow rate, whereas higher levels of plasma resulted in increased polyphenols and flavinoids. It was postulated that this could be due to the activation of antioxidant compounds as a result of oxidative distractions caused by reactive species from plasma.

2.3 OHMIC HEATING TECHNOLOGY

Ohmic heating is one of the emerging thermal processing technologies, which is a thermal- electrical method, where food is heated in direct contact with the electrodes. Ohmic heating is also known as electro-conductive heating, joule heating, electro heating and electrical resistance heating in the literature. Ohmic heating is widely applied in the sterilisation/ pasteurisation of liquid food products resulting in better quality.

The concept of ohmic heating technology is not new and dates back to 1897 (Jones, 1897). In 1841, James Prescott Joule discovered that the passage of electric current generates heat due to the resistance of the medium of conduct, hence this process is also known as Joule heating. Ohmic technology was firstly used in the 19^{th}century (Anderson and Finkelsten, 1919; Prescott, 1927) for milk heating, and was then investigated regularly in the earlier part of the last century. The electro-process used for pasteurisation of milk had been named as “electropure”. Ohmic heating is an electro heating technique based on the passage of electrical current through a food product having electrical resistance (Reznick, 1996; Sastry and Salengke, 1998; Icier, 2003). Heat is generated instantly inside the food, and its amount is directly related to the voltage gradient, and the electrical conductivity (Sastry and Li, 1996).

The advantage of ohmic treatments over conventional methods is the lack of high wall temperatures and limiting heat transfer coefficients requirements. It is also maintains the colour and nutritional value of food, short processing time, and higher yield (Wang and Sastry, 2002; Leizerson and Shimoni, 2005a). Ohmic treatment is used in a wide

range of applications such as preheating, blanching, pasteurisation, sterilisation, extraction of food products (Lima and Sastry, 1999; Leizerson and Shimoni, 2005b). Since USDA and FDA suggested the usage of ohmic technology for liquid foods, it is currently being used commercially throughout the world (USA, Japan, UK, and other several European countries) for the pasteurisation of liquid foods (syrups including whole fruits, fruit juices, egg, milk, *etc.*) and aseptic packaging (Icier and Bozkurt, 2009; Zell *et al*., 2011).

2.3.1 Principle of Ohmic Heating

Most foods contain high levels of water and dissolved salts and these solutions can conduct electricity through electrolytic conduction. When electrolytes are placed in an electric field, the ions present within the electrolyte move towards the electrodes with opposite charges. The movement of ions in the electrolyte generates heat. Also the moving ions within it collide with each other, which in turn create resistance for the movement of ions and increase their kinetic energy, thereby heating the product (Singh and Heldman, 2014). Similarly, heat is produced directly within the food itself by Joule heating as alternating electric current is passing through a food material, accomplishing internal heat generation causing the temperature rise (Reznick, 1996). An ohmic heater is an electrical heating device that uses a food's own electrical resistance to generate the heat (Fryer *et al.,* 1993). Heat is generated instantly and volumetrically inside the food materials (joule effect) due to the ionic motion. The amount of heat generated is directly related to the current induced by the voltage gradient in the field and the electrical conductivity of the materials being heated (Icier and Ilicali, 2005). The heat generation rate during ohmic heating is described by Samprovalaki *et al.* (2007):

$$Q = \sigma E^2 \qquad 2.1$$

This is equivalent to the more familiar I^2R. Here Q is the internal energy generation rate (W/m^3), σ is the local electrical conductivity (S/m) and E is the electric field strength (V/m). The voltage distribution is given by

$$\nabla (\sigma \nabla E) = 0 \qquad 2.2$$

The voltage distribution depends on the electrical conductivity within the medium as well as the system geometry. Equation (2) differs from the usual form of Laplace's Equation (3) because it deals with a medium in which the electrical conductivity is a function of both position and temperature.

$$\nabla^2 E = 0 \qquad 2.3$$

The most important parameter in the applicability of ohmic heating is the electrical conductivity of the material. Most foodstuffs, which contain water in excess of 30% and dissolved ionic salts, have been found to conduct sufficiently well for ohmic heating to be applied.

2.3.2 Mechanism of Microbial Inactivation

Thermal effects can be considered as the main mechanism behind the microbial inactivation during ohmic processing. The higher temperatures lead to the destruction of membrane structure and inactivation of the enzymes of the microorganisms (Sun *et al.*, 2008). Recent research indicates that ohmic heating may cause mild non-thermal cellular damage due to the presence of the electric field (Cho *et al*., 1999; Sun *et al*., 2008). The electroporation is one of the most widely accepted mechanisms (Park *et al*., 2013).

Electroporation is the formation of holes in a cell membrane due to individual ion pressure, which cause changes in the permeability of the cell membrane, due to the varying electric field (Weaver and Chizmadzhev, 1996). The attraction of opposite charges built up on the outer and inner surfaces of the cell membrane and development of compression pressure leads to the reduction in membrane thickness. If critical electrical field strength is exceeded, the membrane gets permeabilised by pore formation and it could be irreversible or reversible, contingent upon the treatment time, electrical field strength, membrane surface charge, suspending liquid medium, cell size and cytoplasm (Lojewska *et al*., 1989). Conventionally heated samples reach plate counts of 10,000 cfu/ml compared to the ohmic heated samples with plate counts of 10 cfu/ml after 12 weeks (Reznick, 1996). The principal reason for the additional effect of ohmic treatment might be due to the low frequency of 50 - 60 Hz, which causes accumulation of charges

in the cell wall and pore formations (USA-FDA, 2000). Hence, a reduction in the D value for the microbial inactivation during ohmic heating was observed compared to conventional heating methods. Pereira and Vicente (2010) studied inactivation kinetics of *Bacillus licheniformis* ascospores in cloudberry jam and *Escherichia coli* in goat's milk during ohmic heating and found that minimum thermal death times were required for bacterial inactiation. Similar reduction has also been observed for *Streptococcus thermophilus* (Sun *et al.,* 2008) and *Bacillus subtilis* (Cho *et al*., 1999). This result was explained by the injury effects of the ohmic heating to the cells due to possible electroporation.

2.3.3 Components of Ohmic Heating System

The ohmic heating set up comprised of an ohmic heating chamber, electrodes, AC power source, thermocouples, variable transformer, data logger and volt ampere meter. The power supply is required to conduct electricity through the food products. The food samples are sandwiched between the electrodes in order to conduct the electrical current supplied by power source. The electrode gap can be varied for different system according to the processing capacity per batch. The electric field strength can be varied by changing the distance between the electrodes or increasing the applied voltage between the electrodes (Varghese *et al.,* 2014).

The ohmic heating chamber can be considered as the heart of the ohmic heating system. Various researchers used different materials for the construction of ohmic heating chamber. Marra *et al.* (2009) used stainless steel cell for analysing the ohmic processing of liquid foods. The stainless steel could not be suggested due to the higher conductivity of electrical current. Since Teflon could act as an excellent insulator and higher temperature withstanding capacity, Zareifard *et al.* (2003) designed a static Teflon chamber for ohmic heating. The Pyrex and Acryilic tubes can also be used for the development of ohmic heating chamber (Tumpanuvatr and Jittanit, 2012; Davirshi *et al*., 2012).

2.3.4 Electrodes

One of the important parameter need to be considered is the suitable selection and design of electrodes for an ohmic heating system. Previous studies have used various efficient materials for construction of electrodes such as stainless steel, titanium, graphite, aluminium and platinized titanium (Assiry *et al.*, 2003; Samaranayake *et al.*, 2005; Zell *et al.*, 2011). Generally electrodes are selected based on the cost and capability of corrosion resistance, which influence the efficiency of ohmic heating and ultimate quality of the product. Low carbon electrodes are used in the situations where low product qualities were required for waste water treatment. The stainless steel electrodes are employed for the products that need high quality and safety (Stanel and Zinty, 2010).

2.3.4.1 Transverse configuration

This design provides a simple mechanical construction in which electrodes are arranged in plane or coaxial design. The liquid food would be allowed to flow perpendicular to the electric field and parallel to the electrodes. The major problems during ohmic heating includes the occurrence of electrodes very close to inlet and outlet pipe works can cause great leakage currents to earth through the food product (Sakr and Liu, 2014).

2.3.4.2 Collinear design

This design is more suitable for food materials with high conductivity which offers an extensive spacing between electrodes. The electrodes can be located in the fluid streams or as collars around a pipe which offers a fully unobstructed flow conduit. This design requires higher voltage gradient than parallel plate design. The design also causes a localised arc and boiling in fluid due to uneven distribution of current and areas of high current density located at the leading edges of the electrodes (Sakr and Liu, 2014).

2.3.4.3 In-line field system

In this system, the electrodes are located at different points along the product flow path. A non-uniform distribution of electric field occurs, *i.e.* upstream materials

experience higher field strength than the downstream materials because of the voltage drop throughout the system (Sakr and Liu, 2014).

2.3.4.4 Cross-field system

In this system electric field strength throughout the system is constant and electrodes are positioned at right angles to the flow pathway.

2.3.5 Application of Ohmic Heating

Ohmic heating has great potential in a large number of food processing applications including pasteurisation, sterilisation, microbial inactivation, enzyme stabilisation, extraction, blanching, thawing, starch gelatinisation and fermentation.

2.3.5.1 Pasteurisation and sterilisation of fruit pulp, purees and juice

Ohmic heating is widely applied in pasteurisation/sterilisation of products resulting in high quality and safety. The ohmic heating of orange juices could considerably reduce the process time and retain nutritional and flavour compounds than conventional treatments. Ohmic heated orange juices also showed twice the shelf life of conventional treated (Leizerson and Shimoni, 2005a). The food components containing large particles (up to 2.5 cm), which is difficult to sterilise by other methods can be sterilised by ohmic heating technology. Pasteurisation of fruit juices milk and UHT processing of liquid foods were successfully achieved by ohmic heating. Different fruit juices such as orange, pineapple, guava and coconut juices were pasteurised in a static ohmic heater at 23 V/cm and found to be stored as conventional pasteurised fruit juice but with less changes in flavour and colour (Somboonsilp *et al*., 2011).

2.3.5.2 Extraction

Various studies have been conducted in the ohmic assisted extraction, such as extraction of sucrose from sugar beets, soymilk from soybeans and lipid from rice bran so on (Kim and Pyun 1995; Lakkakula *et al.,* 2004). Ohmic assisted extraction could provide a better yield in minimum time for the product than the conventional method. The higher extraction rate could be owing to the increase in leaching of soluble solids due to the irreversible or reversible electroporation of cell membranes (Mizrahi, 1996; Wang

and Sastry, 2002). Apple, potato and sugar beets were observed a better juice extraction rate when pre treated with ohmic heating. In addition to that ohmic heating need lower pressing energy than microwave heating (Wang and Sastry 2002; Praporscic *et al.,* 2005). About 70-75% reduction in extraction time was observed in ohmic heating assisted oil extraction of rice bran (Nair *et al.,* 2012).

2.3.5.3 Blanching

Blanching with ohmic heating may prevent the amount of solute leaching, than hot water process with a minimum blanching time irrespective of the size and the shape of the food material (Mizrahi, 1996). The ohmic blanching of pea purees at voltage gradients between 20-50 V/cm from 30 to 100°C resulted in the inactivation of peroxidase enzymes in a very short duration of time. The ohmic blanched pea purees above 20 V/cm showed better colour values than conventional water blanching (Icier *et al.,* 2010). Cousin *et al.* (2001) reported that exposure of high direct electric fields on potato tubers during ohmic blanching resulted in a clean cutting due to the tissue softening during ohmic heating.

2.3.5.4 Thawing

Ohmic thawing provides uniform due to volume heating and also reduces the generation of water and waste water associated with thawing (Luzuriaga *et al.,* 1996). Ohmic thawing have less microbial growth and the better quality product compared to the conventional methods (Icier *et al.,* 2010). Rate of thawing and energy utilisation ratio (EUR) of frozen beef cut were significant effects of ohmic heating (Bozkurt and Icier 2012). The ohmic heating system was observed to be efficient on passing critical temperature of tuna fish and beef, which is -3°C during thawing and this process reduces thawing time at the rate by four to three times (Li and Sun, 2002). Thawing of frozen meat and surimi with the help of ohmic heating resulted in fast thawing process when the largest surface of product placed perpendicular to the electrical field and the brine concentration was increased (Wang and Sastry, 2002 ; Miao *et al*., 2007). The increase in voltage gradient resulted in a decrease in ohmic thawing time of beef cuts (Bozkurt and Icier, 2012).

2.3.5.5 Starch gelatinisation

Ohmic heating resulted in immense effects of gelatinisation with short processing period at high voltage (Fernando *et al.,* 2000). The starches generated using ohmic heating process provides various functional characteristics with varied degrees of gelatinisation used in the food industry. The ohmic heating of rice flours and rice starch at different voltages and frequencies resulted in greatest decrease in enthalpy, due to the maximum extension time of pre-gelatinisation during ohmic heating process. The ohmic heating of brown rice flour showed a lowest enthalpy for gelatinisation at 20 V/cm (An and King, 2007). Fernando *et al*. (2000) reported that that 70.0% and 39.1% of jicama starch and cassava starch were found to gelatinise with highest voltage and longer treatment time respectively.

2.3.5.6 Fermentation

Ohmic heating extend the lag phase of microorganism and reduce the fermentation time. The ohmic heating assisted fermentation of cocoa beans resulted in higher degree of fermentation within short duration of time. The fermentation of coca beans at 50°C for 3 days resulted in 81.4% of fermented beans compared to 63.35% in conventional fermentation (Supratomo *et al.,* 2019).

2.3.6 Ohmic Heating of Liquid Foods

Ohmic heating has been studied for various liquid foods such as milk, fruit juices, fruit pulps, purees, liquid and whole egg. Different factors such as electrical conductivity, particle size, viscosity and uniformity of liquid foods affects the efficiency of the ohmic heating process. Liquid foods depicted a rapid and uniform heating during ohmic processing than conventional technologies. The ohmic heated samples gave higher inactivation due to uniform and rapid heating, in addition to non thermal and electric effects. The ohmic heating did not result in any adverse changes in food. The other quality changes during ohmic heating were minimal when compared to the conventional heating methods (Makroo and Srivastava, 2017).

2.3.7 Microbial Inactivation

Microbial inactivation during ohmic heating is highly influenced by various process and product parameters. Various studies have proved that ohmic heating can be an effective method for inactivating bacteria, yeast and bacterial spores.

Yoon *et al.* (2002) studied the structural and cell membrane permeability changes in the cells of *Saccharomyces cerevisae* during ohmic heating. It was observed that ohmic heating treatment resulted in translocation of intracellular protein materials out of the cell wall. An increase in the amount of exuded protein was observed with the increase in electric field from 10-20 V/cm.

Orange and tomato juices were evaluated for the inactivation of *Escherichia coli, Salmonella typhimurium* and *Listeria monocytogenes* during ohmic heating process. In orange juice, more than 5 log cfu/ml reduction of *Escherichia coli* was observed after ohmic processing at electric field strength of 10, 15, and 20 V/cm for 540, 210 and 120 seconds respectively. Whereas, more than 5 log cfu/ml reduction of *Escherichia coli* after 480, 180 and 90 seconds were obtained in tomato juice. The strains of *Escherichia coli* showed highest resistance to the ohmic heating treatments in both juice samples (Sangong *et al.,* 2011). Similarly Park *et al.* (2017) also reported that ohmic heating at 60 V/cm for 20 s resulted in a 5 log reduction in three strains of bacteria *Escherichia coli O157:H7, Salmonella typhimurium*, and *Listeria monocytogenes* in 36°Brix apple juice.

Lee *et al.* (2012) conducted studies on the inactivation of three bacterial strains such as *Escherichia coli, Salmonella typhimurium* and *Listeria monocytogenes* in tomato juice, and observed a 5 log cfu/ml reduction in *Escherichia coli* at electric field strength of 25 V/cm for 30 seconds. Similar reductions in population were observed for other strains also.

An ohmic heating system designed with five sequential electric fields was evaluated for the inactivation of *Alicyclobacillus acidoterrestris* spores in apple juice. The ohmic heating treatment at a voltage gradient of 26.7 V/cm, temperature of 100°C and holding time of 3 seconds resulted in the complete inactivation of spores without any

recovery action, while recovery of spore cells was observed in conventional heat treatment at the same temperature and pressure (Kim *et al*., 2019).

The effect of ohmic and conventional heating on the inactivation of *Bacillus subtilis* spores in a sodium chloride aqueous solution was analysed by Murashita *et al.* (2017). The authors found that ohmic heating was more effective in the inactivation than conventional heating at all electric field strength, and the maximum reduction of spores was observed in the voltage gradient of 20 V/cm for all the holding times (8, 10, 12, 14, and 16 min).

2.3.8 Enzyme Inactivation

The enzymes have a wide application in the food industry, as an accelerator for several food processing operations. But some of the enzymes such as PPO, PME and PG (polyphenol oxidase, pectin methylesterase and polygalacturonase) are found to act negatively on the fruits and vegetables and their products. The ohmic heating treatment could inactivate different range of enzymes. The PPO in grape juice showed a higher deactivation at increased voltage gradient and electrical conductivities during ohmic heating. A lower deactivation temperature was required for PPO at 40 V/cm than other voltage gradients of 20 and 30 V/cm (Icier *et al*., 2008). Similarly the increase in electric field strength and temperature resulted in an efficient inactivation of PPO in sugarcane juice. Stable fractions required higher temperature of 80-90°C whereas viable fractions required only 60-70°C for inactivation (Saxena *et al*., 2016). Makroo *et al*. (2016) observed that ohmic heating at temperature 90°C for 1 min inactivated both PME and PG enzymes in tomato juice, but similar result was obtained for 5 min treatment with conventional heat treatment.

2.3.9 Factors affecting Microbial Inactivation during Ohmic Heating

The microbial inactivation and nutritional qualities of food material during the ohmic heating process depends heavily on various intrinsic and extrinsic factors related to the process. The extrinsic parameters such as the electric field strength, frequency, electric current, processing time and temperature have a decisive action in maintaining

the process conditions specifically for food materials. The intrinsic factors include electrical conductivity, food composition, pH, type and growth stage of microorganism. These factors could be altered to obtain more effective processing by ohmic heating technology (Sakr and Liu, 2014).

2.3.9.1 Current

The electrical current can be considered as one of the important parameters for ohmic heating. The Joules law portraits that heat generated during the ohmic heating process is directly proportional to the square of the electric current. Hence the electric current has a significant impact on the inactivation of micro organism during ohmic heating.

An inverse relationship between decimal reduction time and electric current was reported by Guillou and Murr (2002) during the ohmic heating of phosphate buffer solution inoculated with *Saccharomyces cerevisiae*. Thus, higher reduction in *S. cerevisiae* was obtained with increasing electric current. Studies on ohmic processing for inactivation were mostly based on the variation in voltage gradient not on variation in current.

2.3.9.2 Voltage gradient

During ohmic heating increase in the voltage gradient leads to increase in the heating rate by rapid volumetric heating. An increase in inactivation rate of *Salmonella typhimurium*, *Listeria monocytogenes* and *Eschericha coli* O157:H7 was observed during ohmic heating of orange and tomato juice with the increase in voltage gradient from 10-20 V/cm (Sangong *et al.*, 2011) and 25-40 V/cm (Lee *et al.*, 2012) at the same frequency and treatment time. Murashita *et al.* (2017) reported a higher inactivation of *Bacillus subtilis* spores at a high voltage gradient than that of 5 and 10 V/cm in sodium chloride solution.

2.3.9.3 Frequency

The increase in the electric field frequency resulted in an increase in the reduction of microbial population and reduced the treatment time by ohmic heating. Lee *et al.*

(2012) experienced an increase in electrical conductivity of the sample with the rise in electric field frequency. It was also observed that sine and saw tooth waves showed a higher heating rate than the square wave at 60 Hz. At higher frequency levels, the time required for the ohmic heating treatment to reduce the bacterial population of *Escherichia coli* O157:H7 and *Salmonella enterica* to below detection level was minimised (Lee *et al.,* 2015). The increase in frequency resulted in a higher inactivation of *B. subtilis* spores. The spores were found alive at 20 and 40 kHz for 14 to 16 min and completely inactivated at 60 kHz at same treatment times (Murashita *et al.,* 2017).

2.3.9.4 Heating time and temperature

The combined action of higher temperature and longer heating time could achieve a greater reduction in microorganisms in all thermal processing technologies including ohmic heating. Park and Kang (2013) reported a 4.0, 4.63 and 1.11 log cfu/ml reductions of bacterial strains *Escherichia coli* O157:H7, *Salmonella typhimurium*, and *Listeria monocytogenes* at 58°C for 30 seconds, whereas a higher reduction of 5.62, 6.93 and 4.37 log cfu/ml was observed at 60°C for same treatment time during the ohmic heating process. Ryang *et al.* (2015) found that the number of *Bacillus cereus* surviving spores decreased with the increase in temperature and time. The complete inactivation of *Bacillus cereus* spores (5.4 log cfu/ml reductions) required a very high temperature of 105°C for more than 30 seconds in the ohmic heating treatment.

2.3.9.5 Electrical conductivity (EC)

The electrical conductivity of the food material can be considered as one of the most decisive parameters in ohmic heating. The electrical conductivity, mainly depends on the free water, ionic strength and microstructure of the food materials (Lima and Sastry, 1999).The presence of non polar substance like lipids and fats reduces the EC, whereas, the ionic constituents, salts and acids raise the EC of food material. The conductive nature of the food determines the efficiency of the ohmic heating process. The electrical conductivity of the fruit juices and liquids increased with increase in temperature during ohmic heating at constant voltage gradient. Among different ohmic

heated fruit purees and juices, orange and pineapple juice showed high electrical conductivities, hence found to be highly suitable for ohmic heating (Castro *et al.,* 2004a).

The EC of the juices observed a rise in value when the concentration of the juices increased from 12°Brix to 17°Brix but further increase in concentration was not effective in the increase in EC (Tumpanuvatr and Jittanit, 2012). The voltage gradient of the process also significantly affects the electrical conductivity of the product (Icier and Ilicali, 2005). Darvishi *et al.* (2011) reported that highest electrical conductivity of lime juice was observed at 55 V/cm followed by 45 and 30 V/cm. The authors also found that electrical conductivity at 40 V/cm was higher than 20 and 30 V/cm during the temperature range between 55 and 75°C.

2.3.9.6 Fat content of food materials

The effect of milk fat on inactivation of some bacterial strains such as *Escherichia coli* O157:H7, *Salmonella typhimurium* and *Listeria monocytogenesis* during ohmic heating was studied by Kim and Kang (2015). They found that a higher increase in temperature was observed in the samples with lower milk fat due to the higher electrical conductivity than other samples. The inactivation rate of bacterial strains decreased with an increase in fat content from 0-10%. Hence fat content also showed an inhibitory effect on the inactivation of microorganisms and the highest inhibition was observed in *Escherichia coli.* Similar results were obtained in another study on the effect of ohmic heating on pathogenic organisms in milk fat and lactose media. It was found that the lactose inhibitory effects were prominent in the other strains of bacteria such as *Salmonella typhimurium* and *Listeria monocytogenes* (Kim *et al.*, 2019).

2.3.9.7 pH of the food material

The combined effect of low pH and ohmic heating could effectively inactivate the microorganisms. Higher reduction of bacterial strains such as *Salmonella typhimurium, Listeria monocytogenes, Escherichia coli O157:H7* was observed when the pH decreased from 4.5 to 2.5 during ohmic heating (Lee *et al.,* 2012). Another study of ohmic heating of orange juice observed a rapid inactivation of *Salmonella typhimurium* at pH 2.5,

Escherichia coli O157:H7 and *Listeria monocytogenes* at a pH of 4.5. Hence increasing or decreasing the pH of fruit juice can be considered as an effective method for inactivation during ohmic heating (Kim and Kang, 2015).

2.3.9.8 Species and growth stage of microorganisms

The type and the growth stage of microorganism also affect the ohmic heating behaviour of food materials. It was found that the lag phase in the growth stage of *Lactobacillus acidophilus* was found more susceptible to the electric field strength than that of other stages such as stationary and exponential phase (Lebovka *et al.,* 2007; Loghavi *et al.,* 2008). Sangong *et al.* (2011) reported that *Escherichia coli* was more resistant than other species *Salmonella typhimurium* and *Listeria monocytogenes* during ohmic heating of tomato and orange juice.

2.3.10 Changes in Physico-Chemical Properties during Ohmic Heating

The ohmic heating of fruits and vegetable products showed a significant increase in the reduction of ascorbic acid during high temperature and voltage gradient. The degradation due to ohmic heating at very high voltage gradient was found higher than conventional heating (Poojitha and Athmaselvi, 2018). Lima *et al*. (1999) reported that the type of heating had no significant effect on the ascorbic acid reduction. About 21% degradation of ascorbic acid was observed in ohmic heating of orange juice at 90°C for 30 seconds (Lima *et al.,* 1999). A 15% reduction in vitamin C was reported in orange juice treated with temperatures of 150, 120 and 90°C for 0.68, 0.85 and 1.13 seconds respectively (Leizerson and Shimoni, 2005b). The application of lower electrical field frequency (10 Hz) during ohmic heating resulted in higher degradation of ascorbic acid due to electrochemical reactions (Mercali *et al.,* 2014). Makroo *et al.* (2016) also observed a first order trend in degradation of vitamin C in tomato juice with both ohmic and conventional heating.

An increase in total phenolic content was found in pomegranate juice treated with ohmic heating and which caused a decrease in colour quality (Yildiz *et al.,* 2010). Brochier and Domeneghini (2016) observed a 7 to 12% degradation of total phenolic

content and 11 to 23% degradation of flavinoids in ohmic heated sugar cane juice. It was also observed that phenolic and flavinoids compounds remained constant after 6 and 12 min treatment.

A decrease in pH value was observed with an increase in voltage gradient. The pH of ohmic heated banana pulp ranged between 4.5-4.63. The pH of pulp with varying sugar concentration also increased with voltage gradient (Darvishi *et al*., 2012). Chakraborty and Athmaselvi (2014) reported a 5.2% reduction in the pH value of guava juice treated with 13.3 V/cm for 3 minutes.

The total soluble solids (TSS) of the fruit juices and purees found to be increased during ohmic heating. Chakraborty and Athmaselvi (2014) reported that the TSS value of guava juice increased with the rise in voltage gradient, processing time and storage time. The ohmic heating at 13.33 V/cm for 1, 3 and 5 minutes observed a slight increase of TSS from 7.25 to 7.4. The TSS of the ohmically heated banana pulp varied between 22 to 56°Brix which may due to the loss of water during heating (Poojitha and Athmaselvi, 2018).

Mercali *et al.* (2014) observed that all ohmic heated samples showed a decrease in L*, a* and b* colour values and also recorded a higher colour difference in ohmic heated samples than conventional heated ones. Chakraborty and Athmaselvi (2014) recorded an increase in L value of guava juice from 56.23 to 61.28 on the third day of storage, when treated with ohmic heating at a voltage gradient of 13.3V/cm for 3min. The increase in voltage gradient and storage time resulted in an increase in total colour difference. Poojitha and Athmaselvi (2018) reported that colour values of banana puree were greatly influenced by the voltage gradient during ohmic heating. The L values were found to decrease with an increase in voltage.

2.4 PULSED LIGHT TECHNOLOGY (PLT)

Pulsed light (PL) is a technique to decontaminate surfaces by killing microorganisms using short time pulses of an intense broad spectrum, rich in UV-C light. UV-C is the portion of the electromagnetic spectrum corresponding to the band between

200 and 280 nm. PL is produced using technologies that multiply the power many fold. Power is magnified by storing electricity in a capacitor over relatively long times (fractions of a second) and releasing it in a short time (millionths or thousandths of a second). The emitted light flash has a high peak power and consists of wavelengths from 200 to 1100 nm (Dunn *et al.*, 1995; Dunn *et al.*, 1997). The technique used to produce flash originates high peak power with greater relative production of light with shorter bactericidal wavelengths (Mac-Gregor *et al.*, 1998). This technique has received several names in the scientific literature: pulsed UV light (Sharma and Demirci, 2003) high intensity broad-spectrum pulsed light (Roberts and Hope, 2003), pulsed light (Rowan *et al*., 1999) and pulsed white light (Marquenie *et al*., 2003).

According to Wekhof (2000), the first works on disinfection with flash lamps were performed in the late 1970s in Japan, and the first patent dates from 1984 (Hiramoto, 1984). Bank *et al.* (1990) seems to be the first work published in the scientific literature on the application of PL to inactivate microorganisms using a UV-C light source of 40 Watt peak power and a 6 to 7 log decrease in viable cell numbers was achieved.

The technique of UV-C treatment to preserve foods was discovered in the 1930s (Artes and Allende, 2005). PL is a modified and improved version of delivering UV-C to bodies. The classical UV-C treatment works in a continuous mode, called continuous-wave UV light. Inactivation of microorganisms with continuous-wave UV systems is achieved using low-pressure mercury lamps designed to produce energy at 254 nm (monochromatic light), called germicidal light (Bintsis *et al.*, 2000).

More recently, medium-pressure UV lamps have been used because of their much higher germicidal UV power per unit length. Medium-pressure UV lamps emit a polychromatic output, including germicidal wavelengths from 200 to 300 nm (Bolton and Linden, 2003). Another possibility for UV-C treatments was the use of excimer lasers, which can emit pulsed light at 248 nm (Crisosto *et al.*, 1998). Pulsed light works with xenon lamps that can produce flashes several times per second. The following units are commonly used to characterise a pulsed light treatment.

- Fluence rate: is measured in Watt/meter^2 (W/m^2) and is the energy received from the lamp by the sample per unit area per second
- Fluence: is measured in Joule/meter^2 (J/m^2) and is the energy received from the lamp by the sample per unit area during the treatment
- Dose: used sometimes as a synonym of fluence.
- Exposure time: length in time (seconds) of the treatment.
- Pulse width: time interval (fractions of seconds) during which energy is delivered
- Pulse-repetition-rate (prr): number of pulses per second (Hertz [Hz]) or commonly expressed as pps (pulses per second)
- Peak power: is measured in Watt (W) and is pulse energy divided by the pulse duration

Definition by IUPAC (1996)

2.4.1 Principle

The high energy pulsed light is generated from a low energy source and then highly concentrated energy as broad spectrum released on to the food materials within a very short period of time to ensure microbial decontamination. In a PL system the electromagnetic energy gets stored in the capacitor and is then released in the form of light within a billionth of a second, which results in power amplification and minimum additional energy consumption. The inactivation efficiency of pulsed light depends upon the intensity (measured in J/cm^2) and the number of pulses delivered (Ravi-Shankar *et al.,* 2014). A pulsed light produced using engineering technology multiplies the power many fold. Accumulating electrical energy in an energy storage capacitor relatively over a longer time (whole process within a fraction of a second) and releasing this stored energy to do work in a much shorter time (millionths or thousandths of a second) magnifies the applied power. The result is a very high power during the duty cycle, with the expenditure of only moderate power consumption (Dunn *et al.,* 1995).

2.4.2 Pulsed Light System

The development of a PL system mainly involves the generation of high-power electrical pulses and their transformation into high-power light pulses. The energy storage in a PL system is normally performed using a capacitor bank, *i.e.* a number of high-voltage capacitors connected in parallel, which accumulate energy from the electrical power supplier during the charge phase and release it during the discharge phase thus supplying large amounts of current. As an alternative, Marx generators can be used, which differ from capacitor banks only during the discharge phase when all the capacitors are temporarily connected in series. Therefore, the Marx generators also work as voltage amplifiers supplying large amounts of higher voltage current (Pai and Zhang, 1995).

The conversion of the continuous low power DC into the pulsed high-electric power is obtained by special switches capable of handling very high power and performing opening/closing cycles of very short duration, by instantaneously passing from a perfect insulating condition to a perfect conducting condition. The action of the switches is regulated by a controller that determines the pulse shape and the electrical operating conditions in order to yield the optimum PL wavelength for a particular application (Pai and Zhang, 1995).

2.4.3 Components of PL System

In general, the PL systems consist of several common components (Fig. 2.1). A high-voltage power supply provides electrical power to the storage capacitor which stores electrical energy for the flash lamp. The pulse-forming network determines the pulse shape and spectrum characteristics. A trigger signal initiates discharging of the electrical energy to the flash lamp. The xenon gas discharge flash lamp, then converts 45% to 50% of the input electrical energy to pulsed radiant energy (Xenon Corp., 2005). The flash lamps used to generate the broad-spectrum light are available in a variety of shapes, such as linear or circular, which allows them to uniformly treat substrates of different shapes and sizes (Koutchma *et al.*, 2009).

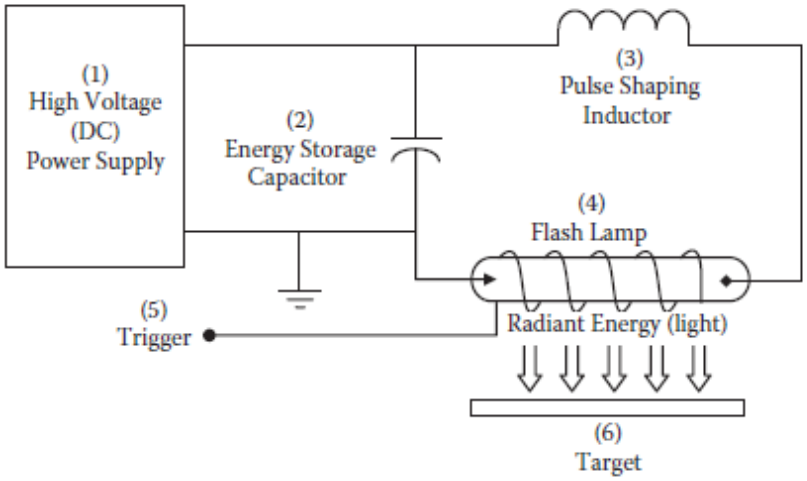


Fig. 2.1 Schematic diagram of pulsed light system

(Source: Koutchma *et al.,* 2009)

2.4.4 Microbial Inactivation by Pulsed Light

The inactivation of microorganisms upon exposure to pulsed light treatment may be attributed to its short duration, its rich, broad ultraviolet spectrum, high peak power and the ability to regulate the frequency output and pulse duration of flash lamps. The emission spectrum of PL includes ultraviolet to near infrared spectrum. The major share of (more than 50%) PL spectrum covered by ultraviolet light, hence it can be considered as the root cause of microbial cell inactivation. The ultraviolet spectrum includes three categories of waves, such as, short-wave ultraviolet-C (200-280 nm), medium-wave ultraviolet-B (280-320 nm) and long-wave ultraviolet-A (320-400 nm) among them UV-C part of the spectrum has a decisive role in the inactivation of microorganisms (Dunn *et al.*, 1995; Takeshita *et al*., 2003).

The pulsed light technology (PLT) exhibits various extents of microbial inactivation with the extremely high power generation. The effect of PL on microorganisms has been explained by different causes and mechanisms by Barbosa-Canovas *et al.* (1998). According to him, the combination of photochemical, photo physical and photo thermal mechanisms considered as the most acceptable hypothesis for microbial inactivation in PL system. Photochemical or photo thermal mechanism or their

simultaneous action contributes a majority of the lethal effect in PL treatment, depending upon the nature of food product and targeted organism.

2.4.4.1 Photochemical Mechanism

The photochemical mechanism can be considered as the most important PL action for the inactivation of microorganism. The UV-C portion of the spectrum action on the DNA of microbial cells is mainly responsible for this effect (Farkas, 1997). The cellular DNA (deoxyribonucleic acid) is an essential component of microorganism for reproduction. DNA composed of highly conjugated carbon-carbon double bonds, which is responsible for the absorption of UV portion of the spectrum. Such absorbed energy can crack the bonds of organic molecules which cause a DNA rearrangement, cleavage and destruction. This leads to the activation of photochemical and electronic reactions to generate some compounds such as pyrimidine dimers, thymine dimers and (6–4) pyrimidone photoproducts *etc.* resulting in the inhibition of DNA reproduction (Hariharan and Gerutti, 1977; Jay, 1996).

Ultraviolet irradiation commonly produces cytosine dimers in low quantity and thymine dimers large quantity, and intermediate level of mixed dimers (Setlow and Carrier, 1966). These dimers prevent DNA from replication and hence inhibit the generation of new chains eventually leading to the chologenic death of the affected microbial cell by ultraviolet light (Bolton and Linden, 2003). The reaction of ultraviolet-C spectrum on bacterial spores may result in the generation of spore photo-product 5-thyminyl-5, 6-dihydrothymine in single-strand breaks and cyclobutane pyrimidine in dimers double-strand breaks (Slieman and Nicholson, 2000). It was also found by experiments that enzymatic repair of DNA was not reported after pulsed light induced damage.

2.4.4.2 Photo Thermal Mechanism

The photothermal mechanism is a process which causes localised heating in the food treated with PL. The surface layers of food exposed to PL absorb most of the energy contained in light pulses penetrated through it and dissipated as heat, which imparts a

temperature increase in such thin layers. As the microbial cells have a higher absorption of the pulsed light than that of the water or surrounding medium, resulting in a rapid localised heating of microbial cells. When PL with very high pulse power values are exposed to food, the temperature of microbial cells increased to a sufficient level (about 130°C) that can cause their overheating, rupture and death. The extremely short pulse duration prevents the microorganism cell from being cooled by the water or surrounding medium (Wekhof, 2000).

2.4.4.3 Photo Physical Mechanism

A third effect called photo physical mechanism was also observed, in the context of vacuole expansion, elution of proteins, cell membrane damage as well as structural deformation of spores and cells during PL exposure (Wekhof, 2000; Takeshita *et al.*, 2003). High intensity pulses of PL system induce photo physical effects, which causes damage similar to that of pulsed electric field. The pulsating effect of high intensity light pulses imparts disturbances in microbial cells and eventually leads to cell structure disruption. Therefore, the efficacy of pulsed UV light process could be enhanced by optimising the number of pulses and pulse width.

2.4.5 Application of Pulsed Light (PL)

Even though pulsed light treatment have been developed long years ago, still the industrial application is limited to the surface sterilisations especially in the food sector. The pulsed light is proved to obtain 5 log reductions for most of the bacteria in liquid foods, which is also capable of inactivating bacterial and fungal spores (Gomez-Lopez *et al.*, 2007). PL is the best sterilisation method in applications where light can access all the surfaces and volumes especially transmissive materials (such as air, water, and many solutions), packaging materials, and many medical or pharmaceutical products (Saikiran *et al.,* 2016). The pulsed light treatment has been approved by the US FDA (Food and Drug Administration) in 1999, and after being analysed for effectiveness and safety (Federal Register, 1999). Application of pulsed light can be categorised as follows.

2.4.5.1 Application of PLT in food industry

Whole eggs were treated with PL at fluence 2.1 J/cm^2 resulted in a 5 log cfu/ml reduction of *Salmonella enteritidis* on the surfaces, higher dose of PL did not show any adverse quality changes in egg (Lasagabster *et al*., 2011). Another study on the surface decontamination of egg observed a 6.7 log cfu/ml reduction in *Salmonella* species. The 1.8 and 3.6 log cfu/ml reduction of salmonella species was obtained in the PL treatment of washed and unwashed eggs respectively. The PL treatment did not affect the cuticle of the egg shell (Holck *et al*., 2018).

Chicken surface inoculated with different species such as *Salmonella typhimurium* and *Listeria monocytogenes* reduced by 2-2.4 log cfu/ml during PL treatment at 5.4 Joule/cm^2 for 200 seconds. A 2 log cfu/ml reductions were obtained in total mesophiles on the surface of meat and no objectionable changes in treated meat were found as compared to raw meat (Paskeviciute *et al*., 2011).

PL treatment at 58 J/cm^2 showed a 7 log reduction of *Saccharomyces cerevisiae* in food powders. The coloured food powders are decontaminated by the thermal effects than the UV decontamination of pulsed light (Fine and Gervais, 2004). The PL decontamination of *Listeria monocytogenes* and *Escherichia coli* on the stainless steel slicing knife was evaluated by Rajkovic *et al*. (2017). They observed a 6.5 log cfu/side reduction was obtained at dosage of 3 J/cm^2 and input voltage of 3000 V for 60 seconds on one side of the knife.

A 2.7 log cfu/ml reduction in population was achieved for paper-polyethylene inoculated with spores of *Aspergillus niger, Aspergillus repens, Cladosporium herbarum*, *Aspergillus cinnamomeus* using pulsed light dosage from 0.244 to 0.977 J/cm^2. The different spores were inactivated at various fluences (Turtoi and Nicolau, 2007). Mushrooms slices treated with pulsed light at fluence rate of 4.8, 12 and 28 J/cm^2 showed extended shelf life by 2-3 days compared to fresh samples. Pulsed light treatment could reduce the natural microflora by 0.6-2.2 log cfu/ml and improve the shelf life to 15 days in refrigerated storages (Oms-Oliu *et al.*, 2010).

Different vegetables such as white cabbage, spinach, celeriac, iceberg lettuce, green bell pepper, soybean sprouts and carrots treated with pulsed light with 2700 pulses/ both sides achieved a 0.56-2.04 log reduction of aerobic and mesophilic microorganisms (Gomez-Lopez *et al.*, 2005a).

Pulsed light application of 16 J/cm^2 with pulse duration of 0.5 milliseconds observed a 1.5 log reduction of *Pseudomonas* species in curds of dry cottage cheese (Dunn *et al.*, 1991). The bulk tank of milk is decontaminated with pulsed UV light at 25 J/cm^2 resulted in a reduction of mesophilic count. Similarly, seven different potential bacterial pathogens, including, *Escherichia, Listeria* and *Salmonella* species were inactivated by PL treatment and no growth was observed up to 21 days (Smith *et al.*, 2002).

PL energy dosages between 116.6-223.6 J/cm^2 could efficiently reduce the allergic proteins in peanuts *viz.* Ara h_1, Ara h_2, Ara h_3. The peanut butter under PL exposure could deactivate the most important allergic protein Ara h_2. Similarly, soy allergens also found to decrease after the PL treatment of soy extracts (Yang *et al*., 2010). PL inactivation of different microorganisms such as viruses, parasites and bacteria inoculated in water was evaluated using the Pure Bright unit at 0.25 J/cm^2 fluence. More than 7.4 log cfu/ml reduction of *Klebsiella terrigena* was reported at 1 flash in 2.4 seconds in PL water treatment (Huffman *et al.*, 2000). The *Saccharomyces cerevisiae* cells suspended in potassium phosphate buffer were exposed to flash lamp at an energy dose of 3.5 J/cm^2 reduced the initial population by 5.8 log cfu/ml (Takeshita *et al.*, 2003). Sauer and Moraru (2009) examined the inactivation of *Escherichia coli* using pulsed light technology at less than 12 J/cm^2 of fluence level and 50 mm distance from the lamp. The authors recorded a 5.5 log cfu/ml and 7 log cfu/ml reduction in apple cidar and apple juice respectively.

2.4.6 Pulsed Light Treatment System for Liquid Products

2.4.6.1 Pulsed light system for pumpable foods (fruit juices)

A system for the sterilisation of liquid foods was patented by Dunn *et al.* (1988). The system consists of two cylinders inner and outer cylinder and liquid allowed pass through the annular space. The inner cylinder contained pulsed UV lamp and the outer cylinder was made of highly reflective material. The liquid to be treated is pumped through the system using a circulation pump based on the frequency for delivering the required number of pulses to the product.

2.4.6.2 Flow through pulsed UV system

Krishnamurthy *et al.* (2007) developed a Pulsed UV flow through system for inactivation of *Staphylococcus aures* in milk. The pulsed UV chamber consisted of UV lamp and a V groove reflector fixed with quartz tube held inside and the whole assembly can be moved to and fro to adjust the distance between the lamp and the quartz tube. Only 28 cm of quartz tube was exposed to light and rest all portions were covered with Aluminium foil. A V groove reflector enhanced the light absorption of milk, since it had a polished surface and angles of approximately 56° and 117°. A peristaltic pump was used to pump the milk inoculated with *Staphylococcus aures* and the flow rate was measured with the help of the flow meter.

A flow through pulsed light system was developed by Uslu *et al.* (2016) evaluate the inactivation of *Escherichia coli* and *Bacillus subtilis* spores in municipal waste water effluent. The developed system was a concentric cylinder set up, in which pulsed UV lamp at the center cylinder and the water was allowed to pass through the annular space between the lamp and outer cylinder. The cylinders were made up of stainless steel with 10.2 cm of outer diameter and length of 40.6 cm. A quartz liner was fixed between the annular gap of UV light and outer wall to improve light transmission. The centrifugal pump was used to pump the water and flow rate was measured by flow meter.

2.4.6.3 The continuous flow PL system

Pataro *et al.* (2011) developed a continuous pulsed UV treatment system for the inactivation of *Escherichia coli* and *Listeria innocua* in apple and orange juice. The system composed of a peristaltic pump to pump the fruit juices, two quartz tubes held in a grooved hollow metal enclosure, an external refrigerated bath, and lamp assembly with xenon lamp.

The juice inoculated with the bacterial strains was pumped through a food grade tygon tube and then allowed to flow through two quartz tubes placed below 1.9 cm from quartz window. The quartz tubes were aligned in line with xenon lamp. Only 20 cm of quartz tubes was exposed to the xenon lamp and rest were covered with Aluminium foil. The metal enclosure containing the quartz tubes was cooled by recirculation water – ethylene glycol solution as a coolant.

2.4.6.4 Bench top system

Xenon Corporation in USA and SteriBeams systems in Germany are pioneers in the production of xenon flash lamp in the world (Wekhof, 2000). The basic systems provided by them composed of the treatment chamber and control module. The stainless steel treatment chamber consisted of a lamp house at the top center containing the xenon flash lamp and a sample shelf for holding the food samples. The sample shelf could move from the initial position to top and bottom in order to adjust the distance from the light source (Jun *et al.*, 2003). A control cable is used to connect the control module with light source, for modulating the electric current to specific pulse width, pulse repetition rate and peak power (Ghasemi *et al.*, 2003).

Preetha *et al.* (2016a) developed a static pulsed light system to inactivate *Escherichia coli* in pineapple juice. The treatment chamber was made of plywood with 12 mm thickness with a dimension of 30×30×30 cm. For ensuring maximum incidence inner surface of the chamber was covered with Aluminium foil. The xenon lamp with an arc length of 75 mm and envelop made of clear fused quartz was fixed with clamps to the bottom of the top.

2.4.7 Process Parameters

The efficacy of PL processing is highly influenced by various operating parameters, which determine the rate of microbial inactivation, retention of quality and other characteristics of food material. These factors include process parameters such as fluence rate or number of pulses, distance of sample from the light source, exposure time and flow rate (in case of fluids) etc. and product parameters like product thickness, properties of the material and initial population of micro organisms (Oms-Oliu *et al.,* 2010).

2.4.7.1 Fluence rate

Fluence rate can be considered as the most important factor affecting the efficiency of the pulsed light process. The lethality due to PL treatment primarily relies on the obtained PL intensity (Rowan *et al.,* 1999; Anderson *et al.,* 2000; Wekhof *et al.,* 2001; Sonenshein, 2003). This depends on various interconnected factors such as pulse duration, exposure time, number of pulses, distance of sample from light and pulse frequency. Higher levels of microbial inactivation were obtained in the samples processed with higher total fluence (Gomez-Lopez *et al.,* 2007).

A number of studies have proven the effect of pulse dosage on microbial inactivation. When the pulse number raised from 270 to 540, a corresponding increase in percentage reduction of *Clostridium sporogenesis* spores (22.1% to 89.4%) was observed in clover honey treated with PL (Hillegas and Demirci, 2003). Similar trends in percentage reduction of patulin from 9.4% to 45% were also reported in PL treatment with dosage ranges from 14.2-99.4 mJ/cm^2 (Ferrario *et al.,* 2018). Apart from the batch system the flow through PL systems also reported a higher reduction (6 log cfu/ml) of *Listeria innocua* in water at a PL dosage of 10 J/cm^2 compared to lower dosage levels with less than 3 log cfu/ml reductions (Artíguez *et al.,* 2011).

2.4.7.2 Distance from the sample

Higher inactivation of microbial population was attained in samples kept nearer to the light source (Hillegas and Demirci, 2003; Ozer and Demirci, 2006). PL treatments for inactivation of microorganisms are usually carried out by keeping the samples directly

below the light source. Gomez- Lopez *et al.* (2005b) studied the effect of different positions of samples on the inactivation of microorganism in PL treatment. It was found that when different samples were kept directly below the light source maximum decontamination was observed, whereas the samples kept at different positions exhibited a minimum decontamination. The decontamination level also varied according to the variation in vertical distance between the sample and light source.

The effect of shelf height on the percentage spore reduction of the *Clostridium sporogenes* in clover honey was evaluated by Hillega and Demirci (2003). Shelf heights of 8, 13 and 20 cm demonstrated an increase in spore reduction of 87.6, 49.5 and 39.5 percent, respectively. Similar studies were carried out by Sharma and Demirci (2003) to inactivate *Escherichia coli* O157:H7 in alfalfa seeds and Jun *et al. (*2003) to inactivate *A. niger* spore in corn meal.

2.4.7.3 Exposure time and flow rate

The exposure time in the batch PL systems is one of the crucial factors determining the total fluence exposed to the sample. Sharma and Dermirci (2003) observed a significant reduction in *Escherichia coli* O157:H7 with increase in exposure time in alfalfa seeds. When the exposure time was highest (90 s), more than 4 log cfu/ml reduction was exhibited in alfalfa seeds for all seed thickness, whereas only less than 2 log cfu/ml reduction in seeds was found in exposure times below 60 s. Similarly PL treatment of corn meal for inactivation of fungal spores (*Aspergillus niger*) showed a maximum reduction of 4.93 log cfu/ml for the highest treatment time (100 s) at 3 cm distance and voltage of 3800 V (Jun *et al.*, 2003).

The flow rate and exposure times are mutually interconnected parameters in case of flow through PL systems. Krishnamurthy (2006) reported that an increase in the flow rate of milk from 20 to 40 ml/min resulted in a substantial reduction in the destruction of *St. aureus* from 7.23 to 0.55 and 7.26 to 0.63 at 8 and 11 cm distance from the lamp respectively.

2.4.7.4 Product thickness

The microbial inactivation of PL is significantly affected by the thickness of the sample, which is considered as one of the limiting factor. Since the PL have a limited penetrability, overlapping of opaque samples prevent the inner layers from decontamination, and liquid samples with suspended matter attenuates the light waves. Keyser *et al.* (2008) reported that 90 percentage of the absorption of light occurs at minimum depth in fruit juices.

PL treatment of water inoculated with *L. innocua* showed a decrease in log reduction of bacterial population when the sample thickness increased from 2.15 mm to 6.23 mm for same treatment variables (Artiguez *et al.,* 2011). In the same way an increase in the depth of the fluid layer from 6 to 10 mm resulted in a corresponding reduction in the inactivation of *Penicillium expansum* from 3.21 log cfu/ml to 1.88 log cfu/ml in apple juice (Mafeti *et al.,* 2014). PL processing of pineapple juice portraits a maximum reduction of 5.8 log cfu/ml of *Escherichia coli* at minimum depth of 5 mm with maximum fluence of 19.70 J/cm^2. However the increase in the depth to 20 mm resulted in a decrease in the log reduction to a value of 4.7 log cfu/ml (Preetha *et al.,* 2016b).

2.4.7.5 Food properties

The efficiency of PL treatment significantly depends on the composition of food samples exposed to the sample. Gomez-Lopez *et al.* (2005b) studied about the decontamination of bacterial populations, such as *Candida lambica*, *Listeria monocytogenes*, *Photobacterium* and *phosphoreum* inoculated in agar samples added with the food components. This found that when starch or water was supplemented to agar no trend in decontamination efficiency was observed, whereas agar with oil and proteins reduced the efficiency of decontamination. Roberts and Hope (2003) also reported a decrease in efficiency of virus inactivation in buffered saline solution added with protein. Hence it was concluded that food samples containing high amount of fat and protein have reduced susceptibility to PL treatment.

The initial microbial population also showed some effects on the decontamination of micro organism. Mafeti *et al.* (2014) reported 2.66 log cfu/ml and 3.76 log cfu/ml reduction in population of *P. expansium* in apple juice inoculated with a population of 3 $x10^5$ cfu/ml and $2.3x10^4$ cfu/ml respectively. Similar result was also observed in *Listeria monocytogenes* in nutrient agar, a higher reduction of 4 log cfu/ml was observed in the samples inoculated with lower population of $1x10^5$ cfu/ml, whereas the samples with higher population($1 x10^8$ cfu/ml) resulted in only one log reduction of bacteria (Gomez-Lopez *et al.,* 2013).

Several studies point out that the effect of initial population was pronounced when the samples treated with PL at low levels of intensities, whereas high intensity levels showed a negligible effect on initial population. It was hypothesised that this could be due to the shadow effect or layered distribution of high density populations in samples, hence the inactivation is limited to superior layers and rest are not affected (Gomez-Lopez *et al.,* 2005a).

The microbial inactivation in PL treatment can be considered as a linear function of the energy dose absorbed by microorganism. The liquids like drinking water and high purity water have a high degree of transparency to a wide range of spectral waves including UV, visible and infrared waves. Most of the liquid food like fruit juices exhibits a uniform distribution of suspended solids in the samples, which increase the absorption properties of liquids and eventually reduce the penetration of light pulses to the liquids (Cacace and Palmieri, 2014).

Pataro *et al.* (2011) studied the inactivation of microorganism using a continuous PL system in apple and orange juices. The absorption coefficient of both juices varies widely and the absorptivity of orange juice was found to be more than apple juice. Accordingly a higher reduction of 4 and 2 log reductions *in Escherichia coli* and *Listeria innocua* were observed in apple juice than 2.98 and 0.93 log reductions in orange juice respectively. Choi *et al.* (2010) also observed a variation in microbial reduction with variation in colour and viscosity.

2.4.8 Effect of PL Treatment on Microbial Inactivation

The susceptibility of PL treatment of different organisms was reported as in the following order by gram-negative bacteria > gram-positive bacteria > bacterial spores > fungal spores (Rowan *et al.,* 1999; Anderson *et al.,* 2000; Levy *et al.,* 2012). Different studies found out various observations which are contradictory to each other. Ferrario *et al.* (2015) found that the most resistant strain of PL treatment is *S. service* compared to the bacterial strains like *Salmonella enteritidis, Escherichia coli* and *Listeria innocua.* Whereas bacteria was found to have more resistance than yeast during PL treatment in another study conducted by Nicorescu *et al.* (2013). Huang *et al.* (2017) reported a higher reduction of the virus population than the bacterial population.

Another factor which greatly influences the effectiveness of PL treatments is the type of microorganism. The sensitivity of each microorganism to the exposure of light pulses varies to a large extend. The PL treatment of juices inoculated with *Escherichia coli* and *Listeria innocua* strains at constant process parameters showed that *Escherichia coli* cells were more susceptible to treatment than *Listeria innocua.* It was also found that gram-negative organisms are more susceptible than that of gram-positive bacteria (Mac-Gregor *et al.,* 1998; Rowan *et al.,* 1999; Anderson *et al.,* 2000; Sharifi-Yazdi and Darghahi, 2006). The microbial inactivation by PL treatment is related to the effects on cell envelopes. Hence the variation in the resistance to the PL treatment can be attributed to the structural and compositional changes of gram positive and negative bacteria (Sharifi-Yazdi and Darghahi, 2006).

2.4.8.1 PL inactivation of bacterial spores and fungal spores

Bacterial or fungal spores are considered as the most resistant form of microorganisms which can survive extremely unfavourable conditions. Different studies have proven that even UV-C resistant bacteria such as *Bacillus subtilis, Aspergillus niger* and *Cryptosporidium* spores are efficiently inactivated by pulsed light (Dunn *et al.,* 1997). Significantly higher reduction in *Bacillus subtilis* spores was observed on surfaces and aqueous solutions treated with pulsed UV light than with continuous light (McDonald *et al.,* 2000).

Dunn *et al.* (1997) conducted experiments with single light pulse at 1-2 J/cm^2 fluence demonstrated that 6 log cfu/cm^2 reduction of bacterial spores and total fluence of 4-6 J/cm^2 could kill 9 log cfu/cm^2 of vegetative cells. Jun *et al.* (2003) carried out a study on the inactivation of *Aspergillus niger* spores in corn meal, and found that a 4.95 reduction in *A. niger* spores were obtained with the input voltage of 3800 V and at 8 cm distance from the quartz window to the sample.

It was reported that about 4.8 log cycles of *Aspergillus niger* spore inactivation was obtained at 5 pulsed light flashes at 1 J/cm^2 (Wekhof *et al.,* 2001). *Aspergillus niger* spores exhibited a 3.0-3.5 log reduction in buffer solution after PL processing at 71.6 J/cm^2 for 60 seconds. Electron micrographs of PL treated samples showed that *A. niger* spores was ruptured and the deformed and collapsed spores revealed deep craters during the light pulse (Wekhof *et al.,* 2001). Chaine *et al.* (2012) revealed that PL treatment at 1.8 J/cm^2 under static conditions resulted in a 3 log cfu/ml inactivation of *B. subtilis* spores in sugar syrup than in distilled water**.**

Keyser *et al.* (2008) reported that apple juice treated with a continuous UV-C light treatment resulted in a 3 log reduction for overall yeasts and mould counts. A 4 log cfu/ml reduction in *S. cerevisiae* was reported in fruit juice inoculated with cell densities such as 5×10^4, 7×10^5 (99.99%) and 5.4×10^6 at 266 nm wavelength with energy dose of 30 J/cm^2 (Sm *et al.,* 2011). Ferrario *et al.* (2013) conducted a study on pulsed light treatment on the *Saccharomyces cerevisiae* KE162 inoculated in apple juice and peptone water using flow cytometry. The authors found that a PL dosage of 71.6 J/cm^2 for 60 s induced a 3.9 and 6-7 log cfu/ml reduction in apple and peptone water respectively. Another study by Ferrario *et al.* (2015) showed a higher resistance of *S. cerevisiae* in apple juices compared to other bacterial strains and *S. cerevisiae* strains were able to recover after the PL processing at refrigerated storage conditions.

2.4.8.2 PL inactivation of fungi

Marquenie *et al.* (2003) studied the inactivation of fungi species such as *B. cneria* and *M. favagena* using PL and observed that a maximum of 3 and 4 log reduction was

obtained for exposure time of 250 s. Anderson *et al.* (2000) also reported a 4.5 log cfu/ ml reduction of fungi when treated with pulsed UV light.

PL processing of apple juice inoculated with 3 and 2.3 cfu/ml *P. expansium* showed a 1.30 log cfu/ml and 3.2 log cfu/ml reductions at dosage of 16 J/cm^2 (Maftei *et al.*, 2014). Patulin is a mycotoxin produced by *Pencilium expansium* and it was found that PL treatment with 2.4 and 35.8 J/cm^2 resulted in significant reduction in the concentration of patulin, whereas a 22% decline in the patulin was observed at the highest dose of PL treatment. Patulin inoculated apple puree showed a 51% decline during PL treatment with 12 J/cm^2 (Funes *et al.,* 2013).

2.4.8.3. Effect of PL on bacterial population

Krishnamurthy *et al*. (2007) evaluated the inactivation of *Staphylococcus aureus* in milk using a flow through pulsed light system. Milk was processed with various distances from UV strobe (5, 8, or 11 cm) and different flow rates *viz.* 20, 30 or 40 ml/min by recirculating the flow through the system thrice. The log reduction of bacteria varied between 0.55 and 7.26 log cfu/ml according to the different process parameters fixed.

Hwang *et al.* (2015) studied the effect of PL treatment on inactivation of *Pseudomonas aeruginosa* in various liquid products. The authors found that the absorptive properties affect the inactivation of microorganism. About 7 log cfu/ml reduction of bacterial population was observed in mineral water and isotonic solutions at a total fluence of 0.97 J/cm^2. Whereas liquid samples such as carbonated beverages, apple juice and plum juice showed 7 log cfu/ml reductions at a PL dosage of 12.17-24.35 J/cm^2. The properties like transmittance and extinction coefficients showed significant effect on the PL treatment of the sample.

Preetha *et al.*(2016b) evaluated the effect of different process parameters such as the number of pulses, depth of juice layer and shelf height on the inactivation of *Escherichia coli* inoculated (10^7 cfu/ml) in orange juice. A 5.38 log reduction of bacteria and 4.14 log reduction of fungi in the lowest shelf height of 5 cm from the lamp and with 5 mm thickness of the juice could be obtained. The authors concluded that increase in

number of pulses, decrease in sample thickness and reduced shelf height from sample resulted in higher reduction of microorganism.

Pollock *et al.* (2017) revealed that PL treatment of agar plates or liquid media inoculated with 5 different strains of *Listeria monocytogenes* from marine sources (lobster, shrimp, salmon and crab) and found that strains of *Listeria monocytogenes* was more resistant which required 20 seconds for 5 log reduction at 800 V and 5 cm distance from PL source.

Salvador *et al.* (2018) studied the effect of PL treatment on the lag phase and the specific growth rate of *Listeria innocua* in culture media maintained at 7°C using time-lapse microscopy. The increase in fluence resulted in an increased lag phase of survivors and 8.7 times increase was obtained at 0.525 J/cm^2 whereas the maximum specific growth rate was decreased by 38%.

2.4.9 Effects of PL on Food Properties

So far very limited studies are conducted on the effect of PL on different nutritional and sensorial qualities of fruit juices. No studies were found reported on any adverse effect of PL on nutritional compounds or the formation of any toxic by-products after treatment (Dunn *et al.,* 1995). Since the wavelength range of PL fall under non ionising radiation of electromagnetic spectrum, the presence of radioactive by products are not expected (Gomez-Lopez *et al.,* 2007).

The PL treatment did not show any significant difference in the pH, TSS and colour values in apple, pineapple, orange and mulberry juices compared to the fresh juice (Maftei *et al.,* 2014). Similarly, Palgan *et al.* (2011) revealed that the apple juice treated with PL at 14 J/cm^2 showed no significant variations in TSS, pH, total antioxidant capacity, total phenol content, colour, acidity, odour and sweetness of apple juice.

Even though colour values observed a significant effect on PL treatments, some minor changes in the colour values were observed in PL treated fermented mulberry, pineapple and apple juices. The fermented mulberry juice showed a significant colour difference in treatment with higher process times (8 seconds) (Kwaw *et al.*, 2018).

Vitamin C is a highly sensitive vitamine very susceptible to exposure of light, oxygen and heat treatment (Kabasakalis *et al.,* 2000). Preetha *et al.* (2016a) observed a significant reduction of vitamin C in PL treated pineapple juice. The PL treatment at 120 flashes (9.6J/cm^2), 15 cm distance and 20 mm depth of sample retained 25.3 mg/100ml vitamin C content, but the increase in dose from 120 to 240 flashes with minimum depth of the sample reduced the vitamin C content to 19.2 mg/100 ml. Similar significant reduction in vitamin C was also reported in apple juice treated with high intense pulse lights (Orlowska *et al.,* 2013). The reduction in vitamin C in food product during the intense pulse radiation exposure could be attributed to the oxidation reactions (Tikekar *et al.,* 2011b).

The pulsed light processing of fermented mulberry juice showed an increase in total phenolic content and total flavour components in the fermented mulberry juice, whereas a significant reduction in total anthocynine content was noted. The increase in total phenolic content and total flavinoid content was in line with the increase in pulsed light exposure (Kwaw *et al.,* 2018).

2.5 COMBINED TECHNOLOGIES

An emerging thought of combination technology has been proposed in the food sector recently, to improve the processing conditions and quality of liquid, solid and multiphase foods. The hybrid technology can provide a better microbial safety and quality of the product by the synergic action of the both techniques together, and it could also reduce the adverse effect of the each technology by lowering the level of application. This technology also would help to improve the energy efficiency and reduce the operating cost.

Different researchers have conducted studies on various combination technologies so far. The ohmic heating treatment depends on the electrical conductivity of products in a large extend. Hence pre-treatments are required in such conditions. Microwave heating may induce cold spot development and overheating of food products. Therefore a combination of ohmic and microwave heating could achieve thermal uniformity in solid and liquid food products. Similarly microwaves penetrate to the solid particles which

could help in the volumetric heating of product independent of the electrical conductivity of the product (Lee, 2014).

It has been reported that during high pressure processing (HPP), some undesirable changes were exhibited in enzymes activities. A combination of high pressure processing with thermal processing could achieve the reduction of enzyme activities. A minimum temperature of 50°C and pressure of 600 Mpa and pH value of product significantly affected the PME activities. Castro *et al*. (2004 b) reported that antagonistic effect of temperature, above 54°C and pressure below 300 Mpa has a positive impact on protein denaturation and enzyme inactivation. The combination of thermal and PEF system was applied for the inactivation of *Lactobacillus plantarum* present in salad dressings and found a higher reduction of more than 6 log cfu/ml in combined thermal and medium electric field treatments (Krebbers *et al.,* 2019).

Thermal Treatment-HPP combination treatments were applied for microbial inactivation of strawberry juice, carrot and tomato purees. Thermal treatment of 80-90°C and high pressure processing at 700 Mpa for 30 seconds resulted in a 4.5 log cfu/ml reduction of *B. stearothermophilus* spores in tomato purees when the final temperature reached 121°C in 30 seconds (Krebbers *et al.,* 2003).

Kim *et al*. (2019) studied the effect of UV combined with ohmic heating against *Escherichia coli* O157:H7, *Salmonella typhimurium* and *Listeria monocytogenes* in tomato juice. The authors observed a 0.48, 1.84 and 3.83 log cfu/ml reduction of *Escherichia coli* O157:H7 in juice treated with UV-C radiation, ohmic heating and combined treatments respectively. A synergistic bactericidal effect was observed in combined treatment such as acceleration of lipid oxidation, which imparts an additive effect in pore formation in cell membranes.

Caminiti *et al.* (2011) reported that the combination of high intensity pulsed light (5.1 J/cm^2 or 4.0 J/cm^2) and pulsed electric field (24 kV/cm, 34 kV/cm) achieved a minimum microbial reduction of 5 log cfu/ml as recommended by FDA. Quality attributes were not affected by these treatments and sensory evaluation was most acceptable for the selected non-thermal treatments.

Munoz *et al*. (2012) reported that the combination of high intensity pulsed light (HIPL- high (H) 5.1 J/cm^2, low (L) -4.03 J/cm^2) and thermosonication (TSH- 5 min residence time, 50°C, TSL-2.8 min residence time, 40°C) resulted in an inactivation range from 1.10 (TS-H) to 2.42 (HIPL-H) for the individual treatments, and from 2.5 (HIPL-L and TS-H) and 3.93 log cfu/ml (HIP L- H and TS- L) for the combined treatments. Munoz *et al.* (2012) found that the combination of both pulsed light (PL- 4.03 J/cm^2- low and 5.1 J/cm^2 - high) and thermosonication (TS – 24 kHz, 100 μm) resulted in a 6 log cfu/ml inactivation of *Escherichia coli* in apple juice. Individual treatment had only 2.7 and 4.9 log cfu/ml inactivation for thermosonication and pulsed light respectively. All the treatment changed the colour of the apple juice significantly ($p < 0.05$).

A blend of orange juice and carrot juice was processed by combining treatments such as pulsed electric field (24 kV/cm, 18 Hz, 93 μs), ultraviolet light (UV dose -10.6 J/cm^2) and high intensity light pulse (3.3 J/cm^2) with manothermosonication technology (400 kPa, 35°C, 1000 W, 20 kHz). No significant changes were found in non-enzymatic browning or antioxidant activity, but the colour value increased and total phenolics were significantly decreased (Caminiti *et al.,* 2012).

Birmpa *et al.* (2014) evaluated the effectiveness of the combination of three non-thermal light technologies such as non UV-visible, continuous UV and high intensity light pulse (HILP) on their ability to inactivate *Escherichia coli K12* and *Listeria innocula*. Among the treatments, high intensity pulsed light effectively reduced the microorganisms to 3.07 and 3.77 log cfu/ml. Ferrario and Guerrero (2016) reported that the combination of pulsed light (0.73 J/cm^2) and ultrasound (30 min) treatment of apple juice showed a 3.7-6.3 log cfu/ml reduction of inoculated microorganism. Individually pulsed light treatment alone resulted in an inactivation of only up to 1.8 to 4.2 log cfu/ml. Combined treatments delayed yeast and mould growth and also prevented the apple juice from browning during storage.

Gouma *et al.* (2015) evaluated the resistance of yeast population in apple juice against the combined UV and heat treatment. The combined UV and heat treatment

between temperatures of 52.5 and 57.5°C observed a higher inactivation in *S. Cerevisiae* than individual UV or heat treatment, which proves the synergic action. A 5 log cfu/ml reduction of *S. Cerevisiae* was obtained with lower UV doses and treatment times than required for individual treatments.

The effect of PL treatment in combination with heat treatment on the resistance of *B. subtilis* spores were evaluated by Artiguez *et al.* (2015). A PL dosage of 0.4 and 0.5 J/cm^{2}and thermal processing at 90°C with different treatment times of 5, 10, 15 and 20 min were selected for the study. A 1.8 log reduction of *B. subtilis* spores were obtained in pre heating of the product at 90°C for 5 min and exposure to the PL at a dosage of 0.4 J/cm^2, whereas PL dosage at 0.5 J/cm^2 with the thermal treatment showed a 4 log reduction in *B. subtilis* spores. Whereas the thermal treatment alone resulted in a 0.3 and 0.6 log reduction in *B. subtilis* spores treated with 90°C for 5 min. It was demonstrated that the combination of PL treatment with thermal treatment could be a promising technology for inactivation of microorganism.

A study was carried out to evaluate the effect of combined UV and thermal treatment on inactivation of pathogens *viz. Listeria monocytogenes, Escherichia coli, Staphylococcus aureus* and *Salmonella typhimurium*. All strains of bacteria showed a significant reduction in population when exposed to UV-C rays at a temperature range of 50 to 60°C. *Escherichia coli* was the most resistant organism during UV-heat treatment. The combined treatments could reduce the process time from 49.6% to 89.1% than with UV-C treatment alone (Gouma *et al.,* 2015).

UV treatment in combination with mild ohmic treatment (1485 V/m) at 65°C resulted in more than 6 log cfu/ml reduction of *Escherichia coli* K12 in apple juice (Lee *et al.,* 2013). Sean *et al.* (2016) found that 5 log cfu/ml reduction in bacterial population was obtained in pineapple juice treated with a UV assisted ohmic heating process at 30 v/cm and 50°C temperature and 1200 mJ/cm^2 of UV radiation dose. It was also reported that the synergic action of ohmic heating and UV effectively improved the microbial reduction of the UV penetration and damage of microbial cell structure through the pores developed by the electroporation during ohmic process.

Materials & Methods

CHAPTER III

MATERIALS AND METHODS

This chapter describes the materials used and the methodology adopted for the development of ohmic heating assisted pulsed light treatment system for fruit juices, and the procedure followed for the evaluation of the developed system leading to the standardisation of the operating parameters towards the preservation of pineapple and cashew apple juice. The procedures adopted for the evaluation of the physiochemical, microbial and sensory qualities of the pineapple and cashew apple juice are also explained in detail.

3.1 DEVELOPMENT OF OHMIC HEATING ASSISTED PULSED LIGHT TREATMENT SYSTEM

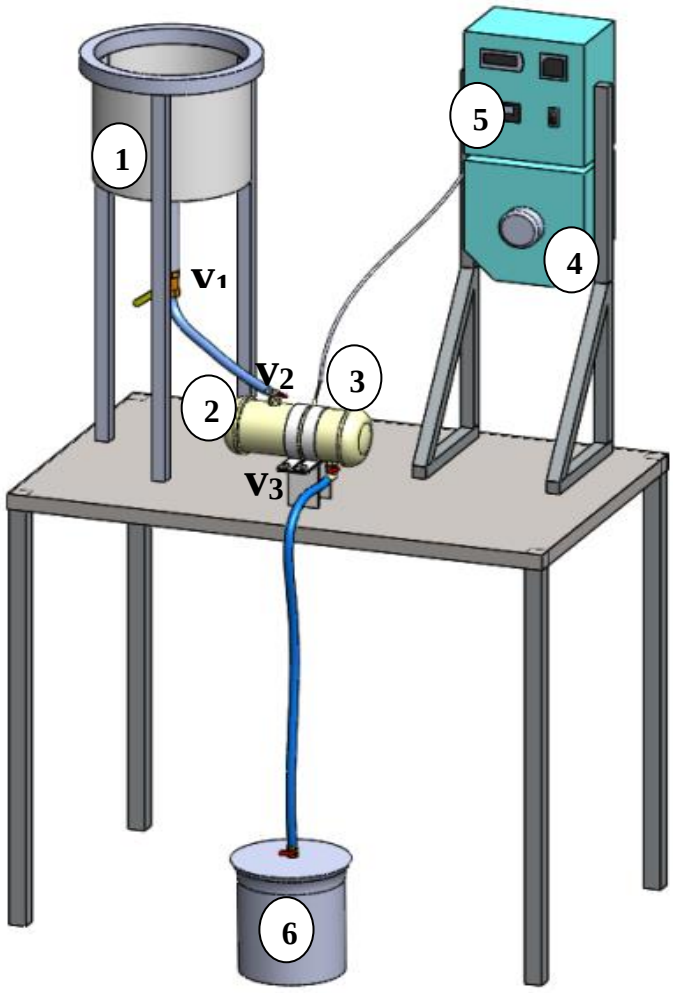
Ohmic heating and pulsed light system are proved to have bactericidal effects but the intensity of process parameters need to be high, if applied separately. The development of a combined system could reduce the intensity of process parameter doses, thereby improving the quality of the processed juice along with increasing its shelf life. In order to list the hypothesis explained in Chapter 1, an ohmic heating assisted pulsed light system for pumpable fruit juices was designed and fabricated.

3.1.1 Development of Ohmic Heating System

Ohmic heating is the process of heating food materials sandwiched between two electrodes. The food materials offer resistance to the flow of current and hence heat the food volumetrically. Ohmic heating technology is highly suitable for liquid and particulate foods especially fruit juices, due to its acidic nature. The acidity and ions present in food systems improve the electrical conductivity, and hence aid in the rapid increase of the process temperature (Sarang *et al.,* 2008). An additional non thermal effect due to electroporation during ohmic heating also facilitates rapid processing and retention of nutrients in food.

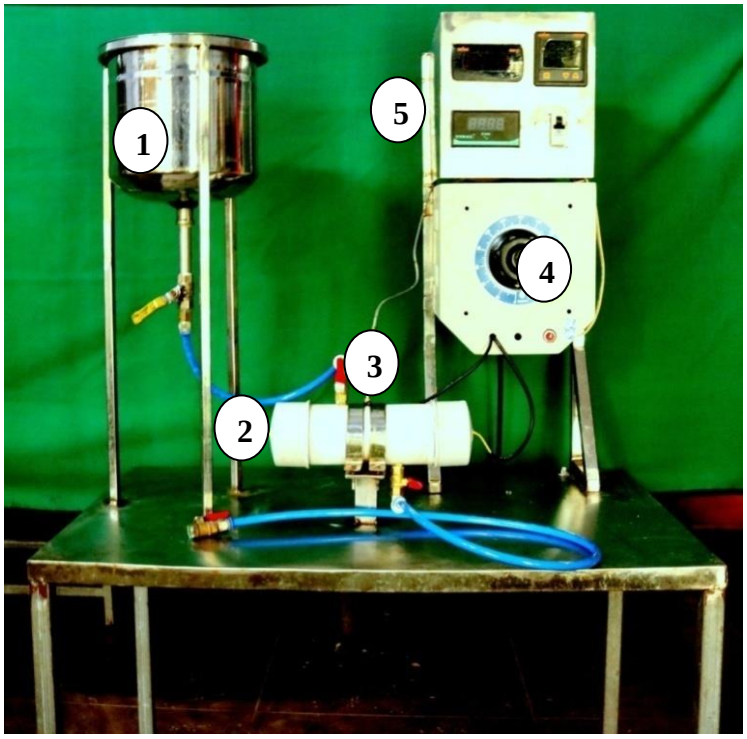
The experimental ohmic heating system consists of a feed tank (1), ohmic heating chamber (2), varying power source (variable transformer) (3) volt and ampere meter (4)

and temperature measuring system (5). The schematic of the experimental ohmic heating set up is shown in Plate 3.1 and the developed experimental ohmic heating set up is shown in Plate.3.2.



1. Feed tank
2. Ohmic heating chamber
3. Temperature measuring probe
4. Variable transformer
5. Measuring instruments
6. Sample collector

Plate 3.1 Schematic of ohmic heating system



1. Feed tank
2. Ohmic heating chamber
3. Temperature measuring probe
4. Variable transformer
5. Measuring instruments

Plate 3.2 Developed ohmic heating system

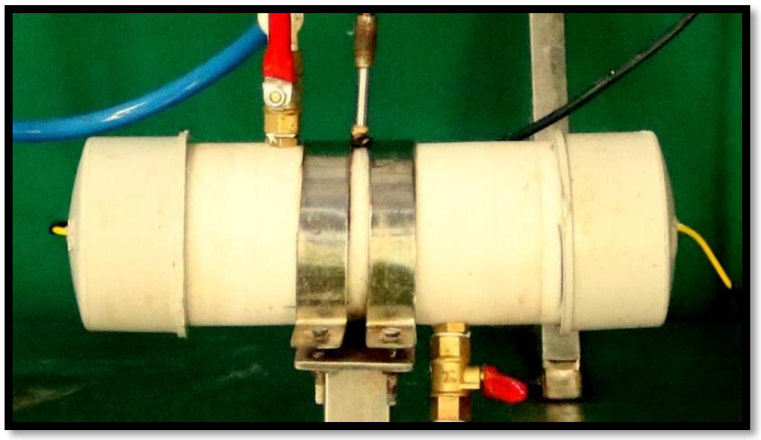


Plate 3.3 Ohmic heating chamber

Plate 3.4 Stainless electrode

3.1.1.1 Feed tank

A feed tank made up of 0.6 mm thick food grade SS 304 stainless steel would supply the fruit juice to the ohmic heating chamber by gravity discharge. The diameter and height of the tank are 200 mm and 200 mm, respectively, with a capacity of five litres. About 10% head space volume was preferred for the possible process actions such as thermal expansion or foaming and filling of the fruit juices. The feed tank is connected to the ohmic heating chamber via a SS 304 stainless steel pipe of diameter 1.5 cm and a flexible hose of diameter 1 cm. This arrangement would ensure free flow of fruit juices to the chamber.

3.1.1.2 Ohmic heating chamber

Ohmic heating chamber consists of a cylindrical chamber made up of Teflon and two stainless steel electrodes fixed on opposite sides of the chamber. The material to be heated would be filled in the space provided between the two electrodes.

The ohmic heating chamber should be designed considering three important criteria; that is, it should be electrically nonconductive, able to withstand process temperature and should not impart off flavour to the product. Teflon rod is an excellent insulator this can withstand high treatment temperature and does not react with food materials. Accordingly, a cylindrical Teflon block of 80 mm diameter and 150 mm length was used for the construction of the chamber. A bore of 70 mm diameter was drilled so that the wall thickness of the chamber is 5 mm. The ends of the cylinder were closed with the Teflon end caps (Plate 3.3) which perfectly seals the hollow Teflon cylinder on either side. Holes of diameter 3 mm were drilled on both the end caps for power connection to the electrodes installed on the either side of the chamber; another hole of 5 mm diameter was drilled in the middle of the chamber to insert the thermocouple for the detection of temperature inside the chamber. The ohmic heating chamber with overall dimensions of 150 mm length and 70 mm inner diameter was used for conducting experiments in batches with each batch being enabled to handle a volumetric capacity of 300 ml of fruit juice per treatment.

3.1.1.3 Electrode

The electrode material should be a non porous surface that does not absorb odour and flavour and does not provide breeding ground for bacteria and fungi. Also it should be capable of withstanding high temperature. The electrodes for the ohmic heating system are made of food grade, non-corrosive SS 304 material of 2 mm thickness. The electrodes were cut into circular shape with an outer diameter of 70 mm so that they fit perfectly inside the cylinder (Plate 3.4). The surface of the SS electrodes was polished to a smooth finish using a fine emery sheet. The ohmic heating rate depends on the spacing between the electrodes and the voltage gradient applied. Based on the preliminary studies conducted, the spacing between the electrodes was fixed at 150 mm. The circular electrodes at both ends were provided with 2.5 mm dia and 3 cm length copper rods welded to the centre of discs which serves as a terminal for electric supply. A Teflon end cap is provided at both ends which cover the electrodes. To ensure safety, the ohmic heating chamber ends are provided with polypropylene couplings so that all the electrical terminals are perfectly covered (Plate 3.3).

The accuracy and efficiency of the developed ohmic heating chamber was roughly tested by determination of the electrical conductivity of sodium chloride solution at 1.5% concentration and comparing the calculated values with reference values (Parmar *et al.*, 2016).

3.1.1.4 Variable transformer

A variac (M/s. Delta control, Mumbai) capable of varying the voltage in the range of 0 to 270 V (8 A capacity) was used to supply power to the ohmic heating chamber (Plate 3.1). A 230 V, 50 Hz electrical supply would serve as input to the variac.

3.1.1.5 Measuring instrumentation

A digital type Voltmeter (500 V, 20 A) was used to monitor potential difference between the electrodes throughout the process and a digital type ammeter (230 V, 20 A) was used to monitor changes in current during the process. The temperatures of the samples were recorded using a SS 304 stainless steel resistance temperature detector

(RTD), which can measure temperature up to 300°C (M/s. Heatran Services, Coimbatore). A temperature indicator unit is attached to the RTD probe to read the temperature in the system.

3.1.2 Measurement of Electrical Conductivity

The electrical conductivity of fruit juices is considered as an important parameter in ohmic heating treatment. The electrical conductivity of pineapple and cashew apple juice at different voltage gradients and at constant intervals of time were determined using the Equation 3.1.

$$\sigma = \frac{IL}{VA} \qquad 3.1$$

where, V is voltage, I is current, L is length of ohmic heating chamber and A is the area of cross section of the stainless steel electrodes.

The electrical conductivity was determined with the help of the data collected from the ammeter and volt meter attached to the ohmic heating chamber. Fresh fruit juices were supplied to the ohmic heating chamber from the feed tank through the valve V_1 and V_2 once the chamber was filled, constant voltage gradient was set with the help of variac whose value was noted from the voltmeter. The current and temperature readings were noted with the help of ammeter and RTD temperature probe at regular intervals of time. This experiment was allowed to continue until the temperature of the sample in the ohmic heating chamber was 60°C. Experiment was performed for five different voltage gradients and readings were taken in triplicate for each voltage gradient and the average is reported.

3.1.3 Design of Pulsed Light Treatment System

A pulsed light treatment system for efficient inactivation of microorganisms as per the hypothesis mentioned in Chapter I, was conceived and developed. The components of the developed pulsed light treatment system are xenon flash lamp, contactor relay, resistors, rectifier, capacitors, micro processors, trigger transformer, thyristor and timer

relay *etc.* In order to produce pulsed light, a circuit with these components was developed.

3.1.3.1 Xenon flash lamp

Xenon flash lamps are generally filled with inert gases *viz.* xenon and krypton. xenon as a filling gas is more efficient due to its higher overall conversion capability (www.perkinelmer.com/opto). Xenon flash lamp have several other advantages including its mercury free nature, higher microbial inactivation rates and reduced rate of formation of organic chemicals due to the pulsed nature of light (Grapperhaus *et al.*, 2005; Wekhof, 2003; Hillegas and Demirci, 2003). Also, the pulsed light generated using the xenon flash lamp produces higher total radiative output for a given electrical input than other gases like krypton. Hence, the xenon flash lamp was selected for generation of pulsed light in this study and the circuit was designed based on the xenon lamp specifications.

The xenon flash lamp (Make: Heraeus Noblelight ltd., U.K) as shown in Plate 3.5. comprises of a sealed tube with a thickness of 3 mm, bore diameter of 8 mm and arc length of 70 mm made of clear fused quartz (CFQ) filled with xenon gas at a pressure of 59 kPa. The selected CFQ tube also consists of electrodes to carry electrical current to the gas, and a trigger electrode. The xenon flash lamp with a clear fused quartz envelope was used to produce the broad spectrum flash from 100 to 1100 nm. CFQ transmits short wave ultra-violet (UV) light, while the other forms of quartz restrict wavelengths in the UV region.

A high voltage power source is necessary to energise the gas which is stored in a capacitor for allowing accelerated delivery of high voltage electrical current once the lamp is triggered. For this the capacitor which is charged to a relatively high voltage is connected to the electrodes fixed at both ends of the tube. These tubes are flashed through the trigger electrode, which is a thin wire made of nickel wound around the length of the lamp. A high voltage of approximately 12 kV is applied as a pulse to the trigger electrode which ionises the xenon gas and initiates conduction. A current (5 A) flowing through the lamp at this stage produces more ionisation of gas and hence conduction of electron occurs inside the tube. The electron surrounding the xenon atoms gets excited, and results

in jumping to higher energy levels. When the electrons drop back to a lower orbit, energy is released in the form of photons (high intensity light pulses).

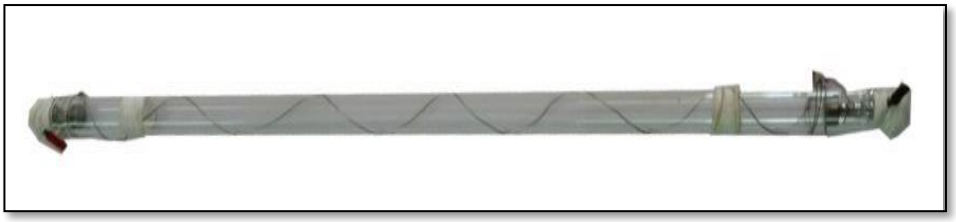


Plate 3.5 Xenon flash lamp

3.1.3.2 Relay

Relay is a switch which performs ON/OFF function. In the circuit, time gap between the triggering is achieved using the relay. The contactor relay provided has a capacity of 32 A.

3.1.3.3 Resistors

In order to protect the rectifiers, transformers and other components in the circuit, current must be limited. A number of metallic film resistors are used in different circuits act as a current limiting component in the circuit.

3.1.3.4 Rectifier

Pulsed light treatment works on a DC current and hence a full wave rectifier was used to convert the incoming 230 V alternating current to direct current of 5 V to provide input voltage to microcontroller and other sensors. The rectifiers are also provided in the flashing circuit.

3.1.3.5 Capacitor

Capacitor is a passive two-terminal electrical component used to store electrical energy temporarily in an electric field. Capacitor is the main component of the xenon lamp and totally 31 capacitors from C_1 to C_{31} is attached in different circuits.

3.1.3.6 Microcontroller

Microcontrollers are generally employed in automatically controlled devices and products. Microcontroller is a single low cost micro computer on a single metal oxide

semiconductor integrated circuit chip. The microcontrollers include processers and programmable input output peripherals. The control of pulsed light process parameters are achieved through the microcontroller (PIC16F877a) as depicted in Plate.3.6.

The microcontroller store the program for the efficient working of PL system in its memory and the output signals are displayed in the LCD display board provided in the machine. The controller consists of five 8 bit ports which include total 40 numbers of analog input pins and digital input and output pins.

These pins provide different control functions for the generation of pulsed light. The analog pin 1 was connected to the input supply of 5 V for the working of micro controller. The pin 2, 3, and 4 are programmed for the analog input signals from the three knobs provided in display panel for varying the distance from the xenon lamp, flow rate and pulsed dosage respectively. Pins 8, 9, and 10 are featured for the control of stepper motor drive which helps in varying distance from the xenon lamp using a mechanical assembly. The pins from 34 to 40 were programmed for the digital output display to the LCD display board provided in machine.

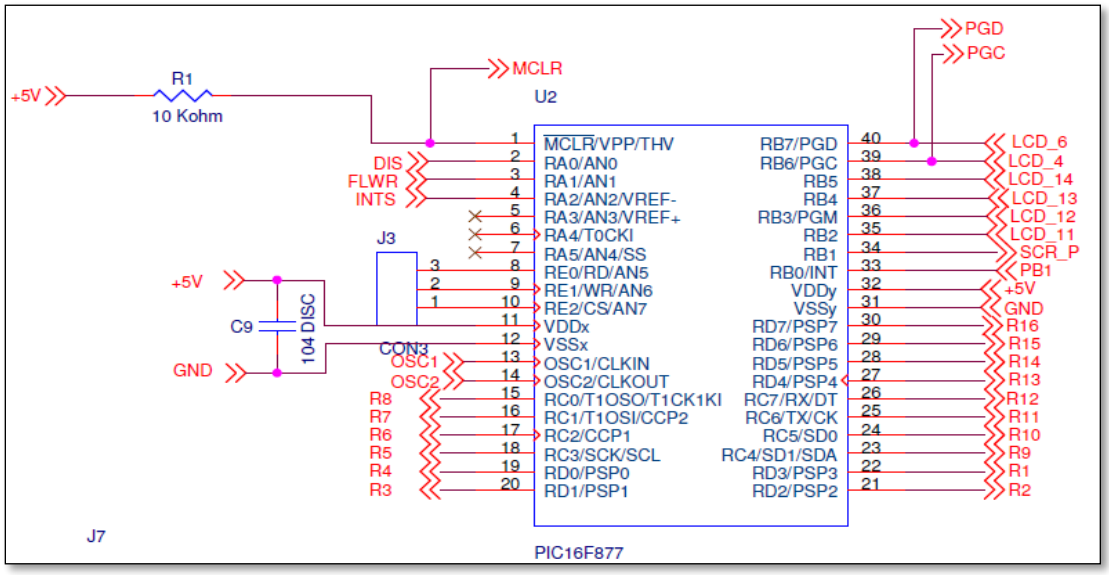


Plate 3.6 Connection diagram of microcontroller

3.1.3.7 Stepper motor driver

The stepper motor driver is a micro stepping driver used for controlling bipolar stepper motor employed for moving the xenon lamp assembly to 5 cm, 10 cm, and 15 cm so as to change the distance between the lamp and the fruit juice flowing through the quartz tube. The connection diagram of stepper motor is depicted in Plate 3.7.

The driver system is connected to two pins of the microcontroller. The micro controller defines the direction of rotation of motor and the number of steps the motor would proceed when operated through the external knobs provided on the front display board. The steps of motor movement are adjusted as 5 cm, 10 cm, and 15 cm distance of the lamp from quartz tube. The driver requires a voltage range of 8-35 and a capacitor for protecting the driver board from voltage variations.

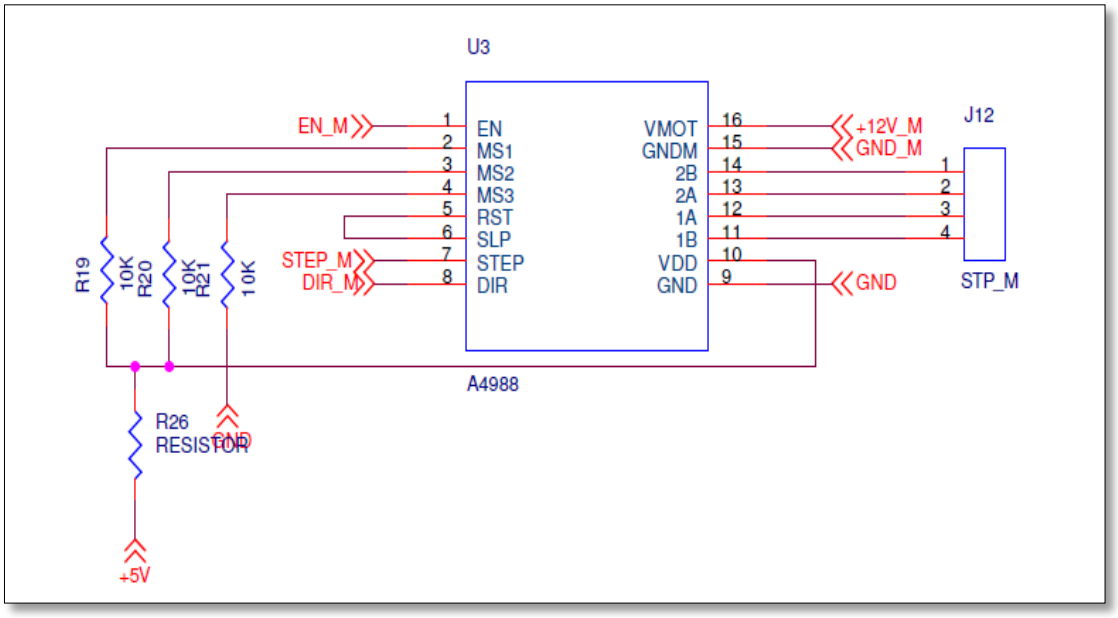


Plate 3.7 Connection diagram of stepper motor driver

3.1.3.8 Transistor arrays

The transistor arrays (ULN283A) employed in this circuit is a high current high voltage transistor array used with microcontrollers where high power load is required. This includes 8 N-P-N Darlington transistors to provide proper amplification of current with respect to applied load. Darlington pins are used in circuit to execute required

amplification. When the base voltage is absent, no signal is given to the input pins of the integrated circuit and the transistor remains in off stage.

3. 1. 3.9 Trigger coil Transformers

The conventional PCB-mount trigger transformer with specification ZS-1052-AC was used in the flash generator circuit. This is mainly employed in intense pulsed light generators for generating high intensity flashes by ionisation. The trigger pulse transformer is used to supply the high voltage of 12 kV required for ionisation of the xenon gas as a very sharp pulse in a very short duration of time. The trigger transformer was designed in such a way to step up 400 V DC current to 12 kV DC and to produce a sharp pulse. This resulted in a very high impulse voltage across the secondary coil of the trigger pulse transformer which is conducted into the trigger electrode wire that surrounds the flash lamp.

3.1.3.10 Opto- coupler

An opto-coupler integrated in to the circuit for the purpose of isolation of two basic circuits in xenon flash lamp circuits having different voltage and transfers the electrical signals from one to other by using light. The basic design of an opto-coupler, also known as an opto-isolator, consists of an infrared emitting diode that produces infrared light and a semiconductor photo-sensitive device that is used to detect the emitted infrared beam. The main advantage of photo-triacs is to provide a complete isolation from any noise or voltage spikes present on the AC power supply.

In this circuit the opto-coupler is employed to isolate the SCR pulse circuit from thyristor in the flash lamp circuit. Based on the microcontroller program, the SCR pulse is generated at specified interval. The optocoupler conducts when SCR pulse is received on it and provides the gate voltage (400 V) for thyristor to start working as a switching device.

3.1.3.11 Thyristor

A thyristor is a solid-state semiconductor device with four layers of alternating P and N type materials. It acts exclusively as a bistable switch, conducting when the gate

receives a current trigger, and continues to conduct until the voltage across the device is reverse biased, or until the voltage is removed. It requires a gate pulse to start and gets self-latched, and stays on until the supply get interrupted. For this, we have used a switching circuit (SCR) across the thyristor to turn it off. In this circuit thyristor functions as a switching device in the xenon flash lamp circuit. When the thyristor gates receive supply voltage from optocoupler, the thyristor starts conducting and provides the supply to run the flash lamp circuit.

3.1.3.12 Generation of pulsed light

The generation of flash is initiated by the ionisation of the gas and large pulses of current are then transferred through the ionised gas. Ionisation is important phenomena which could reduce the electrical resistance of gas and hence make ease of the transfer of as much as thousands of amperes through the xenon flash tube. When this current pulse travels, it excites electrons surrounding the xenon atoms causing them to jump to higher energy levels. The electrons immediately drop back to a lower orbit, producing photons in the process. Thus conversion of electrical energy in to light energy is achieved.

3.1.3.13 Circuit for the Pulsed light treatment

The pulsed light circuit includes different components as explained in the section 3.1.3 portrayed in Plate 3.8 and the circuit diagram is shown in Plate 3.9. The circuit includes a DC-DC convertor to convert 5 V to 400 V for xenon lamp, a main capacitor which store and discharge electrical energy (10 μF, 450 V), thyrister (which act as a trigger switch) and a trigger transformer to generate high voltage in order to ionise the xenon lamp (400 V to 12 kV). The input signals are sending through the step pin connected to the micro controller and then the opto-coupler will trigger the thyristor according to the input signal received and eventually the thyristor actuates discharge through trigger transformer. This generates a short high voltage (12 kV) pulse that triggers the flash tube. The main flash capacitor C_{30} (10 μF) of the flash tube circuit gets discharges, which generates a bright flash of xenon lamp. The charging and discharging of C_{30} (10 μF) continues through the relay circuit in very short time intervals.

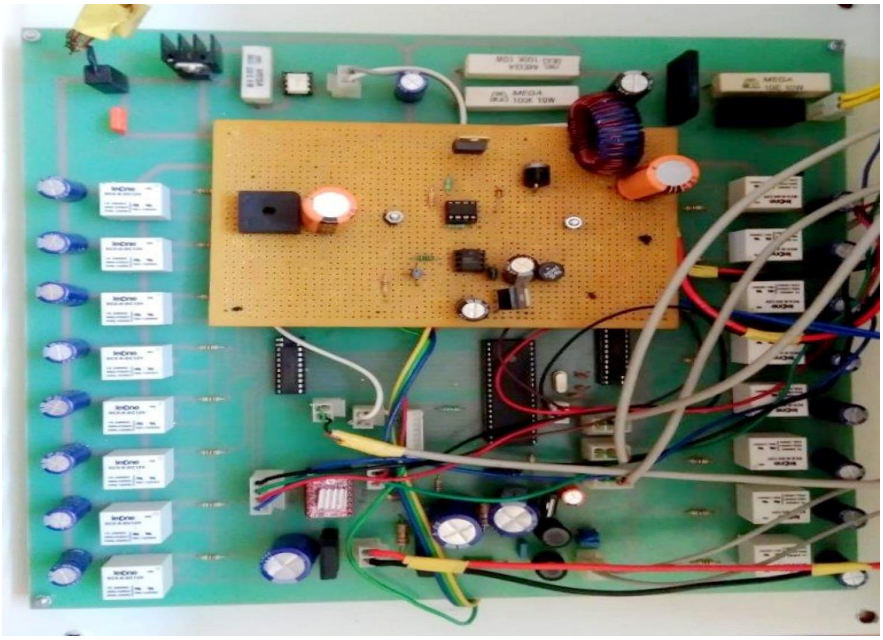


Plate 3.8 Circuit board of PL system

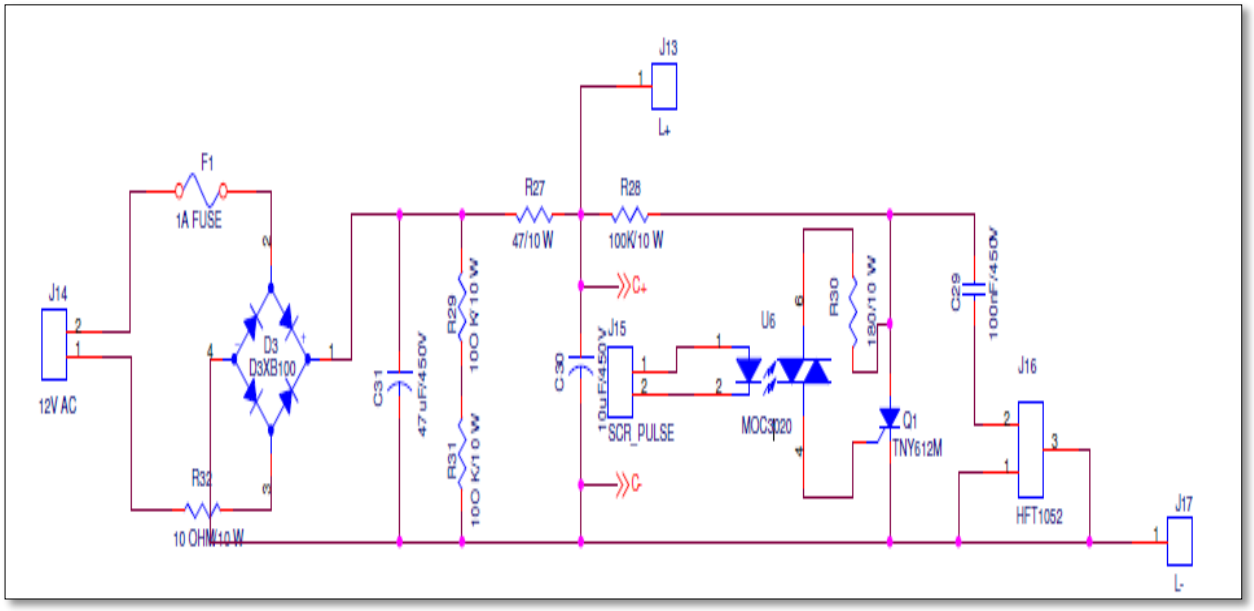


Plate 3.9 Circuit diagram for xenon flash lamp

3.1.3.14 Pulsed light radiation dose

The Bunsen-Roscoe reciprocity law for photochemical processes states that the magnitude of a photochemical reaction, such as the effects of UV-C radiation on nucleic acid of micro-organism is directly related to the total dosage of radiant energy that hits the target (E). The intensity of the pulsed light treatment is expressed as pulsed light dose. The overall effect of the radiation depends on combination of applied intensity and exposure time. The applied dosage was calculated using the following formula (Chang *et al.,* 1985; Morgan, 1989; Stevens *et al.,* 1999).

$$E = \frac{C\mathrm{V}^2}{2} \qquad 3.2$$

where,

E= Energy delivered from pulsed light, kJ

C- Capacitance of storage capacitor, μF

V- Voltage across flash lamp, V

The area of quartz tube exposed to pulsed light,

$$A = \frac{\pi r^2 \mathrm{L}}{2} \qquad 3.3$$

The pulsed light dosage (J/cm^2),

$$D = \frac{\mathrm{E}}{A} \qquad 3.4$$

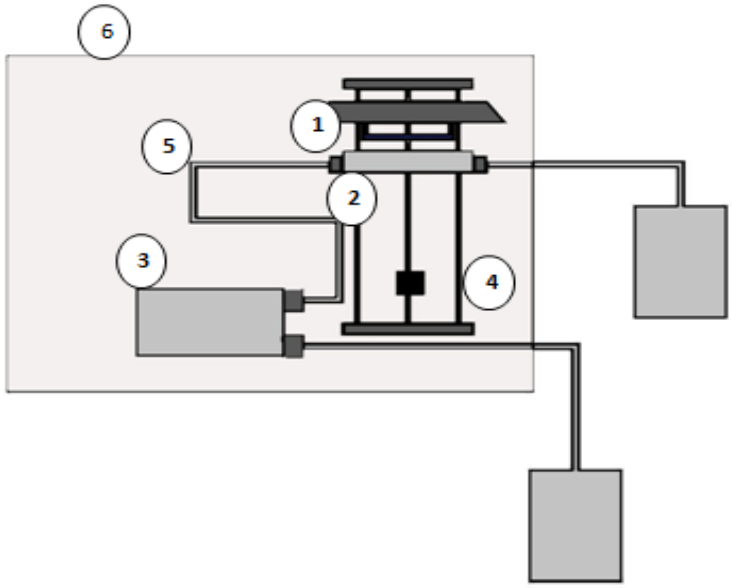
L-Length of quartz tube, cm

3.1.4 Design of Continuous Flow Pulsed Light System

Batch pulsed light system are being employed for carrying out research in fluid systems. Research on continuous flow PL systems is not found reported widely. In this study an indigenous continues flow PL system towards preservation of fruit juices was designed and fabricated for lab scale research.

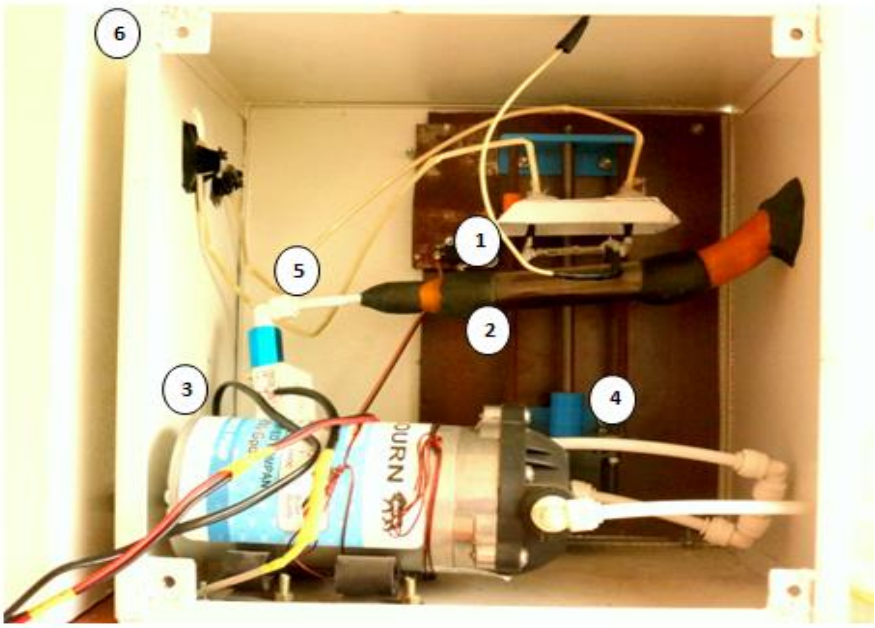
The developed experimental model PL system integrating the electronic circuits and PL generation and monitoring components mentioned in earlier sections are shown in Plate 3.10 and Plate 3. 11 respectively.

The PL treatment experimental set up consists of the feed tank, pulsed light treatment chamber, circulation system, collection chamber, and a display panel.



1. PL treatment chamber
2. Quartz tube
3. Pump
4. Stepper motor
5. Flow pipe routing
6. Xenon flash lamp

Plate 3.10 Schematic diagram of pulsed light treatment system



1. PL treatment chamber
2. Quartz tube
3. Pump
4. Stepper motor
5. Flow pipe routing
6. Xenon flash lamp

Plate 3.11 Pulsed light treatment system

3.1.4.1 Feed tank

A 1000 ml glass beaker was used as feed tank for the system. A flexible polypropylene tube of 8mm diameter connects the feed tank and PL treatment chamber through a circulation pump as shown in Plate. 3.11.

3.1.4.2 Pulsed light treatment chamber

The pulsed light treatment chamber consists of the pulsed light lamp, mechanical assembly of backward and forward movement of PL lamp, quartz tube of 15 mm dia and 70 mm length, circulation motor and associated electrical and electronic connecting terminals and circuits (Plate 3.11). These components are enclosed in the chamber made of stainless steel (SS 304) with dimensions of 400 mm length 200 mm breadth and 300 mm height. The quartz tube for fluid flow was installed at the centre of the chamber horizontally and parallel to the PL lamp. The quartz tube would permit 90 percent of the

light spectrum radiant on it from PL lamp (Pataro *et al.*, 2011). The distance between the PL lamp (source) and the quarts tube can be varied by the movement of stepper motor and associated mechanical assembly.

3.1.4.3 Circulation system

The liquid food has to be circulated through the treatment chamber based on the required time. The circulation system consists of a 24 V DC booster pump (Make: Bourn) and associated flow routing pipes. The flow rate of the fruit juices through the quartz tube was controlled by a flow sensor attached to the flow routing pipes. A flow diversion valve was provided in the outlet pipe line to collect the treated fruit juices after treatment.

3.1.4.4 Display panel

The main control board includes two switches with different colour codes of Red, and Black for starting the PL process and initiation of the device respectively. Besides, three rotory control knobs are provided on the display panel to vary the pulsed light dosage, distance of lamp from the quarts tube and flow rate. The LCD screen displays the distance of lamp from the quartz tube, PL dosage and flow rate of juice as programmed or set according to the treatment.

3.2 SELECTION OF LIQUID FOODS

Fruit juices are rich source of vitamins and minerals and are commonly marketed as ready to serve beverages, squashes, or cordials. Fruits mainly preferred for preparing juices are orange, lemon, pineapple, grapes, apple, mango, pomegranate, *etc.* Among these, pineapple juice is becoming more popular due to its freshness, aroma and tasty flavour. Cashew apple is found to be an underutilised fruit crop. Usually the fruits are left over the field after the harvest of nuts. Goa is the only state in India where cashew apples are utilised to prepare "*feni*" a fermented cashew apple beverage. Some conventional products like jam, candy, syrup, vinegar and wine are prepared using cashew apple but commercial production is still limited, deserving exploitation of unique processing technologies. Recently the products from cashew apple are found to be

accepted by consumers. Considering all these factors as detailed in Chapter I, cashew apple and pineapple juices were selected for the study.

3.2.1 Pineapple Juice

Pineapple fruits of *'kew'* variety purchased from the local market (Tavanur, Malappuram) were visually inspected for bruises and blemishes and sound fruits were selected. Fruits were washed in running water and the crown portion and the outer skin of the pineapple was removed with a sterilised serrated knife. The peeled slices were cored and then cut in to small pieces and were passed through a centrifugal extractor, (Kenstar food processor, Kitchen appliances Ltd., India) and the juice was collected in sterile stainless steel vessel. The extracted juice was filtered using muslin cloth to remove the coarse tissues, collected in a sterile PET bottle and stored in refrigerated condition (4±2°C) for conducting the ohmic and PL treatments.

3.2.2 Cashew Apple Juice

Fully ripe firm fruits of cashew apples without bruises were collected from farm of Cashew Research Station, Kerala Agricultural University, Madakkathara, Thrissur. Fruits were washed in running water after discarding immature, rotten, and damaged fruits. Juice was extracted in juice extractor (Plate 3.12), strained through muslin cloth, clarified with addition of powdered and cooked sago @ 2g per litre to remove the astringency caused due to the tannin compounds present in cashew apple (Jayalekshmy and John, 2004). Stirred juice was kept still for 12 hours at refrigerated condition (4±2°C) to allow the tannin to settle and the upper layer of clear juice was decanted carefully without mixing with the sediments. This clarified juice was stored in well sterilised air tight stainless steel barrels at below -18°C and then used for treatments as required.

Plate 3.12 Juice extractor

3.3 EXPERIMENTAL DESIGN

The whole research work is divided in to four major sections such as experimental design of ohmic heating, pulsed light treatment, ohmic assisted pulsed light treatment, characterisation studies of fruit juices with ohmic assisted pulsed light treatment and *Escherichia coli* and *Listeria monocytogens* species inoculation studies.

Experiments were framed using Box-Bekhen Response Surface Methodology using Design expert software version 7.0.0 to optimise the voltage gradient, process temperature, and holding time combination of ohmic heating process with respect to the microbial and biochemical quality of the fruit juices under study. Similarly, process parameters of pulsed light treatment such as pulsed light dose, sample source distance, and flow rate were optimised using response surface methodology the software for the effective pasteurisation with respect to the biochemical and microbial quality of the fruit juices.

3.3.1 Independent Variables

Table 3.1 independent variables of ohmic heating and pulsed light treatment

Independent variables of ohmic heating treatment	Independent variables of pulsed light treatment
a) Fruit juices ➢ Cashew apple juice ➢ Pineapple juice	a) Fruit juices ➢ Cashew apple juice ➢ Pineapple juice
b) Voltage gradient (10 to 15 V/cm)	b) Pulse dosage (8-32 J/cm^2)
c) Treatment temperature (50 to 60°C)	c) Sample-source distane (5-15cm)
d) Treatment time (1 to 5 minutes)	d) Flow rate (150 to 300 ml/min)
e) Storage period (30 days)	e) Storage period (30 days)
f) Storage temperature (4±2°C)	f) Storage temperature (4±2°C)

3.3.2 Dependant Variables

3.3.2.1 Physico-chemical characteristics

The physicochemical characterestis *viz.* pH, TSS, titrable acidity, ascorbic acid content, total sugars, total phenols, tannin content and total colour difference of treated fruit juices are analysed to optimise the process parameters of ohmic heating and pulsed light process.

3.3.2.2 Microbial analysis

The microbial properties such as bacterial reduction and yeast and mould reduction were analysed for treated fruit juices to optimise ohmic heating and pulsed light operating parameters.

3.3.2.3 Sensory Properties

The sensory attributes of the optimally treated fruit juices were analysed using fuzzy logic quality ranking.

3.3.3 Design of Experiments Using Response Surface Methodology

Response surface methodology (RSM) was used to determine the optimal processing condition, to estimate the interaction between the parameters, and to provide data for a predictive regression model. Among several approaches used to apply the response surface method, the Box-Behnken design, an independent quadratic design was chosen because the safe operating zone for the process is known. Central composite designs in general, have axial points outside the cube, which may not be in the region of interest, or may be impossible to run because they are beyond safe operating limits. The Box-Behnken design, however, does not have axial points, thus all points designed can fall within the safe operating zone. Furthermore, the Box-Behnken design also ensures that no factors are set at high levels simultaneously. The treatment combinations are at the midpoint of edges and the centre of the factor.

The Box-Behnken design with the three experimental factors such as A, B and C requires twelve data points in the middle of each two factor combination and five replications at the centre of the cube, totaling seventeen data points in contrast to 3^3 runs required for a full factorial design. It can be seen that three levels of each factor, a minimum, a maximum, and a medium should be pre-determined in order to use the Box-Behnken Design.

3.3.3.1 Experimental design of ohmic heating treatment

In the study of ohmic heating treatment of the cashew apple juice and pineapple juice, the process parameters such as voltage gradient, treatment temperature, and time are to be optimised. Each process parameter needs to be specified with the ranges of the parameter when a Box-Behnken design is applied to the response surface method. The minimum and maximum values of voltage gradient were selected as 10 and 15 V/cm, treatment temperatures as 50 and 60°C and treatment time as 1 and 5 minutes specifically given as in Table 3.2 based on the preliminary studies conducted and a thorough review of literature. The details of the RSM experimental runs for ohmic heating process are given in Table 3.3.

Table 3.2.Independent variables and its coded and actual values

Independent variables	Units	Code levels		
		-1	0	+1
Voltage gradient (A)	V/cm	10	12.5	15
Treatment temperature (B)	°C	50	55	60
Holding time (C)	min	1	3	5

Table 3.3 Coded and Decoded Levels of Factors used in RSM for Ohmic Heating of Fruit Juices

	Coded factors			Decoded factors		
Treatment Run	A	B	C	Voltage gradient (V/cm)	Treatment temperature (°C)	Holding Time (min)
1	+1	-1	0	15	50	3
2	0	0	0	12.5	55	3
3	0	+1	+1	12.5	60	5
4	0	0	0	12.5	55	3
5	0	0	0	12.5	55	3
6	+1	+1	0	15	60	3
7	0	0	0	12.5	55	3
8	0	-1	+1	12.5	50	5
9	-1	-1	0	10	50	3
10	+1	0	-1	15	55	1
11	0	0	0	12.5	55	3
12	-1	0	-1	10	55	1
13	0	+1	-1	12.5	60	1
14	-1	+1	0	10	60	3
15	-1	0	+1	10	55	5
16	+1	0	+1	15	55	5
17	0	-1	-1	12.5	50	1

The responses obtained from the experimental runs of Box-Behnken Design were modeled by the following polynomial model:

$$Y = b_0 + b_1A + b_2B + b_3C + b_{11}A^2 + b_{22}B^2 + b_{33}\ C^2 + b_{12}AB + b_{13}AC + b_{23}\,BC \qquad 3.5$$

where,

Y = response calculated by the model

A = Voltage gradient, V/cm

B = Treatment temperature, °C

C = Holding time, min

b_0 = intercept

b_1, b_2 and b_3 = linear effects

b_{11}, b_{22} and b_{33} = quadratic effects

b_{12}, b_{13} = interaction coefficient

The mathematical models were evaluated for each response by means of multiple linear regression analysis. The significant terms in model were found by analysis of variance (ANOVA) for each response.

3.3.3.2 Experimental design of pulsed light treatment

The pulsed light treatment need to be optimised with respect to three parameters such as pulsed light dosage (which determines the number of pulses), distance of quartz tube from the pulsed light lamp and the flow rate. Ranges of each parameter (minimum and maximum values), are to be defined when a Box-Behnken design is applied to the response surface method. The minimum and maximum values of pulsed dosages were selected in the range from 8 to 32 J/cm^2. The distance of quartz fluid tube from the pulsed light lamp was selected in the range from 5 to 15 cm (Hillegas and Demirci, 2003). The minimum and maximum flow rates (number of passes) were chosen in the range 150 to 300 ml/min (Preetha *et al.,* 2016 a and b) as specifically given in Table 3.4. The ranges were confirmed through preliminary studies also. The details of the RSM experimental runs for pulsed light treatments are given in Table 3.5.

Table 3.4 Independent variables and their coded and actual values

Independent variables	Units	Code levels		
		-1	0	+1
PL dose (A)	J/cm^2	8	20	32
Sample-source distance (B)	cm	5	10	15
Flow rate (C)	ml/min	150	225	300

Table 3.5 Coded and decoded levels of factors used in RSM for pulsed light treatment of fruit juices

	Coded factors			Decoded factors		
Treatment Run	A	B	C	PL dose (J/cm^2)	Sample-source distance(cm)	Flow rate (ml/min)
1	-1	0	-1	8	10	150
2	+1	0	-1	32	10	150
3	+1	0	+1	32	10	300
4	-1	-1	0	8	5	225
5	-1	+1	0	8	15	225
6	+1	-1	0	32	5	225
7	0	0	0	20	10	225
8	0	0	0	20	10	225
9	-1	0	+1	8	10	300
10	0	0	0	20	10	225
11	0	0	0	20	10	225
12	+1	+1	0	32	15	225
13	0	+1	-1	20	15	150
14	0	+1	+1	20	15	300
15	0	-1	-1	20	5	150
16	0	0	0	20	10	225
17	0	-1	0	20	5	300

The responses obtained from the experimental runs of Box-Behnken Design were modeled by the following polynomial model:

$$Y = b_0 + b_1A + b_2B + b_3C + b_{11}A^2 + b_{22}B^2 + b_{33}\ C^2 + b_{12}AB + b_{13}AC\ + b_{23}\ BC \quad 3.6$$

where,

Y = response calculated by the model

A = Pulsed light dosage J/cm^2

B = Source-sample distance, cm

C = flow rate ml/min

b_0 = intercept

b_1, b_2 and b_3 = linear effects

b_{11}, b_{22} and b_{33} = quadratic effects

b_{12}, b_{13} = interaction coefficient

The mathematical models were evaluated for each response by means of multiple linear regression analysis. The significant terms in model were found by analysis of variance (ANOVA) for each response. The model adequacies were checked by R^2, adjusted R^2 and coefficient of variation (CV). Maximisation and minimisation of the polynomials thus fitted was performed by the desirability function method and mapping of the fitted responses using Design-Expert version 7.0.0 (Statease Inc., Minneapolis, USA). Response surface graphs were generated based on the highest interaction between the variables in order to visualise the relationship between response and experimental levels of independent variables as well as to deduce optimum conditions.

3.3.3.3 Experimental design of ohmic assisted pulsed light treatment.

The experimental design of the ohmic assisted pulsed light treatment were carried out at optimised process variable values of ohmic and pulsed light treatments conducted separately as mentioned in section 3.3.3.1 and 3.3.3.2. For better and rigorous comparison, values of highest desirability next to that of optimised condition were also selected for conducting combined treatment studies.

The ohmic heating treatment operating parameters for pineapple juice:

1. OH_1- Voltage gradient: PA_1; Process temperature: PB_1; Holding time: PC_1
2. OH_2 -Voltage gradient: PA_2; Process temperature: PB_2; Holding time: PC_2

The pulsed light treatment operating parameters for pineapple juice are

1. PL_1- PL dosage: PX_1; Sample-source distance:PY_1; Flow rate: P Z_1
2. PL_2 -PL dosage: PX_2; Sample-source distance: PY_2; Flow rate: P Z_2

The ohmic assisted PL treatment combinations for pineapple juice:

1. P_1 (OH_1PL_1):$PA_1PB_1PC_1$-PX1PY1PZ1
2. P_2 (OH_1PL_2): $PA_1PB_1PC_1$-$PX_2PY_2PZ_2$
3. P_3 (OH_2PL_1): $PA_2PB_2PC_2$-PX1PY1PZ1
4. P_4 (OH_2PL_2): $PA_2PB_2PC_2$- $PX_2PY_2PZ_2$

The ohmic heating treatment operating parameters for cashew apple juice:

- OH_1- Voltage gradient: CA_1; Process temperature: CB_1; Holding time: CC_1
- OH_2- Voltage gradient: CA_2; Process temperature: CB_2; Holding time: CC_2

The pulsed light treatment parameters for cashew apple juice:

- PL_1- PL dosage: CX_1; Sample-source distance:CY_1; Flow rate: CZ_1
- PL_2 -PL dosage: CX_2; Sample-source distance: CY_2; Flow rate: CZ_2

The ohmic assisted PL treatment combinations for cashew apple juice:

1. C_1 (OH_1CL_1):$CA_1CB_1CC_1$-$CX_1CY_1CZ_1$
2. C_2 (OH_1CL_2): $CA_1CB_1CC_1$-$CX_2CY_2CZ_2$
3. C_3 (OH_2CL_1): $CA_2CB_2CC_2$-$CX_1CY_1CZ_1$
4. C_4 (OH_2CL_2): $CA_2CB_2CC_2$- $CX_2CY_2CZ_2$

3.4 EXPERIMENTAL PROCEDURES

The experiments of ohmic heating pulsed light treatment and combined ohmic pulsed light treatments of pineapple and cashew apple juice samples were carried out by adopting the procedures as mentioned below.

3.4.1 Ohmic Heating of the Fruit Juices

The filtered fresh pineapple and cashew apple juices separately, was supplied to the ohmic heating chamber from the feed tank through the connected pipes and valves. Approximately 300 ml of juice could be filled in a batch of ohmic treatme nt. The supplied juice would fill the ohmic heating chamber without any air bubbles between electrodes. The electric supply was put on and the predetermined voltage gradient was set. The system was switched on and the juice samples were heated ohmically from ambient temperature to 60°C by applying three different voltage gradients (10, 15 and 20 V/cm), temperature (50, 55 and 60°C) and holding time (1, 3 and 5 min) by adjusting the voltage supply using the variac. Once the fruit juice reach the required temperature, the sample was maintained at that temperature for different holding times (1, 3, and 5 min) in the ohmic heating chamber for each voltage gradient under study. The system was manually operated by adjusting power on and off in order to maintain the sample at the required temperature for the prescribed holding time. The treated samples were then subjected to pulsed light treatment maintaining aseptic conditions. The treated samples were also collected in amber coloured PET bottles and stored at refrigerated condition (4-8°C) for further analysis.

3.4.2 Pulsed Light Treatment of Fruit Juices

The fresh pineapple juice and cashew apple juice were fed in to the feed tank and were pumped through the connecting pipe in to the quartz tube where the juice is exposed to the pulsed light. The samples were treated by varying the process parameters such as PL dosage and sample-source distance based on the Box Behnken design of response surface methodology as shown in Table. 3.4. The treated samples were collected in the amber coloured PET bottle, stored at 4±2°C, and were used for further studies.

3.4.2.1 Number of passes

Preliminary studies revealed that the residence time in a single pass through the quartz tube (treatment chamber) was found to be less to achieve the required reduction in

the microbial count. Therefore, in this study the number of passes for the fruit juice was fixed as three for the entire experiment.

3.4.2.2 Cleaning of the pulsed light unit

The developed pulsed light unit was cleaned before and after the treatment using standard procedures. The unit was rinsed with warm water at 90°C for 10 minutes followed by alkaline (1.5%) wash for 15 minutes and sterile distilled water wash for 2 min to maintain sterile condition (Keyser *et al.,* 2008).

3.4.3 *Ohmic assisted PL treatment of fruit juices*

In this study, the ohmic and pulsed light treatments were combined in order to achieve a higher microbial reduction with minimum changes in juice quality characteristics as per the hypothesis explained in Chapter I. The optimised process operating parameters of ohmic heating and pulsed light treatments were combned for the effectiveness of fruit juice treatment. For a rigorous comparison, the treatment process parameters with statistically second highest desirability for both ohmic and pulsed light treatments were also analysed to check the effectiveness of combined process. The fruit juice was treated with the optimised process parameters in ohmic heating section at frst stage and then maintaining aseptic conditions and good hygienic practices the ohmic heated fruit juice was treated with the optimised process conditions in pulsed light section in the second stage.

3.5 QUALITY EVALUATION OF THE FRUIT JUICE

Physicochemical properties such as absorbance, turbidity, colour, total soluble solids (TSS), pH, titrable acidity, ascorbic acid content, total sugar, phenolic content, tannin content, antioxidant activities, total colour difference and rheological properties of the fresh pineapple juice and cashew apple juices were analysed.

3.5. 1 Absorbance of Fruit Juices

The absorbance of liquid foods plays an important role in the inactivation of microorganisms during the exposure of UV radiation (Kotchuma *et al.,* 2004). Hence it is essential to determine the absorbance of UV light passing through the fruit juices.

As stated in Bear Lambert's law, the concentration of the solution is in linear relationship with the absorbance of light passing through it. The penetration of UV light is influenced by the absorbance of emitted light (Muller *et al.,* 2011).

The absorbance was measured using a spectrophotometer (UV–VIS spectrophotometer, Systronics, Ahmedabad, India) as depicted in Plate 3.12. Distilled water was selected as the reference sample. The absorbance of blank solution was determined by placing the cuvette with distilled water in the analysis chamber of the spectrophotometer and measuring absorbance at a wavelength of 254 nm. The absorbance of fruit juice samples were then measured at 254 nm.

3.5.2 Turbidity

Turbidity of pineapple and cashew apple juice were obtained by using a UV-VIS spectrophotometer (Systronics, Ahmedabad, India) at a wavelength of 610 nm (AOAC, 2000) (Plate.3.4). The absorbances of the samples were determined in relation to the reference sample (distilled water) as described in section 3.5.1. The transmittance (Tr) and turbidity (Tur) of samples were analysed using the Equation (3.7) and (3.8) respectively.

$$Tr = 100(10^{-Ab}) \quad 3.7$$

$$Tur = 100 - Tr \quad 3.8$$

where, Ab- Absorbance at 610 nm

3.5.3 pH

pH is a measure of active acidity, which influences flavour or palatability of a product and affects processing requirements. The pH value was determined by using a digital pH meter as depicted in Plate. 3.14 (Elico pH meter, Model LI120). The pH meter was standardised with distilled water of pH 7.0 and buffer standards of pH 4, 7, and 9.0. Three different samples of each fruit juice were analysed for pH measurement and the average value was recorded as pH of the specimen (AOAC, 2000).

3.5.4 TSS

The TSS of fruit juices was determined using a digital refractometer (Erma, Italy) (Plate 3.15). A drop of fruit juice was placed on the measuring port of the refractometer to read the value of total soluble solids (TSS) in °Brix (Ranganna, 1991).

3.5.5 Titrable Acidity

Total acidity of fruit juices were determined and expressed in malic acid equivalent percentage (AOAC, 2000). Five millilitres of fruit juice samples were collected in 250 ml conical flask containing 100 ml of distilled water. Few drops of phenolphthalein were added to the solution as an indicator and shaken well. The burette was filled with 0.1 N NaOH. The solution was titrated against the solution in burette until the sample solution showed a faintest discernible pink colour which persisted for 30 seconds. Acidity was estimated using the following equation:

$$Acidity\ (\%\ malic\ acid) = \frac{volume\ of\ titrant\ (ml)\ \times\ Normality\ of\ titrant\ \times\ 0.067}{Sample\ weight\ (g)}\ x\ 100 \quad 3.9$$

where, 0.067 - milli equivalent of malic acid

3.5.6 Ascorbic Acid

Ascorbic acid content in fruit juices were estimated using the 2, 6-dichlorophenol indophenols titrimetric method as described by Sadasivam and Manickam (1992). Dye solution was prepared by dissolving 52 mg of 2, 6 dichloro phenol indophenols, and 42 mg of sodium bicarbonate in 200 ml distilled water. Standard solution was prepared by adding 100 mg of ascorbic acid to 100 ml of 4% oxalic acid. To prepare working standard solution, 10 ml of standard solution was pipetted out and was diluted to 100 ml using 4% oxalic acid. The 5 ml fruit juice samples were made up to 50 ml using 4 percent oxalic acid. To find dye factor, 10 ml of working standard solution was pipetted out into a 50 ml conical flask and 10 ml of 4% oxalic acid was added and titrated against the dye. The end point was the appearance of pink colour which persisted for a few minutes. The

titration was repeated to get concordant values. The amount of dye consumed was equal to the amount of ascorbic acid present in the working standard solution (V_1). Ten millilitre of sample extract was pipetted out to which 10 ml of 4% oxalic acid was added. It was then titrated against the dye. The titration was replicated for each sample until the concordant values were obtained (V_2).

$$\text{Dye factor} = \frac{0.5}{\text{Titrable value}(V_1)} \quad 3.10$$

$$\text{Ascorbic acid}\frac{\text{mg}}{100\text{g}} = \frac{0.5\text{mg}}{V_1\text{ml}} \text{x} \frac{V_2}{5\text{ ml}} \text{x} \frac{100\ ml}{\text{Wt. of the sample}} \text{x}100 \quad 3.11$$

V_1 - Amount of dye consumed by ascorbic acid present in the working standard solution, ml.

V_2 - Amount of dye consumed by the liquid sample, ml.

3.5.7 Total Sugar

The total sugar of the fruit juices was determined according to Ranganna *et al.* (1991). To prepare the glucose standard curve, the stock standard was prepared by dissolving 100mg of glucose in 100 ml of distilled water. The working standard solution was prepared by diluting 10 ml of stock solution in 100 ml of distilled water so that the final concentration become 100 µg/ml. Different concentrations of 0, 0.2, 0.4, 0.6 and 0.8 ml of working standard was pipetted out in test tubes and was made up to 1ml using distilled water. Five millilitre of anthrone reagent was added into each test tube. The contents in the test tubes were heated for 8 minutes in a boiling water bath. These were cooled rapidly and kept in dark room and the absorbance was measured using spectrophotometer at 620 nm. A standard graph was drawn by plotting concentration of the standard vs. absorbance.

One millilitre of fruit juice was diluted with 5 ml of the 80% ethanol. The supernatant was collected and evaporated to dryness in the water bath at 80°C. The residue was then diluted by adding 10 ml distilled water to dissolve the sugars. Half millilitre of aliquots was pipetted out in to test tubes and made up to 1 ml in each test tube. Then 5 ml of anthrone reagent was added to the test tubes containing juice samples.

The content in the test tube was heated for 8 minutes in a boiling water bath. The test tubes were allowed cool in a dark room. The absorbance was measured at 620 nm. The glucose content with respect to the absorbance was determined from glucose standard curve.

3.5.8 Total Phenolic Content

Total phenolic content of pineapple and cashew apple juice was determined by Folin-ciocalteu method as described by Sadasivam and Manickam (1992). The production of blue coloured complex during the reaction of phenols with phosphomolibdinic acid in presence of Folin-ciocalteu reagent in alkaline medium is the basis of the estimation. One millilitre of fruit juice samples were diluted with 10 times volume of 80% ethanol and then centrifuged at 10000 rpm for 20 minutes. The supernatants were collected and re-extracted with five times volume of 80% ethanol. The supernatants were collected and allowed to evaporate to dryness. The residue was collected and diluted to 5 ml using distilled water. About 0.2 ml aliquots of cashew apple juice and 1 ml aliquots of pineapple juice were pipetted out in to the test tubes and the volume was made up to 3 ml using distilled water. The standard curve was plotted with different concentration of gallic acid (0.2 -2 ml). The gallic acid was diluted to 3 ml in all test tubes with distilled water. Half millilitre of Folin-ciocalteu reagent was added to all test tubes and after 3 minutes 2 ml of 20% sodium carbonate was added to each tube. The test tubes were shaken thoroughly and placed in boiling water bath for exactly one minute. The test tubes were then allowed to cool and absorbance was measured at 650 nm against blank as reagent.

3.5.9 Tannin Content

The tannin content in fresh and clarified cashew apple juice was estimated by the Folin-Denis method as explained by Sadasivam and Manickam (1992). One millilitre of cashew apple juice was taken in a conical flask containing 75 ml of distilled water. The solution was boiled for 30 minutes. Boiled samples were centrifuged at 20,000 rpm for 20 minutes. The supernatants were collected in a 100 ml standard flask and made up the volume.

One millilitre of sample extracts was pipetted out in to 100 ml volumetric flask containing 75 ml distilled water. Five millilitre of Folin-Denis reagent and 10 ml of sodium carbonate solution were added and volume was made up to 100 ml by adding distilled water. The standard flasks were shaken well and absorbance was measured at 700 nm after 30 minutes. A standard curve of tannic acid was plotted with different concentration of tannic acid from 0.2 -2 ml at interval of 0.2 ml. For each concentration, the volume was made up to100 ml by following the procedure for sample extracts and the absorbance was measured at 700 nm. The tannin content of the sample was measured as the tannic acid equivalent.

3.5.10 Total Colour Differenc

Colour flex meter (Model: 45°/0°, M/s Hunter Lab, Reston, Virginia, USA) was used for measurement of colour (Plate 3.16). It works on the principle of collecting the light and measuring the energy from the sample reflected across the entire visible spectrum. The meter uses filters and mathematical models rely on "standard observer curves" that define quantity of red, green, and blue primary lights required to match a series of colours across visible spectrum. The mathematical model used is called Hunter model. It provides reading in terms of L*, a*, and b*. Where L* indicates whiteness and darkness. Chromatic portion of the solids is defined by: +a (red), -a (green), +b (yellow), and -b (blue). These data may be sensed using the sensor (colour flex 45°/0°) and it is supported with Universal software V 4.10 package. It produces functions that define hue and chroma. ΔL, Δa and Δb value represents the deviations of the individual values of treated juices as compared to fresh samples and the total colour difference (TCD) are estimated by Equation (3.12). Colour meter is calibrated by fixing the defined colours like white and black tiles on the colour flex meter. Calibration is performed through necessary changes in sample. Measurement of colour was done as per above principle by determining L*, a* and b* values.

$$TCD = \sqrt{(L^* - L_0)^2 + (a^* - a_0)^2 + (b^* - b_0)^2} \qquad 3.12$$

where, L_0, a_0, b_0 are initial colour values.

3.5.10.1 Yellowness Index

Yellowness index (YI) indicates the degree of yellowness. Yellowness is one of the important colour characterestics in the case of fruit juices. This value represents the colour variation due to scorching, soiling, and general product degradation by light, chemical exposure and processing. Yellowness indices are used mainly to quantify these types of degradation with a single value. They can be used when measuring clear, near-colourless liquids or fruit juices. The yellowness index is estimated from the L* and b* values using Equation (3.13).

$$\text{Yellowness Index} = \frac{142.86\ b*}{L*} \qquad 3.13$$

3.5.11 Measurement of Anti-oxidant Activity

A spectrophotometer was used to measure the antioxidant capacity of the fruit juices. A stock solution of DPPH (2, 2-Diphenyl-1-picrylhydrazyl) was prepared by dissolving approximately 15 mg DPPH in 100 ml methanol and stored at −20ºC until further use. The working solution was prepared by mixing 10 ml of stock solution with 45 ml of methanol to adjust absorbance at 517 nm wavelength to unity, which was then kept in dark. The change in colour of the DPPH solution from purple to yellow, resulting from the addition of different quantities of ascorbic acid standards and ethanolic extract of juices (20 to 200 µl) was measured at 517 nm after allowing the solution to stand in the dark for 20 min. The decrease in absorbance of DPPH after 20 min was calculated and expressed as mg of ascorbic acid equivalents antioxidant capacity per 100 g. The control sample was prepared as above without any sample extract and methanol was used for the baseline correction. The inhibition rate is calculated by using the Equation 3.13(Nazir *et al.*, 2013).

$$\text{Percentage radical scavenging activity (\%)} = \frac{Absorbance\ of\ control - Absorbance\ of\ sample}{Absorbance\ of\ control} \qquad 3.14$$

Plate 3.13 Spectrophotometer

Plate 3.14 pH meter

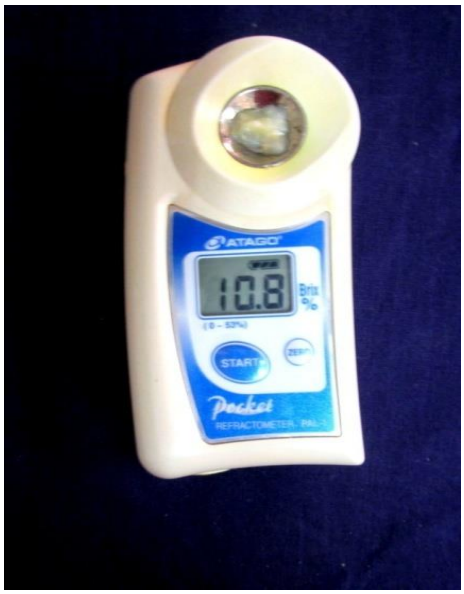


Plate 3.15 Refractometer

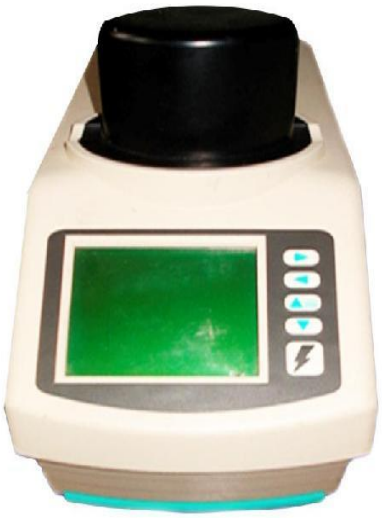


Plate 3.16 Colour flex meter

3.5.12 Non Enzymatic Browning Index

The browning index was measured using the method described by Meydav *et al.* (1977) supplemented by others (Fustier *et al.*, 2011 and Guerrouj *et al.*, 2016). Ten millilitre of juice sample was centrifuged at 3000 rpm for 10 min in order to remove coarse particles from the sample. Five millilitre of supernatant was collected and added with 5 ml of ethyl alcohol and again centrifuged. A UV spectrophotometer (UV-1800, SHIMADZU) (Plate 3.13) was used to read the absorbance of the supernatant at 420 nm. Distilled water was used as blank. Measurements were taken in triplicate and mean values were reported

3.5.13 Minerals

Minerals in the liquid foods were estimated using the Atomic Absorption Spectrophotometer (AAS) (Plate 3.17) as per AOAC (2000). Five hundred milligram of sample was taken in a pyrex flant bottomed flask and digested with a mixture of concentrated nitric acid, sulphuric acid and perchloric acid (9:2:1 v/v). Initial digestion was carried out in a cold state and then digested over sand both until an ashy white digest was obtained. The digest was filtered and made up to a known volume. The triple acid aliquot was used for the estimation of minerals.

Flame atomic absorption technique was used for analysis of minerals. The liquid is injected in to the inlet of the AAS. The liquid samples were aspirated, aerosolized, and mixed with combustible gases such as acetylene and nitrous oxide. The mixture was ignited in a flame whose temperature ranges from 2100 to 2800˚C. During combustion, atoms of the element of sample were reduced to free unexcited state atoms, which absorb light at characteristic wavelength. The amount of light absorbed would be measured against a standard curve. The minerals such as calcium, potassium, and sodium were determined by comparing the atomic spectroscopic signal for each with that for standard solution of same ion.

3.6 RHEOLOGICAL PROPERTIES OF FRUIT JUICE SAMPLES

The rheological behavior of the fresh and treated fruit juices were analysed in a rheometer (Plate 3.18). (Physica MCR 52, Anton Paar, GMBH, Ostfildern, Germany). The PP50 probe (Plate and plate geometry with 50 mm diameter) was used for the flow curve measurements. Test gap for the sample was fixed at 1.0 mm. The shear rate tests were conducted over a shear rate range of 1-100 s^{-1}. The data was recorded at 30±3°C. The shear stress and viscosity changes were recorded. The data of all rheological measurements were analysed with supporting software Rheoplus /322 v 2.81 (Anton Paar). A sample volume of approximately 2 ml of fruit juice samples were placed in between the plates and the samples were subjected to a programmed shear rate linearly increasing from 1 to 100 s^{-1}. The experimental data were fitted to different rheological models *viz.* Herschel bulkley and Ostwald model as shown below.

3.6.1 Herschel Bulkley Model

$$\tau = \tau_0 + K\gamma^n \qquad 3.15$$

where, τ - Shear stress (Pa)

τ_0 - Yield stress (Pa)

k - Consistency coefficient (Pa s^n)

γ- Shear rate (s^{-1})

n - F low behavior index

3.6.2 Ostwald Model

The Ostwald model (Equation 3.15), also known as the Power Law model, is applied to shear thinning fluids which do not present a yield stress (Pevere *et al.*, 2006). The n-value gives fluid behaviour information according to:

$$\tau = K\gamma^{n-1} \qquad 3.16$$

n < 1 → Pseudoplastic behaviour

n = 1 → Newtonian behaviour

n > 1 → Dilatant behavior

Plate 3.17 Atomic Absorption Spectrophotometer

Plate 3.18 Antonpar Rheometer

3.7 MICROBIOLOGICAL ANALYSIS

The microbiological quality characteristics of the pineapple and cashew apple juice samples were determined both for fresh and treated samples. The growth of bacteria, yeast, and mould were found through standard plate count method. The PL (Pulsed light) treated, ohmic heated and ohmic assisted PL treated pineapple and cashew apple juice samples were analysed for the growth of microorganisms for a period of 15, 20 and 25 days respectively at an interval of 5 days for each treatment.

3.7. 1 Enumeration of the Total Bacterial Count and Yeast and Mould Count in Fruit Juices

The bacterial and yeast and mould population in fruit juices were analysed by different microbiological methodologies, that includes enumeration of the micro organism in selective media for different dilutions of samples, incubation of plates and counting the number of colonies present. The media generally used for enumeration of bacteria is nutrient agar medium, whereas, for yeast and mould enumeration chloramphenicol yeast glucose agar media was used (Allen, 1953). The fruit juice sample of 1 ml was pipetted using a sterile pipette into a test tube containing 9 ml of sterile water which gave a 1:10 (10^{-1}) dilution. The test tubes were shaken well for 10-15 minutes for uniform distribution of microbial cell in the water blank. Then 10^{-2} dilution was prepared by pippetting out 1 ml of (10^{-1}) dilution to 9 ml of sterile water in test tube with a sterile one ml pipette. The process was repeated up to 10^{-6} dilutions with the serial transfer of the dilutants. One millilitre of aliquots from 10^{-1} and 10^{-6} dilutions were transferred to the sterile petri dishes for the enumeration of bacteria and one millilitre aliquots from 10^{-1} and 10^{-3} dilutions were transferred to the sterile petri dishes for the enumeration of yeast and mould. For ohmic heated samples, dilutions of 10^{-2} and 10^{-3} were selected for enumeration, whereas for pulsed light samples, dilutions of 10^{-4} and 10^{-5} were selected for enumeration of bacterial colonies. The dilutions were so selected based on the preliminary studies conducted and a thorough review of literature. Since the ohmic treatment is a mild thermal treatment, the chance of survival of microorganisms is limited

and therefore higher dilutions can be omitted. The experiments were carried out in triplicate and the mean value is reported.

Approximately, 15-20 ml of molten and cooled (45ºC) respective agar medium was added to each petridish containing the sample dilutions and the plates were rotated in clockwise and anticlockwise direction for thorough mixing of the dilutants and the medium. The plates were then incubated at 35°C (room temperature) for 24-48 hours for bacteria and at 20 to 25°C for two to four days for yeast and mould, respectively. After the incubation period, the colonies were counted and the number of organisms (total bacteria and yeast and mould) per gram of sample was calculated by using the Equation (3.16).

$$Number\ of\ coloby\ forming\ units(cfu)per\ gram\ of\ the\ sample = \frac{\text{Mean number of cfu x Dilution factor}}{Quantity\ of\ sample\ on\ weight\ basis} \quad 3.17$$

3.8 QUALITY EVALUATION OF OHMIC HEATED FRUIT JUICES

The ohmic heated pineapple and cashew apple juice samples were analyesd for various physiochemical properties such as pH, TSS, titrable acidity, ascorbic acid, total sugars, total phenols, tannin content and antioxidant activities, total colour difference and rheological properties as described in section 3.5. The microbiological analyses were carried out according to the methodologies explained under section 3.7.

3.9 QUALITY EVALUATION OF PULSED LIGHT TREATED FRUIT JUICES

The pulsed light treated pineapple and cashew apple juice samples were analyesed for various physiochemical properties such as pH, TSS, titrable acidity, ascorbic acid, total sugars, total phenols, tannin content and antioxidant activities, total colour difference and rheological properties as described in section 3.5. The microbiological analyses were carried out according to the methodologies explained under section 3.7.

3.10 QUALITY EVALUATION OF OHMIC ASSISTED PL TREATED FRUIT JUICES

The physico-chemical characteristics of combined ohmic and pulsed light treated fruit juices such as pH, TSS, titrable acidity, ascorbic acid, total sugars, total phenols, tannin content, antioxidant activities, browning index (Section 3.9.1) and total colour difference, rheological characteristics, and microbiological characteristics were evaluated by the methodologies as explained under section 3.5, 3.6 and 3.7. These characteristics were then compared with that of standardised ohmic treated and pulsed light treated samples. A comparison of quality parameters of the combined treated fruit juices with that of thermally processed fruit juices were also carried out.

3.11 STORAGE STUDIES

Storage studies of control (fresh juice) and optimised PL treated, ohmic processed and combined ohmic and PL treated fruit juices bottled in amber coloured PET bottles, and stored at refrigerated temperature (4±2°C) were conducted for a period of 35 days. The changes in the physical, biochemical and microbial qualities of optimised fruit juice samples were analysed (as explained under section 3.5 and 3.7) at regular intervals of 5 days. All the experiments were carried out in triplicate and the mean values were reported.

3.12 SENSORY ANALYSIS

Sensory analysis is a scientific study used to measure, analyse and interpret reactions to those characteristics of foods and materials as they are perceived by the senses of sight, smell, taste, touch and hearing. In general, sensory quality of liquid foods is the consumer's reaction to the physical nature and chemical constituents of the food in its prepared and formulated form. Organoleptic evaluation of the product was carried out by a panel of twenty untrained judges for colour, flavour, taste and overall acceptability using 9 point hedonic scale (Ranganna, 1995). Comparison was made with treated and fresh samples after 20 days of storage. The score card model is given in appendix A.36.

3.12.1 Fuzzy Logic

The sensory analysis performed subjectively was ranked based on its attribute preference mathematically using fuzzy comprehensive model (Sana *et al.*, 2016). The sensory preference score given by judges were taken and their linguistic judgment was converted to numerical ranking using fuzzy model. The attributes were assigned with respective values based on the preference given by sensory panels. Scores assigned for the pineapple and cashew apple juice were colour and appearance – 0.27, flavour – 0.26, taste - 0.24 and overall acceptability – 0.23. The sensory analysis using fuzzy logic involved formation of three sets.

Table 3.6 Different sets involved in sensory analysis using fuzzy logic

I.	Factor set (F_f)	Quality attributes of pineapple and cashew apple juice (colour and appearance, taste, flavour and overall acceptability)
II.	Evaluation set (E_f)	Scale factors for quality attributes (Excellent (EX), Good (GD), Medium (MD), Fair (FR) and Not Satisfactory (NS))
III.	Transformation set (T_f)	Numerical values for the evaluation set (EX =1, GD = 0.9, MD = 0.7, FR = 0.4, NS = 0.1)

The fuzzy model for sensory analysis was done through the membership functions as represented below:

Fuzzy Membership Function (FMF) - Value obtained by dividing the added individual scale factors with total number of judges.

Normalised Fuzzy Membership Function (NFMF) - It is the function obtained by multiplying FMF and scale factor allotted to respective membership function.

Normalised Fuzzy Membership Function Matrix – It is the matrix formulated by adding NFMF with its respective scale factors.

Judgment Membership Function Matrix (JMFM) – This would be deciding matrix for ranking. It could be obtained by adding the column values of all matrix and divide with highest total column value.

Judgment Subset (JS) – It is the final ranking of samples evaluated along with attributes preference of judges.

3.13 MICROBIAL INACTIVATION STUDIES

Water used in food production has been recognized as a vector for the transmission of pathogenic *Escherichia coli*. It is a non-spore forming bacterium that has been implicated as the causative agent in numerous food borne outbreaks of contaminated, non-pasteurised fruit juices. The bacteria *Listeria monocytogenes* also found to be the causative organism for numerous food born outbreaks (EFSA, 2015).

In order that the fruit juice disinfection to be accurate and complete, information about the inactivation behavior of pathogenic micro-organisms causing food borne diseases is inevitable.

In this research, the microbial inactivation studies were conducted to test the effectiveness of ohmic heating assisted pulsed light treatments of pineapple and cashew apple juices subjected to standardised operational parameters. The detailed procedure of inoculation and enumeration of the *Escherichia coli* and *Listeria monocytogenes* in the fruit juice system before and after treatment is detailed below.

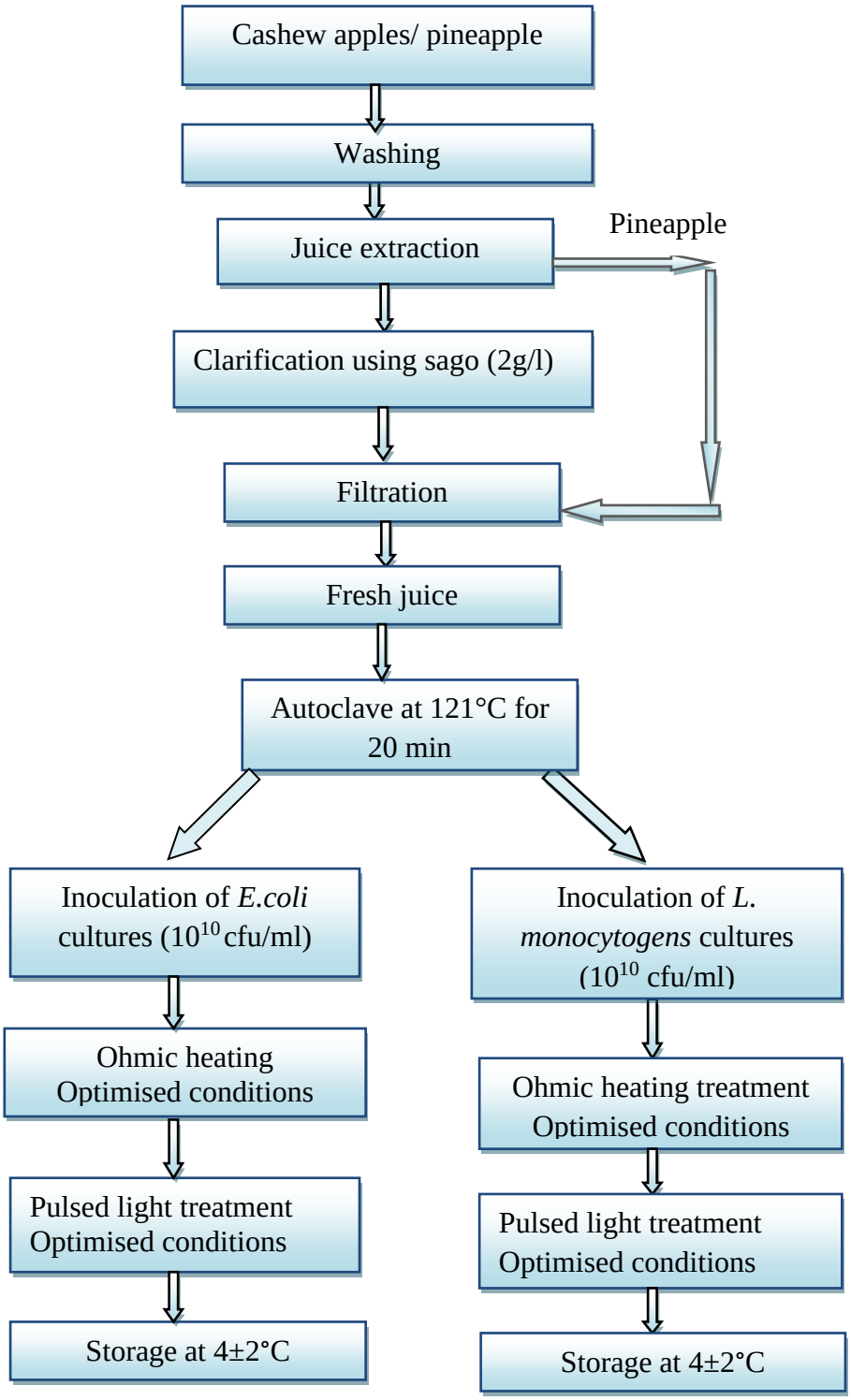


Fig. 3.1 Flowchart of inoculation studies of *Lsteria monocytogensis* and *Escherichia coli* strains in pineapple and cashew apple juices

3.13.1 Preparation of Inoculum

The gram negative bacteria *Eschericha coli* MTCC 433 as well as the gram positive bacteria *Listeria monocytogenes* 839 were used as model organisms in this study. Both strains were collected from Microbial Type Culture Collection and Gene Bank (MTCC), Chandigarh, India. Pure cultures of each strain were supplied in freeze-dried form and stored under 0°C until the start of the experiment.

The *Escherichia coli* and *Listeria monocytogens* Strains were initially grown in 100 ml of Luria-bertaini (Sambrook *et al.,* 1989) and Fraser broth containing amphicillin (100 μg/ml) respectively, at 37°C for 16–18 h in a shaking bath. Luria-bertani and Fraser agar plates were subsequently inoculated with the respective bacterial suspension using an inoculating loop. These plates were incubated for 24 h at 37°C and stored at 5°C as stock culture. Working cultures were made by inoculating 1000 ml of Luria-bertani and Fraser broth supplemented with ampicillin (100 μg/ml), with cell material from the agar surface and following incubation for 16–18 h at 37°C in a shaking bath until early stationary phase (Ferrario *et al.,* 2015).

The required bacterial cells alone were separated from the respective medium by consecutive pelletisation and cleaning methodologies. The bacterial cells were pelletised by centrifugation at 5000 rpm for 20 min at room temperature and cell pellets were re-suspended in 100 ml of phosphate buffered saline (PBS) and centrifuged.

The final cell count was determined using McFarland standards as reported by Zapata and Ramirez-Arcos (2015). McFarland Standards are employed in standardisation of the approximate number of bacteria in a liquid suspension by comparing the turbidity of the test suspension with that of the standard. McFarland standards of 10 were used as a reference to adjust the microbial cell concentration to the range of 10^{10} based on the turbidity of bacterial cell suspension. The turbidity of test suspension was compared with that of the McFarland standard by employing a spectrophotometer. The turbidity of cell suspensions were adjusted with addition or removal of the cell pellets.

The fresh juices of cashew apple juice and pineapple juice were autoclaved at 121°C at 1.5 kg/cm^2 for 20 min (Awua *et al.,* 2011). The sterilised juice samples were inoculated with *Escherichia coli* and *Listeria monocytogens* of 10^{10} cfu/ml by adding required amount directly in to the fruit juices (Ngadi *et al.,* 2003). The process flow chart of inoculation study of the ohmic assisted pulsed light treated pineapple and cashew apple juices is shown in Fig. 3.1.

3.13.2 Enumeration of Bacteria

The fruit juices inoculated with *Escharichia coli* and *Listeria monocytogens* strains were treated with combined ohmic and PL treatment at the optimised treatment condition arrived at ohmic heating process parameters such as voltage gradient of 14.02 V/cm, holding time of 2.31 min and treatment temperature of 55.26°C and pulsed light treatment parameters *viz.* PL dosage of 13.69 J/cm^2, sample-source distance of 10.26 cm and flow rate of 165.06 ml/min for pineapple juice. Whereas in cashew apple juice, optimum ohmic assisted PL treatment conditions arrived at ohmic heating operating parameters *viz*. voltage gradient of 14.53 V/cm, process temperature of 55.25°C with holding time of 2.77 min and pulsed light operating parameters such as PL dosage of 12.49 J/cm^2, sample-source distance of 8.63 cm with a flow rate of 164.01 ml/min. The treated juice samples were immediately pour plated in respective media to determine the total log reduction in microorganism. About 1 ml of fruit juice sample was taken immediately after treatment and pipetted into a test tube containing 9 ml of sterile water. The test tubes were shaken well for 10-15 minutes for uniform distribution of microbial cell in the water blank. This gave a dilution of 1:10 (10^{-1}). One ml from (10^{-1}) dilution was transferred to 9 ml of sterile water with a sterile one ml pipette, which gave a dilution of 10^{-2}. The process was repeated up to 10^{-4} dilutions with the serial transfer of the dilutants. One ml aliquots from 10^{-1}, 10^{-2} and 10^{-3} dilutions were transferred to the sterile petriplates. The *Escherichia coli* and *Listeria monocytogens* were enumerated in Maconckey agar and Fraser agar respectively. The 15-20 ml of selective media (45°C) was added in to the respective petriplates and was incubated at 37°C for 48 h. Colonies

on the plates were enumerated and colony counts in 1 g or 1 ml of samples were counted using a digital colony counter and expressed as cfu/ml.

3.14 STATISTICAL ANALYSIS

The statistical analysis for optimising the process variables of ohmic heating system and pulsed light treatment system was performed through Response Surface Methodology (RSM) as explained in section 3.3 using Design Expert (Version 7.0.; Stat-Ease Inc., Minneapolis, MN, USA) software. Both the variables and responses were fitted to the model by performing the analysis of variance (ANOVA). The effect of independent parameters on the dependent parameters of fruit juices and their interactions was analysed to determine the significance of the model. The optimised process parameters of ohmic treatment, pulsed light treatment and combined treatment were chosen for conducting the shelf life studies and fresh fruit juices were taken as control. Statistical analysis of the shelf life study was carried out using Duncan's multiple range tests (DMRT) employing SPSS software version 16.0 (Berney *et al.*, 2006).

Results & Discussion

CHAPTER IV

RESULTS AND DISCUSSION

This chapter details in brief the developed system for ohmic assisted pulsed light treatment for pumpable fruit juices. The results of the experiments conducted towards optimisation of the process parameters of ohmic heating process, pulsed light treatment, and combined ohmic and pulsed light treatment of pineapple and cashew apple juice are also elaborated. The chapter also discusses the results of the experiments conducted towards the characterisation of the optimally treated pineapple and cashew apple juices and their storage studies. The results of the microbial inactivation studies to determine the effectiveness of the combined treatment is also discussed in detail.

4.1 DEVELOPMENT OF OHMIC HEATING ASSISTED PULSED LIGHT TREATMENT SYSTEM

In order to test the hypothesis of the study as explained in chapter I, an ohmic heating assisted pulsed light treatment system suitable for pumpable fruit juice was fabricated. The fruit juices were subjected to ohmic heating process followed by pulsed light treatment in the developed system to evaluate the effectiveness of the system.

In the first phase of treatment, the fruit juices were subjected to mild thermal processing in ohmic heating chamber. During the ohmic heating process, fruit juices were sandwiched between the stainless steel electrodes and exposed to heating during the passage of electricity. As detailed in section 3.1.1 the main components of the ohmic heating system were feed tank, ohmic heating chamber and associated valves, regulators and switches and measuring instruments for parameters such as temperature, voltage, current etc.

The second part, the pulsed light treatment system, is a non thermal processing technology wherein, the fruit juices were subjected to high intense short pulses of Xenon flash lamp to achieve the recommended microbial destruction. The fruit juices were circulated through the quartz tube, which is exposed to the PL irradiation inside the treatment chamber. The system is attached into instrumentations and associated

electronic and electrical components and their circuits for controlling flow rate, distance of xenon flash lamp (source) from the quartz tube and pulsed light dosage. The components of the developed pulsed light treatment system are xenon flash lamp, relay, resistors, rectifier, capacitors, micro processors, trigger transformer, thyristor and timer relay etc. as detailed in section 3.1.3.

In general, the developed system could be widely applicable to all pumpable fruit juices, the biomaterial present in each fruit juice behaves differently with the varying process parameters of the system. Thus it is essential to validate the process parameters of the developed system towards the unique fruit juices. This study aims at standardising the operational parameters of two fruit juices such as pineapple and cashew apple for the reasons as stated at earlier.

4.2 CHARACTERISATION OF THE FRESH FRUIT JUICES

In order to assess the effectiveness of the ohmic heating, pulsed light and combined treatment, the changes in quality attributes of the fresh juices needs to be assessed. The characterisation of fruit juices in the form of optical, biochemical, rheological and microbiological properties were determined. The properties of the fresh pineapple and clarified cashew apple juices are presented in Table 4.1.

Table 4.1 Properties of the fresh pineapple and cashew apple juice

Properties	Pineapple juice	Cashew apple juice
Optical properties		
Absorbance (cm^{-1})	0.43±0.01	0.07±0.0018
Transmittance (%)	31.10±0.31	94.9±0.92
Turbidity (NTU)	553.74±2.80	154.54±0.91
Physico-chemical properties		
pH	4.45±0.04	4.32±0.11
TSS	10.82±0.28	7.73±0.33
Titrable acidity (mg /100 ml)	0.382±0..002	0.422±0.01
Ascorbic acid (mg /100 ml)	31.18±0.59	168.46±1.55
Total sugar (mg /100 ml)	10.53±0.27	9.96±0.35
Total phenols (mg /100 ml)	67.25±0.76	169.46±1.02
Tannin content (fresh %)		0.96±0.04
Tannnin content (clarified %)		0.57±0.01
Antioxidant activity (%)	75.36±0.92	82.66±1.03
Electrical conductivity (Sm^{-1})	0.23±0.03	0.54±0.08
Colour values		
L*	39.86±0.71	1.56±0.05
a*	-3.75±0.13	0.28±0.007
b*	22.93±0.15	-0.94±0.03
Minerals (mg/100 ml)		
Total mineral content (%)	26.56±1.43	42.64±1.65
Calcium	10.53±0.22	5.05±0.22
Pottassium	123.78±1.67	65.34±1.10
Sodium	19.34±0.18	17.68±0.25
Initial microbial population		
Total bacterial count (cfu/ml)	6.52×10^5	7.05×10^5
Yeast and mould (cfu/ml)	5.32×10^4	5.47×10^4

Cashew apple juice showed an absorbance value of 0.07±0.0018 and pineapple juice depicted a higher absorbance value of 0.43±0.01. Higher absorbance in pineapple juice may be due to the scattering of light by the suspended particles present in the fruit juice. Qualls and Johnson (1983) reported that the presence of suspended solids negatively impact disinfection efficacy due to additional absorbance, scattering and or blocking of UV-C light. The absorbance is a key factor to be considered when assessing the effect of UV light on microbial inactivation in a liquid system (Christensen and Linden, 2003; Keklik and Demirci, 2014; Sauer and Moraru, 2009; Unluturk *et al.*, 2004). Transmittance, an optical characteristic inversely proportional to absorbance, was 31.10±0.31% for pineapple juice. Whereas, in clarified cashew apple juice it was 94.9±0.92%. This is because cashew apple juice had lower suspended solid particles and therefore is more transparent. Presence of suspended particles in pineapple juice resulted in higher turbidity value of 553.74±2.80 NTU whereas, in cashew apple juice, it was only 154.54±0.91NTU.

The pH of the pineapple and cashew apple juice were 4.45±0.04 and 4.32±0.11 respectively. Both juices have a medium acidic pH value as they are comparatively rich in organic acids (Tasnim *et al.*, 2010). Since, the minimum growth pH of most of the microorganisms is 4.2, both juices are found to be a susceptible for microbial growth (Meng *et al.*, 2001).

Apart from the kind of sugar and other solid fraction, the type of cultivar and maturity of fruit also are the determining factors of TSS. It is an important factor that contributes to the typical flavour of juice (Matthews, 1994). The TSS content of pineapple juice and cashew apple juice were 10.82±0.28 and 7.73±0.33°Brix respectively. The penetration of the light into the juice mainly depends on the absorbance, turbidity, and total soluble solids. The higher soluble solids will tend to have lower penetration (Keyser *et al.*, 2008).

The titrable acidity values of juices revealed that pineapple juice showed a lower titrable acidity. This could be due to lower organic acid levels in pineapple juice compared to cashew apple juice. The two major acids present in pineapple juice are citric

and malic acids representing 60% and 36%, respectively (Saradhuldhat *et al.,* 2007). Whereas, the cashew apple juice contains organic acids such as acetic acid, oxalic acid, citric acid, tartaric acid and fumaric acid in comparatively higher amounts (Adou *et al.*, 2012).

The fresh cashew apple registered an ascorbic acid content of 168.46 mg/100 ml, whereas pineapple juice showed an ascorbic acid content of 31.18 mg/100 ml indicating approximately 6 times higher vitamin C content in cashew apple juice. A higher antioxidant activity of 82.66% was found in cashew apple juice compared to that of 75.36% in pineapple juice. The antioxidant activities of both juices are due to the inherent vitamin C, polyphenolic compounds, and other bioactive compounds in them (Ferreira *et al.*, 2016).

Table 4.1, showed that the fresh pineapple and cashew apple juice contain 10.53 and 9.96 mg of total sugars respectively, per 100 ml. Fructose, sucrose, and glucose are the major sugars present in both juices (Cordenunsi *et al.*, 2010). The higher sugar content results in the decrease of electrical conductivities of juices having similar concentrations, which may impart changes in the ohmic heating behavior of juices (Icier, 2003).

The phenolic contents of pineapple and cashew apple juices are depicted in Table 4.1. Cashew apple registered a higher polyphenolic content of 168.46 mg /100 ml in comparison to 67.25 mg/ 100 ml in pineapple juice. This could be due to bioactive phenolic compounds such as quercetin, naringenin, and phenolic acids (caffeic acid, tannic acid, p-coumaric acid, ferulic acid, gallic acid) in cashew apple juice (Marc *et al.*, 2012).

Tannin, a phenolic compound is responsible for the astringent flavour of cashew apple juice which makes the juice undesirable in spite of its high nutritional benefits as a RTS beverage. Therefore, the astringent causing tannin needs to be removed through a clarification process. The tannin content of fresh cashew apple juice and the clarified juice are also presented in Table 4.1. A 46% reduction in tannin content was observed after the clarification using cassava starch. Jayalekshmy and John (2004) and Talasila *et*

al. (2012) have reported similar reduction in tannin using sago as clarifying agent. The removal of tannin from juice could be attributed to the process of flocculation which is accelerated by the higher affinity of starch to tannin compounds. In this work the clarified cashew apple juice was used for all experimental studies.

The ohmic heating process highly depends on the electrical conductivity of food under treatment. The cashew apple juice showed higher electrical conductivity (0.54 Sm^{-1}) when compared to 0.23 Sm^{-1} for pineapple juice. Though ohmic heating process is the result of inherent resistance of the food, the heating process initiates only once the food material conducts the electric current to pass through the food system permitting the resistance to take effect. Therefore, an increase in electrical conductivity within certain limits has a positive influence on ohmic heating process. The electrical conductivity of the fruit juices can be considered as a function of soluble solids, sugar, and pH. Higher soluble solids in juices result in lower electrical conductivity values during ohmic heating (Castro *et al.*, 2003; Icier and Ilicali, 2005). The higher electrical conductivity of cashew apple might be due to its higher mineral contents and lower soluble solids (Lamsal and Jindal, 2014).

Colour of juice is a key factor that influences consumer sensory acceptance. For liquid foods, colour is typically measured by the colour parameter such as 'L*' indicating lightness,'a*' indicating chromomaticity on green (-) to red (+) axis and 'b*' indicating chromomaticity on blue (-) to yellow (+) axis (Chutintrasria and Noomhorm, 2007). The L*, a* and b* values of pineapple and cashew apple juice were 39.86 and 1.56, -3.75 and 0.28 and 22.93 and -0.94 for respectively. The a* and b* values of cashew apple juice were recorded to be less compared to pineapple juice. This might be due to translucent and clear nature of clarified cashew apple juice. The higher b* value represents yellowish colour in pineapple.

Cashew apple juice had higher mineral content with a total mineral content of 42% compared to 26% in pineapple juice. Both juices are found to be rich sources of minerals with potassium in higher amounts. The cashew apple juice was found to possess a high total mineral content compared to pineapple juice, which might be due to the presence of more

number and higher amount of minerals such as copper, zinc, sodium, potassium, calcium, iron, phosphorous and magnesium (Lowor and Agyente-Badu, 2009).

The initial microbial population of liquid foods depends on many factors such as cleanliness of fruit, storage period, processing conditions and application of GAP, GMP etc. (Laorko *et al.,* 2013). The total bacterial count of fresh pineapple and cashew apple juice were 6.52×10^5 cfu/ml and 7.05×10^5 cfu/ml respectively. Yeast and mould population in fresh pineapple and cashew apple juice were 5.32×10^4 and 5.47×10^4 cfu/ml respectively. The higher microbial content of the cashew apple juice could be attributed to the high water, sugar and nutritional content of the cashew apple in comparison to pineapple apart from other factors (Sivagurunathan *et al.,* 2010).

The rheological studies of the fresh pineapple and cashew apple juices were carried out as described in section 3.6 using an Antonpar rheometer. Flow curves were generated and data was fitted into different models to categorise the juice's viscous behaviour. Steady-state flow curves were obtained by increasing the shear rate (0-100 s^{-1}) for fruit juices. Viscosity vs. shear rate of the cashew apple and pineapple juice is represented in Fig. 4.1 and 4.2. It was observed that, viscosity increased initially and then gradually decreased with increasing shear rate. From the figure, the blue line in represents the experimental values and red curve is used to fit the modeling equation. Therefore a shear thinning behavior could be observed. As mentioned in section 3.6. Two models such as Herschel bulkley and Ostwald models were evaluated for the suitability. It was found that both cashew apple and pineapple juice fits well with the both model. Pineapple juice showed higher correlation ratio (0.87) for Herschel bulkley model whereas cashew apple juice recorded higher correlation ratio (0.89) for Ostwald rheological model.

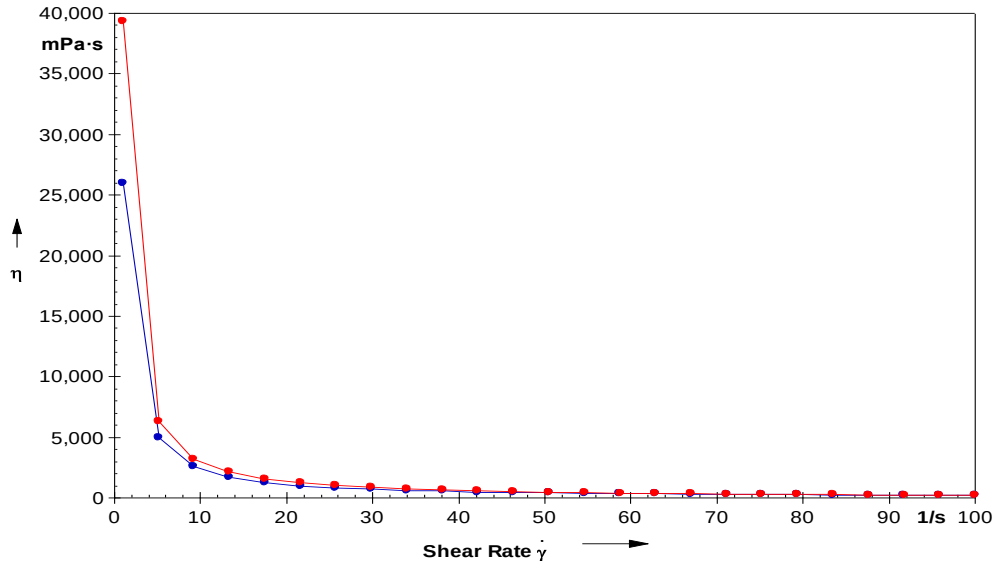


Fig. 4.1 Viscosity v/s shear rate curve of pineapple juice

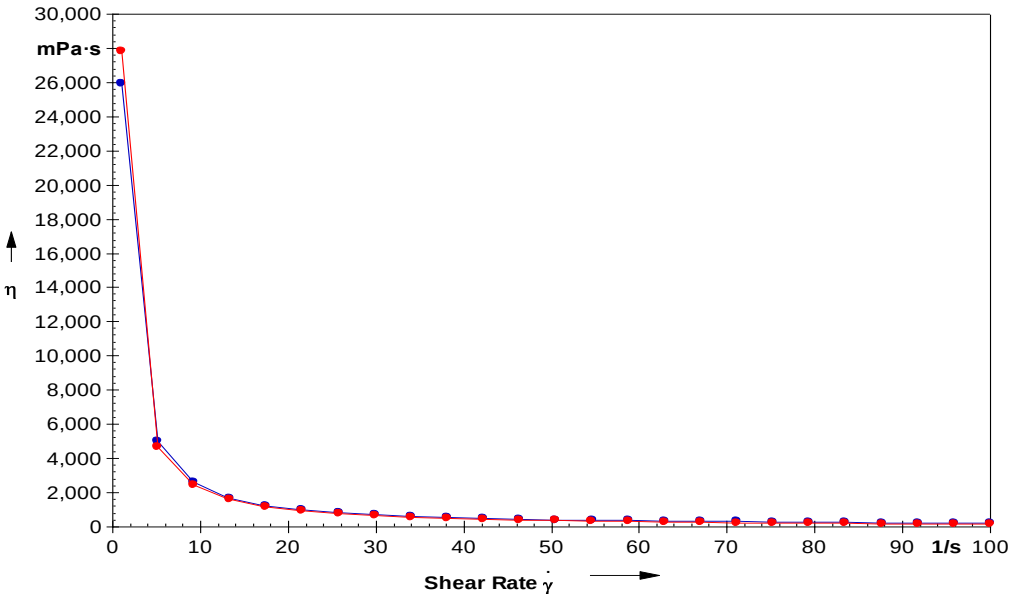


Fig. 4.2 Viscosity v/s shear rate curve of cashew apple juice

4.2 EFFECT OF ELECTRICAL CONDUCTIVITY ON OHMIC HEATING PROCESS

The electrical conductivity (EC) is one of the driving forces of ohmic heaing. The variation in electrical conductivity during ohmic heating of pineapple and cashew apple juice at different voltage gradients are depicted in Fig. 4.3 and 4.4 and Appendix A.1and A.2.

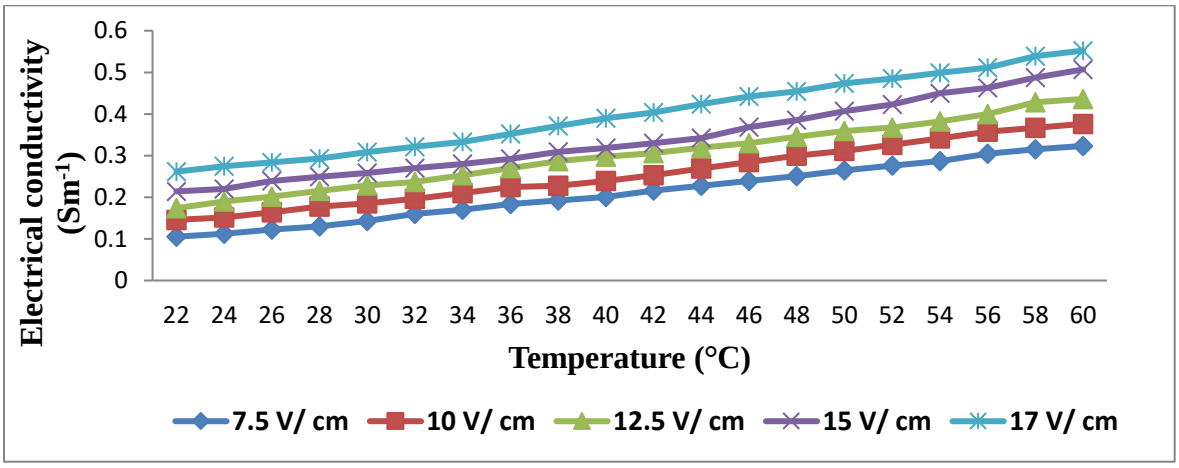


Fig.4.3 Changes in EC during ohmic heating of pineapple juice

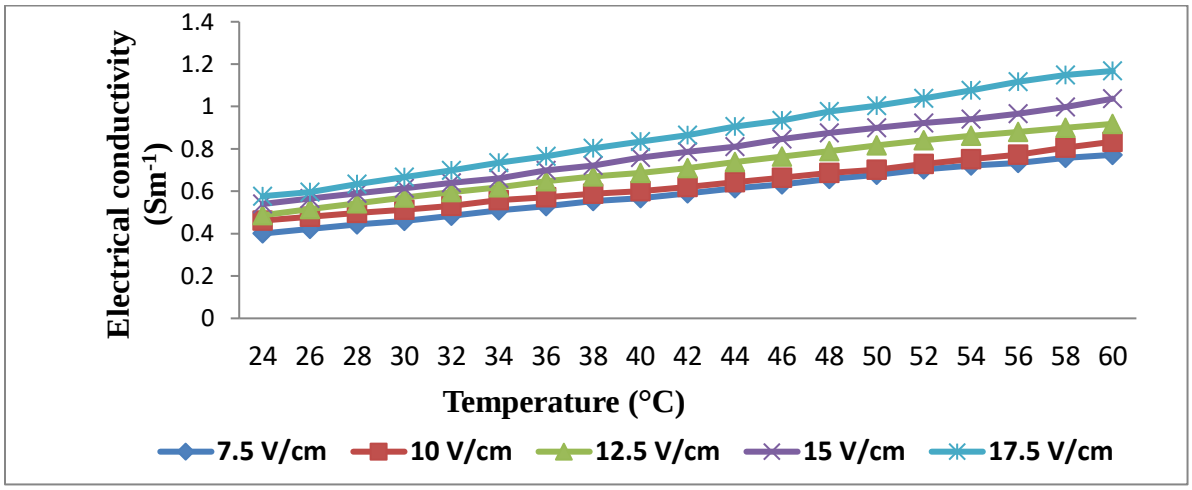


Fig.4.4 Changes in EC during ohmic heating of cashew apple juice

It is evident from figures that electrical conductivity of fruit juices observed a linear relationship with the temperature. It could also be noted that voltage gradient also have a

positive influence on electrical conductivity of fruit juices. Both increase in voltage gradient and increase in temperature showed a significant ($p<0.05$) increase in the electrical conductivity. The heating times decreased as a result of higher heating rates resulting from the higher voltage gradient applied. The electrical conductivity of pineapple juice ranged from 0.105 to 0.323, 0.145 to 0.376, 0.173 to 0.435, 0.214 to 0.506 and 0.261 to 0.552 Sm^{-1} for voltage gradient of 7.5, 10, 12.5, 15, 17 V/cm respectively. Whereas in cashew apple juice EC ranged from 0.384 to 0.771, 0.445 to 0.833, 0.460 to 0.918, 0.519 to 1.03 and 0.562 to 1.167 Sm^{-1} for 7.5, 10, 12.5, 15 and 17 V/cm respectively. The highest electrical conductivity value of 0.552 and 1.167 Sm^{-1} for pineapple juice and cashew apple juice respectively were obtained at the highest voltage gradient of 17.5 V/cm and highest temperature of 60°C.

Similar trend of increase in electric conductivity with increasing temperature during ohmic heating was observed by Assiry *et al*. (2010), Zell *et al.* (2010), Akbarpour *et al.* (2009), Marra *et al.* (2009), and Amiali *et al.* (2006). The increase in average kinetic energy of molecules during the increase in temperature could have intensified the mobility of molecules, which might have increased the electrical conductivity of fruit juices (Kaushal and Muchomba, 2013). Icier (2003) also explained that the rise in the EC values during temperature increase might be due to the reduced drag for mobility of ions.

It may be revealed from the Fig. 4.3 and 4.4 that, the voltage gradient had a positive influence on the electrical conductivity of fruit juices. As Ohm's law states, at higher voltage gradient a higher current would pass through the ohmic heater, which in turn increase the generation of heat increasing the EC of the fruit juice (Kaushal and Muchomba, 2013).

As may be seen from Fig. 4.1 cashew apple juice had higher electrical conductivity when compared to pineapple juice. This might be due to the higher amount of minerals and ion of calcium, sodium, magnesium, manganese etc. (Talasila and Shaik, 2013). The amount of ionic species (acids and salts) in food has reported a direct correlation with electrical conductivity. It could also be due to the difference in soluble solid content of pineapple and

cashew apple juice. The clarified cashew apple juice had a lower TSS value of 7.23°Brix compared to 10.83°Brix that of pineapple juice. The solid content and particle size present in pineapple juice could have obstructed the electric current movement and eventually leads to lower electrical conductivity values (Castro *et al.*, 2004a). Tumpanuvatr and Jittanit, (2012) also reported a negative influence of solid content and particle size on electrical conductivity of fruit juices. The boiling or bubbling of pineapple juice at lower temperature during the process of ohmic heating may cause increase in solid content due to evaporation which could also be cited as a reason for reduced electric conductivity (Palaniappan and Sastry, 1991).

4.2.1 Temperature dependence of the electrical conductivity of juice during ohmic heating

As has been revealed from the above discussion, the electrical conductivity of fruit juices was found to be a linear function of temperature. Thus a linear model was employed to fit the electrical conductivity values of pineapple and cashew apple juice. The experimental values of electrical conductivity within a temperature range of 22-60°C during ohmic heating are fitted in the Equation 3.1 for different voltage gradients (7.5, 10, 12.5, 15, 17.5 V/cm). It was found that the voltage gradient and temperature had significant effect on electrical conductivity of the pineapple and cashew apple juice ($p < 0.05$). The regression analysis was performed using SPSS software. The regression equations and R^2 values representing the electrical conductivity at different voltage gradients for both pineapple and cashew apple juice are presented in Table 4.2.

Table 4.2. Regression analysis on modeling of electrical conductivity with temperature

Fruit juice	Voltage gradient (V/cm)	Electrical conductivity – Temperature model (Sm^{-1})	Regression coefficients	
			R^2	Adj. R^2
Pineapple juice	7.5	$\sigma = 0.06T-0.31$	0.998	0.998
	10.0	$\sigma = 0.06T-0.03$	0.995	0.995
	12.5	$\sigma = 0.026T-0.007$	0.998	0.998
	15.0	$\sigma = 0.08T-0.027$	0.981	0.980
	17.5	$\sigma = 0.08T-0.076$	0.996	0.996
Cashew apple juice	7.5	$\sigma = 0.012T +0.151$	0.999	0.999
	10.0	$\sigma = 0.010T+0.216$	0.994	0.994
	12.5	$\sigma = 0.012T+0.203$	0.998	0.998
	15.0	$\sigma = 0.014T+0.207$	0.998	0.998
	17.5	$\sigma = 0.017T+0.169$	0.998	0.998

It could be revealed from the Table 4.2 that for majority of fitted linear models the R^2 value was around 0.995, representing a very good fit of the model. Hence a linear regression equation could be used effectively to predict the electrical conductivity values of pineapple and cashew apple juice during ohmic heating between temperature ranges of 22-60°C. Icier and Ilicali (2005) and Darvishi *et al.* (2011) had reported a similar relationship in ohmic heating of orange juice and lemon juice respectively.

4.3. OPTIMISATION OF PROCESS PARAMETERS

The developed ohmic heating assisted pulsed light treatment system was evaluated for its microbial inactivation potential and preservation of the natural quality attributes of pineapple and cashew apple juice as raw materials. The juices were selected based on justification mentioned earlier. As the biomaterial behaves differently to the process parameters of ohmic heating and pulsed light system and the parameters were optimised separately for individual system and then for the combined treatment. The quality parameters were then observed and analysed to conclude the best treatment condition.

4.3.1 Optimisation of Ohmic Heating Process Parameters for Pineapple and Cashew apple Juice

The pineapple and cashew apple juices were subjected to ohmic heating alone at voltage gradients of 10, 12.5 and 15 V/cm, process temperatures of 50, 55, and 60°C and treatment time 1, 3 and 5 min in the ohmic heating section of the developed system as mentioned in section (3.4.1). A come up time was required in all ohmic heating treatment to attain the specified temperature for each voltage gradient. The come up time required to attain 60°C during ohmic heating of pineapple juce was 2.54 min, 1.68 min and 42 seconds at voltage gradient of 10, 12.5 and 15 V/cm respectively, whereas, for cashew apple juice it was 2.35 min, 1.56 min and 36 seconds at voltage dradient of 10, 12.5 and 15 V/cm respectively. The higher come up time during ohmic heating results in a negative influence on quality parameters. The physico-chemical and microbial quality characteristics were analysed. Fresh fruit juice samples of pineapple and cashew apple were taken as control for comparison. The results obtained are presented in Table 4.3 and 4.4.

The variation in physiochemical properties such as pH, TSS, ascorbic acid, tritrable acidity, total sugar, total phenolic content, and total colour difference during ohmic heating with different combination of process parameters for pineapple and cashew apple juice are discussed in detail in the following sections.

Table 4.3 Effect of ohmic heating process parameters on the physico-chemical and microbial properties of pineapple juice

Run	Voltage gradient (V/cm)	Process temperature (°C)	Holding time (min)	pH	TSS (°Brix)	Titrable acidity (mg/100ml)	Ascorbic acid (mg/100ml)	Total sugar (mg/100ml)	Total phenolic content (mg/100ml)	Total colour difference (ΔE)	Bacterial log reduction (log cfu/ml)	Yeast and mould reduction (log cfu/ml)
1	15.0	50	3	4.2	10.95	0.391	27.52	10.56	68.61	0.62	2.79	1.98
2	12.5	55	3	3.98	11.28	0.393	28.08	10.57	68.61	0.89	3.49	2.65
3	12.5	60	5	3.89	11.42	0.392	27.48	10.56	69.19	1.12	3.89	2.89
4	12.5	55	3	3.96	11.36	0.392	27.96	10.56	68.89	0.83	3.54	2.51
5	12.5	55	3	4.02	11.25	0.391	28.13	10.54	68.85	0.86	3.42	2.68
6	15.0	60	3	4.31	11.4	0.393	27.46	10.54	69.31	1.21	4.03	3.04
7	12.5	55	3	4.04	11.37	0.391	27.87	10.54	68.52	0.91	3.58	2.56
8	12.5	50	5	3.95	11.36	0.392	28.16	10.55	67.13	0.73	2.48	1.65
9	10.0	50	3	3.45	10.85	0.391	28.32	10.56	67.65	0.58	1.98	1.32
10	15.0	55	1	4.47	11.05	0.392	28.24	10.57	68.79	0.75	2.75	2.04
11	12.5	55	3	3.96	11.35	0.392	27.98	10.54	68.76	0.84	3.55	2.63
12	10.0	55	1	3.69	10.62	0.392	28.42	10.56	67.72	0.53	1.89	1.21
13	12.5	60	1	4.32	11.32	0.392	27.75	10.54	68.37	0.78	3.13	2.34
14	10.0	60	3	3.47	10.83	0.393	27.52	10.54	68.79	0.71	2.63	1.84
15	10.0	55	5	3.38	10.89	0.391	27.72	10.57	68.63	0.63	2.14	1.45
16	15.0	55	5	4.03	11.39	0.392	27.58	10.54	69.24	1.17	3.92	2.96
17	12.5	50	1	4.28	10.93	0.392	28.32	10.56	67.52	0.68	2.37	1.54

Table 4.4 Effect of different ohmic heating process parameters on the physico- chemical and microbial properties of cashew apple juice

Run	Voltage gradient (V/cm)	Process temperature (°C)	Holding time (min)	pH	TSS (°Brix)	Titrable acidity (mg/100 ml)	Ascorbic acid (mg/100 ml)	Total sugar (mg/100 ml)	Total phenolic content (mg/100 ml)	Tannin content (%)	Total colour difference (ΔE)	Bacterial log reduction (log cfu/ml)	Yeast and mould reduction (log cfu/ml)
1	10	60	3	4.22	7.36	0.423	165.35	9.95	166.74	0.538	0.49	3.32	2.14
2	12.5	55	3	4.26	7.53	0.423	165.24	9.97	166.97	0.509	0.69	3.96	2.75
3	15	60	3	4.32	7.91	0.424	155.72	9.94	168.15	0.496	0.88	4.13	3.05
4	10	55	1	4.24	7.25	0.422	158.62	9.97	165.42	0.521	0.3	1.71	1.5
5	12.5	60	5	4.24	7.72	0.424	166.85	9.96	168.23	0.498	0.84	4.26	3.17
6	12.5	50	1	4.33	7.3	0.422	159.21	9.94	166.12	0.534	0.37	2.21	1.66
7	12.5	60	1	4.32	7.42	0.424	160.29	9.96	166.78	0.502	0.58	3.53	2.27
8	10	55	5	4.21	7.31	0.422	156.79	9.96	165.56	0.494	0.44	2.45	1.79
9	12.5	50	5	4.25	7.41	0.423	167.23	9.95	165.42	0.526	0.51	2.72	1.88
10	15	55	1	4.34	7.52	0.423	159.06	9.97	166.21	0.542	0.54	2.94	1.86
11	15	55	5	4.26	7.86	0.424	161.42	9.97	168.18	0.538	0.86	4.24	2.95
12	12.5	55	3	4.27	7.64	0.424	156.32	9.94	167.47	0.512	0.68	3.94	2.39
13	12.5	55	3	4.31	7.69	0.423	161.34	9.95	167.89	0.536	0.61	3.6	2.5
14	10	50	3	4.23	7.22	0.423	160.15	9.95	166.21	0.54	0.34	1.94	1.6
15	15	50	3	4.31	7.48	0.422	161.29	9.96	166.46	0.514	0.41	2.56	1.99
16	12.5	55	3	4.28	7.55	0.422	162.35	9.94	167.65	0.539	0.64	3.86	2.69
17	12.5	55	3	4.29	7.62	0.422	159.58	9.98	167.23	0.507	0.66	3.88	2.42

4.3.1.1 Effect of ohmic heating process parameter on pH of fruit juices

The variation in pH value of pineapple and cashew apple juice upon ohmic heating treatment at different operating variables combinations are shown in Table 4.3 and 4.4 and the ANOVA tables are presented in Appendix-A.4 a and b. It may be observed from the table that the pH values of pineapple juice ranged from 3.38 to 4.47, whereas that of cashew apple juice ranged between 4.21 and 4.34. The highest pH value of 4.47 and 4.34 was obtained at a voltage gradient of 15 V/cm, temperature of 55°C and holding time of 1 min for both pineapple and cashew apple juice. The minimum pH value was recorded when the juices were treated with voltage gradient of 10 V/cm, process temperature of 55°C and holding time of 5 min for both pineapple and cashew apple juice.

The 3D response surface plots of variation in pH values of pineapple and cashew apple juices at different combinations of ohmic heating operating parameters are shown in Fig. 4.5 and 4.6. It may be observed from the figures that, voltage gradient and holding time had a significant effect on the pH values of pineapple and cashew apple juice ($p < 0.001$). For both juices, with the increase in holding time, the pH values decreased, whereas increase in voltage gradient resulted in a lower reduction in pH value. Makroo *et al.* (2017) and Boldaji *et al.* (2014) also reported such trends during ohmic heating of watermelon juice and tomato paste. A similar trend was also reported in ohmic heating of sapota juice and pomegranate juice (Thangalakshmi *et al.,* 2018; Darvishi *et al.,* 2013).

The increase in pH value of fruit juices during ohmic heating could be attributed to the different chemical reactions like hydrolysis of fruit juices and corrosion of electrodes (Assiry *et al.,* 2010; Assiry *et al.,* 2003). The conduction of current through the fruit juices could have resulted in hydrolysis of fruit juices due the presence of mineral ions and acidic salts in them. The stainless steel electrodes used in this study is less susceptible to corrosion and therefore the change in pH values due to corrosion may be ruled out. When the voltage gradient increased from 10-15 V/cm the change in pH value was found to decrease which could be attributed to the long come up time required for lower voltage gradients to reach the predetermined temperature. The long time of

exposure to the electric fields and contact with the electrodes might have intensified reactions leading to pH change of fruit juices (Darvishi *et al.*, 2012).

A second order regression model was developed relating the experimental pH values of pineapple and cashew apple juices with the coded value of the corresponding combination of the independent variables and presented in the Equation 4.1 for pineapple and 4.2 for cashew apple.

For pineapple juice,

$$pH = 3.99+0.38\ A+0.014B-0.19\ C+0.023\ A\ B-0.032AC-0.025\ B\ C-\ 0.18A^2+\ 0.042\ B^2+0.076\ C^2 \qquad R^2= 0.98 \qquad 4.1$$

For cashew apple juice,

$$pH = 4.282+\ 0.04A-\ 0.002B-\ 0.032C+\ 0.005AB-0.01AC\ +0BC-0.018A^2\ +\ 0.006B^2-\ 0.003C^2 \qquad R^2=\ 0.91 \qquad 4.2$$

Where,

A = Voltage gradient, V/cm

B = Treatment temperature, °C

C = Holding time, min

It may be revealed from equations that the pH values of pineapple and cashew apple juice were in positive correlation with voltage gradient and process temperature and in negative correlation with the holding time. From the coefficients of the independent variables, it was observed that the pH values were highly influenced by the voltage gradient followed by holding time and then by process temperature.

From the ANOVA table (A.4. a and b), it could be noted that the regression model was significant as p-value was too low (<0.001) and the lack of fit was not significant indicating the adequacy of the model. The regression coefficients of both linear and quadratic term of voltage gradient were highly significant. Since the R^2 and adj. R^2 values are close to one and adequate precision value was more than 4, the model could be adjudged adequately fit.

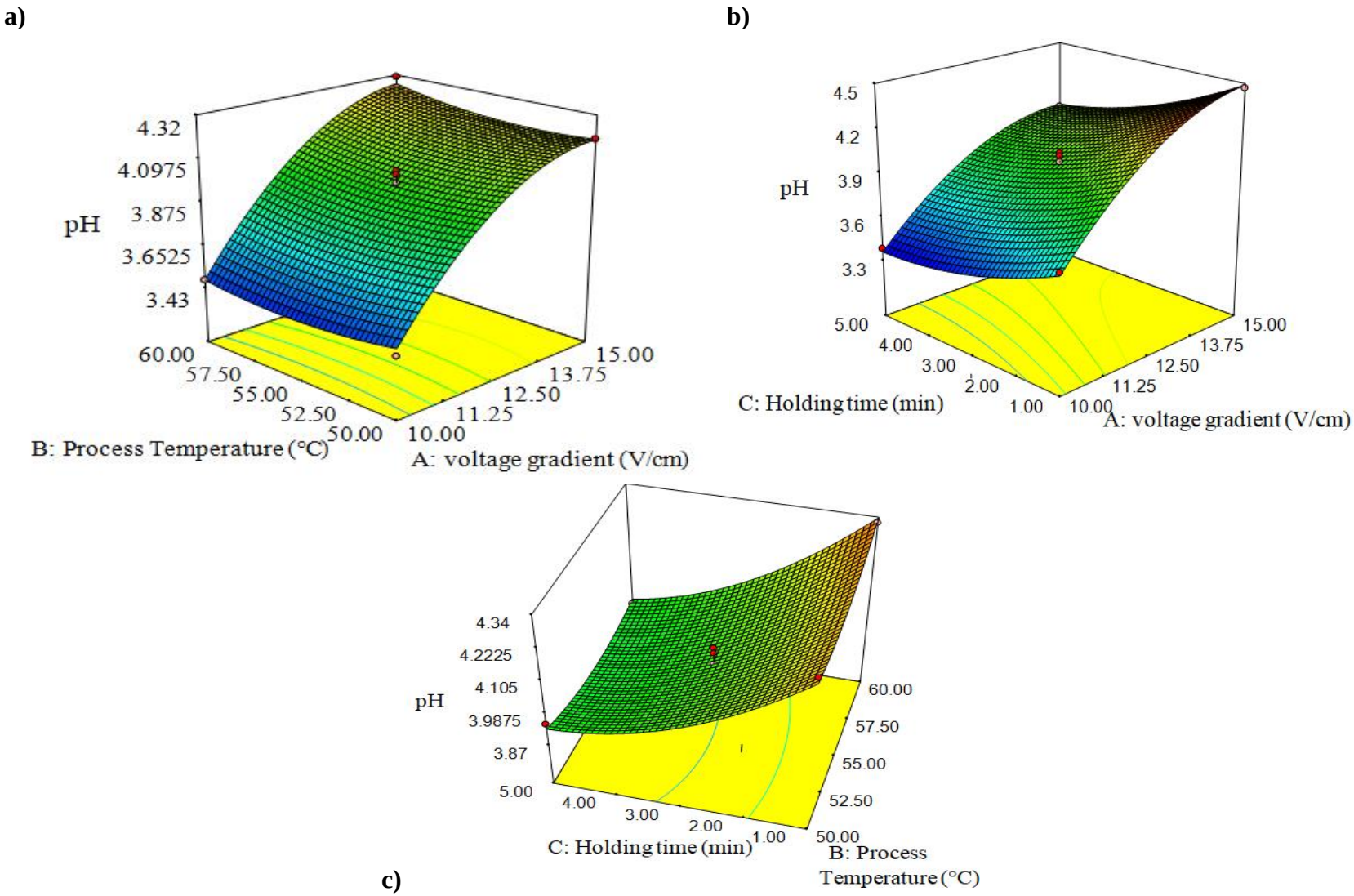


Fig. 4.5 Effect of ohmic heating parameters on pH of pineapple juice

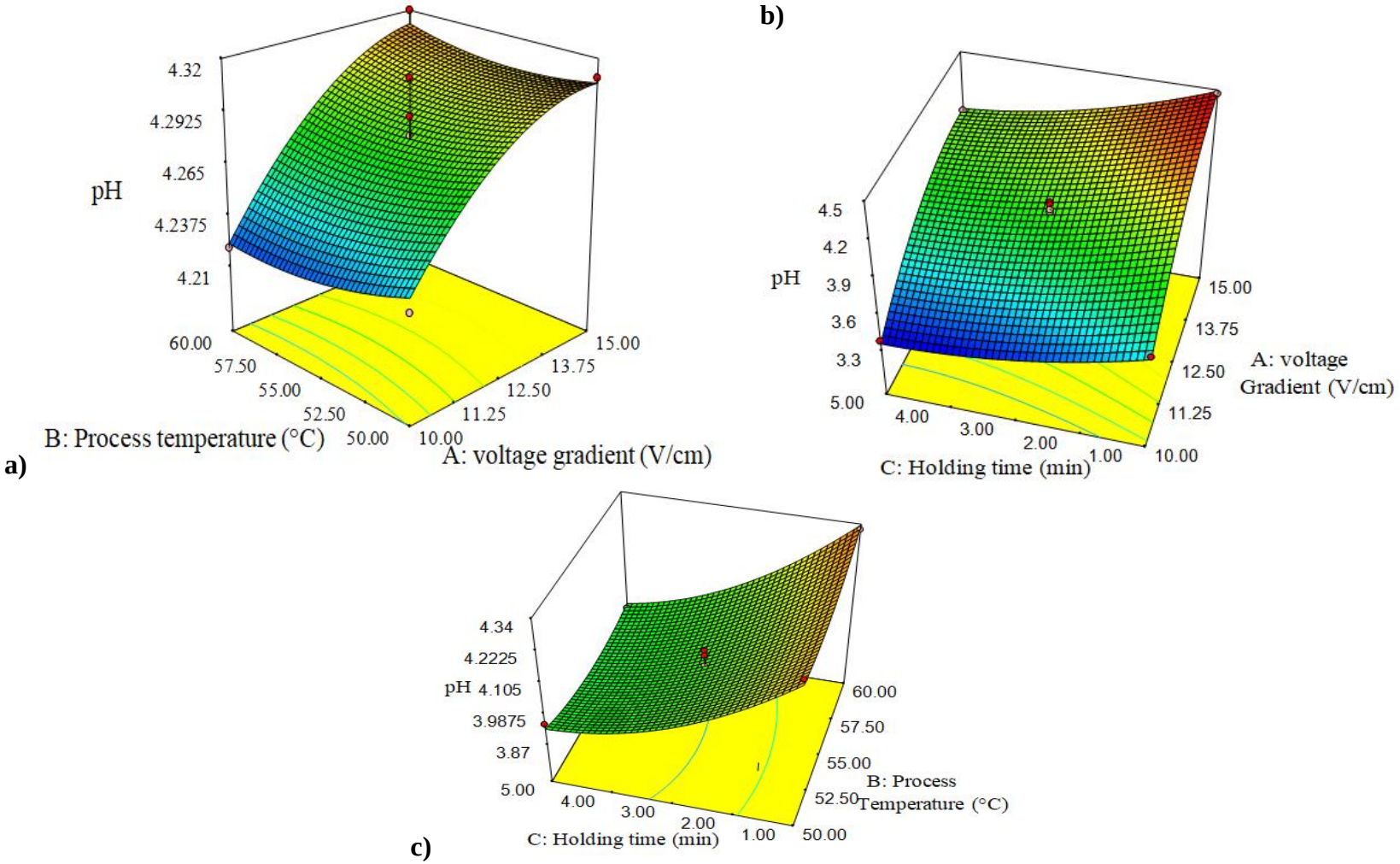


Fig. 4.6 Effect of ohmic heating parameters on pH of cashew apple juice

4.3.1.2 Effect of ohmic heating process parameters on TSS of fruit juices

The variation in TSS content of pineapple and cashew apple juice treated with different combinations of ohmic heating operating variables are summarised in Table 4.3 and 4.4. The ANOVA tables are presented in Appendix A.5 a and b.

It may be observed from the results that the TSS value of pineapple and cashew apple juice ranged between 10.62 to 11.42°Brix and 7.22 to 7.91°Brix respectively. The lowest variation in TSS values of ohmic processed juice with that of fresh juice were observed in samples treated with voltage gradient of 10 V/cm, holding time of 1 min and process temperature of 55°C in the case of pineapple juice and voltage gradient of 10 V/cm, holding time of 3 min and process temperature of 50°C in the case of cashew apple juice.

The effects of different ohmic heating process parameters on TSS content of fruit juices are presented as 3D surface plots in Fig. 4.7 and 4.8. From the figures it may be observed that TSS content increased with increase in voltage gradient, holding time and process temperatures for both juices. It was found that all ohmic heating parameters had significant effect on the TSS value of both the juices ($p<0.001$). Similar results were also reported by Abhilasha and Pal (2018); Poojitha and Athmaselvi (2018); and Chakraborty and Athmaselvi (2014) in ohmic heated sugar cane juice, banana purees and guava juice respectively. This could be attributed to the loss of water content with increase in heating and evaporation which in turn increased the solute concentration (Purvis, 1983). The conversion of organic acids to sugars could also be a reason for the modification of TSS content (Echeverria and Valich, 1989).

The R^2 value of the quadratic model for pineapple and cashew apple juice was 0.986 and 0.960, respectively. The Equation 4.3 and 4.4 represent the quadratic equations for the effect of process parameters on TSS value of both fruit juices. The higher R^2 value and non significant lack of fit of the model indicates the suitability of the model

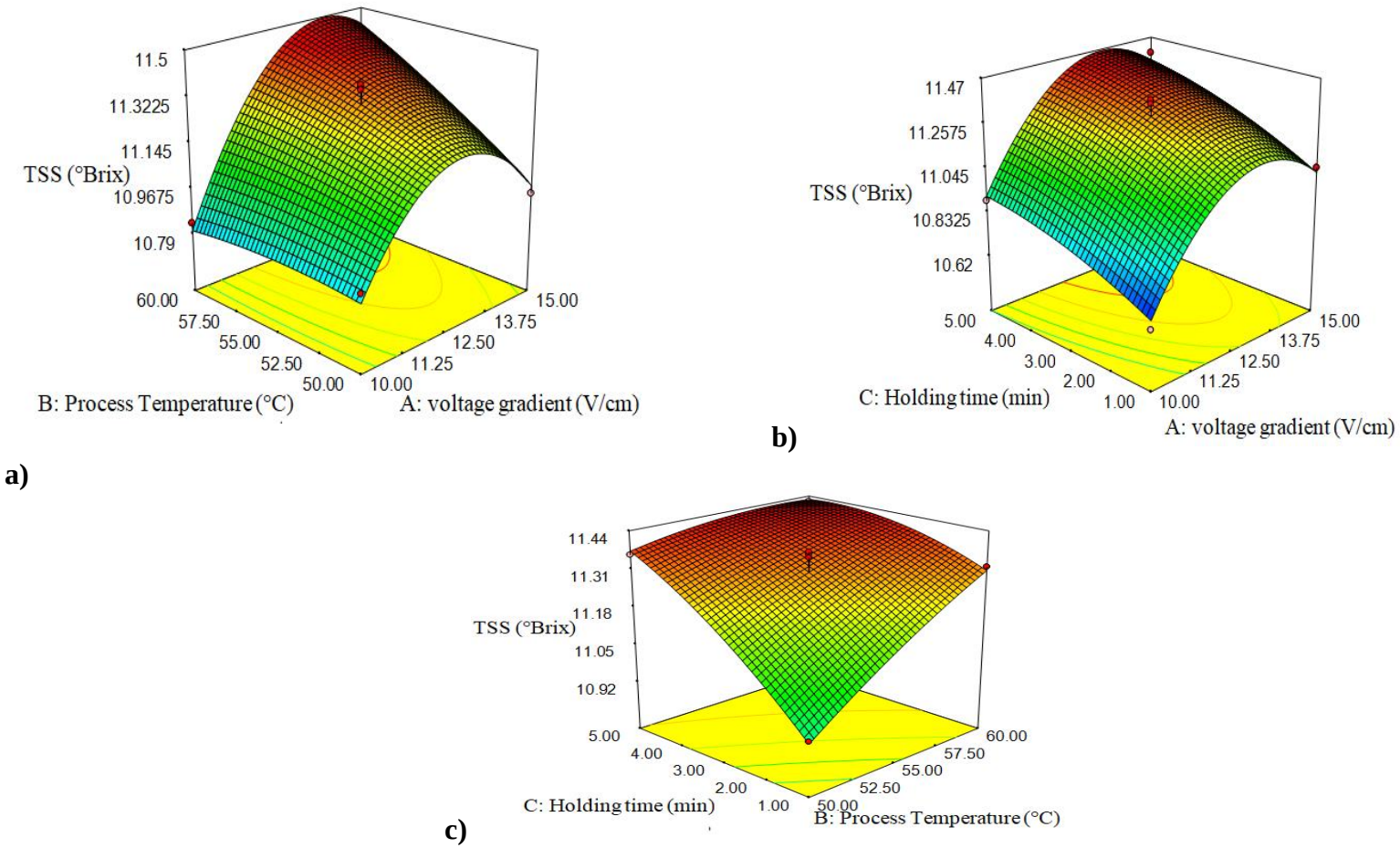


Fig. 4.7 Effect of ohmic heating parameters on TSS of pineapple juice

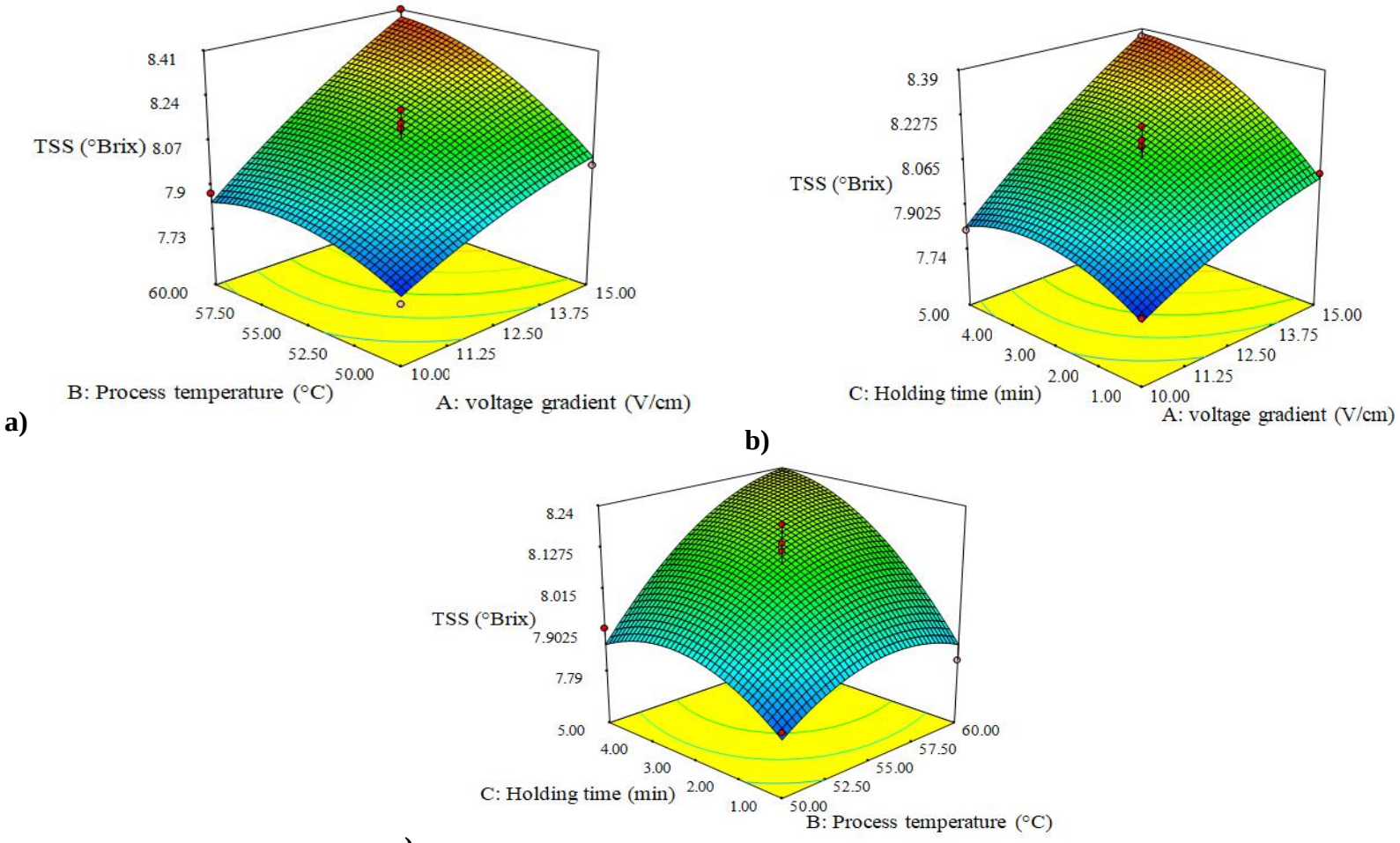


Fig. 4.8 Effect of ohmic heating parameters on TSS of cashew apple juice

For pineapple juice

$$TSS = 11.31+0.26\ A+5.000E-003B+0.19C+0.012A\ B+0.025\ A\ C-0.18A^2-0.016B^2-0.13C^2 \quad R^2 = 0.96 \quad (4.3)$$

For cashew apple juice

$$TSS = 11.606+0.20375A+0.125B+0.101C+0.072AB+0.07AC+0.047BC-0.045A^2-0.068B^2-0.075C^2 \quad R^2 = 0.97 \quad (4.4)$$

Where,

A = Voltage gradient, V/cm

B = Treatment temperature, °C

C = Holding time, min

The Equation 4.3 and 4.4 indicates that the TSS value of pineapple and cashew apple juice were in positive correlation with process temperature and holding time. The ANOVA tables (A 5.a and b) showed that the regression model was significant as p-value was too low (< 0.001) and the lack of fit was not significant indicating the adequacy of the model. Since the R^2 and adj. R^2 values are near to one and adequate precision value was more than 4, the model could be termed adequately fit.

4.3.1.3 Effect of ohmic heating process parameters on titrable acidity of fruit juices

The effect of ohmic heating treatment combinations on tirable acidity of both pineapple and cashew apple juices are depicted in Table 4.3 and 4.4 and the analysis of variance (ANOVA) is presented in Appendix-A.6 a and b.

It may be revealed from the table that titrable acidity of pineapple juice varied between 0.422 and 0.424 mg/100 ml and that of cashew apple juice between 0.391 and 0.393 mg/100 ml. It may be inferred that the titrable acidity did not show any significant changes after ohmic heating treatment as no typical trends were observed in the titrable acidity values of both fruit juices. A slight decrease in titrable acidity values were observed in all treated juice samples irrespective of treatment combinations. The

observed minimum variation in the titrable acidity values might be attributed to the conversion of organic acids in to sugar at elevated temperatures (Chattopadhyay *et al.*, 1992). Thangalakshmi *et al.* (2018) found that electro chemical reactions such as hydrolysis of sample or corrosion of electrodes might have also accounted for the variation in titrable acidity values.

The 3D response surface plots Fig.4.9 and 4.10 of the effect of ohmic heating process parameters on titrable acidity presented that ohmic heating treatment have no significant effect on the titrable acidity value of both the juices. It could be seen from the ANOVA tables Appendix A. 6. a and b, that regression model and lack of fit are non significant with low R^2 values for both fruit juices.

4.3.1.4 Effect of ohmic heating process parameters on ascorbic acid of fruit juices

The ascorbic acid contents of pineapple and cashew apple juices subjected to different ohmic heating variable combinations are shown in Table 4.3 and 4.4 respectively, and the ANOVA table is presented in Appendix-A.7 a and b.

Ascorbic acid is considered as one of the most unstable compound in fruits and vegetables. A significant reduction of ascorbic acid content was observed during ohmic heating of pineapple and cashew apple juice ($p < 0.001$). The ascorbic acid content varied between 27.46 and 28.42 and 155.72 and 167.23 mg/100 ml respectively. The maximum reduction of ascorbic acid was noted at a voltage gradient of 15 V/cm, process temperature of 60°C and holding time of 3 min for both juices. Pineapple juice treated at a voltage gradient of 10 V/cm and process temperature of 55°C with 1 min holding time resulted in minimum reduction of ascorbic acid content. Similarly the cashew apple juice treated at a voltage gradient of 12.5 V/cm and process temperature of 50°C with 1 min holding time showed lowest reduction in ascorbic acid content.

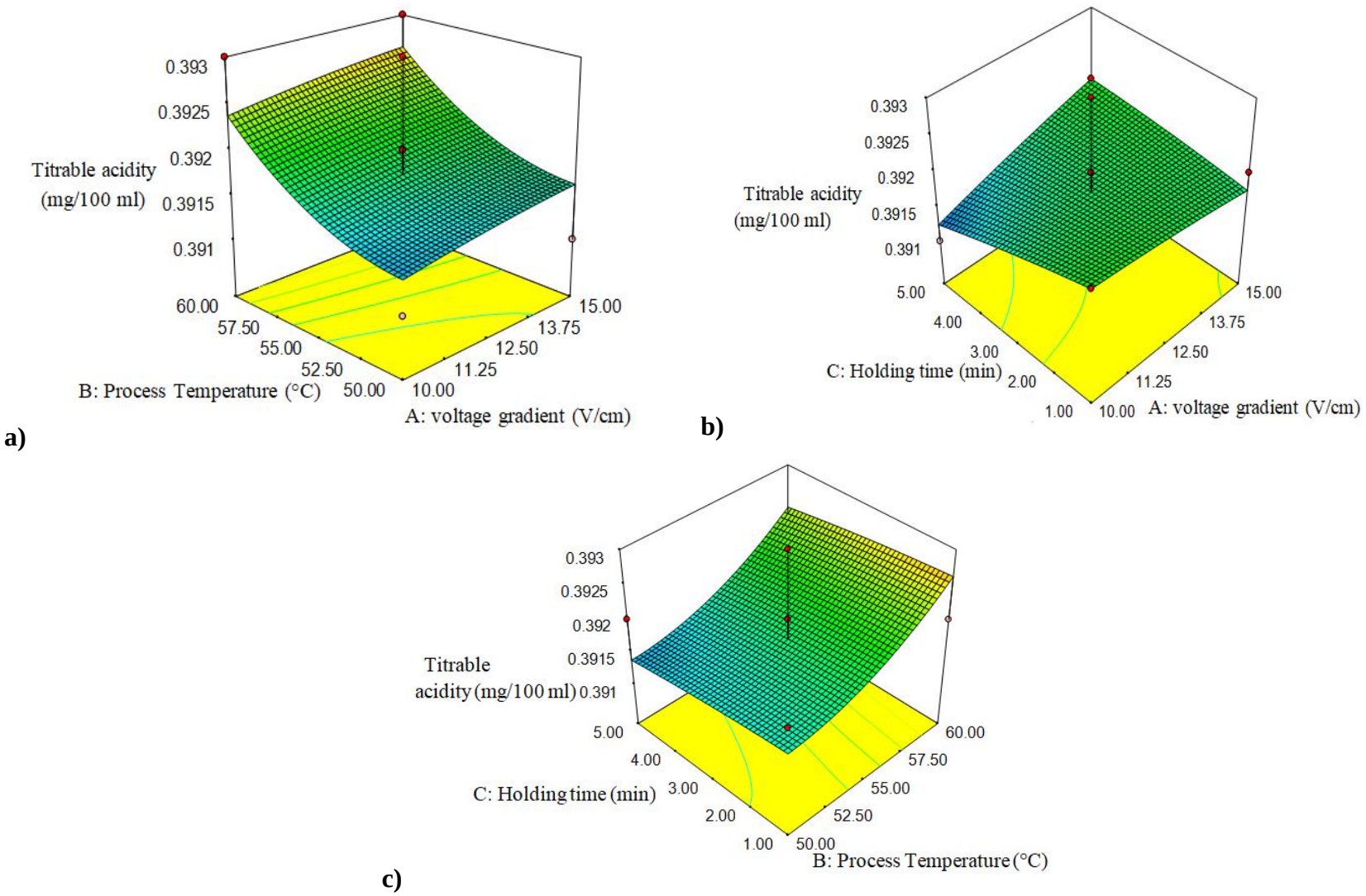


Fig. 4.9 Effect of ohmic heating parameters on titrable acidity of pineapple juice

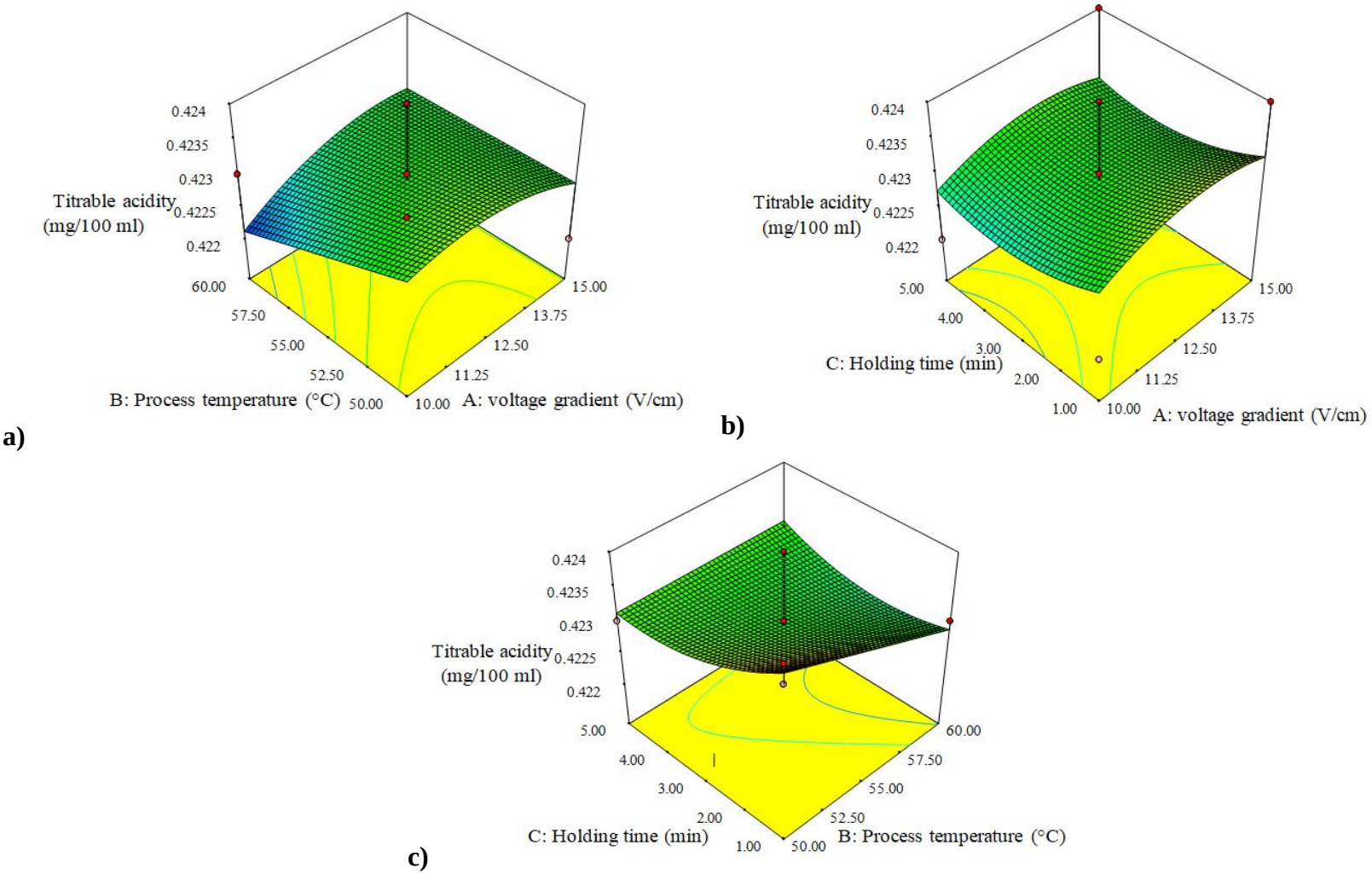


Fig. 4.10 Effect of ohmic heating parameters on titrable acidity of cashew apple juice

The 3D surface plots of variation in ascorbic acid with different combinations of process parameters of pineapple and cashew apple juices are depicted in Fig. 4.11 and 4.12 respectively. It can be noted from the figures that increase in temperature and holding time resulted in a significant reduction of ascorbic acid content of pineapple and cashew apple juice. A similar line of ascorbic acid degradation was reported in orange and pomegranate juice (Paul and Ghosh, 2012). This could be due to the heat sensitive nature of ascorbic acid compounds. Several studies have reported that ascorbic acid follow a first order kinetic of degradation during exposure to above normal temperatures (Paul and Ghosh, 2012).

As may be seen from the Fig. 4.11 and 4.12, the voltage gradient was found to possess a significant effect on the ascorbic acid content in pineapple and cashew apple juices. Increase in the reduction of ascorbic acid was noted with increase in voltage gradient. These results are in agreement with the findings of Thangalakshmi *et al.* (2018) and Poojitha and Athmaselvi (2018). The degradation of ascorbic acids might be attributed to several chemical reactions such as oxidative and corrosive reactions during ohmic heating. The electrolysis of water during ohmic heating might have generated oxygen molecules which in turn resulted in the oxidation of ascorbic acid compounds. In addition to this, the reaction between product of electrolysis and electrodes may also cause degradation of ascorbic acid (Assiry *et al.,* 2003). In contrast with this justification, it has also been reported by Assiry *et al*. (2006) that no degradation of ascorbic acid was noted in presence of electric fields. In this study the overall reduction in ascorbic acid content of both pineapple and cashew apple juice were minimum, compared to that of other thermal processing technologies, due to the mild conditions such as lower temperature and voltage gradient employed.

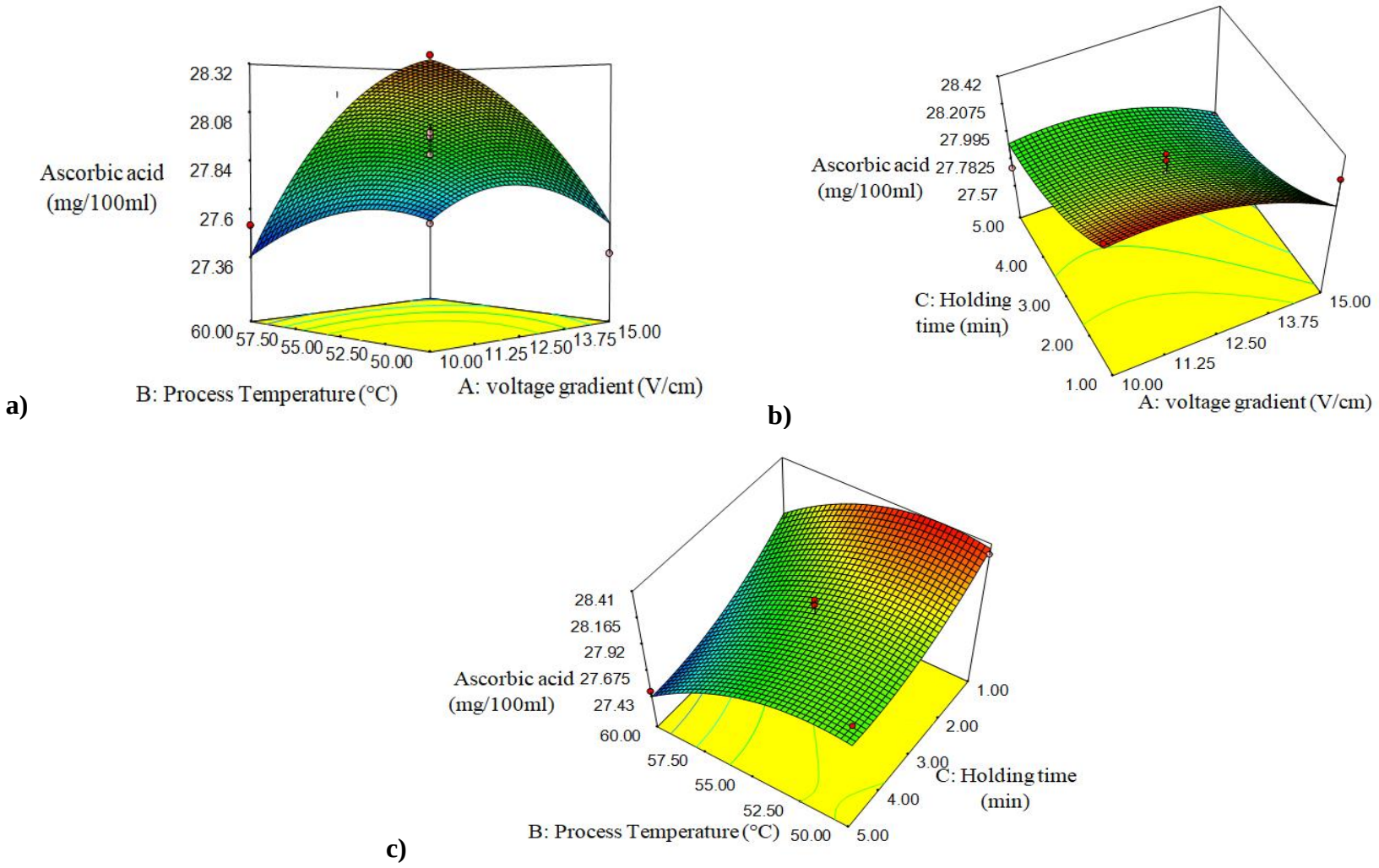


Fig. 4.11 Effect of ohmic heating parameters on ascorbic acid content of pineapple juice

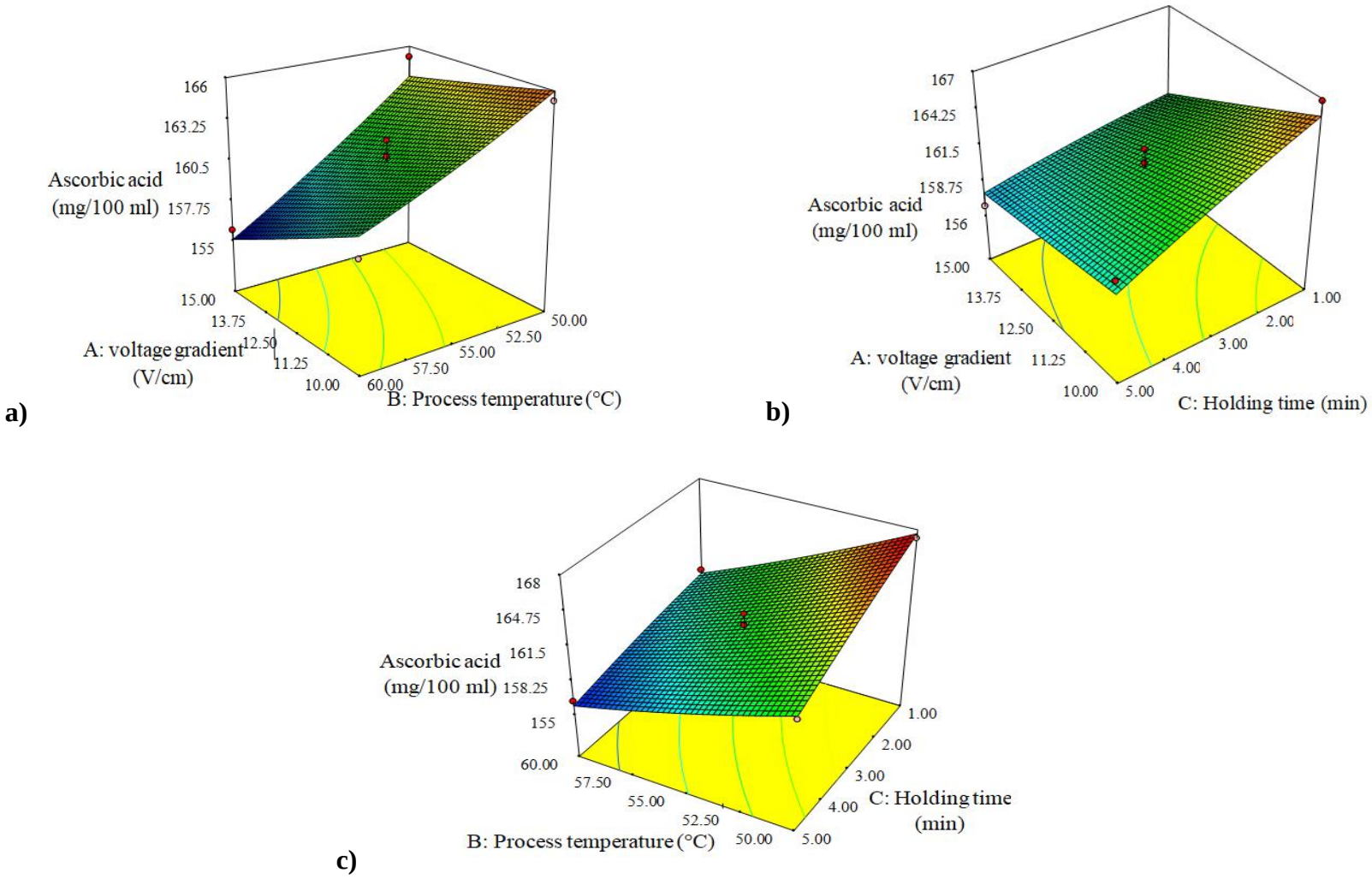


Fig. 4.12 Effect of ohmic heating parameters on Ascorbic acid content of cashew apple juice

For pineapple juice

Ascorbic acid content (mg/100 ml) = 28 - 0.16A - 0.25B- 0.22C + 0.22AB + 0.10AC- 0.27BC - $0.14A^2$+$0.20B^2$+$0.12C^2$ R^2 = 0.86 4.5

For cashew apple juice

Ascorbic acid content (mg/100 ml) = 160 - 1.77A - 3.69B - 2.19C - 0.71AB + 1.035AC + 0.772BC + $0.038A^2$+ $0.26B^2$- $0.19C^2$ R^2 = 0.92 4.6

Where,

A = Voltage gradient, V/cm

B = Treatment temperature, °C

C = Holding time, min

The equations showed that the ascorbic acid content of pineapple and cashew apple juice were in negative correlation with all process parameters. The coefficients of the independent variables revealed that the process temperature had maximum influence on ascorbic acid content followed by holding time and then by voltage gradient. The R^2 value of the quadratic model for pineapple and cashew apple juice was 0.954 and 0.964, respectively. The higher R^2 value and non significant lack of fit of the model indicates that the model is adequately fit.

4.3.1.5 Effect of ohmic heating process parameters on total sugar of fruit juices

The variation in total sugar content of pineapple and cashew apple juice upon ohmic heating treatment at different operating parameter combinations are shown in Table 4.3 and 4.4 and the ANOVA table is presented in Appendix-A.8 a and b.

It may be revealed from the table that the total sugar content of pineapple juice ranged between 10.54 and 10.57 mg/100 ml, and that of cashew apple juice ranged from 9.94 to 9.98 mg/100 ml. It may be concluded that the total sugar did not show any significant variation during ohmic heating treatment and no typical trend was also observed in the total sugar values of both fruit juices. The 3D response surface plots (Fig. 4.13 and 4.14) showing variation of total sugar with changes in process variables between

the selected ranges indicated that the total sugar content is unaffected for both the juices. It could be seen from the ANOVA tables A.8 a and b, that lack of fit was significant and F-value suggested that the model was insignificant at one per cent and five per cent level of significance.

4.3.1.6 Effect of ohmic heating process parameters on phenolic compounds of fruit juices

The effect of various experimental combinations of ohmic heating operating parameters on the total phenolic content of pineapple and cashew apple juices are depicted in Table 4.3 and 4.4 and the analysis of variance (ANOVA) is shown in Appendix-A. 9 a and b.

It could be seen from the table that the phenolic content of ohmic heated pineapple and cashew apple juice ranged from 67.13 to 69.31 and 165.42 to 168.23 mg/100 ml respectively. The highest phenolic content value was found in the pineapple juice treated at voltage gradient of 15 V/cm, at a process temperature of 60°C with 3 min holding time. In cashew apple juice the maximum phenolic content was recorded at a voltage gradient of 12.50 V/cm, process temperature of 60°C and holding time of 5 min. The variation percentage of phenolic content within the operating ranges of variables, were 3.2% and 1.80% in pineapple and cashew apple juice respectively.

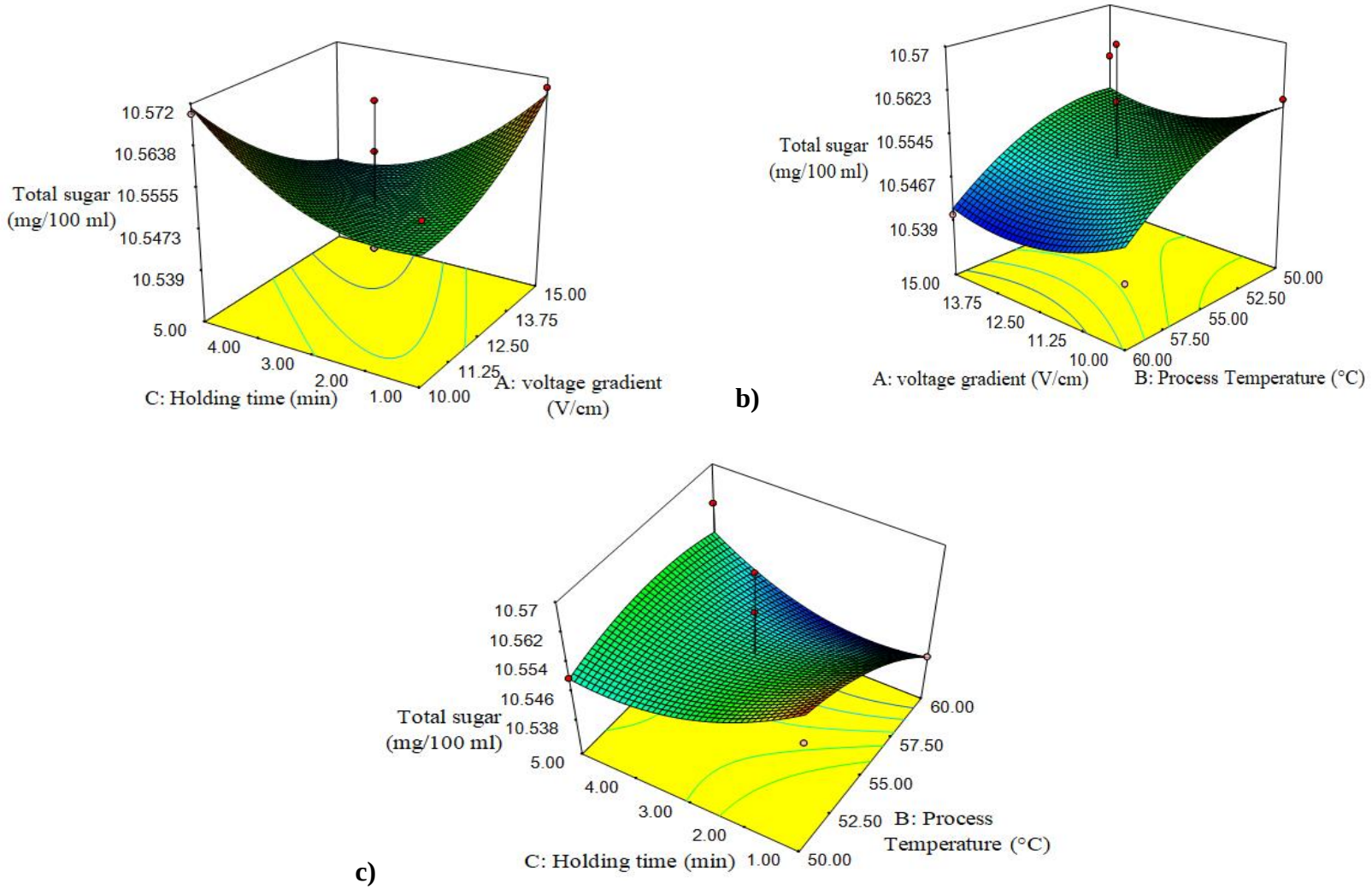


Fig. 4.13 Effect of ohmic heating parameters on total sugar of pineapple juice

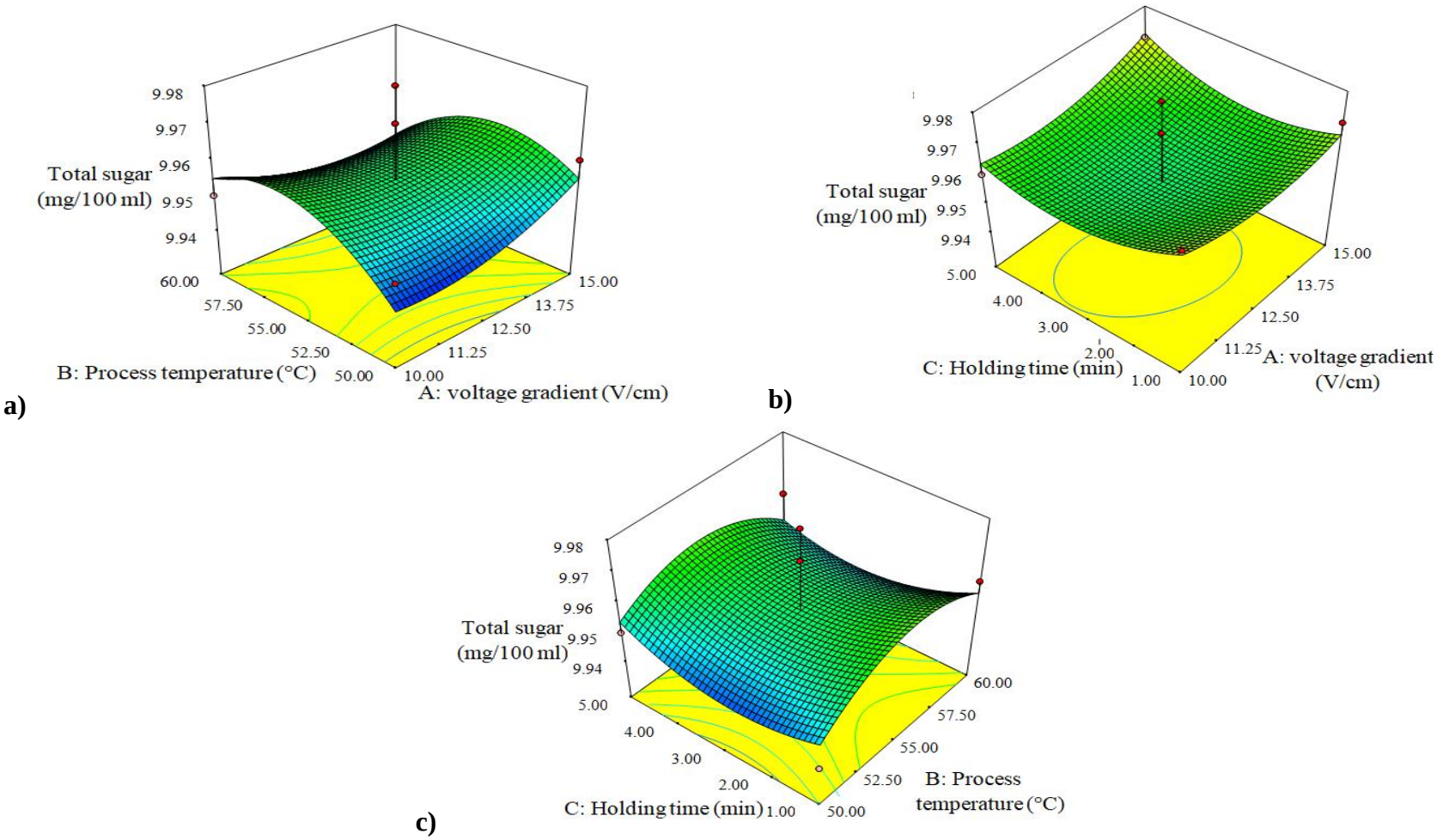


Fig. 4.14 Effect of ohmic heating parameters on total sugar of cashew apple juice

3D surface plots showing the effect of ohmic heating process variables on total phenolic content of pineapple and cashew apple juice are depicted in Fig. 4.15 and 4.16 respectively. Both juice samples showed an increase in total phenolic content during ohmic heating. All ohmic heating process parameters showed a significant effect on the variation of phenolic compounds. Similar observations were also reported in ohmic heated mango juice which illustrated an increase in the phenolic content compared to fresh juice (Abedelmaksoud *et al.,* 2018). The bottle guard juice depicted an increase in phenolic content during ohmic blanching process (Bhat, 2016), whereas ohmic heated cashew apple juice showed a slight decrease in phenolic content from initial value. Similar degradation of phenolic compounds were observed in ohmic heated sugarcane and pomegranate juice (Brochier and Domeneghini, 2016; Brochier and Domeneghini, 2018).

The increase in phenolic content might be attributed to the changes in the tissue structure during heating. This facilitates the movement of phenolics from the cell interior to exterior and eventually resulted in an increased extractability of the phenolic compound. The alternating current at frequencies greater than 40 Hz might have induced a non thermal synergic effect on the liberation of phenolic content during ohmic heating. The ohmic heating may also account for the destruction of associated protein-phenolic acid compounds and release of phenolic compounds (Brochier and Domeneghini, 2016; Bhat *et al.,* 2016). Moreover, ohmic heating of plant materials with higher electric field strength could have imparted cell membrane permeability changes which might have increased the extraction of total flavinoids, carotnoids and polyphenols from plant tissues (Lebovka *et al.,* 2007; Kulshrestha and Sastry, 2010).

A quadratic equation was derived relating the phenolic content of pineapple and cashew apple juice with the coded values of corresponding combinations of ohmic heating process variables and presented as Equation 4.7 and 4.8.

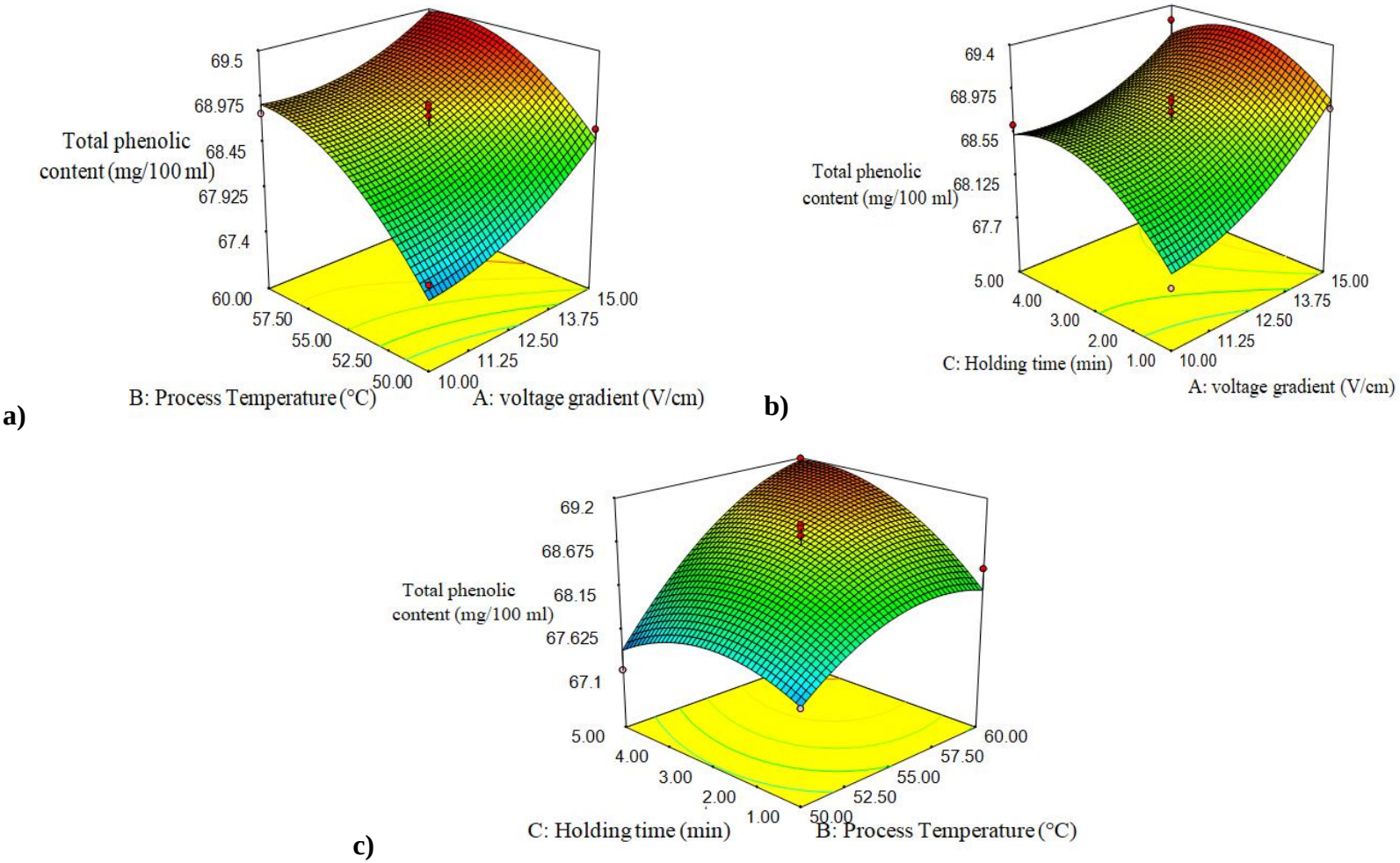


Fig. 4.15 Effect of ohmic heating parameters on total phenolic content of pineapple juice

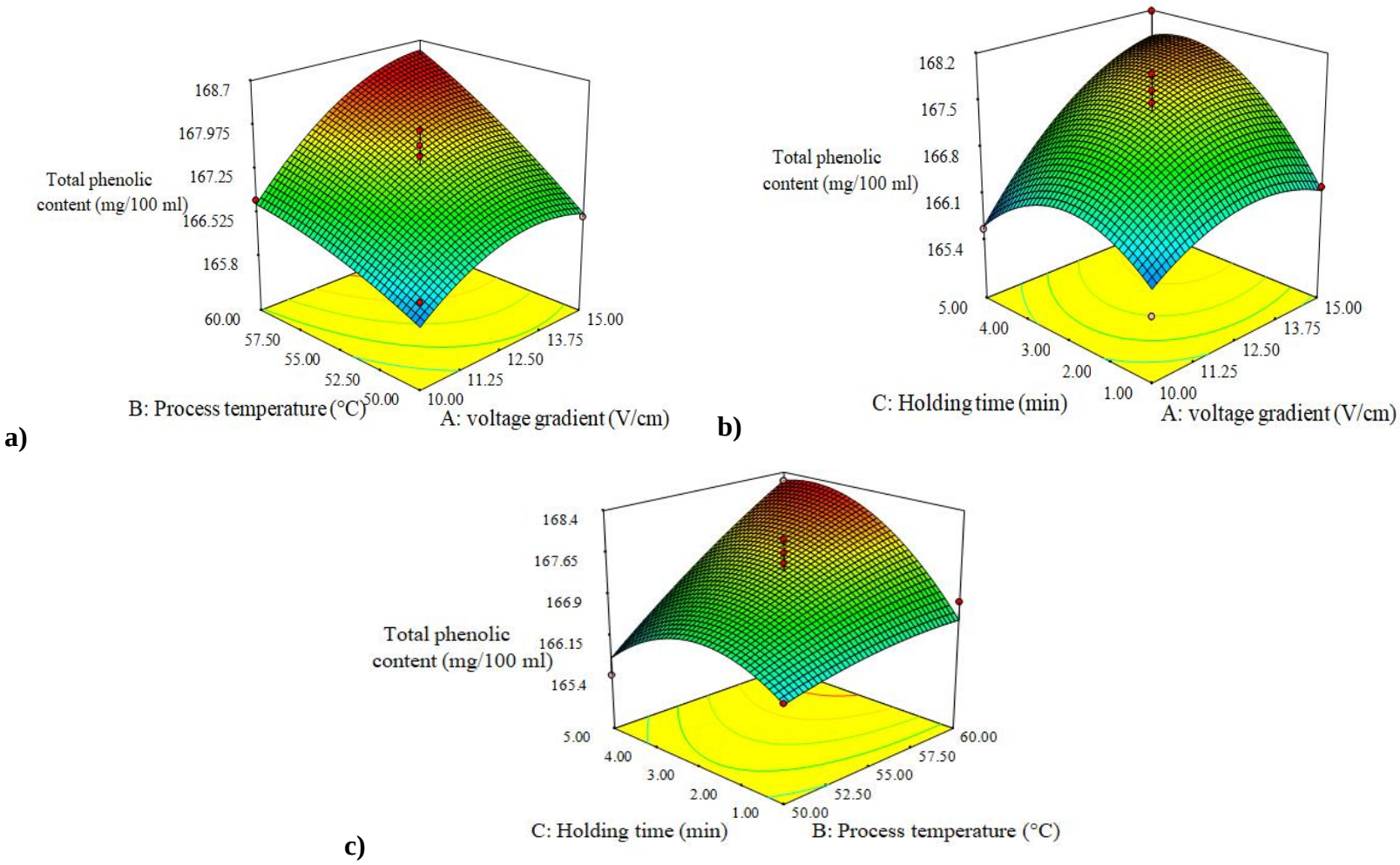


Fig. 4.16 Effect of ohmic heating parameters on total phenolic content of cashew apple juice

For pineapple juice

Phenolic content (mg/100 ml) = 68.728+0.40 A+0.59B+0.22C-0.11AB - 0.13AC+0.30BC+0.20A^2-0.34B^2-0.334C^2 $R^2 = 0.91$ 4.7

For cashew apple juice

Phenolic content (mg/100 ml) = 167.442 + 0.633A + 0.711B + 0.35C + 0.29AB + 0.537BC- 0.423A^2-0.1285B^2-0.676C^2 $R^2 =$ 4.8

Where,

A = Voltage gradient, V/cm

B = Treatment temperature, °C

C = Holding time, min

From Equations 4.7 and 4.8, it may be concluded that the phenolic content of pineapple and cashew apple juice are in positive correlation with all operating parameters of ohmic heating time in the decreasing order of influence in both the fruit juices. From the coefficients of the independent variables, it could also pointed that the phenolic contents were affected by the process temperature, voltage gradient and holding time in the decreasing order of influence in both the fruit juices.

From the ANOVA table (A.8. a and b), it may be found that the regression model was significant as p-value was too low (< 0.0001) and the lack of fit was not significant indicating the adequacy of the model. The R^2 value of the quadratic model for pineapple and cashew apple juice was 0.92 and 0.91 respectively. Since the R^2 and adj. R^2 values are close to one and adequate precision value was more than 4, the model could be considered adequately fit.

4.3.1.7 Effect of ohmic heating process parameters on tannin content of fruit juices

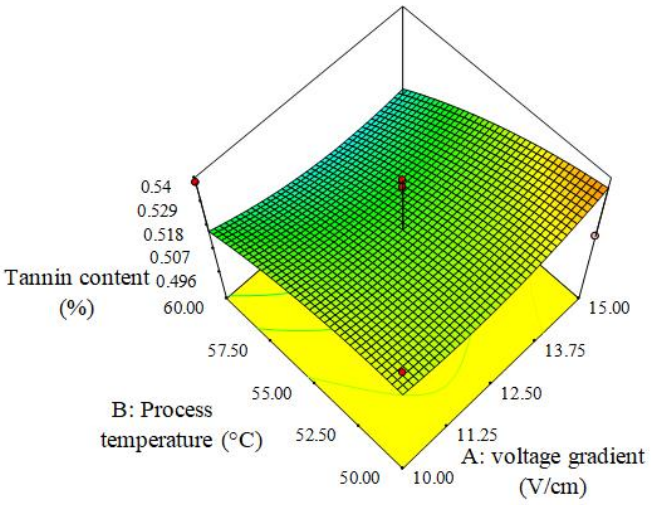
The tannin content in cashew apple is considered as one of the major cause of astringency. The variation in tannin content at different ohmic heating operating variable combinations are depicted in Table 4.3 and 4.4 and ANOVA tables are presented in Appendix-A. 10.

The tannin content of cashew apple juice showed an insignificant ($P > 0.05$) reduction during ohmic heating. Only slight reduction in tannin content was observed in ohmic heated cashew apple juice. It may be revealed from the 3D surface plots (Fig. 4.17) that the process parameters did not exhibit any significant effect on the tanninreduction. Some researchers have reported that heating process such as steaming of cashew apples resulted in the reduction of tannin content. Similarly Emelike *et al.* (2015) also reported that the tannin content of cashew apple juice significantly reduced during hot water treatment. The exposure of increased temperature than that of ambient conditions might have caused phenolic degradation thus the tannin content could have reduced slightly when compared to that of fresh juice. It could be seen from the ANOVA tables (A.9) that regression model and lack of fit are non significant with low R^2 values for both fruit juices.

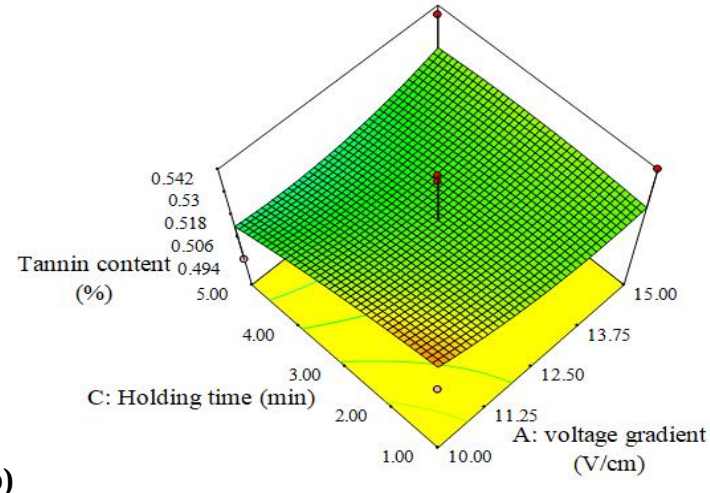
4.3.1.8 Effect of ohmic heating process parameters on total colour difference of fruit juices

Colour is an important attribute of juice quality for the consumer acceptability. The effect of ohmic heating process parameters on colour values (L*, a*, b*) of pineapple and cashew apple juice was evaluated and presented in the Appendix A.11. The L* a* and b* values represents different colour ranges *viz.* lightness to darkness, green to red and blue to yellow respectively.

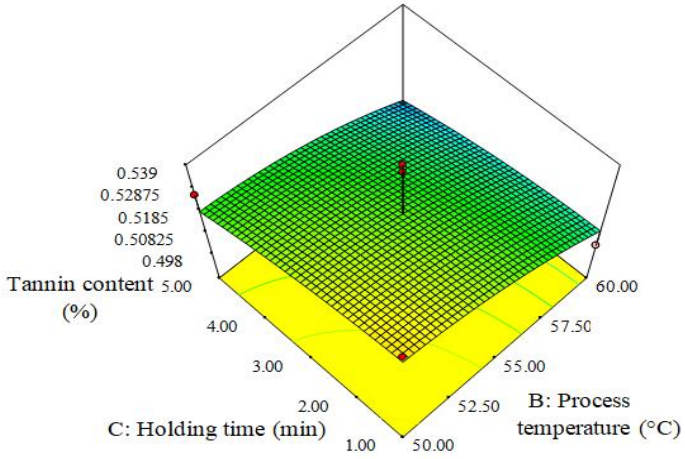
The L*, and b* values showed a slight decrease and a* values noted a slight increase with increase in voltage gradient and treatment temperature. The L* values ranged from 39.33 to 38.88, a* values ranged from -3.46 to -3.20 and b* values between 22.77 and 22.46 in pineapple juice samples. Similarly for cashew apple juice samples the L* values ranged from 1.35 to 1.25, a* values between 0.48 and 0.58 and b* values between -1.34 and -1.44.



a)



b)



c)

Fig. 4.17 Effect of ohmic heating parameters on tannin content of pineapple juice

The reduction in L* values indicates reduction of lightness, the decrease in b* values represents the reduction in yellowness and rise in a* values stand for the increase in redness of the juices after ohmic heating treatment. Similar trends in reduction of L* value was also reported during ohmic heating of banana puree and guava juice (Chakraborty and Athmaselvi, 2014; Poojitha and Athmaselvi, 2018).

Lower voltage gradients resulted in negligible changes in colour values. Brochier and Domeneghini (2018) also reported a minimal colour change at lower voltage gradient of 3.5 V/cm, while higher voltage (20.5 V/cm) gradients resulted in higher variation in colour values. This might be due to the higher temperature accomplished at higher voltage gradients (Thangalakshmi *et al.*, 2018).

The total colour difference increased with the increase in voltage gradient and holding time. The total colour difference is a numerical value calculated as explained in section 3.5.10 which represents how much the colour of a product varies from its reference colour or colour of fresh samples.

A notable visual change in colour will be seen when the total colour difference is above 1.5 (Cserhalmi *et al.*, 2006). The total colour difference of the pineapple and cashew apple juice ohmic treated at various voltage gradients, holding time, and process temperatures are illustrated in Table 4.3 and 4.4 and the ANOVA tables are presented in Appendix A.12 a and b. total colour difference observed fell below 1.5 for all treated fruit juice samples implying slightly or no visible colour change during ohmic heating.

The total colour difference of pineapple and cashew apple juice ranged from 0.53 to 1.21 and 0.3 to 0.88 respectively. The highest ΔE value was observed in pineapple juice treated with voltage gradient of 15 V/cm, holding time of 3 min and process temperature of 60°C. In cashew apple juice, the highest ΔE value was observed in treatment with a voltage gradient of 15 V/cm holding time of 3 min and process temperature of 60°C.

The effects of voltage gradient, holding time and process temperature on the total colour difference of ohmic heated pineapple and cashew apple juice are presented as 3D surface plots in Fig. 4.18 and 4.19. It is evident from the figures that the total colour

difference increased significantly with the increase in voltage gradient and process temperature. Though the changes in process variables were found to have significant effect on the total colour change statistically, the colour change was within a value of 1.5 implying the occurance of a very slight visible colour change noticed in the fruit juices. Chakraborty and Athmaselvi (2014) and Thangalakshmi *et al.* (2018) also reported an increase in total colour difference with increase in voltage gradient or electric field.

In present study very slight variation in the colour values only were observed in pineapple and cashew apple juice. This might be due to the fact that maximum process temperature during the ohmic heating was only 60°C. Considerable changes in colour values were usually observed at higher temperatures during thermal processing. At higher temperatures, substantial changes in colour values were observed in pineapple juice due to the non enzymatic browning and pigment degradation (Rattanathanalerk *et al*., 2005). It was reported that colour changes during ohmic heating could be due to browning in presence of oxygen and metal ions (Icier *et al.,* 2008). The active carbonyl groups released during the degradation of ascorbic acid also aids in enzymatic browning (Leizerson and Shimoni, 2005a). The enzymatic browning will be negligible during ohmic heating since the oxidative enzymes get inactivated at higher temperatures.

The second order regression model was developed relating the total colour difference of pineapple and cashew apple juices with the coded value of the corresponding combination of the independent variables and presented in the Equation 4.9 for pineapple juice and 4.10 for cashew apple juice.

For pineapple juice

$$\Delta E = 0.87+0.16\ A+0.15B+0.11C+0.11A\ B+0.080A\ \ C+0.073BC-0.072A^2-0.014B^2-0.024C^2 \qquad R^2 = 0.97 \qquad 4.9$$

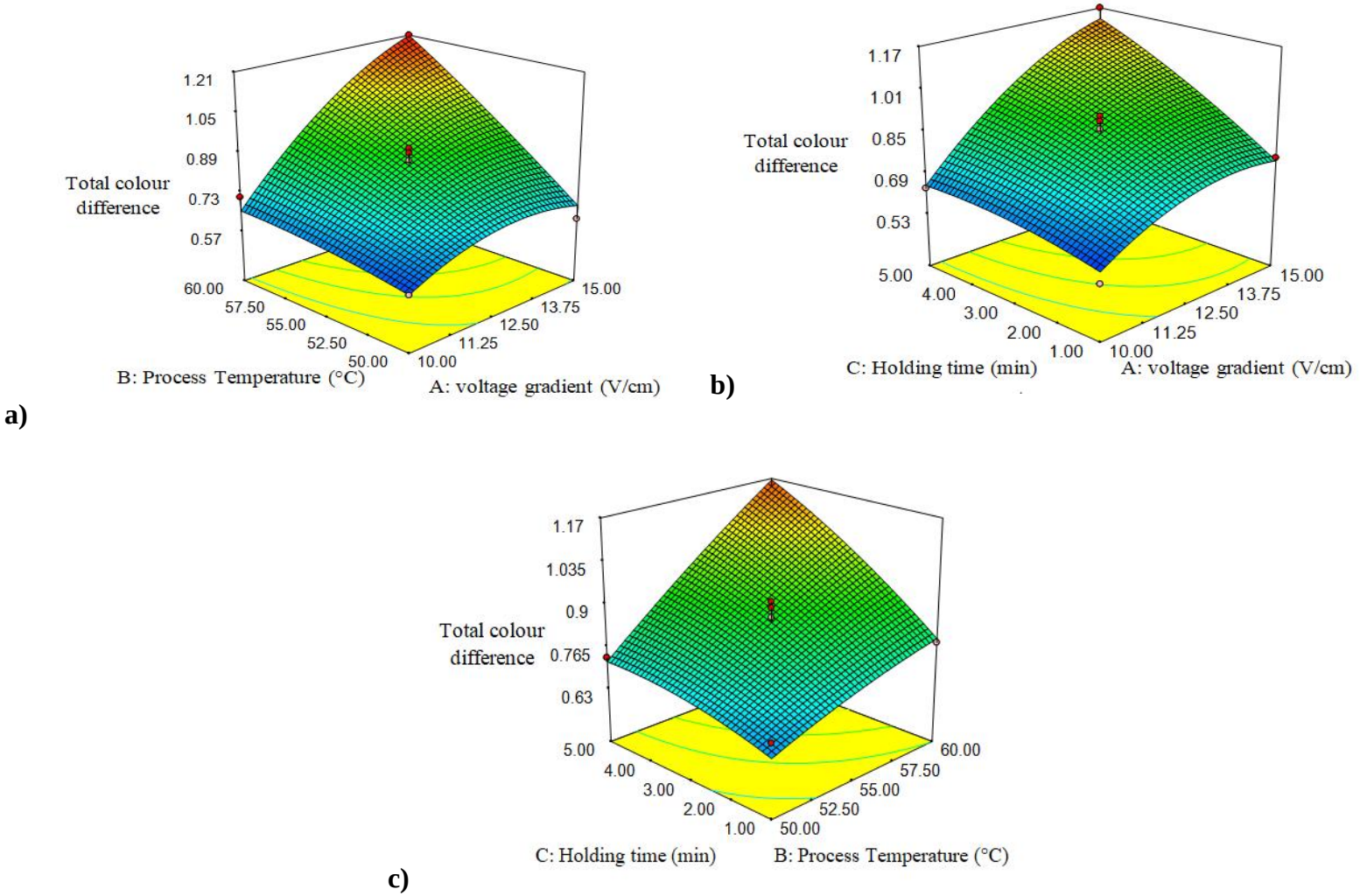


Fig. 4.18 Effect of ohmic heating parameters on total colour difference of pineapple juice

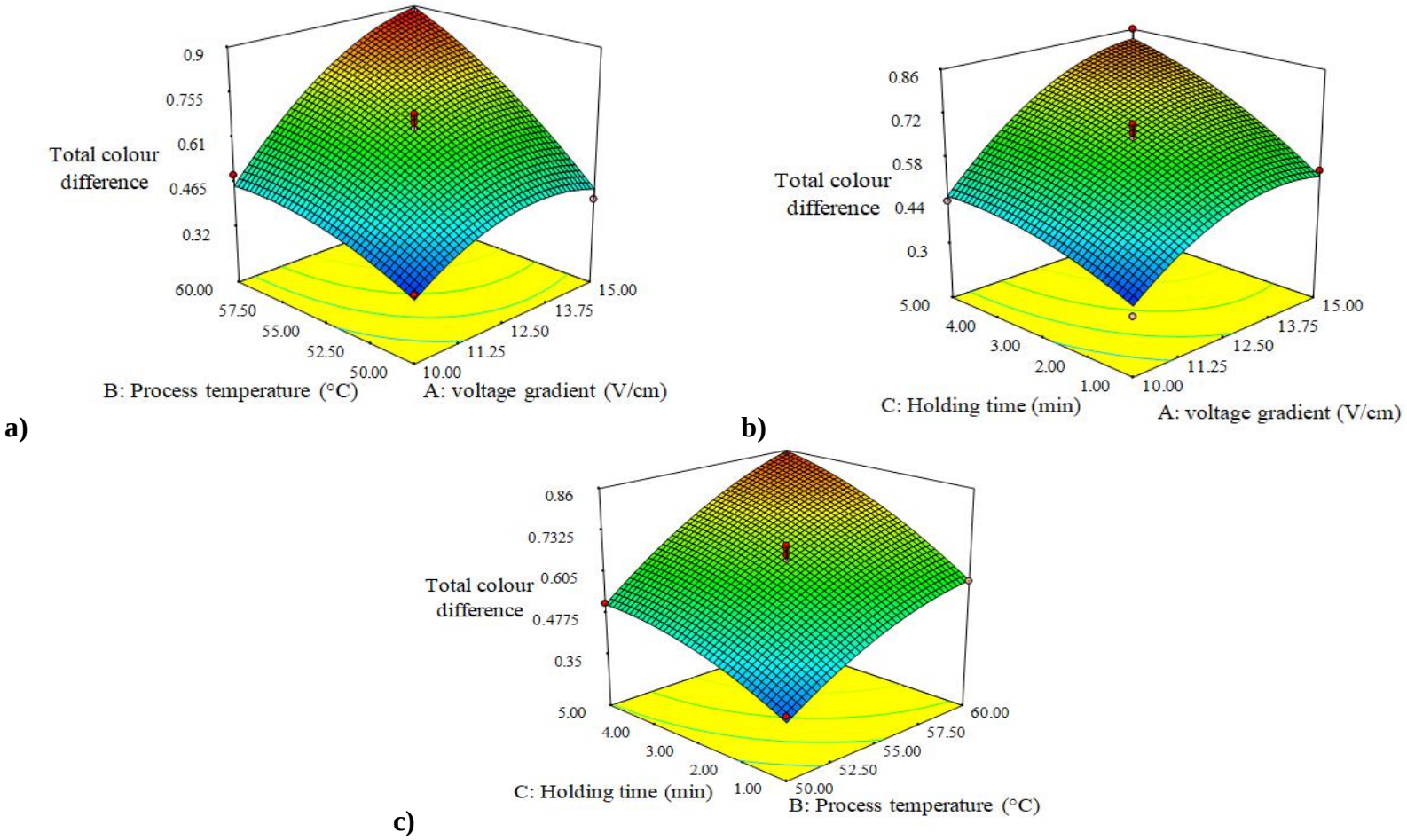


Fig. 4.19 Effect of ohmic heating parameters on total colour difference of cashew apple juice

For cashew apple juice

$\Delta E = 0.656+0.14A+0.145B+0.1075C+0.08AB +0.045AC+0.03BC-0.083A^2-0.043B^2-0.038C^2$ $R^2 =0.97$ 4.10

Where,

A = Voltage gradient, V/cm

B = Treatment temperature, °C

C = Holding time, min

It is perceived from Equation 4.9 and 10 that the total colour difference was in positive correlation with voltage gradient, process temperature, and holding time. ANOVA table A.11 a and b illustrate that, the linear (A, B, C), interactive (AB, BC, AC) terms had a significant effect on total colour difference at 1% ($p<0.001$). The R^2 value (0.97) is in reasonable agreement with the adj. R^2 (0.93) for pineapple juice. Similarly the R^2 (0.97) value of cashew apple juice also is in reasonable agreement with the adj. R^2 (0.95). Lack of fit was insignificant and F-value suggested that the model was significant at 1 and 5% level of significance.

4.3.2 Effect of Ohmic Heating Process Parameters on the Microbial log Reduction of Fruit Juices

The total bacterial and yeast and mould count of both pineapple and cashew apple juices were determined by the method as referred in section 3.7.1. The total bacterial and yeast and mould log reduction of the pineapple and cashew apple juice obtained in various experimental combinations of operating variables are presented in Table 4.3 and 4.4 respectively and the ANOVA tables are presented in Appendix-A.13 a and b and A.14. a and b.

It could be seen from Table 4.3 that the total bacterial, yeast and mould reduction in pineapple juice was found to be in the range of 1.89 to 4.03 and 1.21 to 3.04 log cfu/ ml respectively. The highest bacterial log reduction of 4.03 log cfu/ ml and highest yeast and mould reduction of 3.04 log cfu/ ml were observed in the

treatment at voltage gradients of 15 V/cm, process temperature of 60°C with 3 min of holding time. The treatment with voltage gradient of 10 V/cm, process temperature of 55°C and holding time of 1 min reported a lowest bacterial reduction of 1.89 and yeast and mould reduction of 1.21.

It could be noted from the Table 4.4 that the total bacterial and yeast and mould reduction in cashew apple juice was obtained in the ranges from 1.71 to 4.26 and 1.5 to 3.17 log cfu/ ml respectively. In cashew apple juice the highest bacterial log reduction of 4.26 log cfu/ ml and yeast and mould log reduction of 3.17 log cfu/ ml was observed in ohmic heating treatment at voltage gradient of 12.5 V/cm and process temperature of 60°C with 5 min of holding time. The lowest reduction in bacterial and yeast and mould count was reported in the ohmic heating treatment at voltage gradient of 10 V/cm, process temperature of 55°C and holding time of 1 min. .

The effects of voltage gradient, holding time and process temperature on the total bacterial and yeast count of ohmic heated pineapple and cashew apple juices are presented as 3D surface plots in Figures 4.20 and 4.22 and 4.21 and 4.23 respectively. It could be seen from the figures, that all process parameters had a significant effect on bacterial and yeast and mould reduction for both juices ($p<0.001$). The microbiological reduction is highly dependent on the time-temperature combinations of the ohmic heating process. Higher process temperature and holding time resulted in a significant reduction in microbial load. Among the temperature and time limits under study, higher temperatures and longer heating time during ohmic heating could have successfully created a hostile environment for the microorganism through membrane destruction and inactivation of microbial enzymes (Sun *et al.,* 2008). This result is in conformation with findings of Uemura and Isobe (2003) on inactivation of *B. subtilis* spores and Ryang *et al.* (2015) on inactivation of *Bacillus cereus* spores during ohmic heating.

The microbial reduction during ohmic heating is also significantly influenced by the voltage gradient employed. The voltage gradients have a positive correlation

with microbial reduction. This could be due to the higher temperature attained within a short time period during the application of high voltage gradient. Murashita *et al*. (2017) observed higher reduction of *Bacillus subtilis* spores in sodium chloride solution at 20 V/cm than in treatment with 5 and 10 V/cm during ohmic heating.

It have been reported by different investigators that the microbial reduction during ohmic heating has an additional non thermal mechanism of inactivation (Cho *et al.,* 1999; Sun *et al.,* 2008). During ohmic heating the exposure of the fruit juices to low frequency electric fields might have resulted in electroporation of cell membranes. Electroporation is the formation of pores in the lipid bilayer and proteins of cell membrane due to changes in internal and external cell potentials when subjected to electric fields (Castro and Barbosa-Canovas, 1993; Sitzmann, 1995). The electroporation process occurring in the cell membrane leads to drastic reduction in microbial population.

Among the two fruit juices the cashew apple juice recorded higher reduction in bacterial and yeast and mould count in comparison to pineapple juice. The higher electrical conductivity of cashew apple juice might have improved the ohmic heating behaviour of cashew apple juice which could have resulted in a better microbiological reduction (Jordan *et al.,* 2001; Eribo and Ashenafi, 2003). These results are in confirmation with the findings of Sangong *et al.* (2011) in ohmic heating of orange and tomato juice.

The pH value of both juices implies an acidic nature, which might have facilitated the microbial reduction. Lee (2014) also observed higher reduction of *Escherichia coli* O157:H7, *Salmonella typhimurium*, and *Listeria monocytogenes* at lower pH values of medium during ohmic heating.

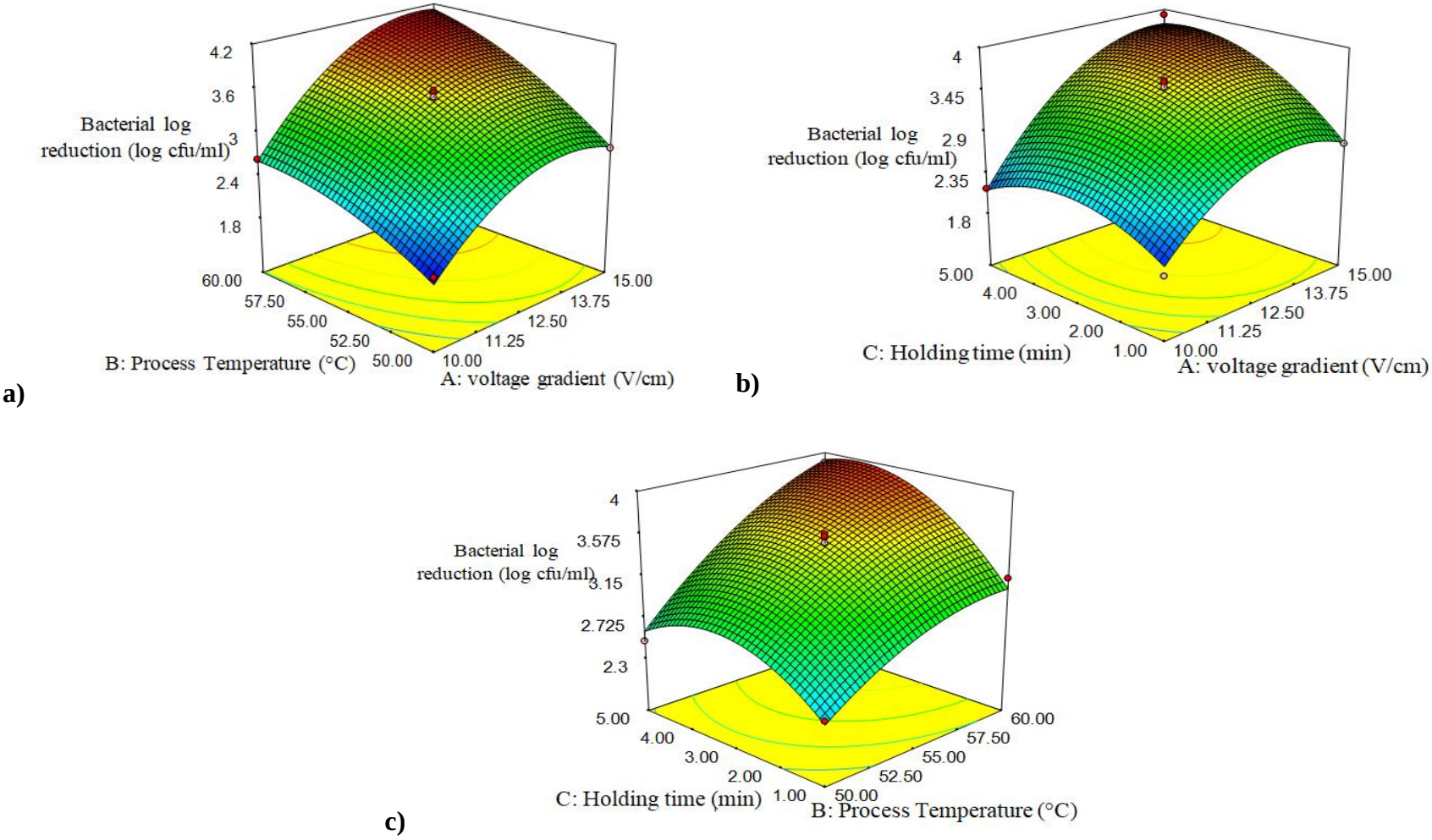


Fig. 4.20 Effect of ohmic heating parameters on bacterial log reduction of pineapple juice

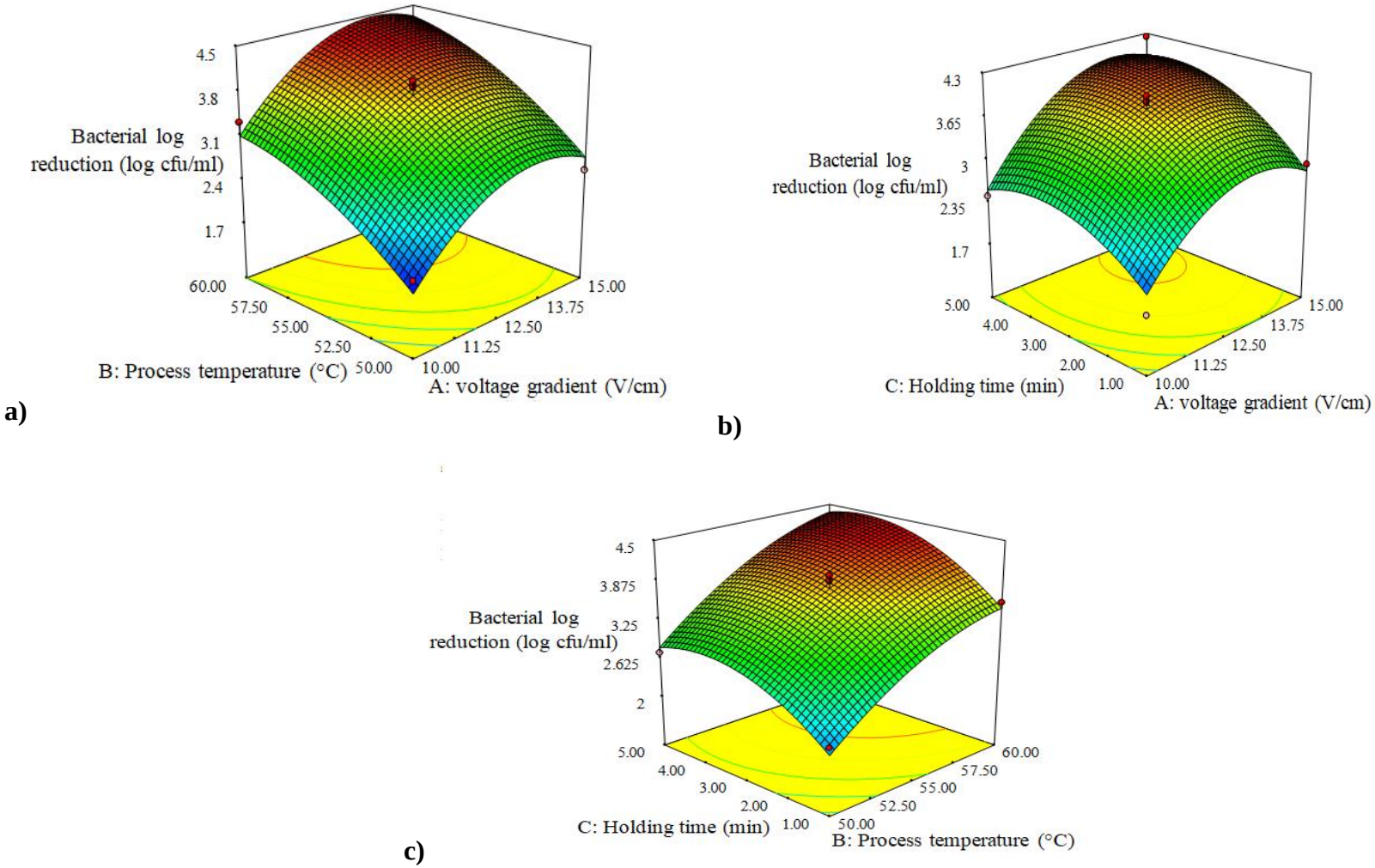


Fig. 4.21 Effect of ohmic heating parameters on bacterial log reduction of cashew apple juice

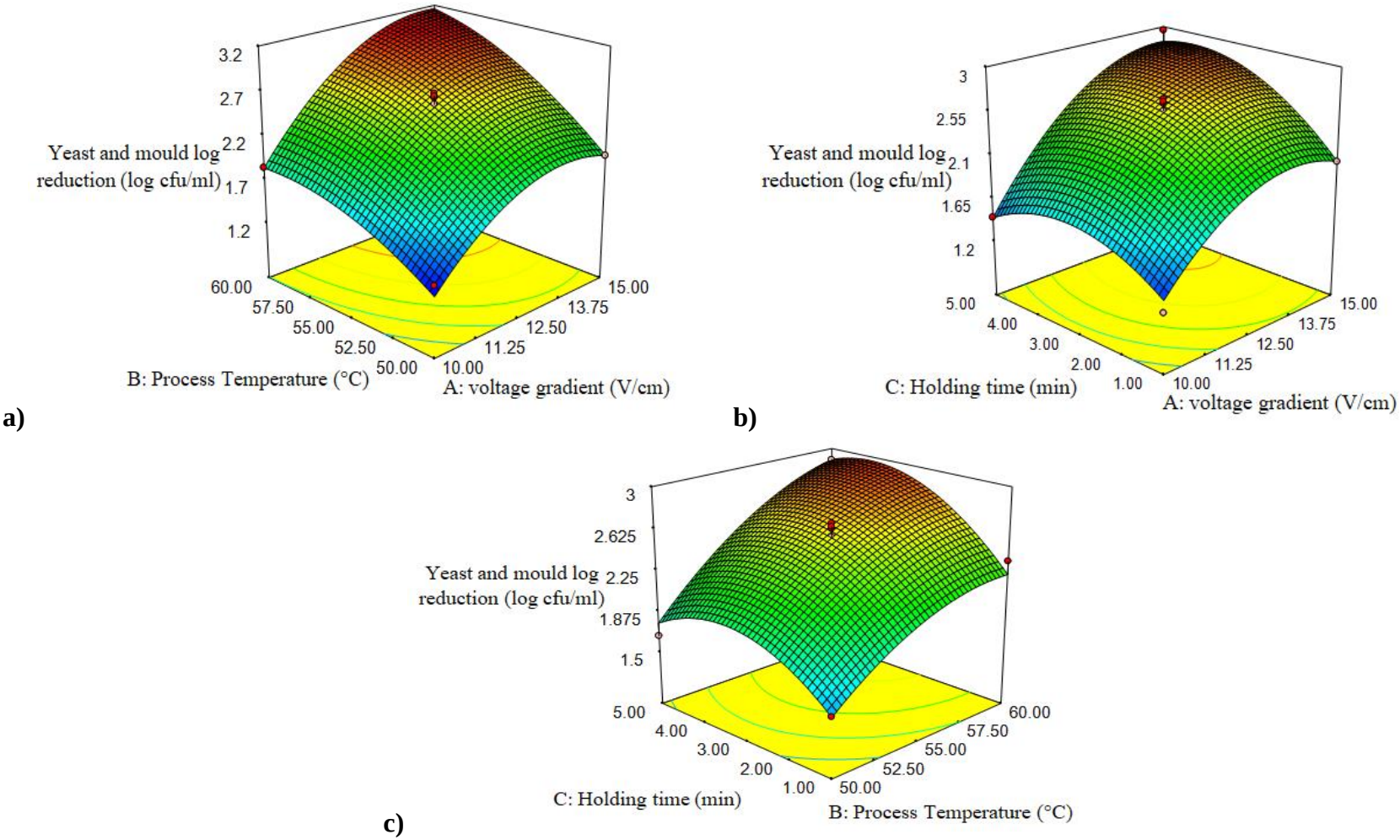


Fig. 4.22 Effect of ohmic heating parameters on yeast and mould log reduction of pineapple juice

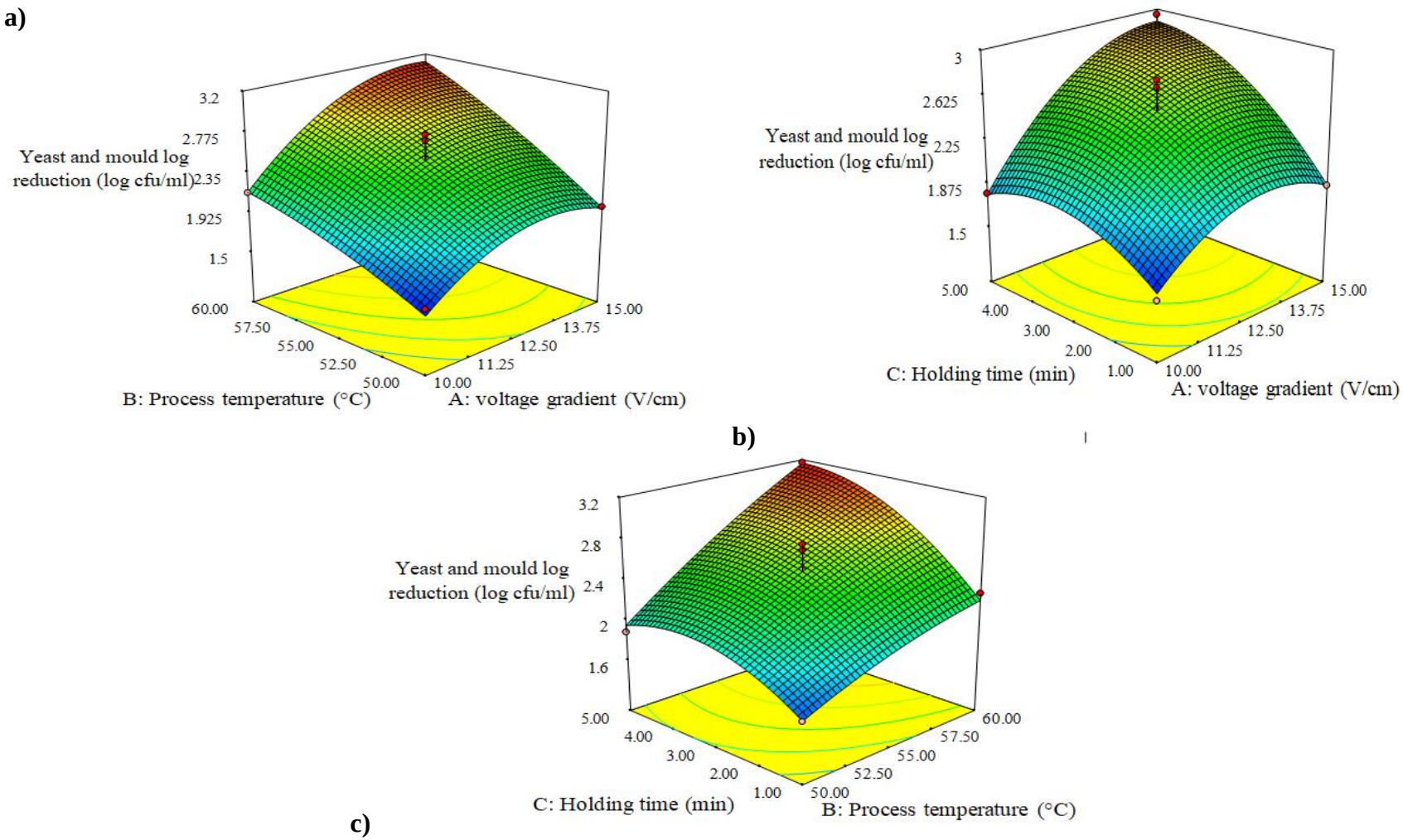


Fig. 4.23 Effect of ohmic heating parameters on yeast and mould log reduction of cashew apple juice

Second order regression equations were derived for both the fruit juices to establish a relationship between the process variables with bacterial and yeast and mould reduction. The Equations 4.11 and 4.12 represent the bacterial and yeast and mould reduction in pineapple juice and Equations 4.13 and 4.14 denotes the bacterial and yeast and mould reduction in cashew apple juice.

For pineapple juice

Bacterial log reduction = $3.52+0.61A+0.51B+0.29C+0.15A\,B+0.23\,AC+0.16B\,C-0.48\,A^2-0.18\,B^2-0.37C^2$ $R^2 = 0.98$ 4.11

Yeast and mould reduction = $2.61+0.53\,A+0.45\,B+0.23\,C+0.13A\,B+0.17\,A\,C+0.11\,BC-0.38\,A^2-0.19\,B^2-0.32\,C^2$ $R^2 = 0.95$ 4.12

For cashew apple juice

Bacterial log reduction = $3.848+0.556A+0.726B+0.41C+0.047AB+0.14AC+0.055BC-0.602A^2-0.257B^2-0.410C^2$ $R^2 = 0.98$ 4.13

Yeast and mould reduction = $2.55+0.352A+0.437B+0.312C+0.13AB+0.2AC+0.17BC-0.287A^2-0.067B^2-0.023\,C^2$ $R^2 = 0.97$ 4.14

Where,

A = Voltage gradient, V/cm

B = Treatment temperature, °C

C = Holding time, min

The Equations 4.11, 4.12, 4.13 and 4.14 indicate that both bacterial and yeast and mould log reduction of pineapple and cashew apple juice were in positive correlation with voltage gradient, holding time and process temperature. From the coefficients of the independent variables, it may be revealed that the influence of process temperature was maximum followed by voltage gradient and then by holding time in case of cashew apple juice, whereas, voltage gradient, process temperature and holding time were found to have influence on bacterial log reduction in their decreasing order in the case of pineapple juice.

From the Appendix A.13 a and b and A.14 a and b, it could be established that the regression models for bacterial and yeast and mould reduction of pineapple and cashew apple juices were significant as p-value was too low (< 0.0001) and the lack of fit was not significant indicating the adequacy of the models. Since the R^2 and adj. R^2 values are close to one and adequate precision value is more than 4, the models were adjudged to be adequately fit.

4.3.3 Optimisation of Ohmic Heating Process Parameters

Optimisation of the three process variables such as voltage gradient (10, 12.5 and 15 V/cm) holding time (1, 3 and 5 min) and process temperature (50, 55 and 60ºC) was performed using the Box-Behnken design in Design Expert Software 7.0. For optimisation of the operating parameters during ohmic heating, the responses were minimised, maximised or kept in target value to get the desired outcome. Higher desirability values were selected as optimum process conditions of ohmic heating treatment. The highest desirability value of 0.788 was obtained at treatment conditions with voltage gradient of 14.02 V/cm, holding time of 2.31 min and treatment temperature of 55.26°C for pineapple juice. Whereas, for cashew apple juice the highest desirability of 0.735 was obtained at a voltage gradient of 14.53 V/cm, process temperature of 55.25°C with holding time of 2.77 min.

The response values were tested using the recommended optimum values of process variables and was used to validate the experimental and predicted values of the responses. The predicted and actual values of the responses and the percentage variation at the optimised conditions of ohmic heating treatment for both the juices are presented in Table 4.5. The predicted values of optimised treatment are comparable with that of the actual values.

Table 4.5 Predicted and actual values of responses at the optimised conditions of ohmic heating

a. Pineapple juice

Sl. No.	Responses	Predicted value	Actual value	Variation (%)
1	pH	4.27	4.26	-0.23
2	TSS (°Brix)	11.05	11.15	0.89
3	Titrable acidity (mg/100 ml)	0.391	0.388	0.76
4	Ascorbic acid (mg/100 ml)	28.01	28.21	-0.70
5	Total sugar (mg/100ml)	10.54	10.50	-0.37
6	Total phenolic content (mg/100ml)	68.33	67.98	-0.51
7	Total colour difference (ΔE)	0.81	0.68	-1.16
8	Bacterial log reduction (log cfu/ml)	3.189	3.24	0.49
9	Yeast and mould log reduction (log cfu/ml)	2.53	2.54	0.39

b. Cashew apple juice

Sl. No.	Responses	Predicted value	Actual value	Variation (%)
1	pH	4.30	4.27	-0.69
2	TSS (°Brix)	7.69	7.52	-0.22
3	Titrable acidity (mg/100 ml)	0.422	0.424	0.473
4	Ascorbic acid (mg/100 ml)	161.63	162.74	0.69
5	Total sugar (mg/100ml)	9.95	9.97	0.20
6	Total phenolic content (mg/100ml)	167.481	168.46	0.584
7	Total tannin content (%)	0.508	0.504	-0.787
8	Total colour difference (ΔE)	0.66	0.63	-0.45
9	Bacterial log reduction (log cfu/ml)	3.71	3.69	-0.53
10	Yeast and mould log reduction (log cfu/ml)	2.51	2.48	-0.11

4.3.4 Optimisation of Pulsed Light Process Parameters for Pineapple and Cashew apple juice

The pineapple and cashew apple juices were subjected to pulsed light (PL) at dosages of 8, 20 and 32 J/cm^2, sample-source distance of 5, 10 and 15 cm and flow rates of 150, 225 and 300 ml/min in the pulsed light treatment section of the developed system as mentioned in section 3.4.2. The physico-chemical and microbial quality characteristics of pineapple and cashew apple juices were analysed. Fresh fruit juice samples were taken as control for comparison. The results obtained are presented in Table 4.6 and 4.7.

The variation in physiochemical properties such as pH, TSS, ascorbic acid, tritrable acidity, total sugar, total phenolic content, and total colour difference during pulsed light processing with different combination of process parameters for pineapple and cashew apple juice are discussed in detail in the following sections.

4.3.4.1 Effect of pulsed light treatment on pH of fruit juices

The variation in pH values of pineapple and cashew apple juices upon pulsed light treatment at different operating variable combinations are shown in Table 4.6 and 4.7 respectively and the ANOVA tables are presented in Appendix- A.15 a and b.

It could be seen from the table that there was no significant change ($p>0.05$) in the pH of fruit juices after PL treatment. Similar trend was reported by Maftei *et al.* (2014) where pH of the apple juice did not show any significant changes with respect to treatments at different PL dosages. Preetha *et al.* (2016a) also observed insignificant variation in pH of pineapple juice treated with PL radiation. Gouma *et al.* (2015) and Noci *et al.* (2008) reported that there occurred no change in pH in apple juice treated with UV-C. Shamsudin *et al.* (2014) reported no significant change in the pH (3.89) between UV treated and untreated pineapple juice.

Table 4.6 Effect of pulsed light treatment process parameters on the physico-chemical and microbial properties of pineapple juice

Run	PL dosage (J/cm^2)	Sample-Source distance (cm)	Flow rate (ml/min)	pH	TSS (°Brix)	Titrable acidity (mg/100 ml)	Ascorbic acid (mg/100 ml)	Total sugar (mg/100 ml)	Total phenolic content (mg/100ml)	Total colour difference (ΔE)	Bacterial log reduction (log cfu/ml)	Yeast and mould log reduction (log cfu/ml)
1	20	10	225	4.45	10.81	0.383	29.18	10.56	66.72	0.72	2.83	2.58
2	32	10	150	4.46	10.8	0.382	28.96	10.54	66.95	0.76	3.27	2.86
3	8.0	10	300	4.45	10.81	0.381	30.83	10.56	66.23	0.34	1.45	1.19
4	20	10	225	4.46	10.81	0.382	29.16	10.56	66.88	0.73	2.99	2.62
5	32	15	225	4.45	10.82	0.384	29.72	10.56	66.84	0.61	2.16	1.94
6	20	10	225	4.46	10.8	0.381	29.51	10.53	66.87	0.65	2.69	2.44
7	8.0	10	150	4.45	10.82	0.382	30.62	10.54	66.75	0.39	1.95	1.58
8	32	5	225	4.46	10.81	0.383	28.98	10.54	66.18	0.78	3.38	2.95
9	32	10	300	4.45	10.81	0.382	30.15	10.56	66.56	0.48	2.38	2.27
10	20	15	300	4.45	10.8	0.381	31.06	10.53	67.23	0.25	1.67	1.42
11	20	15	150	4.46	10.81	0.381	29.84	10.56	66.79	0.56	2.22	2.03
12	8.0	15	225	4.45	10.81	0.381	30.95	10.54	66.34	0.3	1.55	1.22
13	20	5	150	4.46	10.8	0.382	28.96	10.56	66.85	0.76	3.39	2.98
14	8.0	5	225	4.46	10.81	0.382	30.37	10.54	66.43	0.43	1.65	1.33
15	20	10	225	4.46	10.81	0.383	29.32	10.54	66.78	0.71	2.75	2.52
16	20	10	225	4.45	10.81	0.382	29.06	10.54	67.14	0.74	3.14	2.73
17	20	5	300	4.46	10.82	0.383	28.98	10.53	66.56	0.52	2.04	1.64

Table. 4.7 Effect of pulsed light treatment process parameters on the physico- chemical and microbial properties of cashew apple juice

Run	PL dosage (J/cm^2)	Sample-source distance (cm)	Flow rate (ml/min)	pH	TSS (°Brix)	Titrable acidity (mg/100 ml)	Ascorbic acid (mg/ 100 ml)	Total sugar (mg/ 100ml)	Total phenolic content (mg/100ml)	Tannin content (%)	Total colour difference (ΔE)	Bacterial log reduction (log cfu/ml)	Yeast and mould log reduction (log cfu/ml)
1	8	10	150	4.32	11.23	0.423	166.23	9.97	162.73	0.54	0.33	1.74	1.41
2	32	10	150	4.33	11.23	0.424	156.96	9.96	164.12	0.54	0.66	3.49	2.85
3	32	10	300	4.32	11.25	0.423	161.98	9.96	163.95	0.53	0.44	2.24	1.74
4	8	5	225	4.32	11.23	0.425	166.23	9.95	164.42	0.54	0.42	1.59	1.42
5	8	15	225	4.32	11.24	0.424	166.82	9.96	164.24	0.54	0.19	1.45	1.33
6	32	5	225	4.33	11.24	0.423	157.28	9.97	163.98	0.54	0.64	3.43	2.77
7	20	10	225	4.33	11.23	0.426	162.78	9.97	163.89	0.54	0.52	3.1	2.61
8	20	10	225	4.32	11.23	0.425	158.45	9.94	163.97	0.53	0.51	3.15	2.62
9	8	10	300	4.32	11.25	0.424	167.12	9.95	164.12	0.54	0.26	1.56	1.02
10	20	10	225	4.32	11.24	0.423	159.23	9.95	163.92	0.54	0.53	2.82	2.72
11	20	10	225	4.31	11.24	0.425	163.45	9.94	163.88	0.53	0.55	2.85	2.68
12	32	15	225	4.32	11.23	0.425	162.47	9.96	164.12	0.53	0.42	2.18	1.63
13	20	15	150	4.32	11.24	0.423	164.53	9.97	164.12	0.54	0.39	2.76	1.71
14	20	15	300	4.31	11.25	0.424	165.89	9.97	162.56	0.55	0.36	1.95	1.45
15	20	5	150	4.31	11.23	0.425	157.34	9.95	162.66	0.54	0.58	3.56	2.92
16	20	10	225	4.33	11.24	0.425	158.39	9.96	163.91	0.53	0.5	3.06	2.78
17	20	5	300	4.32	11.25	0.424	161.67	9.94	163.42	0.54	0.47	2.46	1.77

Similar results have been reported by Bhat *et al.* (2011) who found that there was no significant change in the pH of star fruit juice treated with UV. It could be inferred that non thermal processing of food provide safer and high quality product with minimum physiochemical changes.

The response surface plots of pH values of fruit juices are shown in Fig. 4.24 and 4.25. It could be revealed from the figures that no typical trend in variation of pH was observed with variation in process parameters. It could also be observed from the ANOVA tables that, model and lack of fit is non significant with low R^2 values for both fruit juices.

4.3.4.2 Effect of pulsed light treatment on TSS of fruit juices

The effect of various experimental combinations of pulsed light treatment parameters on TSS values of pineapple and cashew apple juice are tabulated in the Table 4.6 and 4.7 ANOVA Tables are presented in Appendix A.16. a and b.

Pulsed light treatment did not show any significant ($p>0.05$) effect on the total soluble solid content of fruit juices. The 3D surface plots showing the effect PL process parameters on the TSS content of pineapple and cashew apple juice are shown in Fig. 4.26 and 4.27. It may be revealed from the plots that no significant variations in TSS value of both juices were observed with changes in process parameters. The results of the present study is in agreement with Noci *et al.* (2008); Falguera *et al.* (2011) for apple juice, Pala and Toklucu (2013) for orange juice, Aguilar *et al.* (2016) for nectarine juice and Preetha *et al.* (2016a) for pineapple juice. Apple juice exposed to pulsed UV light treatments at different energy dosages (1850 mJ/cm^2 to 3354 mJ/cm^2) did not cause significant differences compared to the control sample in terms of soluble solids (Kasahara *et al.*, 2004). It could be derived from the ANOVA tables (A.26 a and b), that lack of fit was significant and F-value suggested that the model was insignificant at one per cent and five per cent level of significance.

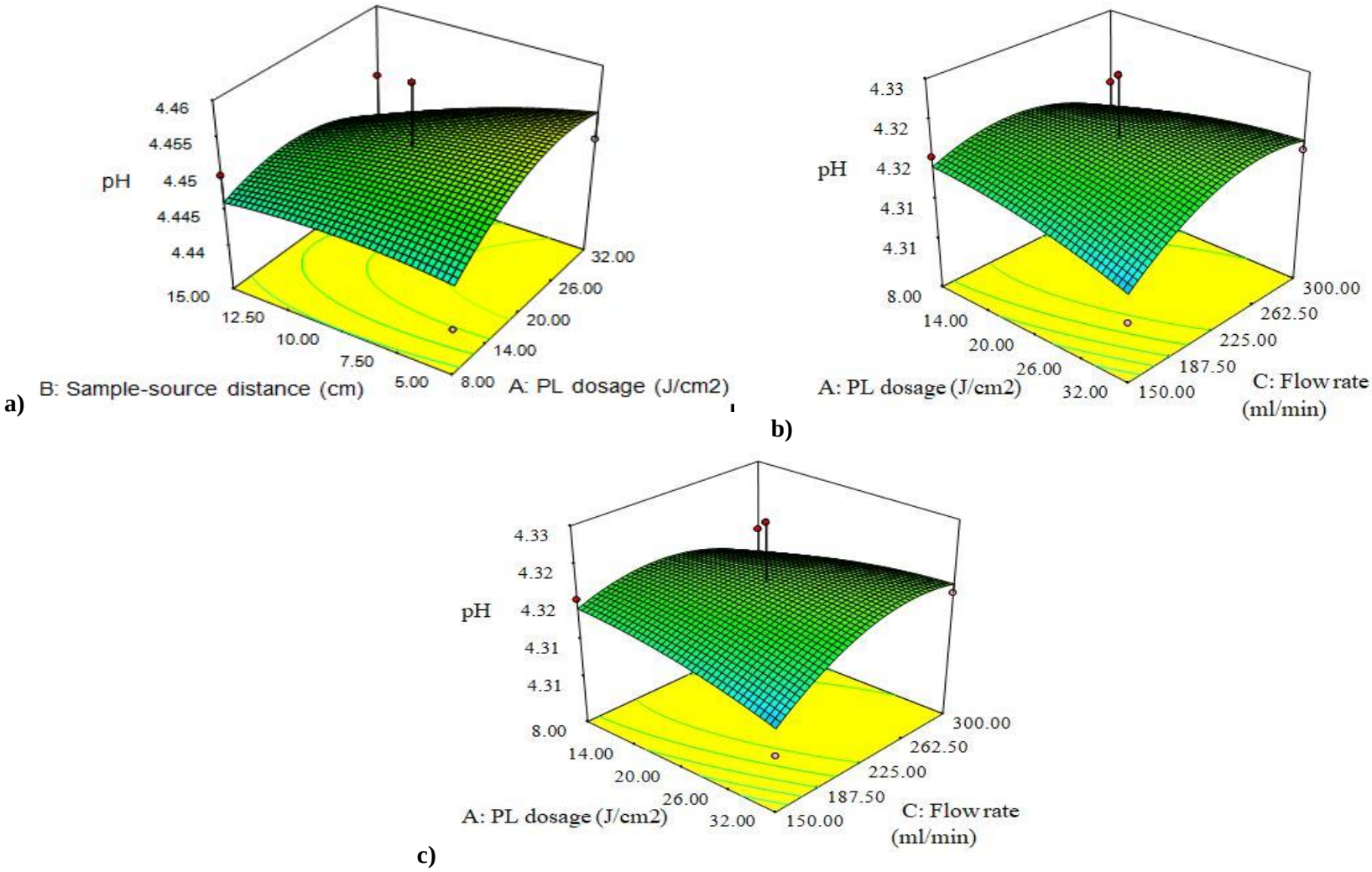
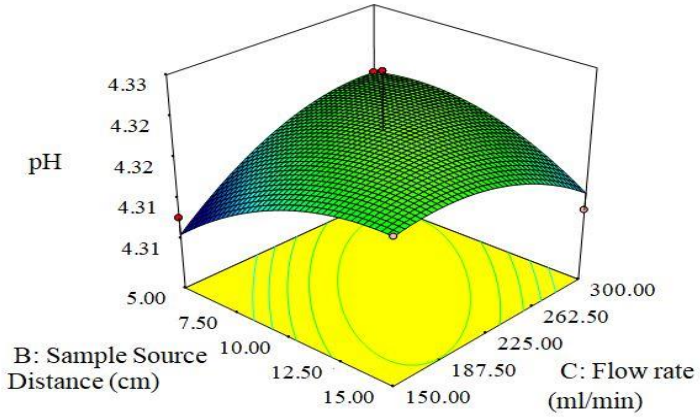
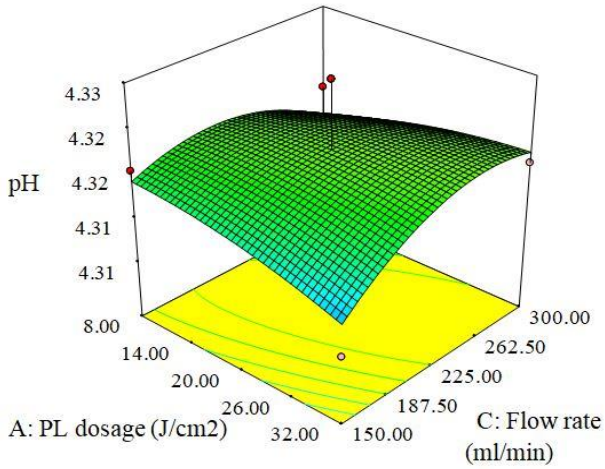


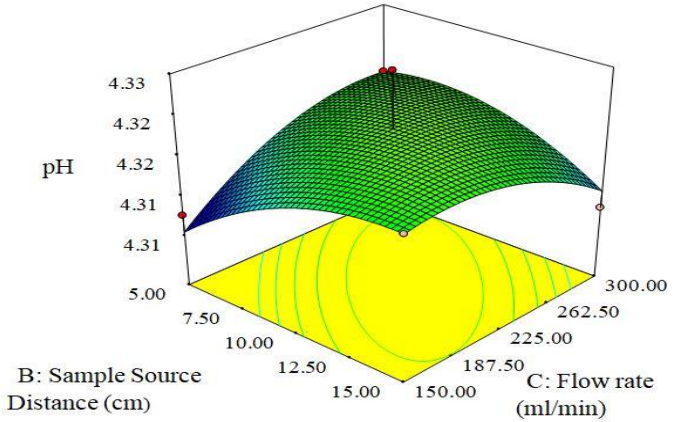
Fig. 4.24 Effect of pulsed light parameters on pH of pineapple juice



a)



b)



c)

Fig. 4.25 Effect of pulsed light parameters on pH of cashew apple juice

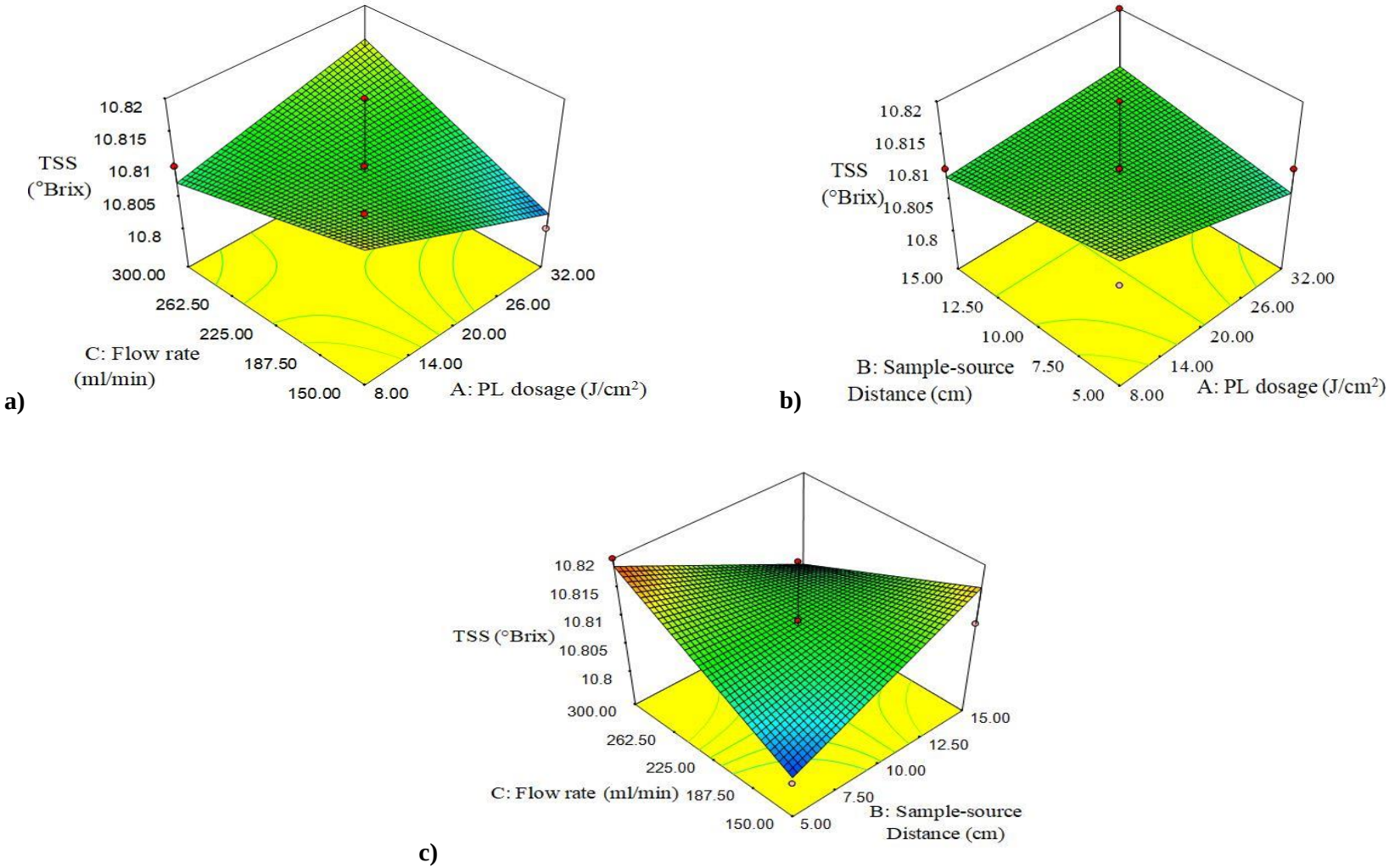


Fig. 4.26 Effect of pulsed light parameters on TSS of pineapple juice

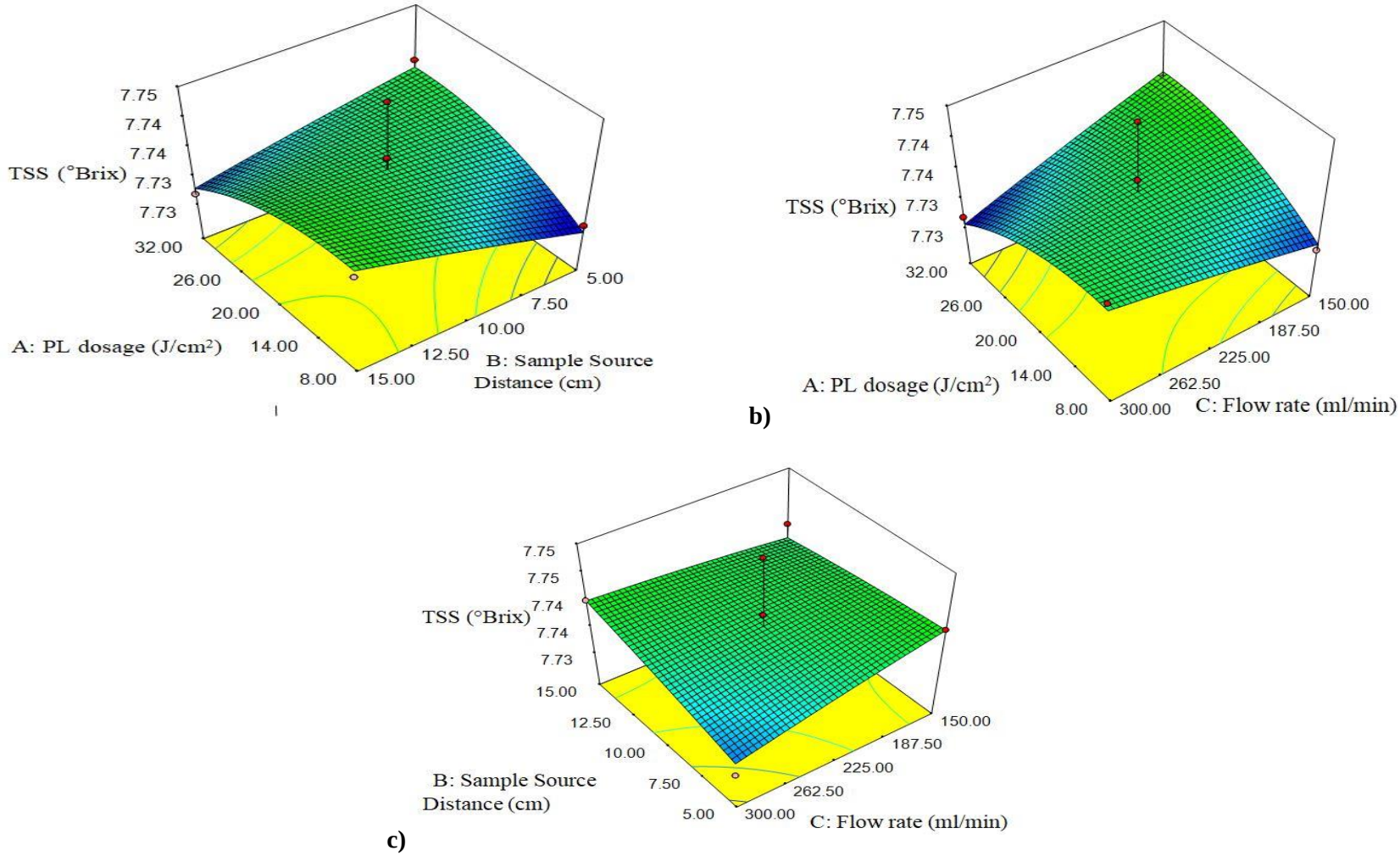


Fig. 4.27 Effect of pulsed light parameters on TSS of cashew apple juice

4.3.4.3 Effect of pulsed light treatment on titrable acidity of fruit juices

The variation in titrable acidity values of pineapple and cashew apple juice treated with different combinations of pulsed light treatment operating variables are presented in the Table 4.6 and 4.7 ANOVA tables are presented in Appendix A. 17 a and b.

It may be observed from the table that there was no significant change ($p>0.01$) in the titrable acidity of fruit juices after treatment. It was reported that orange juice treated with different doses of UV-C from 12.03 kJ/l to 48.12 kJ/l did not cause any significant change in titrable acidity values (Pala and Toklucu, 2013). Similar report of non significant changes in titrable acidity values of pulsed light treated apple juice were also reported by previous researchers (Noci *et al.,* 2008; Walkling-Ribeiro *et al.,* 2008; Falguera *et al.,* 2011; Caminti *et al.,* 2012).

The response surface plots of the effect of process parameters on titrable acidity of juice are shown in Fig. 4.28 and 4.29. It could be concluded from the figures that no significant variation in tirable acidity values could be found with changes in process parameters. Similar trends were also found by Shamsudin *et al.* (2014) and Adzahan *et al.* (2011) for pineapple juice treated by UV irradiation. Kasahara *et al.* (2004) reported that apple juice treated with PL (1850 to 3354 J/cm^2) did not cause any change in the acidity of juice. It could be established from the ANOVA tables (27 a and b) that model and lack of fit is non significant with low R^2 values for both fruit juices.

4.3.4.4 Effect of pulsed light treatment on ascorbic acid of fruit juices

Ascorbic acid (Vitamin C) in fruit juices is a very important characteristic that determines the quality retention of fruit juices upon various treatments as it is very sensitive to treatment conditions (Somogyi *et al.,* 1996). It is an unstable compound, and under desirable condition it decomposes easily (Lee and Coates, 2003). The effect of pulsed light treatment on ascorbic acid content of pineapple juice and cashew apple juice were determined and presented in the Table 4.6 and 4.7 ANOVA tables are presented in Appendix A. 18 a and b.

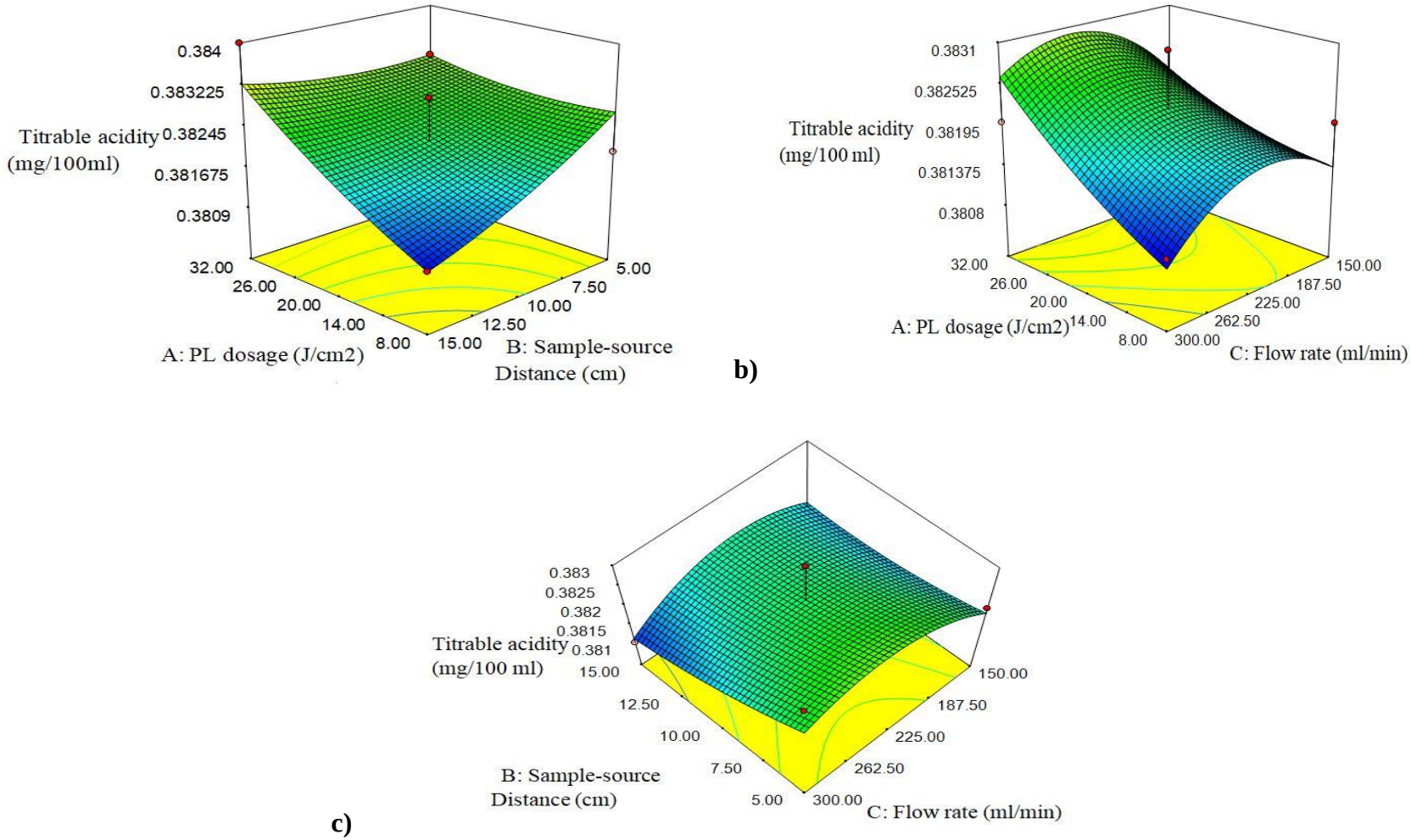


Fig. 4.28 Effect of pulsed light parameters on titrable acidity of pineapple juice

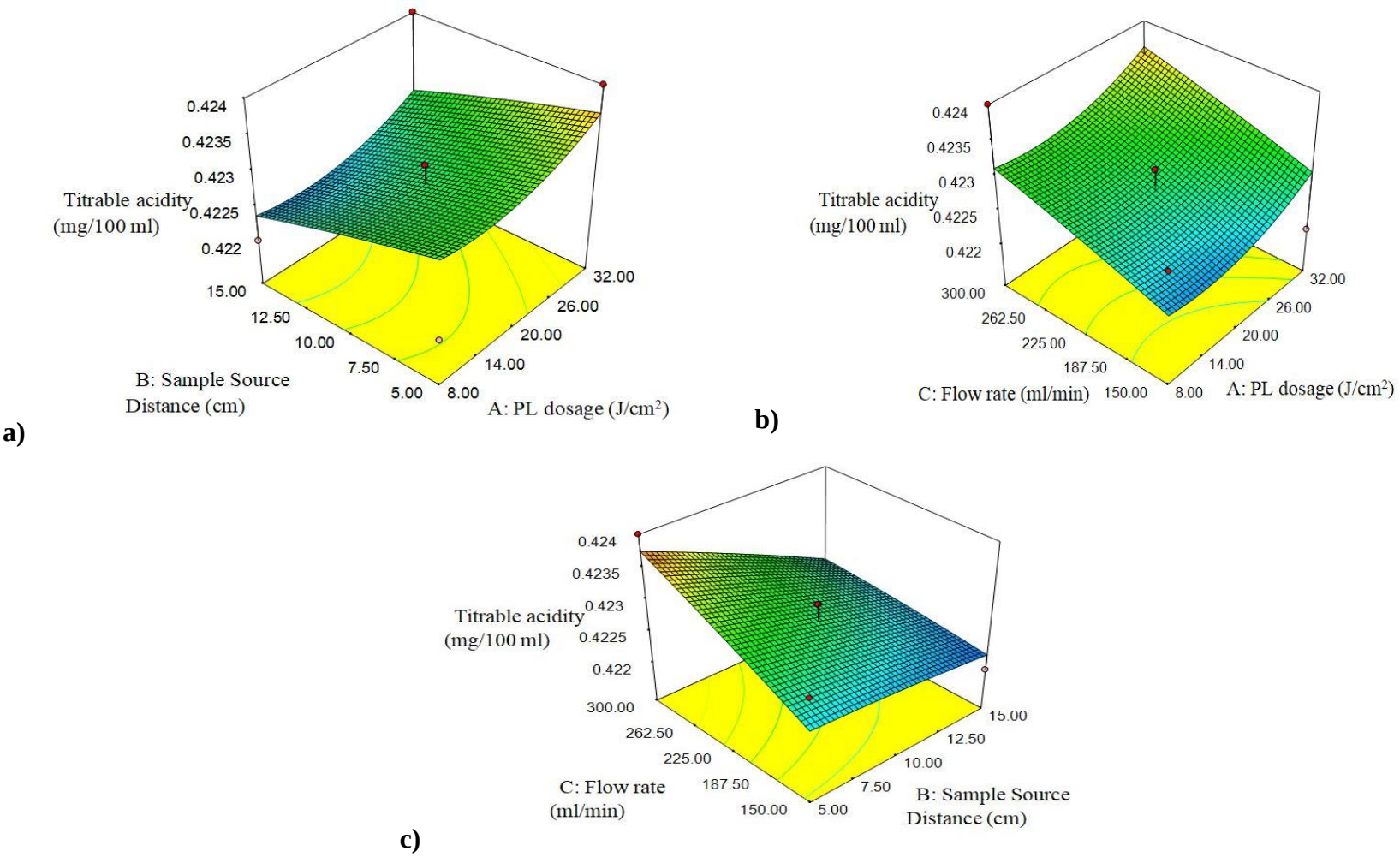


Fig. 4.29 Effect of pulsed light parameters on titrable acidity of cashew apple juice

It may be observed from the table that ascorbic acid content of the pineapple and cashew apple juice had a significant variation during PL treatments ($p < 0.01$). The depletion of ascorbic acid could be due to the formation of free hydroxyl radicals by photochemical reaction associated with oxidative processes (Koutchma *et al.,* 2009). Pineapple juice and cashew apple juice treated at PL dosage of 32 J/cm^2, sample-source distance of 10 cm and flow rate of 150 ml/ min resulted in a higher ascorbic acid reduction of 7.1 and 6.8 per cent respectively. Lowest percentage reduction of 0.38% was observed at pulsed light dosage of 20 J/cm^2, sample-source distance 15 cm and flow rate of 300 ml/min in pineapple juice, whereas, in cashew apple juice lowest reduction of 0.79% was observed at pulsed light dosage of 8 J/cm^2, sample-source distance of 10 cm and flow rate of 300 ml/min.

Batch pulsed light has been reported to cause a reduction up to 12.31% of vitamin C in apple juice (Orlowska *et al.,* 2013). Tikekar *et al.* (2011a) have reported that ascorbic acid degradation occurred more rapidly at higher UV dosage. Bhat *et al.* (2011) also noticed a significant reduction in the vitamin C for star fruit juice after ultraviolet treatment. Tran and Farid (2004) reported that UV treatment of pineapple and orange juice resulted in significant reduction ($p < 0.01$) in the vitamin C content of 19 and 17 per cent respectively. In this study, reduction was much lower than that caused during continuous UV and batch PL treatments. This could be attributed to the short time interval between the successive pulses during PL treatment, whereas continuous UV radiation could have intensified the ascorbic acid reduction due to prolonged direct exposure to UV lamp. Also, in batch PL treatment, the UV radiation is concentrated on the juice for prolonged time resulting in higher reduction of ascorbic acid.

The response surface plots of the effect of PL process parameters on ascorbic acid content of pineapple and cashew apple juice are presented in Fig. 4. 30 and 4. 31. It may be observed that the ascorbic acid content decreased with increase in pulsed light dosage but the reduction was lowered with increase in flow rate of fruit juices.

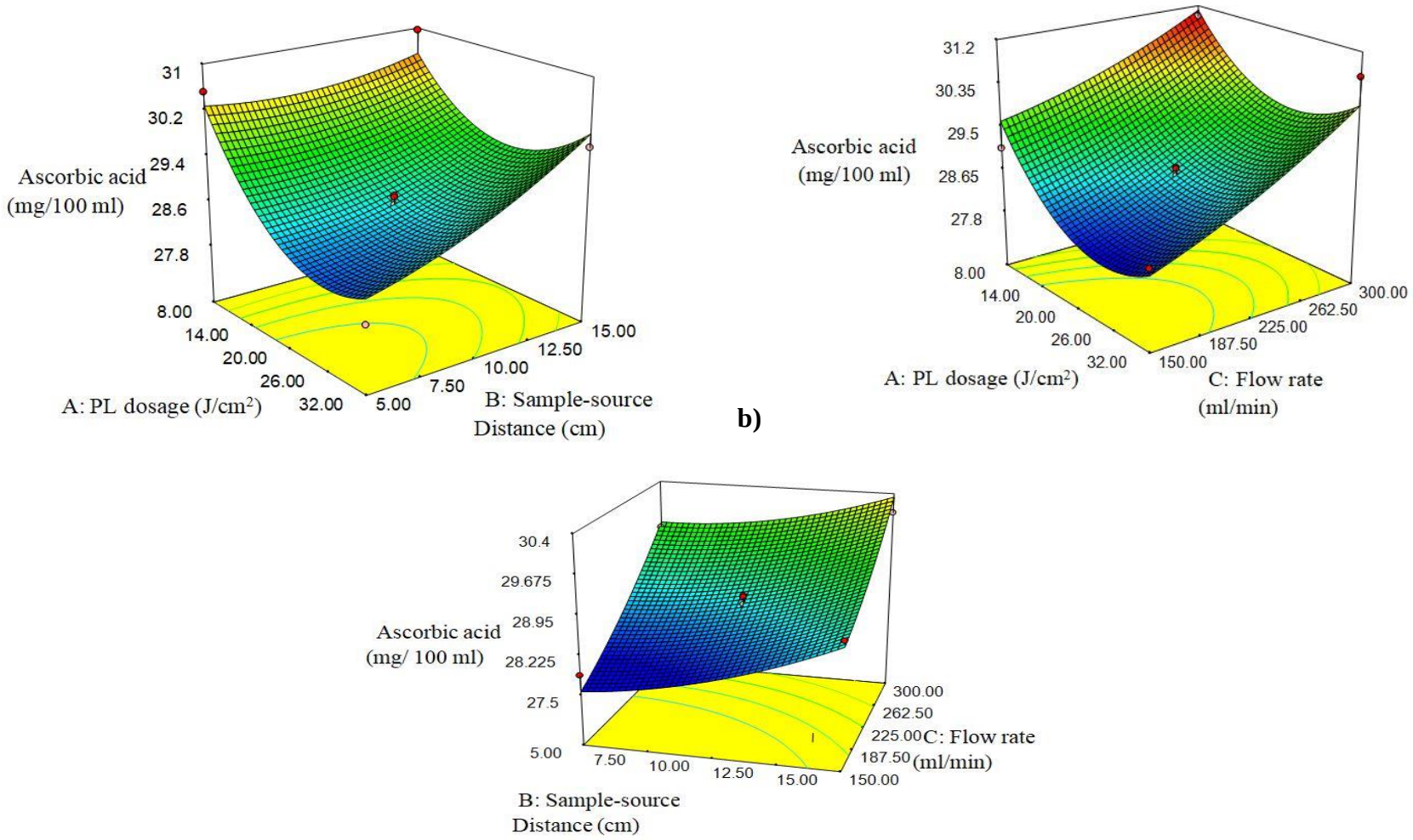


Fig. 4.30 Effect of pulsed light parameters on ascorbic acid of pineapple juice

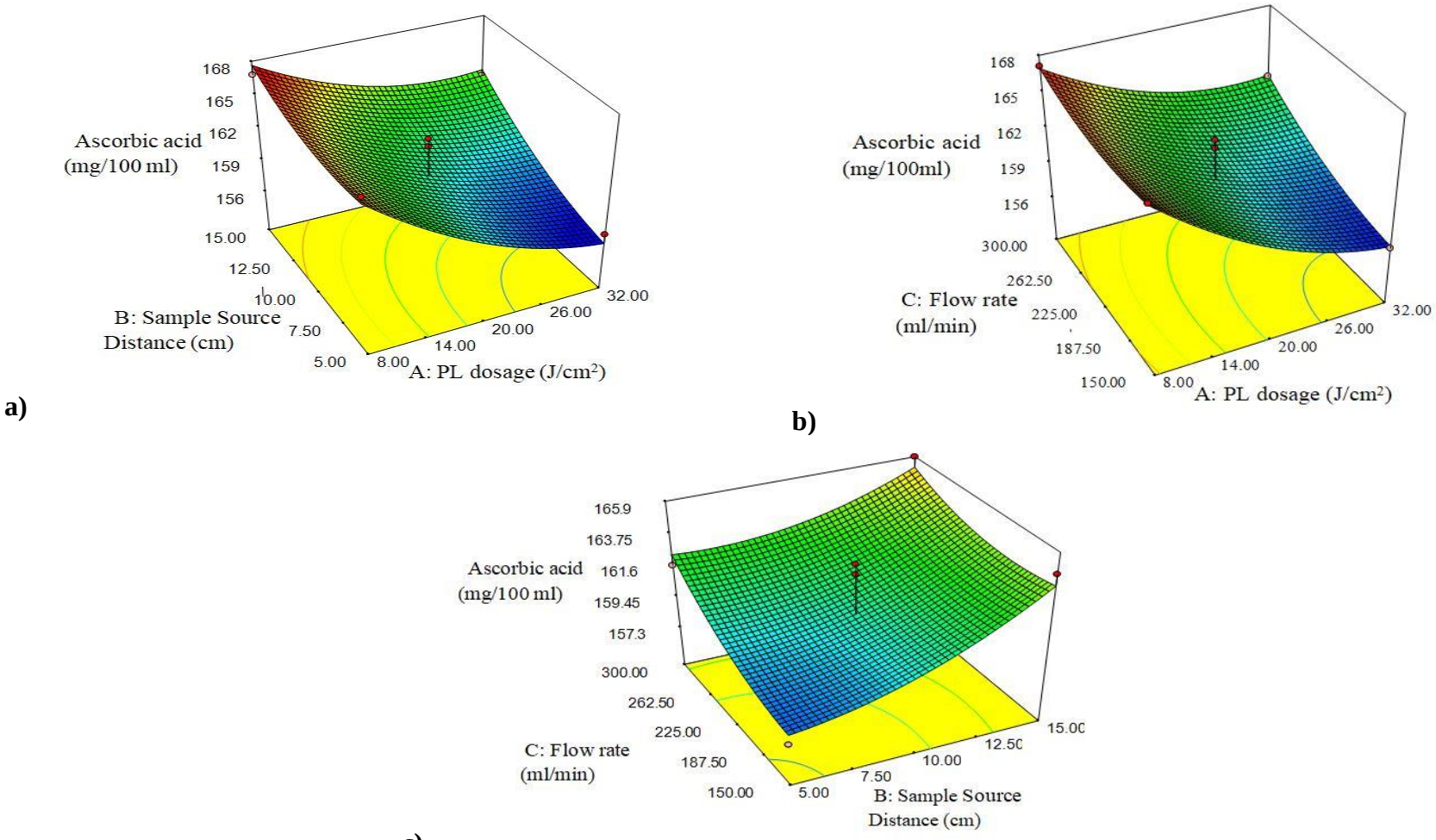


Fig. 4.31 Effect of pulsed light parameters on ascorbic acid of cashew apple juice

It may also be noted that the lower reduction in ascorbic acid content was reported at higher sample-source distance. The results are in consistent with the findings of Preetha *et al.* (2016) where a higher reduction of ascorbic acid was noted with higher dose of 240 flashes and lower sample-source distance of 5 cm. Similarly, Tran and Farid (2004) reported that Vitamin C content in reconstituted orange juice was consistently degraded as UV dose increased. Since UV region accounted for the major portion of pulsed light containing 54% of the total wave length, the degradation of ascorbic acid might be due to the UV action in fruit juices.

It was reported that ascorbic acid degradation is a complex mechanism including several free radical reactions especially in aqouse systems (Gregory and Ortwerth, 2000). During exposure to UV radiation, molecules get excited and could result in photochemical degradation mechanisms in juice samples (Tikekar *et al.,* 2011b). Shah *et al.* (2016) also reported that the light sensitive vitamin C compounds have a high UV light absorbance and therefore prolonged exposure of light may cause the reduction of ascorbic acid in juice. The reduction of ascorbic acid could also be attributed to the oxidation reactions along with activities of peroxidase and ascorbate oxidase enzymes (Davey *et al.,* 2000).

A second order regression model was developed relating the experimental ascorbic acid content of pineapple and cashew apple juices with the coded values of the corresponding combinations of the independent variables and are presented as Equation 4.15 and 4.16.

For pineapple juice

Ascorbic acid (mg/100 ml) = 29.246-0.62A+ 0.5375B+ 0.327C+ 0.045AB +0.24AC+0.3 BC+0.594A^2+0.159B^2+ 0.304C^2 $R^2 = 0.95$ 4.15

For cashew apple juice

Ascorbic acid (mg/100 ml) = 160.46-3.46A+2.15B+ 1.45C+ 1.15AB+ 1.03AC-0.74BC+1.73A^2+1.01B^2+0.89C^2 $R^2 = 0.86$ 4.16

Where, A = Pulsed light dosage J/cm^2

B = Source-sample distance, cm

C = flow rate ml/min

Equations 4.15 and 4.16 indicate that the ascorbic acid values of pineapple and cashew apple juice were in negative correlation with pulsed light dosage and in positive correlation with the flow rate and sample-source distance. From the coefficients of the independent variables, it could be derived that the coefficient value of pulsed light dosage was maximum followed by sample-source distance further followed by flow rate for both the juices.

From the ANOVA table it could be noted that the regression model was significant as p-value was too low (< 0.001) and the lack of fit was not significant indicating the adequacy of the model. The regression coefficients of both linear and quadratic term of ascorbic acid content of pineapple and cashew apple juice were highly significant. Since the R^2 and adj. R^2 values are close to one and adequate precision value was more than 4, the model may be considered as adequately fit.

4.3.4.5 Effect of pulsed light treatment on total sugar of fruit juices

The effect of PL treatment combinations on total sugar content of both pineapple and cashew apple juice are depicted in Table 4.6 and 4.7 and the analysis of variance (ANOVA) is presented in Appendix-A.19 a and b.

It could be seen from the table that there was no significant change ($p>0.05$) in the total sugar content of fruit juices after PL treatment. Similar results were also reported by Sean and Prince (2015) in pineapple juice treated with combined treatment of UV and ohmic heating. However, in contrast, Yang *et al.* (2019) reported a decrease in total sugar content in apple juice treated with UV and thermal processing.

The 3D response surface plots (Fig.4.32 and 4.33) of the effect of PL process parameters on total sugar content revealed that PL treatment have no significant effect on the sugar contents of both the juices. The PL processing have not shown any

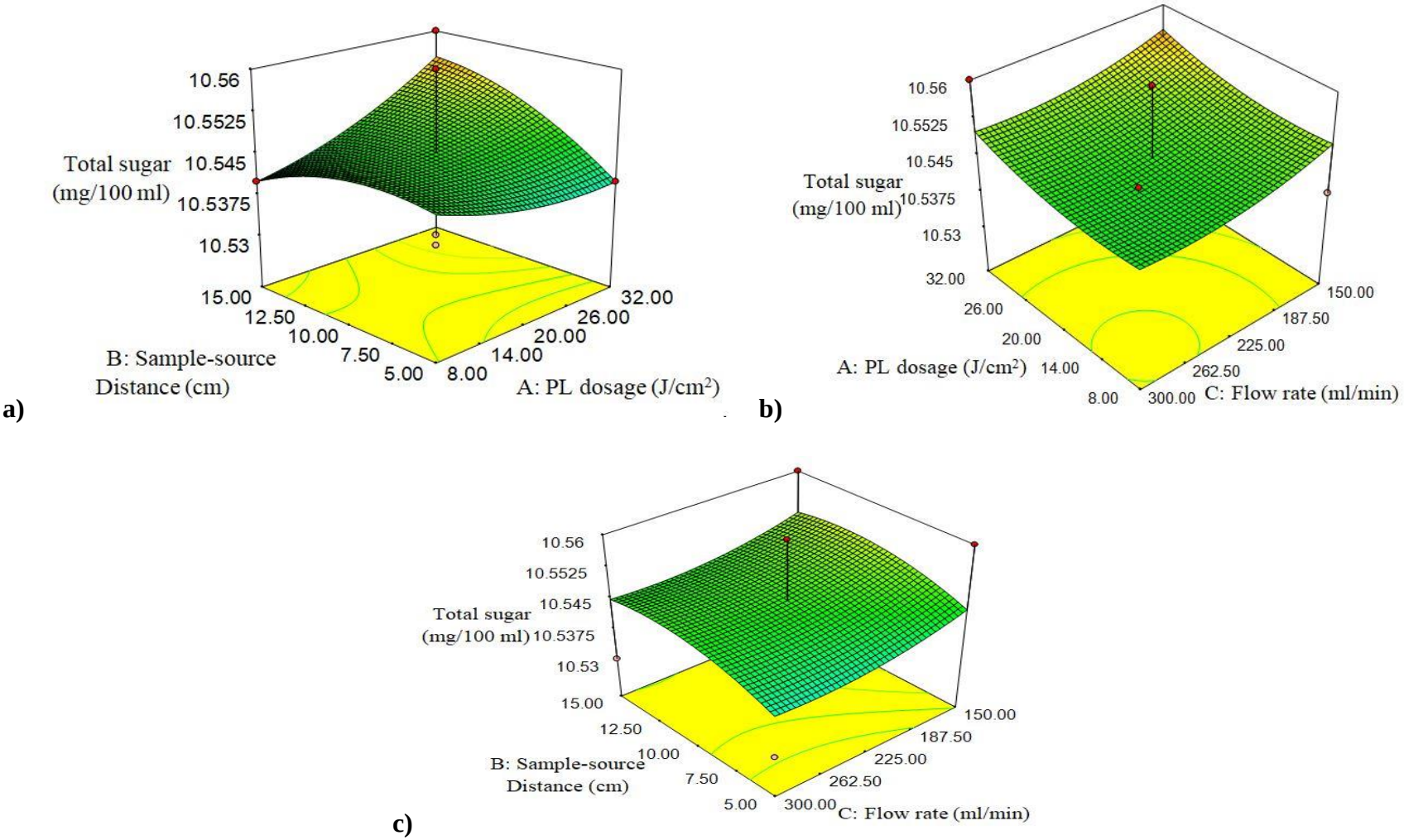


Fig. 4.32 Effect of pulsed light parameters on total sugar of pineapple juice

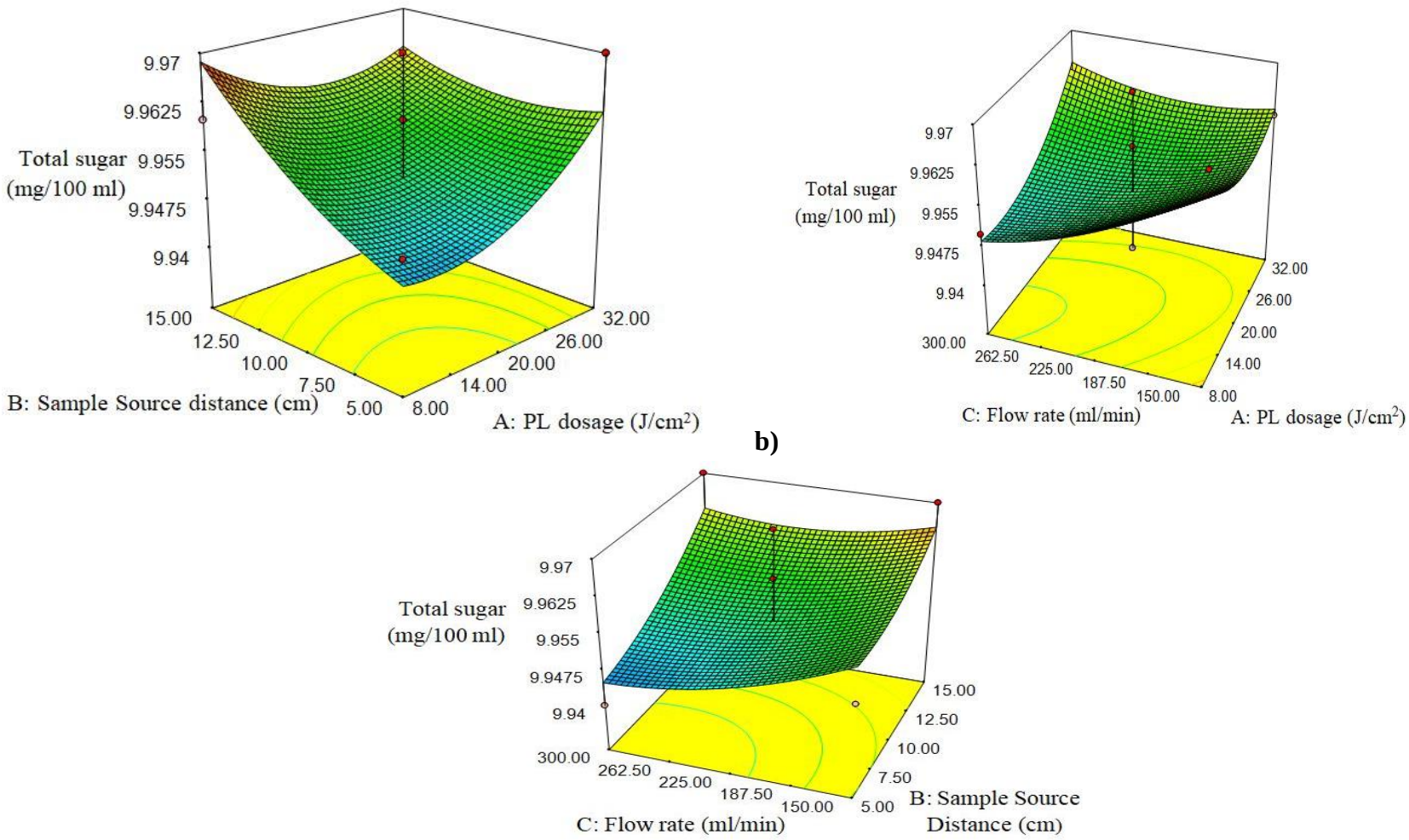


Fig. 4.33 Effect of pulsed light parameters on total sugar of cashew apple juice

significant influence on most of the chemical properties of fruit juices such as pH, TSS, titrable acidity, Phenolic content etc. It could be inferred that PL processing did not cause chemical alteration in the fruit juice system which could be the reason for minimum changes in total sugar content of pineapple and cashew apple juice. It may also be observed from the ANOVA tables (A.18 a and b) that model and lack of fit is insignificant with low R^2 values for both fruit juices.

4.3.1.5 Effect of pulsed light treatment on phenolic compounds of fruit juices

The effect of various experimental combinations of PL treatment parameters on the total phenolic content of pineapple and cashew apple juice are presented in Table 4.6 and 4.7. The analysis of variance (ANOVA) are shown in Appendix-A.20 a and b. No significant ($p > 0.01$) variation of phenolic content was observed among different treatment combinations of PL process. In similar lines Caminiti *et al*. (2010) observed insignificant phenolic content variation in apple juice during UV-C treatment at 5.31 and 53.10 J/cm^2 dosages. Pitaya juice treated with UV treatment showed no significant changes in phenolic compounds with variations in flow rate (Ochoa-Velasco and Quimica, 2012).

The variation in total phenolic content with different treatment combinations of PL process parameters are depicted in Fig.4.34 and 4.35. In this study, though insignificant, slight reduction on phenol content was noted during the PL treatment irrespective of the treatment combinations. A decrease in phenol compounds of apple juice was reported during UV treatment for 30 minutes (Noci *et al*., 2008). Similar reduction was also found in apple juice, blueberry, and raspberry nectars during UV processing (Caminiti *et al*., 2010; Haro-Maza and Guerrero-Beltran, 2016). According to Koutchma (2009) and Ochoa-Velasco and Quimica (2012), the UV-C light affects the structure of phenolic compounds, which could have resulted in the reduction of phenolic compounds. Scientists have reported that phenolic degradation could also be due to the phtoto induced molecular rearrangement or photo-oxidation and the process may depend on many factors *viz.* structure of phytochemicals,

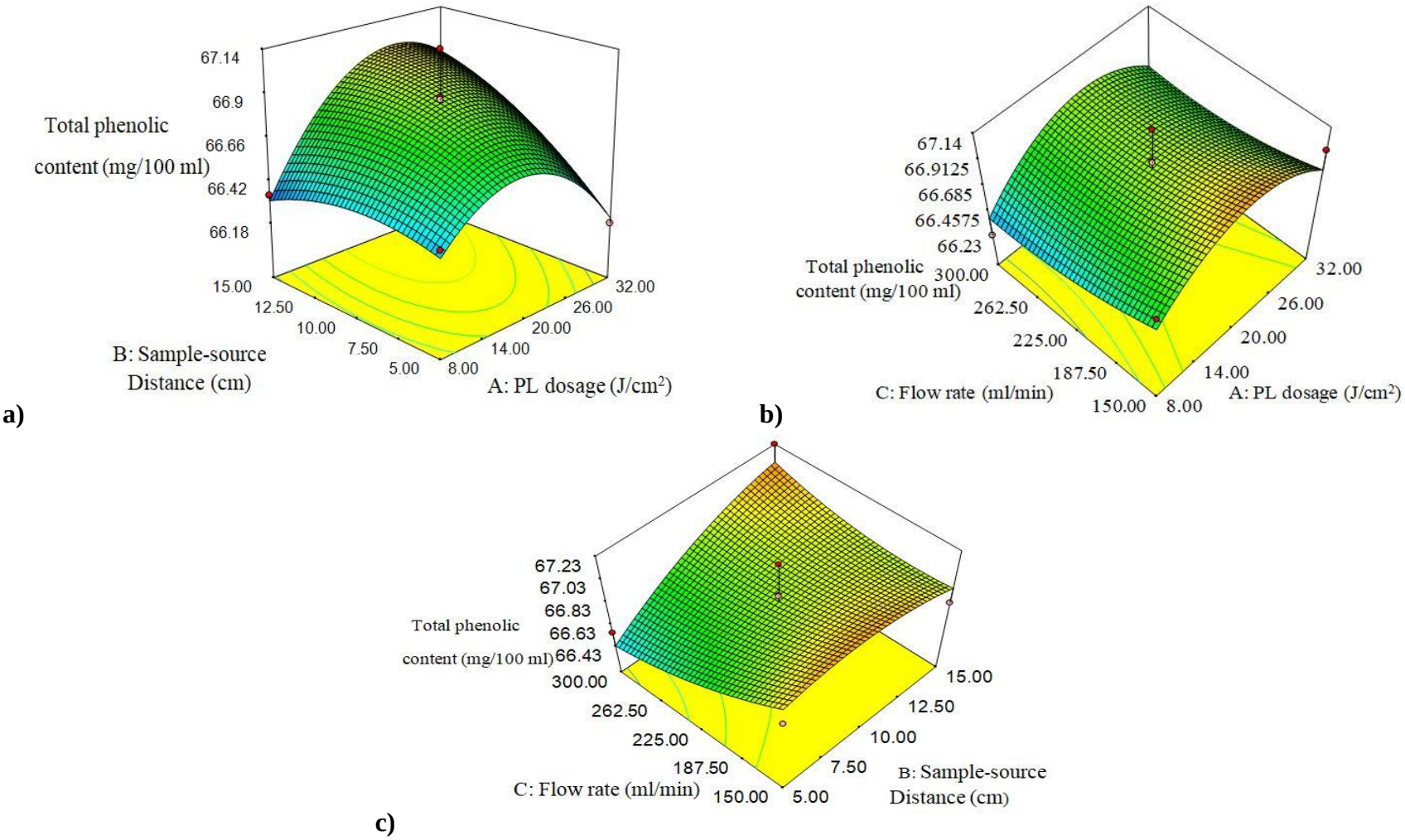


Fig. 4.34 Effect of pulsed light parameters on total phenolic content of pineapple juice

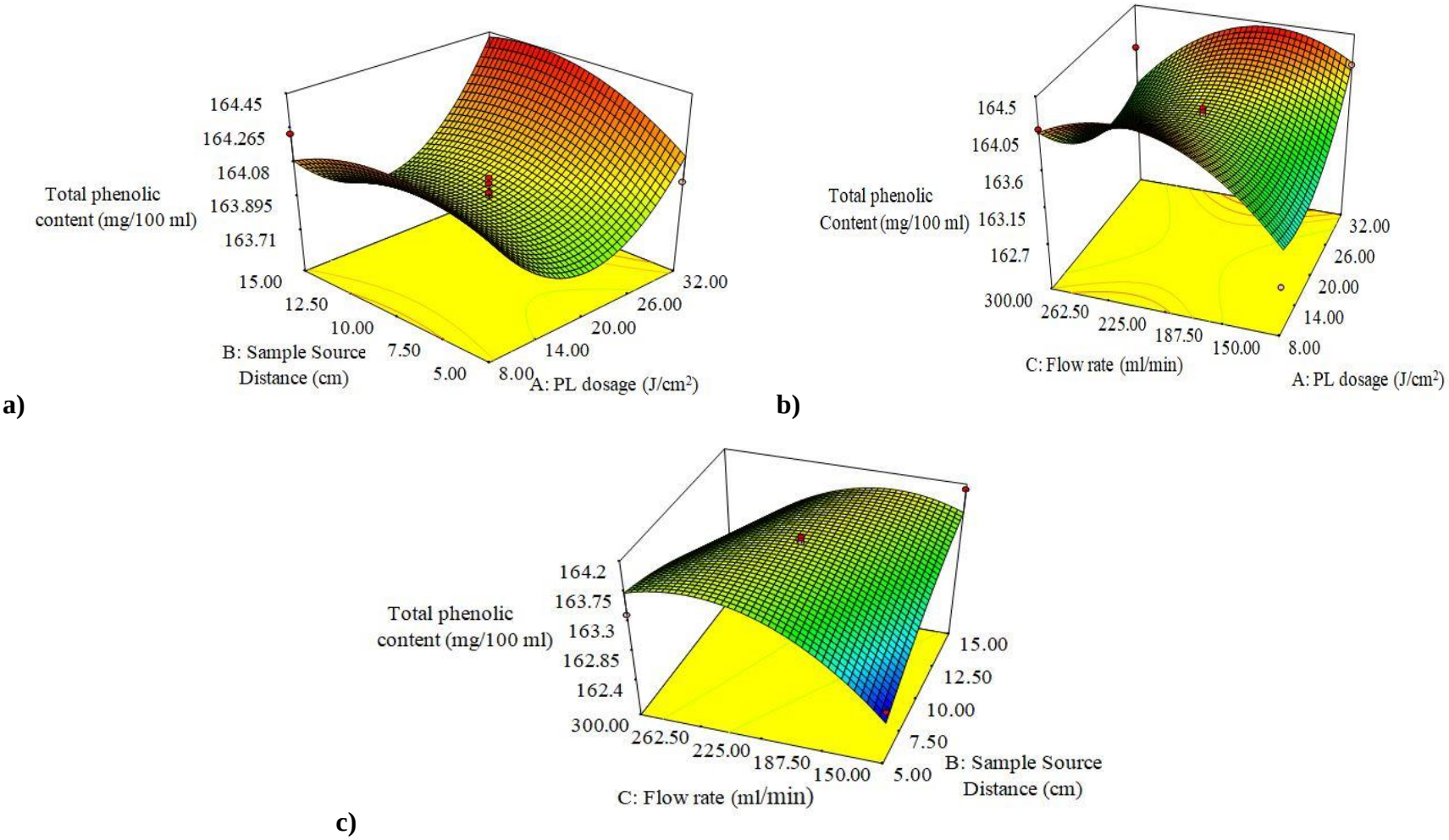


Fig. 4.35 Effect of pulsed light parameters on total phenolic content of cashew apple juice

wavelength of light, pH, presence of oxygen etc. (Aramwit *et al.*, 2010; Bordignon-Luiz *et al.*, 2007). It could be seen from the ANOVA tables (A.19 a and b) that model and lack of fit is non significant with low R^2 values for both fruit juices.

4.3.4.6 Effect of pulsed light treatment on tannin content of fruit juices

The variation in tannin content of pineapple and cashew apple juice upon pulsed light treatment at different operating parameter combinations are shown in Table 4.6 and 4.7 and the ANOVA table is presented in Appendix-A.21.

It may be revealed from the table that the tannin content of cashew apple juice ranged between 0.53 and 0.54. It may be concluded that the tannin content did not show any significant variation during pulsed light treatment and no typical trend could also be observed in tannin content values. The 3D response surface plots (Fig. 4.36) showing variation of tannin content with process variables implies that pulsed light treatment have an insignificant effect on the tannin content of cashew apple juice. It could be seen from the ANOVA table A.20 that lack of fit was significant and F-value suggested that the model was insignificant at one per cent and five per cent level of significance.

4.3.4.7 Effect of pulsed light treatment on total colour difference of fruit juices

The effect of PL treatment variables on colour values (L*, a*, b*) of pineapple and cashew apple juice was evaluated and presented in the Appendix A. 22. The total colour difference of the PL treated pineapple and cashew apple juice at the prescribed PL dosage, flow rate and sample-source distance are illustrated in Table 4.6 and 4.7 and the ANOVA tables are presented in Appendix A.23 a and b.

It could be revealed from results (Appendix. 22) that a slight variation in L*, a* and b* values were observed after PL treatment similar to that observed in other non thermal treatments. The L* values ranged from 39.2 to 39.65, a* values ranged from -3.65 to -3.32, and b* values ranged from 22.6 to 22.82 in pineapple juice.

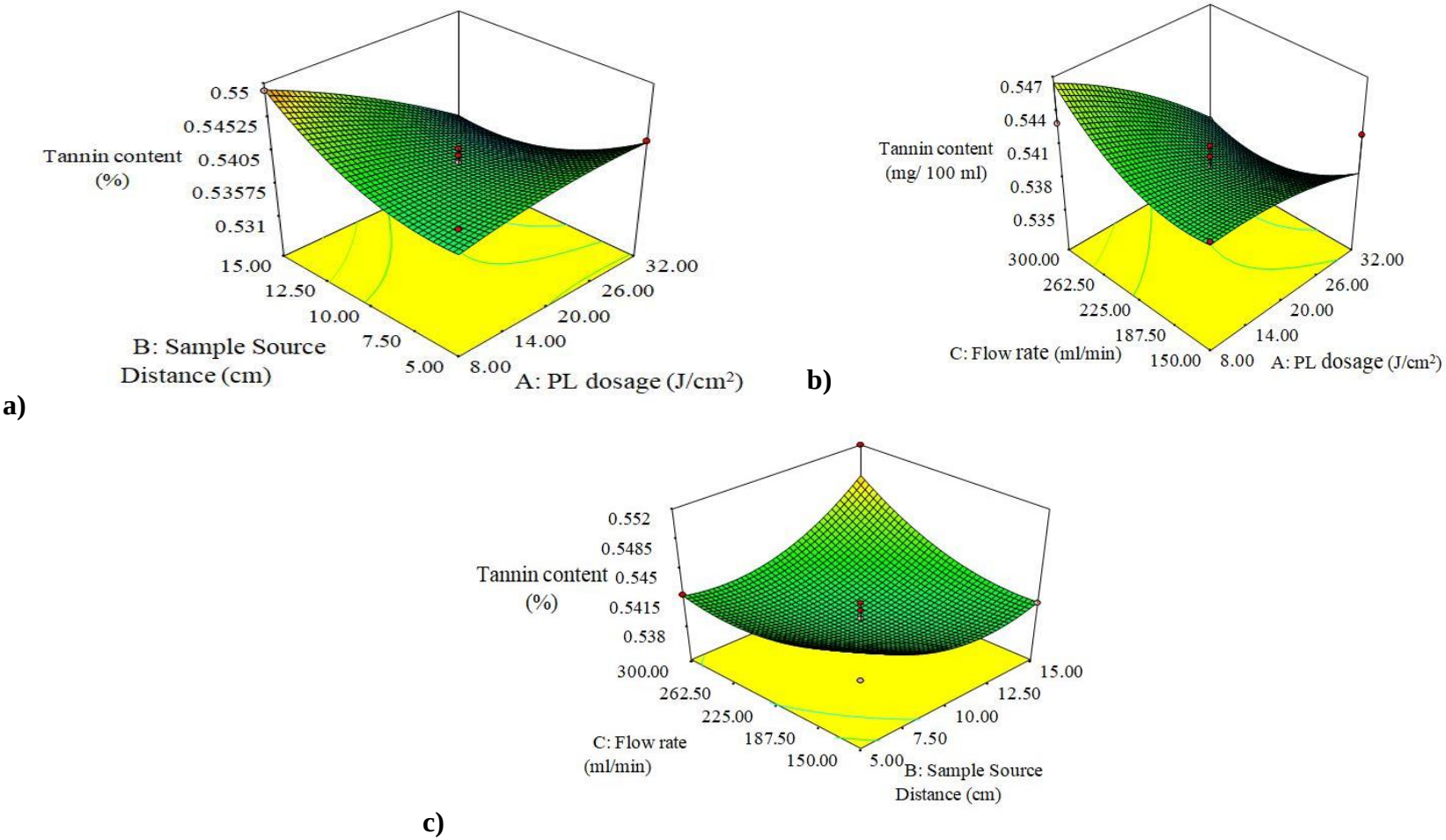


Fig. 4.36 Effect of pulsed light parameters on tannin content of cashew apple juice

In cashew apple juice the L* values ranged between 1.27 and 1.45, a* values between 0.33 and 0.49 and b* values between -1.5 and -1.12. A decrease in L* and b* values were observed in all treated samples whereas a* values showed slight increase. The trend in variation of L*, a* and b* values indicates that lightness and yellowness of juice decreased along with slight increase in redness of juices.

The total colour difference ranged from 0.25 to 0.78 and 0.19 to 0.66 in pineapple and cashew apple juice respectively. The highest percentage increase of colour difference was noted in pineapple and cashew apple juices treated with a PL dosage of 32 J/cm^2, flow rate of 225 ml/min and sample-source distance of 5 cm. The lowest percentage increase was reported in treatment with a PL dosage of 20 J/cm^2, flow rate of 300 ml/min and sample-source distance of 15 cm in pineapple juice. On the other hand, cashew apple juice treated with a PL dosage of 8 J/cm^2, flow rate of 150 ml/min and sample-source distance of 10 cm resulted in lowest colour variation.

The values of total colour difference of all samples ranged between 0.25 and 0.78. According to Cserhalmi *et al.* (2006) total colour difference of 1.5 or above could be considered as a noticeable visual difference. Hence the results indicate that only a very slight variation in colour upon pulsed light treatment with respect to fresh fruit juices. Though the PL parameters statistically showed a significant effect on the total colour difference of pineapple and cashew apple juices ($p < 0.001$), the variation is less than 1.5 manifesting only a slightly visible colour difference. In a study on UV-C treatment of orange-carrot juice blend no noticeable visual colour difference was recorded (Caminiti *et al*., 2012).

The response surface plots of effect of total colour difference with variations in PL process parameters are depicted in Fig. 4.37 and 4.38. It could be revealed from the figures that the total colour difference increased with increase in PL dosage and decrease in flow rate and sample-source distance.

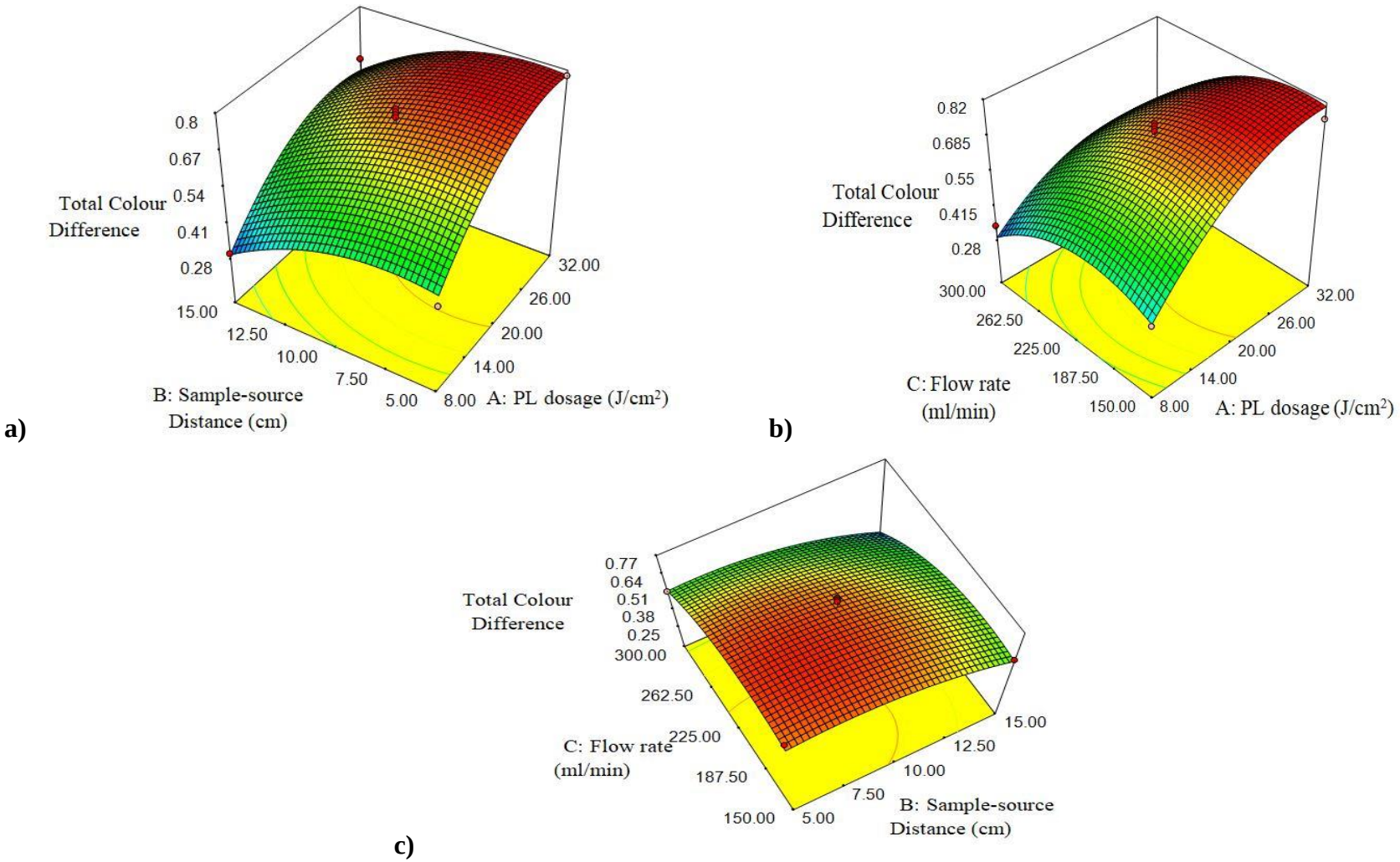


Fig. 4.37 Effect of pulsed light parameters on colour difference of pineapple juice

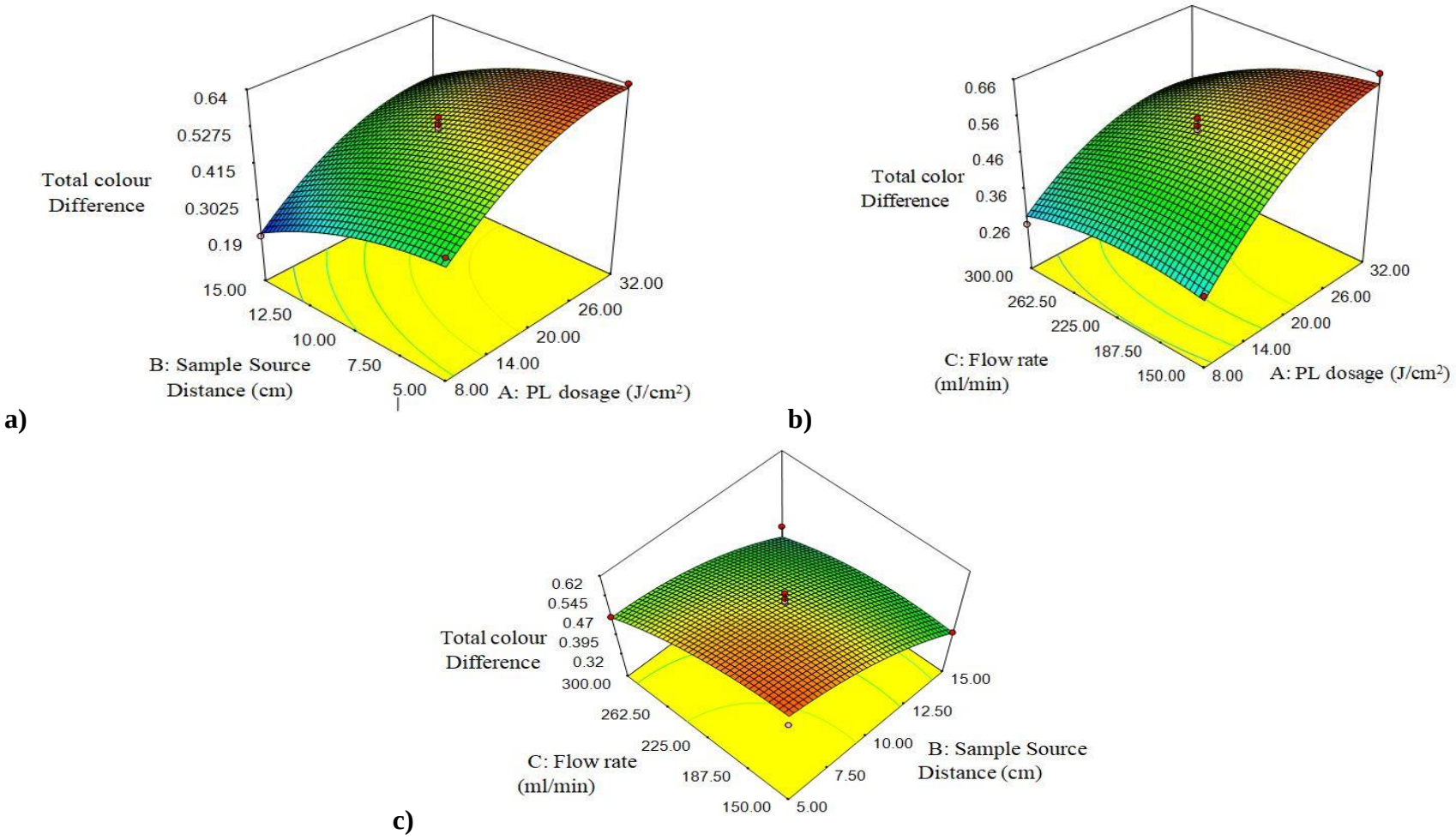


Fig. 4.38 Effect of pulsed light parameters on total colour difference of cashew apple juice

In similar lines only a minimal change in colour difference was observed in pitaya and grape juice exposed to UV light with higher flow rate condition (Guerrero-Beltrán and Barbosa-Canovas, 2005; Ochoa-Velasco and Quimica, 2012). It could be due to the very short exposure time between UV-C light and the fruit juice samples at higher flow rate conditions (Guerrero-Beltran and Barbosa-Canovas, 2005). It may also be noted that the PL treatments at lower sample-source distance and higher time of exposure have influenced the total colour difference compared to other treatments.

A second order regression model was developed relating the experimental colour difference of pineapple and cashew apple juice with the coded values of the corresponding combinations of the independent variables and presented in the Equations 17 and 18.

For pineapple juice

Total colour difference = $0.71+0.15A-0.096B-0.11C-0.010AB-0.058AC-0.018BC-0.11A^2-0.075B^2-0.11C^2$ $R^2 = 0.98$ 4.17

For cashew apple juice

Total colour difference = $0.522+0.12A-0.093B-0.053C+0.002AB-0.037AC+0.02BC-0.066A^2-0.038B^2-0.033C^2$ $R^2 = 0.97$ 4.18

Where, A = Pulsed light dosage J/cm^2

B = Source-sample distance, cm

C = flow rate ml/min

The equations showed that the total colour difference of pineapple and cashew apple juice were in negative correlation with pulsed light dosage and in positive correlation with the flow rate and sample-source distance. From the coefficients of the independent variables, it could be noted that the influence of pulsed light dosage was maximum followed by sample-source distance and further followed by flow rate of the juice.

Data presented in Appendix. A. 23. a and b revealed that the regression models of both juices were significant as p-value was too low (< 0.001) and the lack of fit was not significant indicating the adequacy of the models. The regression coefficients of both linear and quadratic terms of total colour difference of pineapple and cashew apple juice were highly significant. Since the R^2 and adj. R^2 values of total colour difference for both juices are close to one and adequate precision value was more than 4, the models could be considered as adequately fit.

4.5 EFFECT OF PULSED LIGHT PROCESS PARAMETERS ON THE MICROBIAL LOG REDUCTION OF FRUIT JUICES

The total bacterial and yeast and mould count of both pineapple and cashew apple juices were determined by the method as referred in section 3.7.1. The total bacterial and yeast and mould log reduction of the pineapple and cashew apple juice obtained at different experimental combinations of operating variables during pulsed light treatment are presented in Table 4.6 and 4.7 respectively and the ANOVA tables are presented in Appendix- 24. a and b, 25. a and b.

It could be seen from the Table 4.6 that the total bacterial and yeast and mould log reduction in pineapple juice were found to be in the range of 1.45 to 3.39 and 1.19 to 2.98 log cfu/ ml respectively. The highest bacterial log reduction of 3.39 log cfu/ml and highest yeast and mould reduction of 2.98 log cfu/ml were observed in the samples treated at a PL dosage of 20 J/cm^2, flow rate of 150 ml/min with 5 cm sample-source distance. The PL treatment at a dosage of 8 J/$cm^{2,}$, flow rate of 300 ml/min, and 10 cm sample-source distance recorded the lowest bacterial reduction of 1.45 log cfu/ml and yeast and mould reduction of 1.19 log cfu/ml.

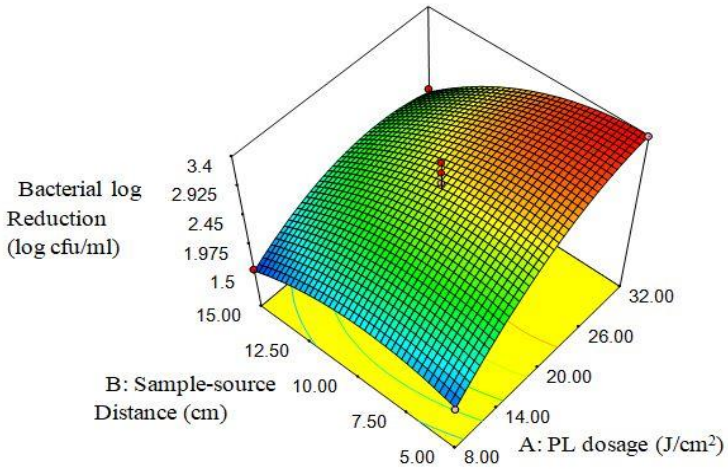
It could be noted from Table 4.7 that the total bacterial and yeast and mould reduction in cashew apple juice ranged from 1.45 to 3.56 and 1.02 to 2.92 log cfu/ml respectively. In cashew apple juice, the highest bacterial log reduction of 3.56 log cfu/ml and yeast and mould reduction of 2.92 log cfu/ml were observed in PL treatment at a dosage of 32 J/cm^2, flow rate of 225 ml/min with 5 cm of sample-source distance. The lowest bacterial reduction of 1.45 log cfu/ml and yeast and mould reduction of 1.02 log

cfu/ml were observed in cashew juice, when treated at a PL dosage of 8 J/cm^2 corresponding to a flow rate of 225 ml/min and 15 cm sample-source distance and PL dosage of 8 J/cm^2 corresponding to a flow rate of 300 ml/min and 10 cm sample-source distance respectively.

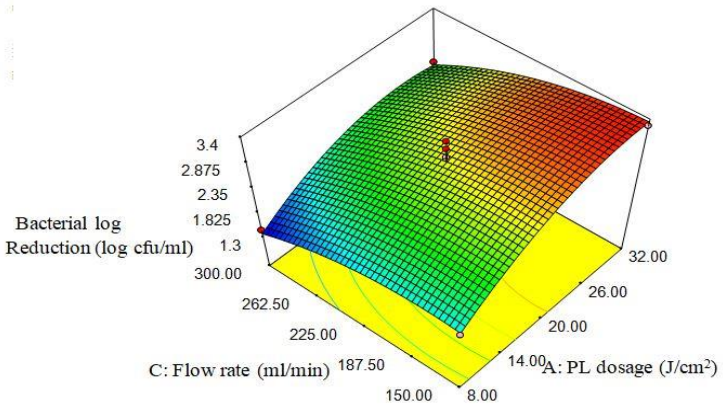
The effects of PL dosage, flow rate and sample-source distance on the total bacterial and yeast and mould log reduction of PL treated pineapple and cashew apple juices are presented as 3D surface plots in Figures 4.39, 4.40, 4.41 and 4.42. It could be inferred from the figures, that all process parameters had a significant effect on bacterial and yeast and mould log reduction for both the juices ($p < 0.01$).

Previous studies on the effectiveness of pulsed light have also shown significant reductions in levels of microbial pathogens, moulds and yeasts (Hillegas and Demirci, 2003; Takeshita *et al.*, 2003; Gomez-Lopez *et al.*, 2007; Turtoi and Nicolau, 2007). The inactivation is prominently due to the absorption of energy rich UV photons by the conjugated proteins and nucleic acid (Jay, 1996; Masschelein, 2002) including DNA (deoxyribonucleic acid) of the organisms, which is essential for their reproduction. The absorbed energy is able to break organic molecular bonds, causing several structural changes in DNA, such as cross linking of strands, rearrangement, cleavage, and breakage of the chain, and thereby activating photochemical reactions that can produce some substances called "photoproducts", that inhibit the reproduction of DNA .

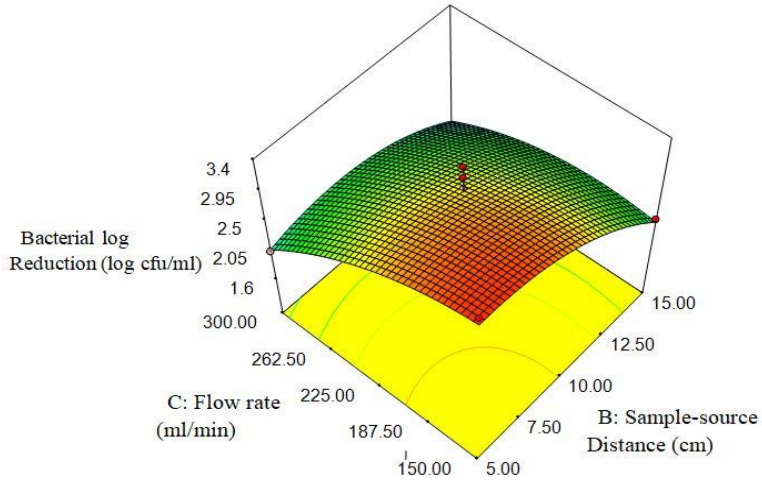
From the figures (4.39, 4.40, 4.41 and 4.42), it may be ascertained that the bacterial and yeast and mould reduction have a linear relationship with PL dosage for both the juices. The microbial reduction increases with increase in PL dosage from 8 to 32 J/cm^2. Similar findings were also reported by Artiguez *et al.* (2011), Funes *et al.* (2013) and Pataro *et al.* (2011). Pataro *et al.* (2011) reported that lethal effect of *Escherichia* and *Listeria* strains in apple and orange juice were found to be intensified with increasing the PL energy dosage.



a)

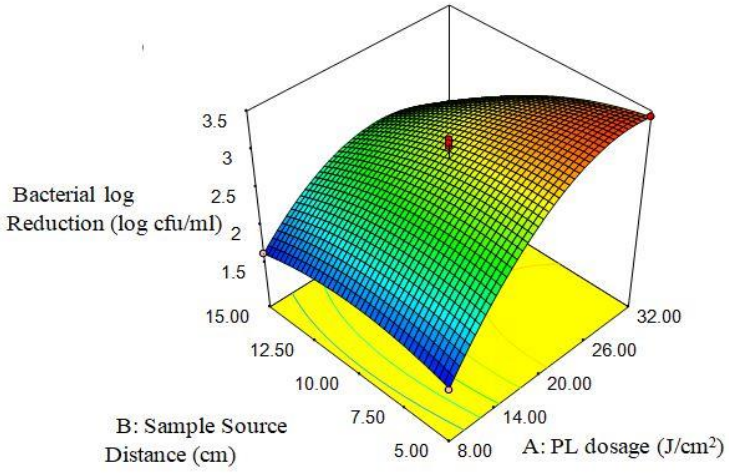


b)

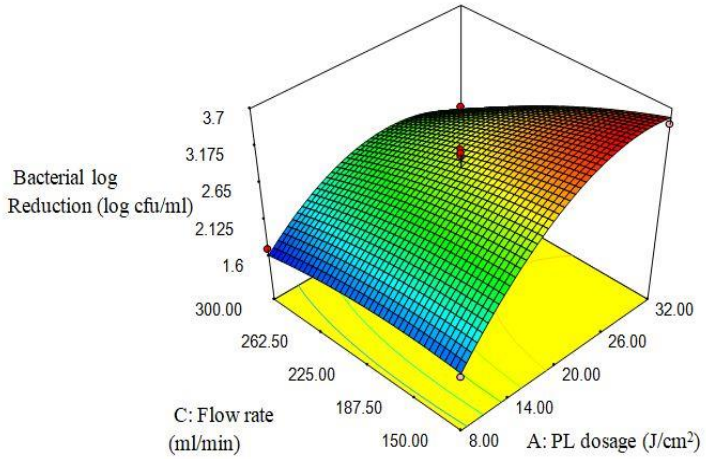


c)

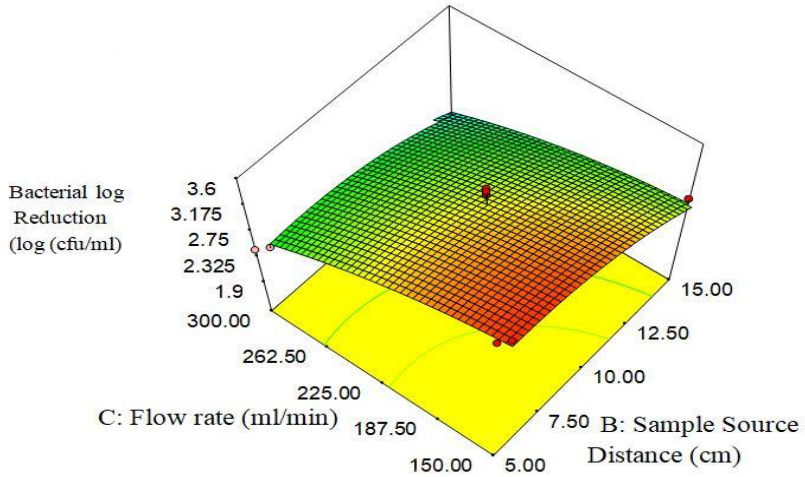
Fig. 4.39 Effect of pulsed light parameters on bacterial log reduction of pineapple juice



a)

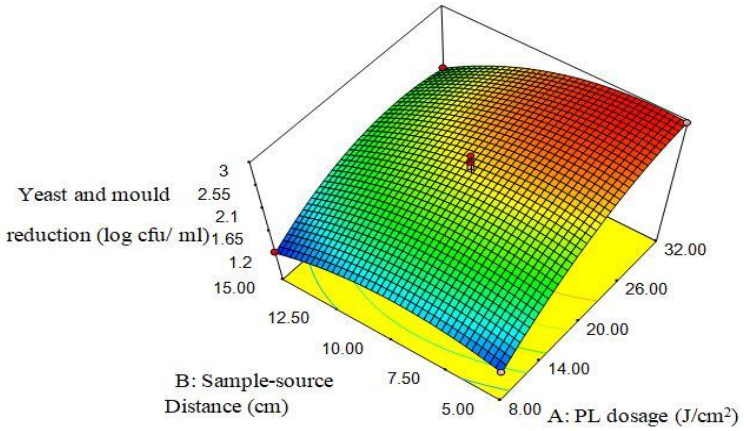


b)

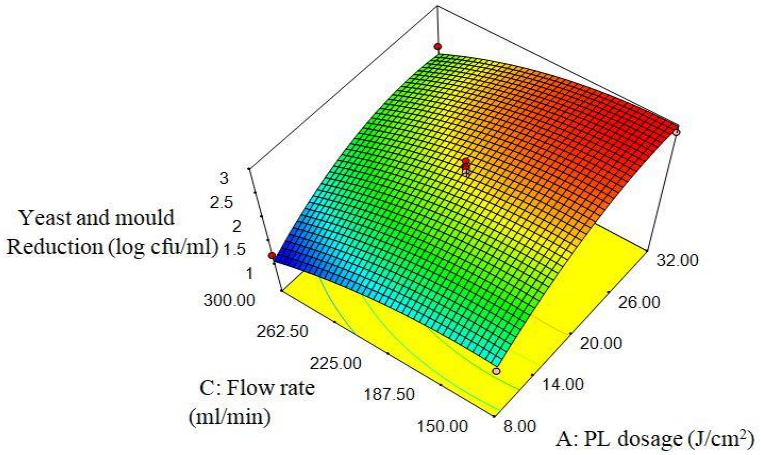


c)

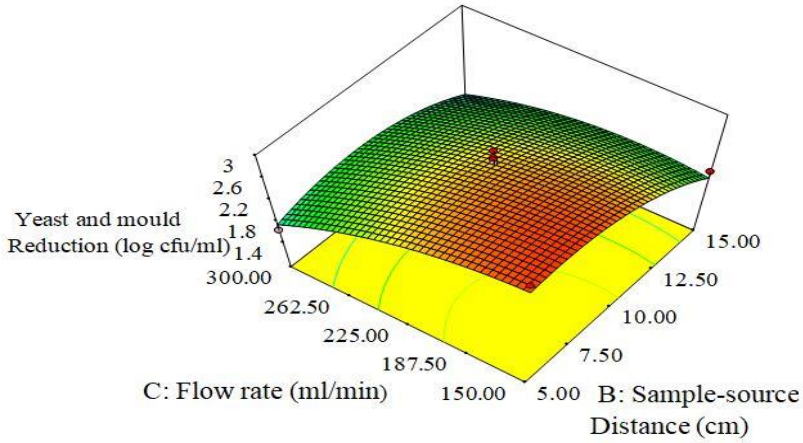
Fig. 4.40 Effect of pulsed light parameters on bacterial log reduction of cashew apple juice



a)



b)



c)

Fig. 4.41 Effect of pulsed light parameters on yeast and mould reduction of pineapple juice

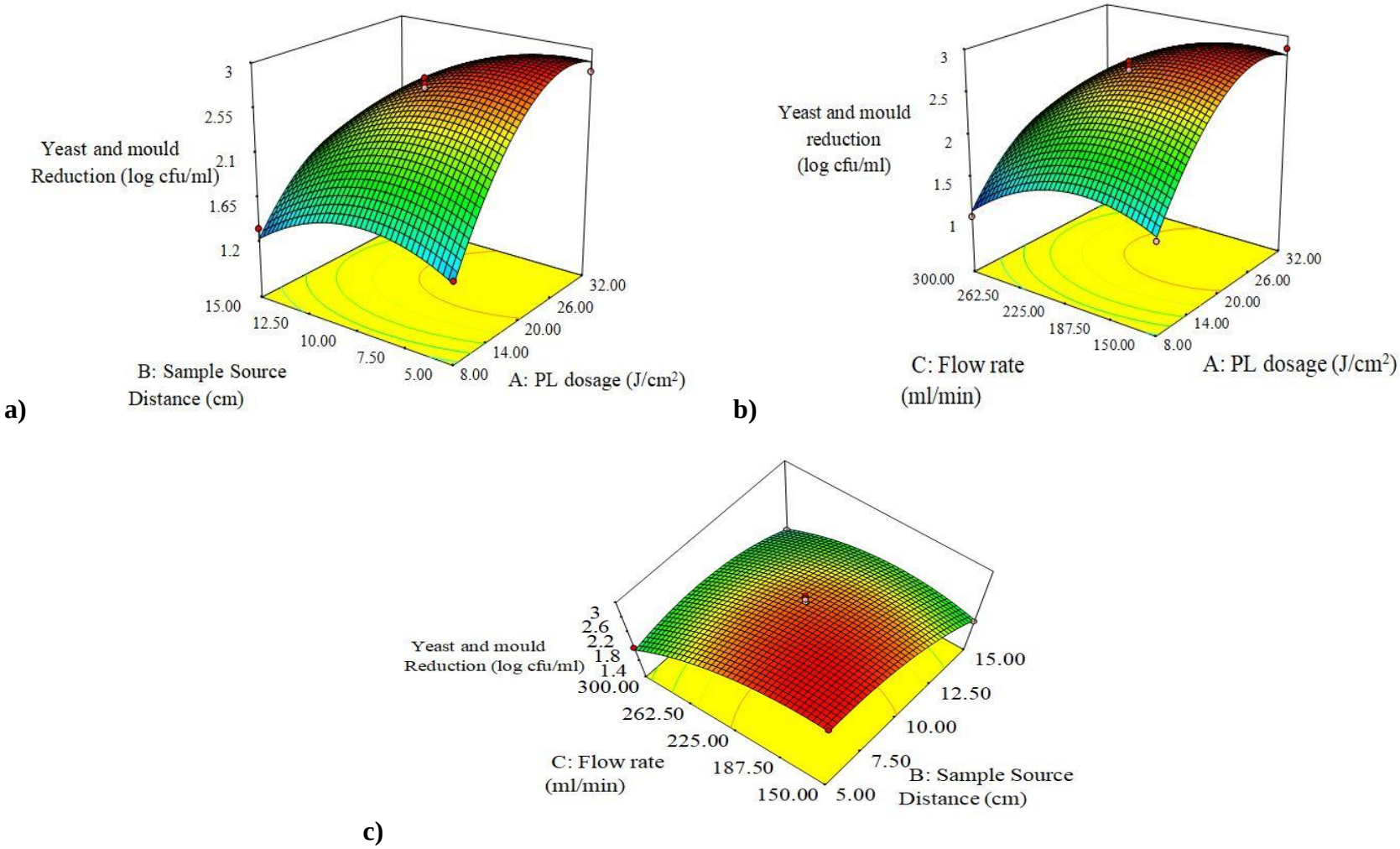


Fig. 4.42 Effect of pulsed light parameters on yeast and mould reduction of cashew apple juice

Microbial reduction by UV-C light could be achieved with low intensity energy for longer period or high intensity energy for shorter time (Bachmann, 1975).

From the figures, it could also be observed that the flow rates of the fruit juices are inversely proportional to the microbial log reduction. In consistent with the results of the study, Krishnamurthy *et al.* (2007) also reported that an increase in the flow rate of milk from 20 to 40 ml/min resulted in the reduction of *St.aureus* destruction at 8 cm sample-source distance. The increase in flow rate of fruit juices might have resulted in insufficient duration of exposure to PL irradiation required for destruction. Krishnamurthy *et al.* (2007) also observed that at lower flow rates, a better inactivation was obtained because of the higher absorption of energy due to the longer residence time.

It may also be revealed from the figures that the microbial reduction increased with the decrease in sample-source distance. Similar results were also reported by Hillegas and Dermici (2003) and Preetha *et al.* (2016 a and b) in batch PL treatment of honey, pineapple juice and orange juice respectively. In consistent with the results of the study, Hillegas and Dermici (2003) also observed a high spore reduction percentage in honey, when the sample-source distance was minimum. About 89.9% reduction in spores was obtained at 8 cm distance between the honey and lamp (Hillegas and Dermici, 2003). Preetha *et al.* (2016a) also found that maximum reduction of *Escherichia coli* strains in pineapple juice was obtained when the shelf height (distance of sample from PL lamp) was minimum (5 cm). This might be due to the higher absorption of the PL radiation when the sample-source distance is minimum. Sharrma and Demirci (2003) reported that higher shelf heights could result in greater dissipation of PL energy during the travel of light pulses from the lamp to the samples.

A lower reduction in bacteria, yeast, and mould was observed in the PL treated pineapple juice when compared to PL treated cashew apple juice. This might be due to the differences in absorption properties of pineapple and cashew apple juice. The higher absorbance value of pineapple juice (0.049 cm^{-1}) indicated limited penetration of PL radiation due to scattering of light by the suspended particles in pineapple juice.

The bacterial reductions in both the juices were found to be higher than the yeast and mould reduction during PL treatment. This could be due to the reduced action of the PL on yeast and mould population. The inactivation of microorganisms during PL process is predominantly due to the chemical action of UV rays. Tran and Farid (2004) reported that yeast and moulds are less affected by UV light than bacteria since DNA molecules of yeast and mould produce less pyrimidine base, especially thymine during the photochemical reactions.

The effect of process parameters on bacteria and yeast and mould reduction of PL treated pineapple juice are shown in Equations 19 and 20. The equations show that microbial reduction was in positive correlation with PL dosage and in negative correlation with flow rate of juice and sample-source distance. From the coefficients of the independent variables, it may be concluded that the influence of PL dosage was highest followed by flow rate of juice and further followed by the sample-source distance.

For pineapple juice

$$\text{Bacterial log reduction} = 2.88+0.573A-0.357B-0.411C-0.28AB-0.097AC+0.2BC-0.381A^2-0.313B^2-0.236C^2 \quad R^2 = 0.97 \quad (4.19)$$

$$\text{Yeast and mould reduction} = 2.578+0.587A-0.286B-0.366C-0.225AB-0.05AC+0.182BC-0.380A^2-0.337B^2-0.222C^2 \quad R^2 = 0.97 \quad (4.20)$$

Where, A = Pulsed light dosage J/cm^2

B = Source-sample distance, cm

C = flow rate ml/min

From the ANOVA tables (A.22a and A.23a), it could be inferred that log reductions of bacteria and yeast and mould in pineapple juice validated that the regression model was significant ($p < 0.001$). The coefficient of regression (R^2) was 0.98 and 0.98 for bacterial log reduction and yeast and mould reduction respectively, indicating that the quadratic model of both bacteria and yeast and mould fits satisfactory with the experimental results.

The regression equations of the effect of process parameters on bacteria and yeast and mould reduction of PL treated cashew apple juice are expressed as Equation 20 and 21. It could be inferred from the equations that microbial reduction was in positive correlation with PL dosage and in negative correlation with flow rate of juice and sample-source distance. From the coefficients of the independent variables, it could be derived that the influence of PL dosage was highest followed by flow rate and then by sample-source distance for bacterial reduction. On the other hand, influence of PL dosage was highest followed by sample-source distance and then by flow rate for yeast and mould reduction.

For cashew apple juice

Bacterial log reduction = $2.996+0.625A-0.3375B-0.4175C-0.2775AB-0.2675AC+0.0725BC-0.62925A^2-0.20425B^2-0.10925C^2$ $R^2 = 0.98$ 4.21

Yeast and mould reduction = $2.682+0.476A-0.345B-0.262C-0.363AB-0.18AC+0.225BC-0.551A^2-0.343B^2-0.376C^2$ $R^2 = 0.99$ 4.22

Where, A = Pulsed light dosage J/cm^2

B = Source-sample distance, cm

C = flow rate ml/min

From the ANOVA tables (A.22 b and 23 b) it could be inferred that log reductions of bacteria and yeast and mould validated that the regression model was significant ($P < 0.001$) for cashew apple juice samples. The coefficient of regression (R^2) was 0.98 and 0.98 for bacterial log reduction and yeast and mould reduction respectively, indicating that the quadratic models of both bacteria and yeast and mould reduction may be considered as adequately fit for the experimental results.

4.3.3 Optimisation of Pulsed Light Process Parameters

Optimisation of the three process variables such as pulsed light dosage of (8, 20 and 32 J/cm^2), sample-source distance (5, 10 and 15 cm) and flow rate (150, 225 and 300 ml/min) was performed using the Box-Behnken design in Design Expert Software

7.0.0. For optimisation of the operative parameters during PL treatment, the responses were minimised, maximised or kept in target value to get the desired outcome. Higher desirability value was selected as optimum process conditions of PL treatment. The highest desirability value of 0.730 were obtained at PL treatment with a PL dosage of 13.69 J/cm^2, sample-source distance of 10.26 cm and flow rate of 165.06 ml/min for pineapple juice, whereas, for cashew apple juice highest desirability of 0.767 was obtained at a PL dosage of 12.49 J/cm^2, sample-source distance of 8.63 cm with a flow rate of 164.01 ml/min.

The response values were tested using the recommended optimum values of process variables and was employed to validate the experimental and predicted values of the responses. The predicted and actual values of the responses and the percentage variation at the optimised conditions of PL treatment for both the juices are presented in Table 4.8. The predicted values of optimised treatment are comparable with that of the actual values.

Table 4.8 Predicted and actual values of responses at the optimised conditions of PL treatment

a. Pineapple juice

Sl. No.	Responses	Predicted value	Actual value	Variation (%)
1	pH	4.45	4.45	0.00
2	TSS (°Brix)	10.81	10.82	0.09
3	Titrable acidity (mg/100 ml)	0.381	0.382	0.26
4	Ascorbic acid (mg/100 ml)	29.02	28.86	-0.55
5	Total sugar (mg/100 ml)	10.53	10.55	0.19
6	Total phenolic content (mg/100 ml)	67.00	67.14	0.20
7	Total colour difference (ΔE)	0.55	0.46	-1.63
8	Bacterial log reduction (log cfu/ ml)	3.46	3.42	-11.56
9	Yeast and mould log reduction (log cfu/ml)	2.14	2.16	0.92

b. Cashew apple juice

Sl. No.	Responses	Predicted value	Actual value	Variation (%)
1	pH	4.32	4.32	0.00
2	TSS (°Brix)	7.24	7.43	2.5
3	Titrable acidity (mg/100 ml)	0.422	0.422	0.00
4	Ascorbic acid (mg/100 ml)	162.75	161.96	-0.48
5	Total sugar (mg/100 ml)	9.95	9.97	0.20
6	Total phenolic content (mg/100 ml)	164.42	167.58	0.584
7	Total tannin content (%)	0.54	0.56	3.57
8	Total colour difference (ΔE)	0.438	0.52	15.0
9	Bacterial log reduction (cfu/ml)	2.62	2.73	0.40
10	Yeast and mould log reduction (cfu/ml)	2.22	2.32	4.3

4.4 OPTIMISATION OF OHMIC ASSISTED PULSED LIGHT TREATMENT PARAMETERS

In this study, the ohmic and pulsed light treatments were combined in order to achieve a higher microbial reduction with minimum changes in juice quality characteristics as per the hypothesis explained in Chapter I. The optimised process operating parameters of ohmic heating and pulsed light treatments were analysed for the effectiveness of combined treatment. For a rigorous comparison, the treatment process parameters with statistically second highest desirability for both ohmic and pulsed light treatments were also analysed to check the effectiveness of combined process. The ohmic heating and pulsed light treatment combinations selected for further studies are as follows:

The ohmic heating treatment operating parameters for pineapple juice:

1. OH_1- Voltage gradient: 14.02 V/cm; Process temperature: 55.60°C; Holding time: 2.31 min
2. OH_2 -Voltage gradient: 12.57 V/cm Process temperature: 58.81°C; Holding time: 1.52 min

The pulsed light treatment operating parameters for pineapple juice:

1. PL_1- PL dosage: 13.69 J/cm^2; Sample-source distance: 10.26 cm; Flow rate: 165.06 ml/min
2. PL_2 -PL dosage: 12.66 J/cm^2; Sample-source distance: 8.83 cm; Flow rate: 163.45 ml/min

The ohmic assisted PL treatment combinations for pineapple juice:

1. P_1 (OH_1PL_1)
2. P_2 (OH_2PL_2)
3. P_3 (OH_1PL_2)
4. P_4 (OH_2PL_1)

The ohmic heating treatment operating parameters for cashew apple juice are:

1. OH_1- Voltage gradient: 14.53V/cm; Process temperature:55.25°C;Holding time : 2.77 min
2. OH_2-Voltage gradient: 14.98V/cm; Process temperature:56.45°C; Holding time : 2.10 min

The pulsed light treatment parameters for cashew apple juice:

1. PL_1- PL dosage: 12.49 J/cm^2; Sample-source distance: 8.63cm; Flow rate: 164.01 ml/min
2. PL_2- PL dosage: 18.04 J/cm^2; Sample-source distance: 11.66 cm; Flow rate:157.08 ml/min

The ohmic assisted PL treatment combinations for cashew apple juice:

1. C_1 (OH_1:PL_1)
2. C_2 (OH_2PL_2)
3. C_3 (OH_1PL_2)
4. C_4 (OH_2PL_1)

4.4.1 Ohmic Assisted Pulsed Light Treatment of Fruit Juices

The effect of ohmic heating and pulsed light technology on enhancing the microbial quality and thus preserving pineapple and cashew apple juice were studied in detail. The influence of process parameters on retaining the quality characteristics of the fruit juices were also studied and optimised statistically. Further, studies were conducted to analyse the effect of the combined process *i.e.* ohmic assisted pulsed light treatment to see the effect of such a system and to validate the hypothesis for the process as manifested in Chapter I. The optimised process variables and the experimental combinations with next highest desirability of each process only were taken for analysing the effectiveness of combined process. The experimental procedure is as detailed in section 3.3.3.3. The results on the effect of ohmic assisted PL treatment on the physicochemical and microbiological qualities of the pineapple and cashew apple juice are detailed in following sections.

4.4.1.1 Effect of ohmic assisted PL treatment on the physicochemical properties of fruit juices

The pH, TSS, titrable acidity, ascorbic acid, total sugar, total phenolics, total tannin, total colour difference, antioxidant activity and mineral content of ohmic assisted PL treated fruit juices at the chosen treatment combinations were compared with the TSS, titrable acidity, ascorbic acid, total sugar, total phenolics, tannin content, total colour difference, antioxidant activity and mineral content of the control (fresh juice), ohmic heated and pulsed light treated fruit juices. The results are tabulated in Appendix A.26 and A.27 and ANOVA tables are presented in Appendix A. 28 and A. 29 for pineapple and cashew apple juice respectively.

The variation in pH of the fruit juices under different treatment conditions are presented in Fig. 4.43 (a) and (b). The control samples of pineapple and cashew apple juice had a pH value of 4.45 and 4.32 respectively. It was found that all ohmic assisted PL treated samples of pineapple and cashew apple juice showed a reduction in pH. The pH of ohmic assisted PL treated samples ranged between 4.27 and 4.29 and 4.26 and 4.28 for pineapple and cashew apple juice respectively. It may be observed from figures that, pH values of PL treated samples were on par with the control sample. Ohmic heated samples reported a higher reduction in pH followed by ohmic assisted PL treatments in both the fruit juices. The reduction in pH during ohmic heating treatment alone for reasons as discussed in section 4.3.1.1 might have contributed to the reduction in the pH of ohmic assisted PL treated samples. No significant variation in pH values were observed among all combined treated samples of fruit juices. Though a difference in pH values was observed between the control and combined treated samples statistically, the pH values were in the range of 3.7-4.5 classified as acid foods (Fraizer *et al.,* 1971) and therefore could not be considered to manifest any significant difference in pH quality. It may be noted that among the combined treated samples of both fruit juice, the optimally treated samples, i.e. P_1 and C_1 showed minimum variation in pH with that of control.

The variation in TSS of pineapple and cashew apple juices subjected to different treatments as listed in section 3.3.3.3 are shown in Fig. 4.44 (a) and (b). All treated

samples showed an increase in TSS value except PL treated samples of pineapple and cashew apple juice. The fresh pineapple and cashew apple juice recorded a TSS value of 10.82 and 7.73°Brix respectively. The ohmic assisted PL treated samples recorded TSS values in the range of 11.14 to 11.16 and 8.08 to 8.12°Brix in pineapple and cashew apple juice respectively. No significant variation in TSS was observed among all combined treated fruit juices. The TSS values of PL treated samples were on par with the control samples for both the juices. Ohmic heated samples showed a higher increase in TSS compared with control followed by combined treated samples. Among the combined treated samples of both fruit juices, optimally treated samples showed minimum variation in TSS with respect to control. The reasons stated for the increase in TSS of fruit juices for ohmic heating alone (section 4.3.1.2) could also be taken for the increase of TSS in combined treated samples of both the juices. It may be noted that the difference in TSS content between the control and optimally treated samples were 0.32°Brix and 0.34°Brix for pineapple and cashew apple juice respectively.

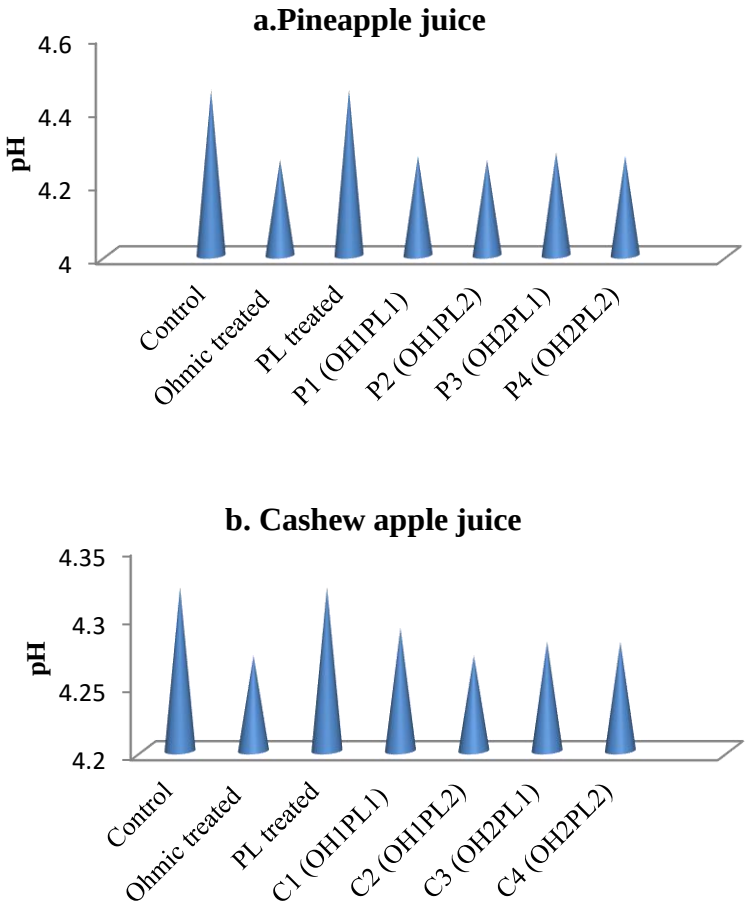


Fig.4.43 Effect of different treatments on pH of the fruit juices

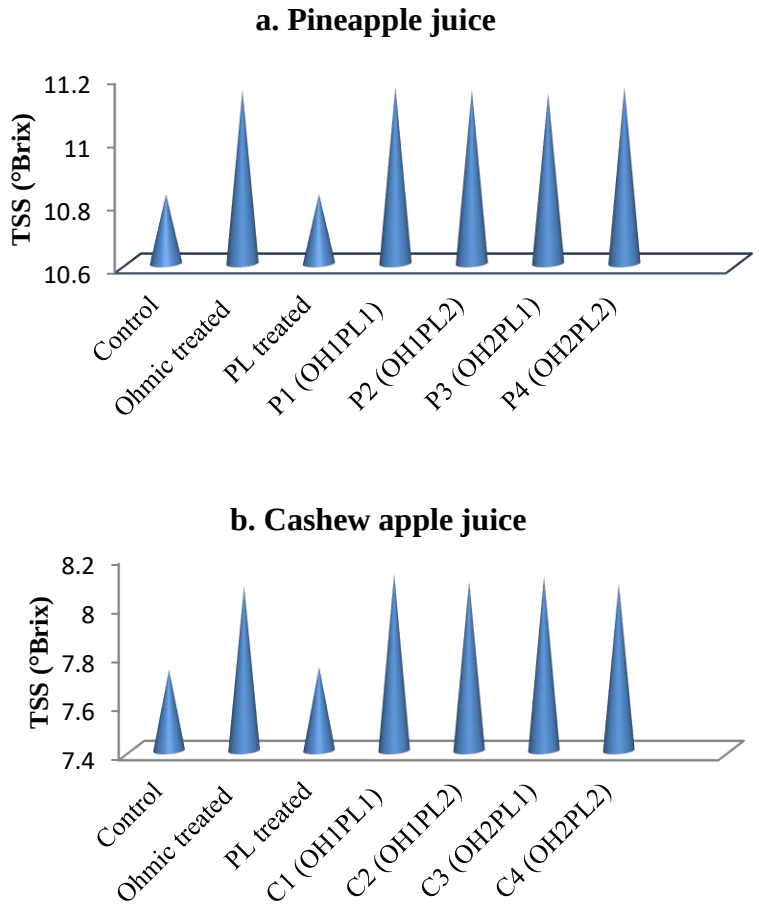


Fig.4.44 Effect of different treatments on TSS of the fruit juices

The effect of the treatments under study on the titrable acidity of the fruit juices are depicted in Fig. 4.45 (a) and (b). The fresh pineapple juice exhibited a titrable acidity value of 0.382 mg/100 ml, whereas that of cashew apple juice was 0.422 mg/100 ml. It was observed that PL treated juices showed titrable acidity on par with that of control sample, whereas all other treatments showed an increase in titrable acidity values. The titrable acidity values of ohmic assisted PL treated samples ranged from 0.384 to 0.387 and 0.422 to 0.424 mg/100 ml for pineapple and cashew apple juice respectively. It could be revealed from the descriptive statistical analysis that the ohmic assisted PL treated pineapple juice samples and PL treated samples did not show any significant variation in titrable acidity with that of control ($p > 0.05$). In the case of cashew apple juice, no significant variation in titrable acidity values was recorded between all treatment conditions and control ($p > 0.05$). Only the ohmic heated pineapple juice showed a statistically significant variation of titrable acidity with that of control ($p < 0.05$). Though the magnitude of variation is negligible, the increased rate of electrochemical reactions such as hydrolysis, imposed due to the inherent chemical composition of pineapple juice during ohmic heating, could have resulted in the increased titrable acidity (Thangalakshmi, 2018).

The variation of ascorbic acid content when the fresh juices were subjected to designated treatment conditions are shown in Fig. 4.46 (a) and (b). The ascorbic acid content of fresh pineapple and cashew apple juice (control) were 31.18 and 168.46 mg/100 ml respectively. It was found that all treated samples recorded a significant reduction in ascorbic acid content ($p < 0.01$). Both ohmic heated and PL treated samples resulted in a reduction in ascorbic acid content due to reasons as explained in section 4.3.1.4 and 4.3.4.4. The ohmic assisted PL treated samples showed an ascorbic acid percentage reduction of 12 to 15% and 10 to 13% in pineapple and cashew apple juice respectively for different treatment combinations.

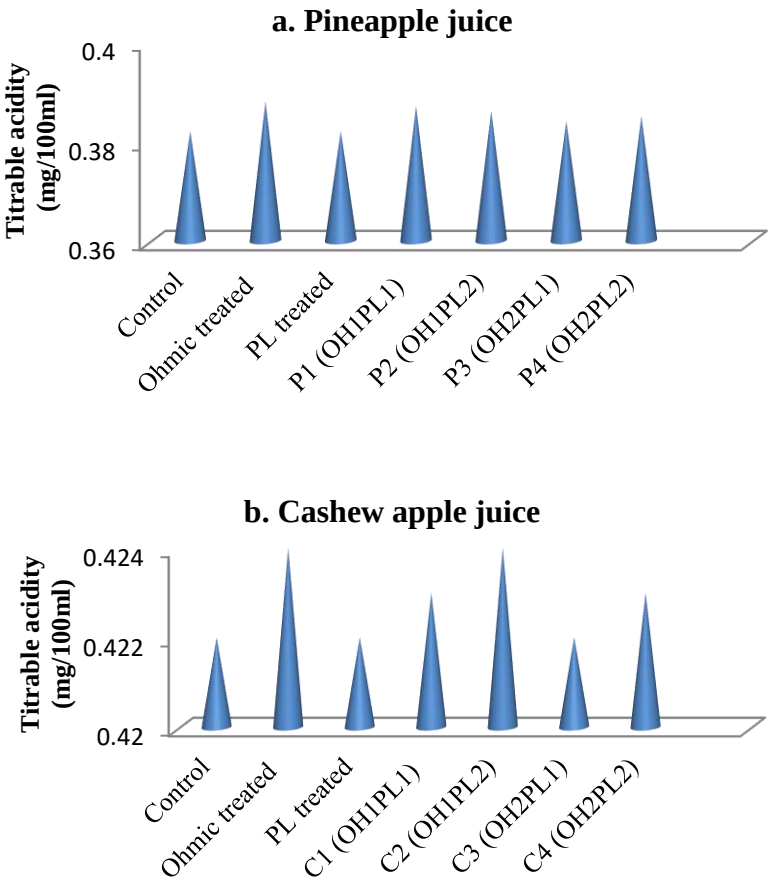


Fig.4.45 Effect of different treatments on titrable acidity of the fruit juices

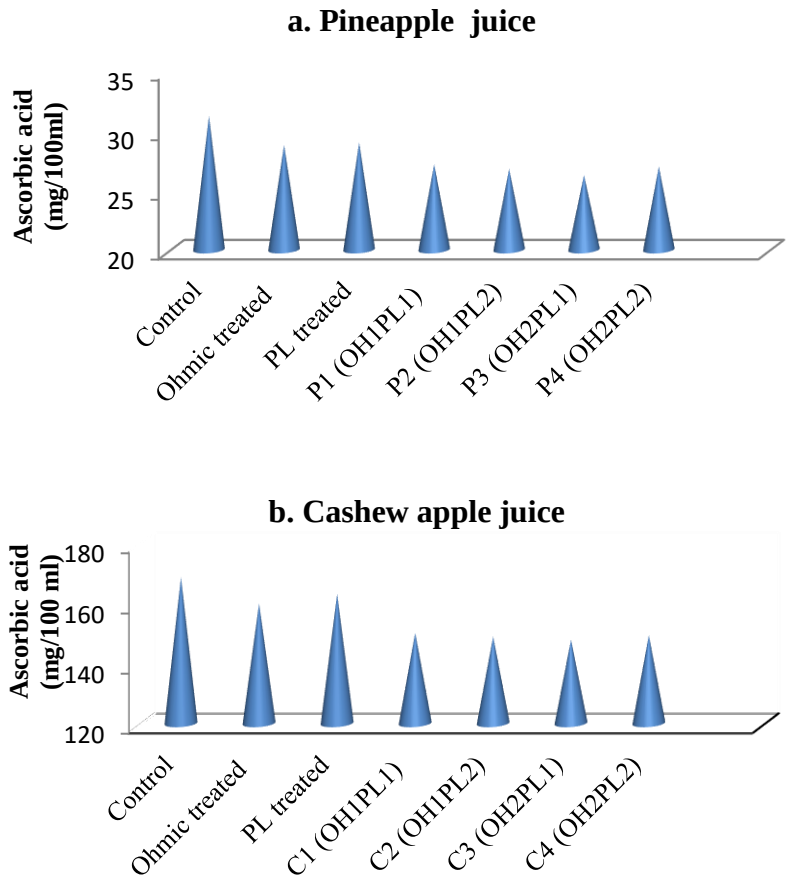


Fig.4.46 Effect of different treatments on ascorbic acid content of the fruit juices

Among the treatment combinations the optimised treatments (P_1 and C_1) showed least reduction in ascorbic acid content. In this study, the reductions in ascorbic acid content during combined treatments were found to be higher than that during individual treatments. During combination treatments, the synergic action of ohmic and PL processing could have resulted in increased reduction of ascorbic acid content in fruit juices. The degradation of ascorbic acid compounds at above ambient temperature, electrochemical reactions during ohmic treatment of water during ohmic heating and photolysis of fruit juice during exposure to UV light during pulsed light treatment might have together contributed for the reduction in ascorbic acid content (Assiry *et al.,* 2003; Shah *et al.,* 2016).

In this study there showed a reduction in ascorbic acid to the tune of 10-15%, in both juices, when compared to fresh juice and 4-5% when compared to ohmic treatment and pulsed light treatment alone. It may be postulated that in order to achieve the same reduction of microbial population, we should have used higher levels of process variables of both treatments which ultimately would have resulted in higher reduction of ascorbic acid content. The combined treatment therefore reduced the intensity of process variables for attaining the safe level of microbial population.

The effect of the treatments under study on the total sugar content of the fruit juices are depicted in Fig. 4.47 (a) and (b).

The fresh pineapple juice presented a total sugar content of 10.53 mg/100 ml, whereas that of cashew apple juice was 9.96 mg/100 ml. The total sugar values of ohmic assisted PL treated samples ranged from 10.53 to 10.55 and 9.96 to 9.97 mg/100 ml for pineapple and cashew apple juice respectively. No significant variations in total sugar content were observed among control and samples under different treatment conditions for both fruit juices ($p > 0.05$). These results are in agreement with the findings of combination treatments of hurdle strategy for preservation of apple juice by Noci *et al.* (2008) and ohmic assisted UV treatments of pineapple juice by Sean *et al.* (2016). Sean *et al.* (2016) observed no significant variations in total sugars among ohmic assisted UV

treated pineapple juice samples under different process conditions and the values were on par with that of fresh pineapple juice.

The variation in total phenolic content of the fruit juices under different treatment conditions are presented in Fig. 4.48 (a) and (b). The control samples of pineapple and cashew apple juice had a total phenolic content of 67.25 and 168.46 mg/100 ml respectively. It was found that PL treated samples and samples under combined treatment of both fruit juices showed a reduction in phenolic content, whereas ohmic heated samples showed an increase in phenolic content. The total phenolic content of ohmic assisted PL treated samples ranged from 67.18 to 67.22, and 167.6 to 168 mg/100 ml respectively. It could be revealed from analysis that the ohmic assisted PL treated pineapple juice samples and PL treated samples did not show any significant variation in phenolic content with that of control ($p > 0.05$). In the case of cashew apple juice, no significant variation in total phenolic contents was recorded between samples under various treatment conditions and control ($p > 0.05$). Only the ohmic heated pineapple juice showed a statistically significant variation of phenolic content with that of control ($p < 0.05$).

The increase in the phenolic content during ohmic heating might be due to the increase in phenolic extractability induced by the changes in tissue structure and the decrease in phenolic content during PL treatment could be due to phenolic degradation during the exposure of UV radiations (Koutchma, 2009; Brochier and Domeneghini, 2016). In case of combined treated samples, the aforesaid two reasons might have worked together ultimately resulting in slight reduction in phenolic content with respect to control. It may be noted that among the combined treated samples of both fruit juice, the optimally treated samples *i.e.* P_1 and C_1 showed minimum variation in phenolic content with that of control.

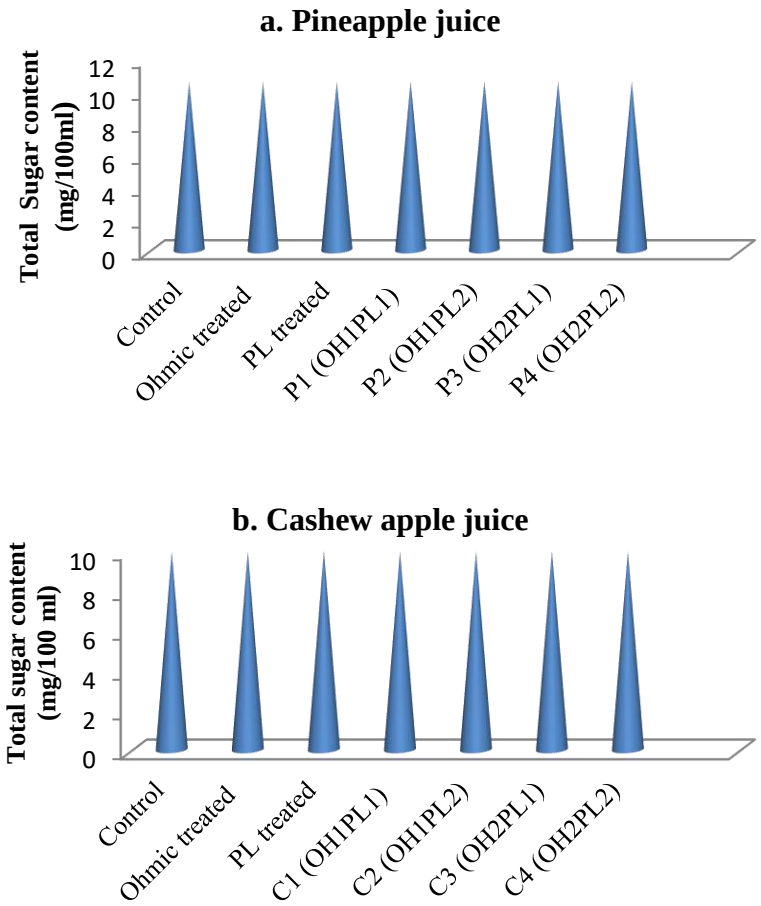


Fig.4.47 Effect of different treatments on total sugar of the fruit juices

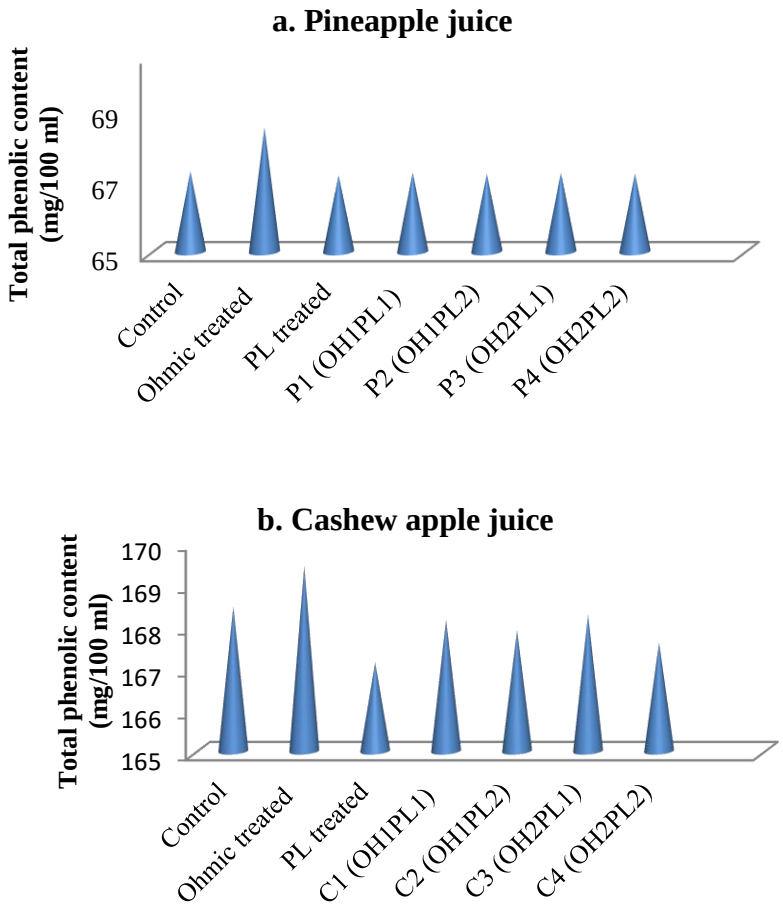


Fig.4.48 Effect of different treatments on total phenolic content of the fruit juices

The variation in tannin content of cashew apple juice subjected to different treatments as stated in section 3.3.3.3 are shown in Fig. 4.49. The fresh clarified cashew apple juice recorded a tannin content of 0.57%. The tannin content of samples under combination treatments varied between 0.51 and 0.53%. No significant variation in tannin content was observed among the combined treated fruit juices, whereas significant variations were observed between control and other treatments. Among the combined treated samples of both fruit juices, optimally treated samples showed minimum variation in tannin content than that of control. The reasons stated for the decrease in tannin content of fruit juices when they are subjected to ohmic heating alone (section 4.3.1.7) could also be taken as justification for the decrease of tannin content in combined treated samples of cashew apple juice. Though the variation in tannin content between control and optimally treated samples was significant statistically the magnitude of the difference is only 0.06%.

The variation of total colour difference of pineapple and cashew apple juice, when fresh juices were subjected to predesigned treatment conditions are shown in Fig. 4.50. (a) and (b). It was found that all treated samples recorded a significant increase in total colour difference ($p < 0.01$). Both ohmic heated and PL treated samples resulted in an increase in total colour difference due to reasons as explained in section 4.3.1.8 and 4.3.4.8. The total colour difference of ohmic assisted PL treated samples ranged from 0.74 to 0.78 and 0.79 to 0.85 for pineapple and cashew apple juice respectively. Among the treatment combinations the optimised treatments (P_1 and C_1) showed least increase in total colour difference. In this study, the increases in colour difference during combined treatments were found to be higher than that during individual treatments. During combination treatments, the synergic action of ohmic and PL processing could have resulted in increased colour variation in fruit juices. The results are inconsistent with the findings of Caminiti *et al.* (2011) in fruit juices treated with the non thermal combinations of HILP+PEF (High Intensity Light Pulse + Pulsed Electric Field) and UV+PEF.

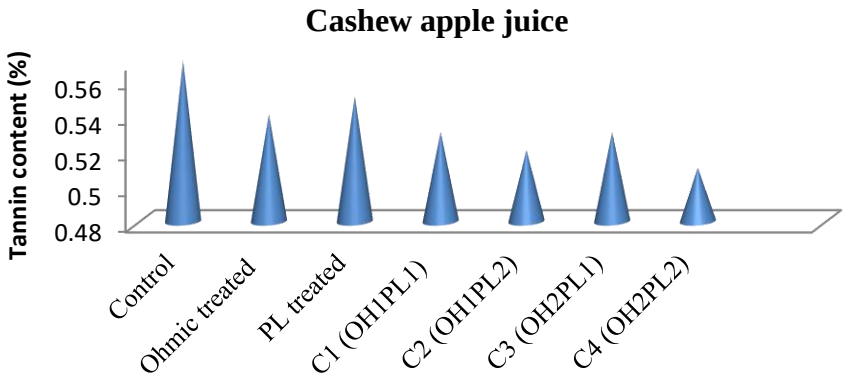


Fig.4.49 Effect of different treatments on tannin content of cashew apple juice

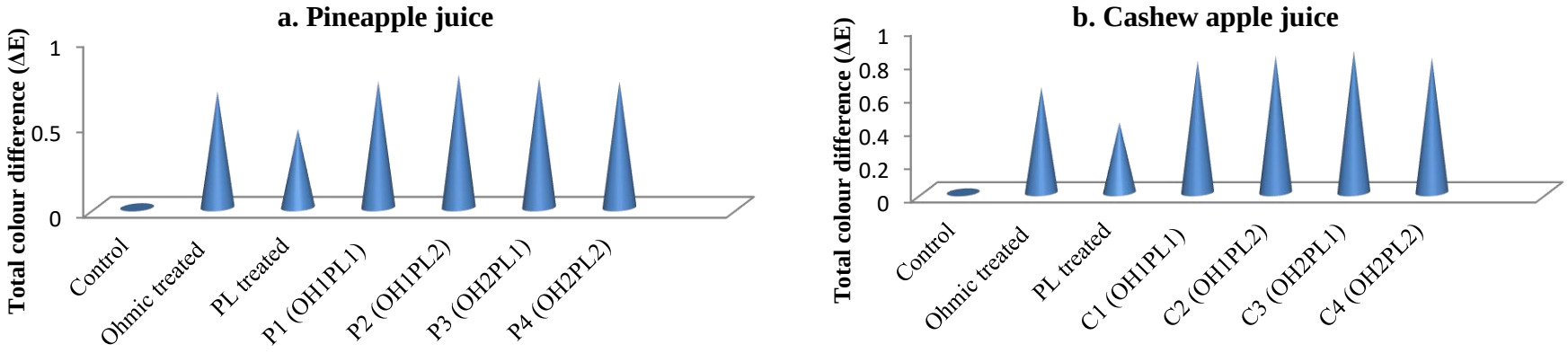


Fig.4.50 Effect of different treatments on total colour difference of the fruit juices

The non enzymatic browning and carotenoid degradation during ohmic heating and the brown pigment formation during ascorbic acid degradation at ohmic and pulsed light treatment together might have contributed to the colour variation (Ibarz *et al.,* 2000).

In the current study, the total colour difference of pineapple and cashew apple juice subjected to the various treatments ranged from 0.46 to 0.78 and 0.42 to 0.85 respectively. A notable visual change in colour will be seen when the total colour difference is above 1.5 (Cserhalmi *et al.,* 2006). Therefore, though, a statistically significant increase in the total colour difference was observed in all treated samples, it could be concluded that only a slightly noticeable visual colour change (0.5-1.5) occurred in the fruit juices during the combined process when compared to that of fresh juices.

The browning index is considered as a quality index that represents the colour variations in product which is linked with various chemical reactions (Caminiti *et al.,* 2011). The variation in browning index of the fruit juices under different treatment conditions are presented in Fig. 4.51 (a) and (b).

All treated samples showed a significant increase in browning index except PL treated samples of pineapple and cashew apple juice ($p < 0.01$). The ohmic assisted PL treated samples recorded browning index in the range of 0.12 to 0.28 and 0.14 to 0.3 for pineapple and cashew apple juice respectively. No significant variation in browning index was observed among all combined treated fruit juices. Ohmic assisted PL treated samples showed a higher value of browning index compared with control followed by ohmic heated and further followed by PL treated samples. Among the combined treated samples of both fruit juices, optimally treated samples showed minimum variation in browning index with that of fresh fruit juices.

In this study, the values of browning index during combined treatments were found to be higher than that during individual treatments. During combination treatments, the synergic action of ohmic and PL processing could have resulted in increased browning index in fruit juices. The magnitude of variation in BI during PL treatment of fruit juices was comparatively less when compared to variation caused during ohmic heating. The

ascorbic acid degradation products such as 5-hydroxymethyl furfural (HMF), furfural or carbonyl compounds during processing might have contributed to browning of juice samples either by reaction with each other or with amino acids (Kennedy *et al.,* 1990; Sawamura *et al.,*1994). These browning reactions could have resulted in slight increase of in BI of pineapple and cashew apple juice subjected to PL treatment. The non enzymatic browning which is triggered at above ambient temperatures might have caused the increase in BI in ohmic heated samples. During ohmic processing, the Maillard reactions could also commence with the reaction of free amino groups with carbonyl group of reducing sugars and results in the formation of brown pigments (Bharate and Bharate, 2014). The synergic effect of the reactions mentioned above would result in the increased browning index during the combined process.

Antioxidant activity of treated pineapple and cashew apple juice were determined by DPPH radical-scavenging assays and the results are presented in Appendix A.26 and A. 27. The variation of antioxidant activity when the fresh juices were subjected to various treatment conditions are shown in Fig. 4.52 (a) and (b) respectively.

It was found that all treated samples recorded a significant reduction in antioxidant activity ($p < 0.01$). The ohmic assisted PL treated samples showed an antioxidant activity reduction of 6.6 to 8.7% and 8.2 to 10.5% in pineapple and cashew apple juice respectively with respect to control. Among the combined treatment combinations the optimised treatments (P_1 and C_1) showed least reduction in antioxidant activity with respect to control. During combination treatments, the synergic action of ohmic and PL processing would have resulted in increased reduction of antioxidant activity in fruit juices.

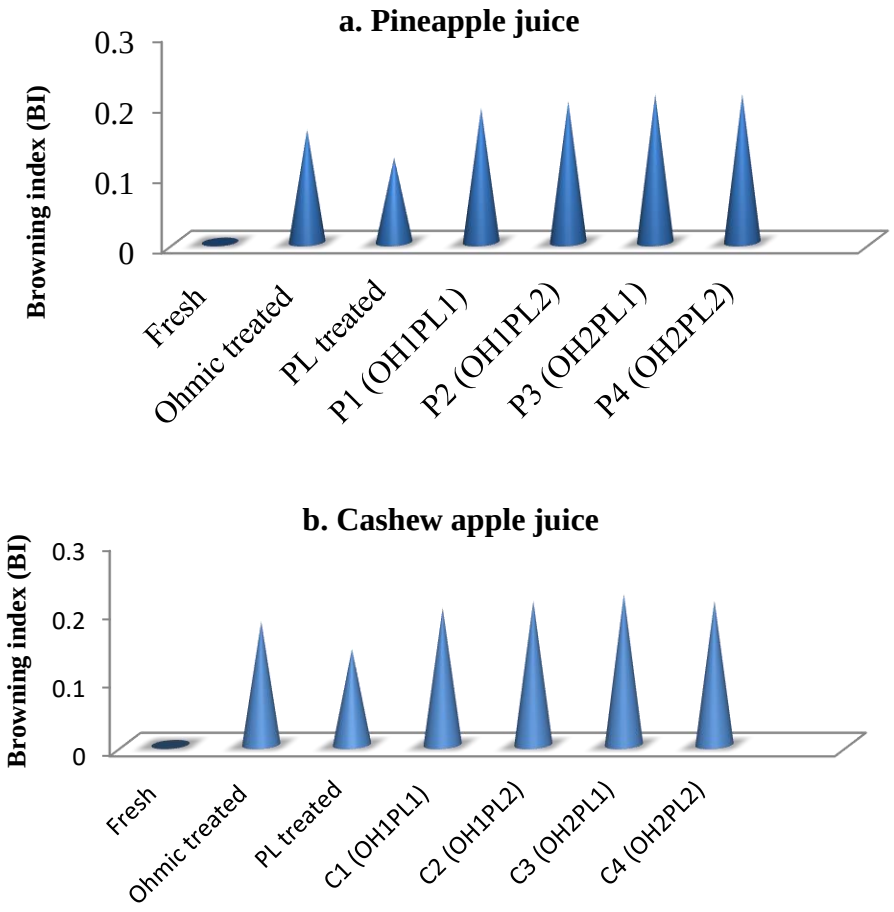


Fig.4.51 Effect of different treatments on browning index of the fruit juices

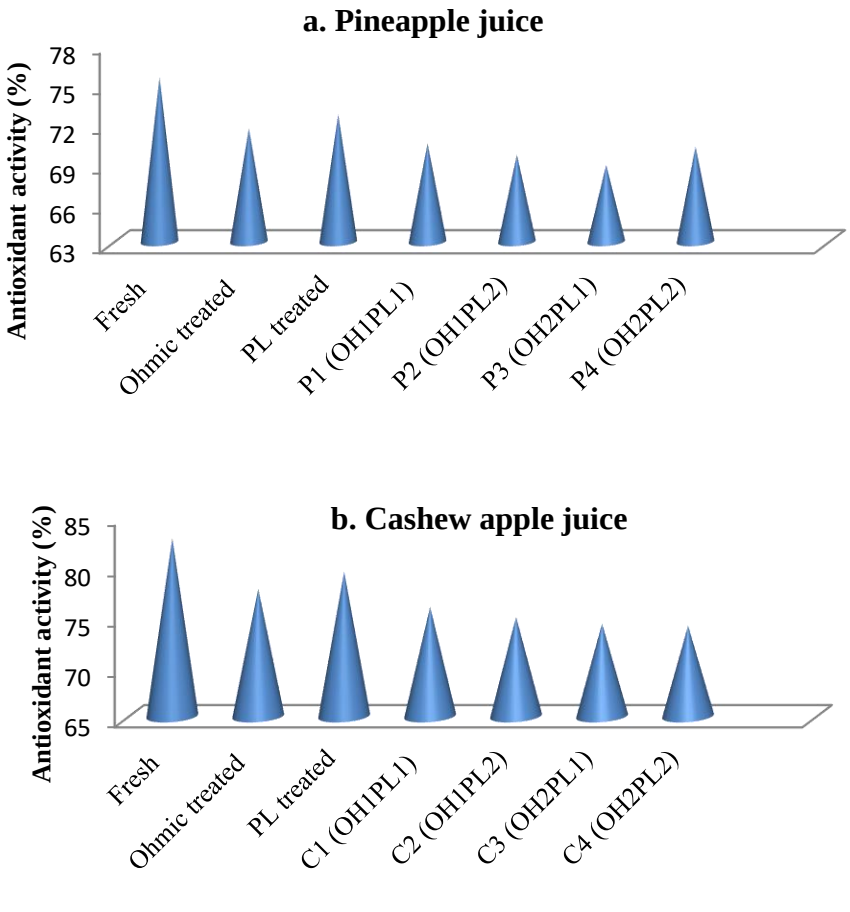
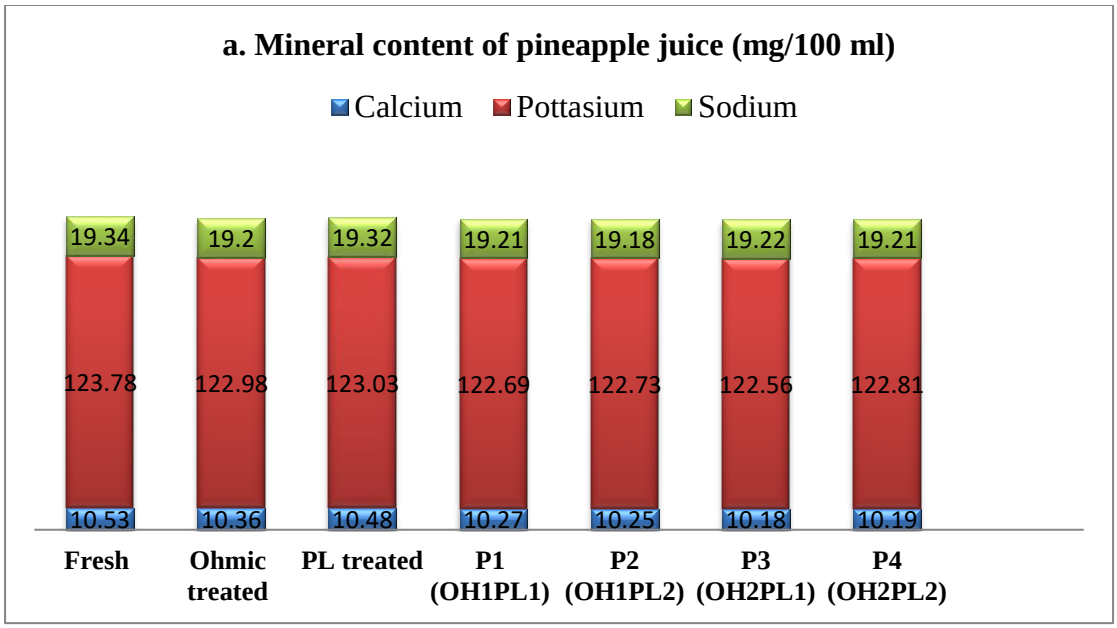


Fig.4.52 Effect of different treatments on antioxidant activity of the fruit juices

A reduction of 3.70% and 4.01% of antioxidant activity was noted in PL treated pineapple and cashew apple juice respectively. Similar reduction pattern was also observed in UV treated apple and pineapple juices (Teja *et al.,* 2017). It could be due to the photolysis of anti oxidant compounds like ascorbic acid, phenols, carotenoids, etc. in presence of UV light (Teja *et al.,* 2017).

An antioxidant reduction of 5.08% and 6.18% was observed in ohmic heated pineapple and cashew apple juice. The exposure to above ambient temperatures during the ohmic processing might have destructed the sensitive components like ascorbic acid and carotenoids. These reduction patterns of antioxidant activity were found to be similar to the findings of Lee *et al.* (2015) during their experiments on combined ohmic and UV treatment of the juice.

The variations in minerals like potassium, calcium, and sodium in pineapple and cashew apple juices subjected to different treatments are shown in Fig. 4.53 (a) and (b). The calcium, potassium, and sodium content in fresh pineapple juice were 10.53, 123.78, and 19.34 mg/100 ml and that in cashew apple juice were 5.04, 65.34 and 17.68 mg/100 ml. The calcium, potassium, and sodium content of ohmic assisted PL treated pineapple samples ranged from 10.18 to 10.27, 122.56 to 122.81, and 19.18 to 19.22 mg/100 ml respectively. In cashew apple juice, calcium, potassium and sodium content varied between 4.96 and 4.99, 64.93 and 65.03, and 17.59 and 17.64 mg/100 ml respectively. No significant variations were observed in all three mineral contents among samples under different treatment conditions with that of fresh for fruit juices ($p>0.05$). The negligible variations in mineral contents in treated samples might be due to mild thermal effects (Khalil, 2001). Jean *et al.* (2007) found that no significant changes were reported in Sodium, potassium, and calcium content of cocoa beans even during storage. It could be concluded that Minerals such as calcium, potassium and sodium are unaffected by ohmic and pulsed light treatments and their combinations under various operating conditions.



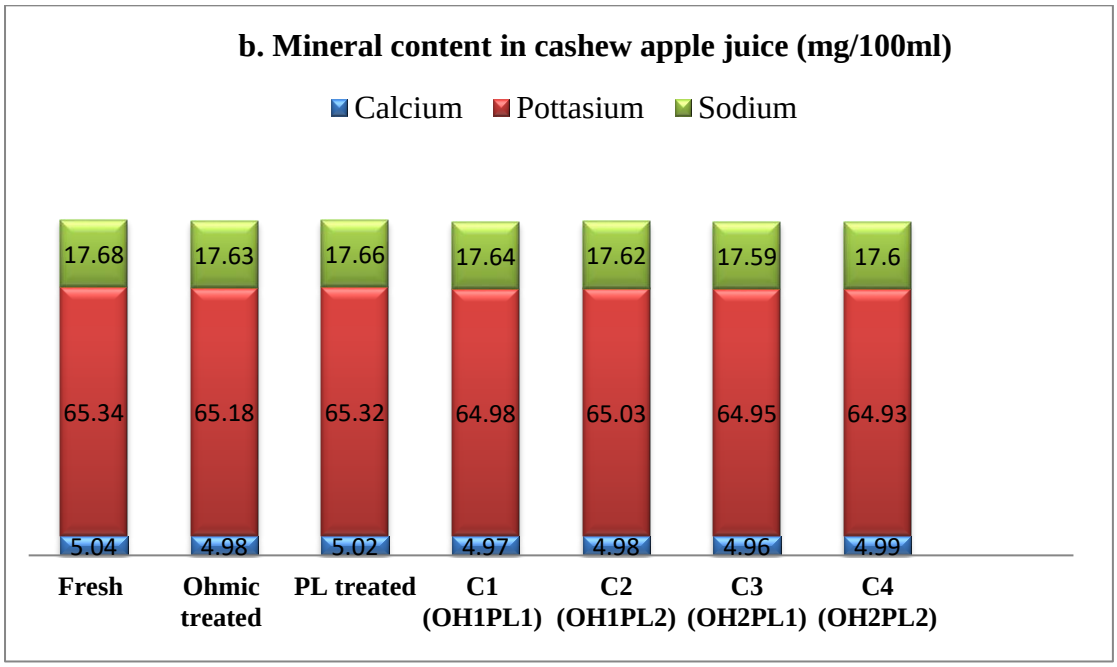


Fig.4.53 Effect of different treatments on mineral content of fruit juices

4.4.1.2 Effect of ohmic assisted PL treatments on the microbiological properties of fruit juices

The reduction in bacterial and yeast and mould population in pineapple and cashew apple juice treated under different treatment variables were evaluated as explained in section 3.7.1 and results are presented in Fig. 4.54 (a) and (b) and 4.55 (a) and (b) respectively. The results are tabulated in Appendix A.26 and A.27 and ANOVA tables are presented in Appendix A.28 and A.29.

A bacterial log reduction of 5.13, 5.07, 4.99 and 4.96 log cfu/ ml was observed in pineapple juice treated with ohmic assisted PL treatments such as P_1, P_2, P_3 and P_4. Similarly, a bacterial log reduction of 5.19, 5.12, 5.04 and 4.97 was obtained in cashew apple juice when treated with combined ohmic and PL treatment of C_1, C_2, C_3 and C_4 respectively. The highest reduction of 5.13 and 5.19 log cfu/ ml was found in treatments P_1 and C_1 which are optimal treatments of combined process of pineapple and cashew apple juice. The highest log reduction in bacterial count was observed in ohmic assisted PL treatments further followed by ohmic treatment alone and then in PL processing alone.

As may be seen from Fig. 4.56 (a) and (b), yeast and mould log reduction of 4.86, 4.85, 4.79 and 4.81 log cfu/ml were observed in pineapple juice treated with ohmic assisted PL treatments such as P_1, P_2, P_3 and P_4. Similarly, yeast and mould log reduction of 4.95, 4.92, 4.89 and 4.9 log cfu/ml were recorded in cashew apple juice when treated with combined ohmic and PL treatment of C_1, C_2, C_3 and C_4 respectively. The highest reduction of 4.86 and 4.95 log cfu/ml were reported in treatment P_1 in pineapple juice and C_1 in cashew apple juice which were combined treatments at optimised process conditions.

The microbial reduction in cashew apple juice was found higher than the pineapple juice for all treatment conditions. This could be due to the high electrical conductivity, minimum suspended particles and a greater transparency of cashew apple juice, which might have improved the effectiveness of ohmic heating and PL treatment. The higher proportion of suspended particles in pineapple juice would have provided a protective shielding effect for the microorganism from pulsed light effect.

As per Food and Drug Administration Codex Standards (FDA), a minimum of 5 log cfu/ml reduction in pathogenic microbial population in fruit juices is recommended for safe consumption and further storage (FDA, 1998). In this study, it was found that the ohmic and pulsed light treatments could not achieve the recommended log reduction of 5 log cfu/ml when applied individually within the specified operating conditions. All ohmic assisted PL treatment combinations could achieve more than 4.95 log reductions in bacterial count and highest reduction was recorded in optimised treatment conditions P_1 for pineapple juice and C_1 for cashew apple juice. The microbial reduction in PL system alone was found to be comparatively lower than ohmic treated samples in both juices. These results indicated that the efficiency of these two systems when applied individually in reducing microbial population to safe levels is limited. On the other hand when applied in a combined mode, the higher microbial reduction compared to individual ohmic and PL treatments could be attributed to the synergic action of ohmic and pulsed light effects on microbial inactivation.

When combined, the ohmic heating stage cause microbial destruction through the mild heat generation due to the resistance of the fluid to the electric field, and formation of pores due to the charges developed in the cell membrane of microorganisms called electroporation. The exposures to pulsed light in the subsequent processes, the microbial cells are further subjected to high intensity PL radiation, wherein, the microbial cell DNA was destructed by the UV- portion of the PL in addition to its photothermal and photophysical actions further ensuring reduction in microbial population. It is also reported that during combined ohmic and UV processing, in addition to the electroporation, thermal impacts and UV exposure might have also caused an irreversible collapse of cellular structures and consequent bacterial inactivation (Leizerson and Shimoni, 2005b; Sun *et al.*, 2008; Lee *et al.*, 2013).

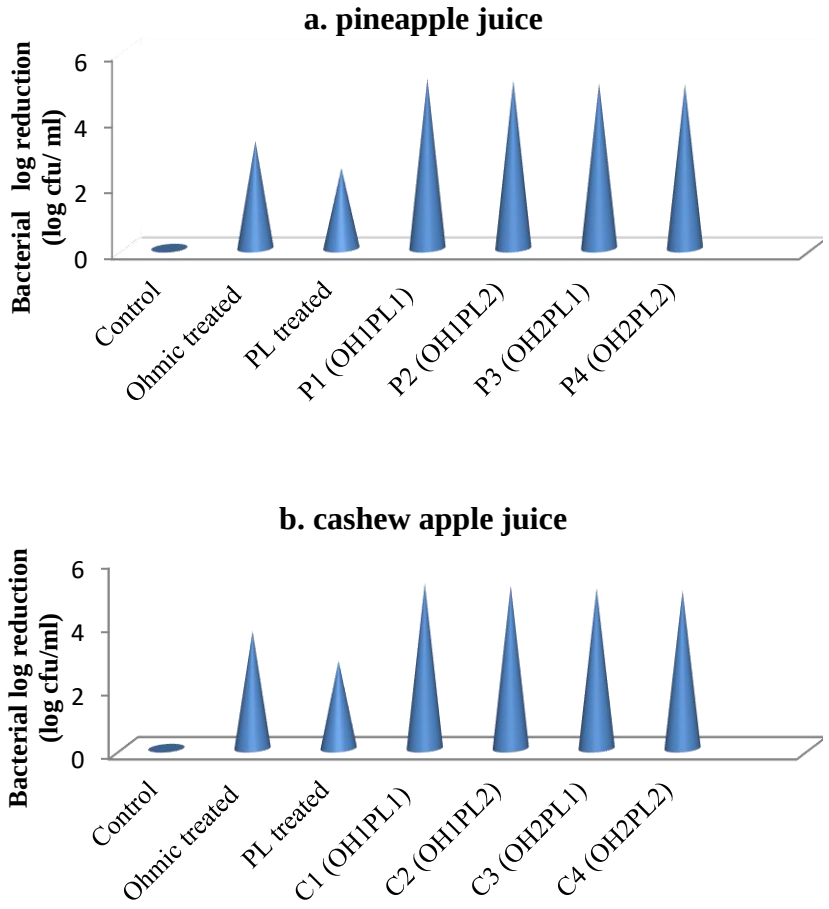


Fig.4.54 Effect of different treatments on Bacterial log reduction of fruit juices

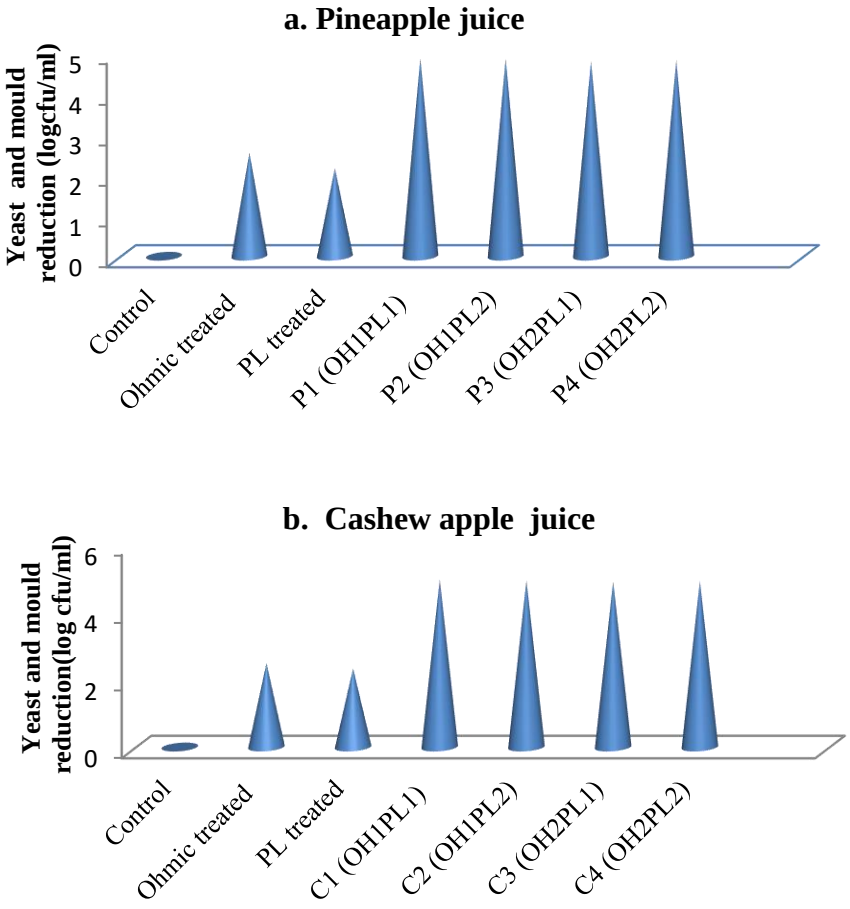


Fig.4.55 Effect of different treatments on yeast and mould reduction of the fruit juices

Similar results were also reported by Sean and Prince (2015) for pineapple juice subjected to combined ohmic and UV treatment. In similar lines, Lee *et al.* (2013) have found above 6 log microbial reduction in apple juice treated with combined ohmic and UV treatment. An additive effect of combined PEF and UV processing resulted in higher reduction of *Escherichia coli* (5.35 log cfu/ml) in apple juice, than in UV (3.46 log cfu/ml) and PEF (4.87 log cfu/ml) treatments alone (Gachovska *et al.*, 2008).

It could be very well concluded from the observation and results of the study that combination of ohmic heating and pulsed light process results in a hybrid effect and was successful enough in effectively reducing the microbial population to a safe level in pineapple and cashew apple juice at the optimised process conditions of both the process as established. The pineapple and cashew apple juice processed at the optimised processing condition could retain their nutritional quality comparable with that of fresh juices at the same time effectively bringing down the pathogenic and other spoilage microorganisms to a safe level for consumption as well as for increasing their shelf life.

4.4.2 Rheological Characteristics of ohmic assisted PL treated fruit juices

Assessment of rheological qualities of the fruit juices that has undergone the treatments under storage is important in analysing the flow behavior and the sensory qualities of the fruit juices. The flow behavior of the fresh, ohmic heated, PL treated and ohmic assisted PL treated pineapple and cashew apple juices were studied as described in section 3.6. Flow curves were generated and data was fitted into different models to categorise the juice sample's rheological behaviour using rheoplus software.

The rheological behaviour of the pineapple and cashew apple juice were analysed at shear rates ranged from 1 to100 s^{-1}. The variation of shear stress with shear rate of both the juices under different treatment conditions are displayed in Fig. 4.56 (a) and (b). It could be seen from the plot 4.56 (a) that all samples showed a nonlinear increase in the shear stress with the increase in shear rate. This non-linear relationship between shear stress and shear rate indicates the typical non-Newtonian behavior of juices.

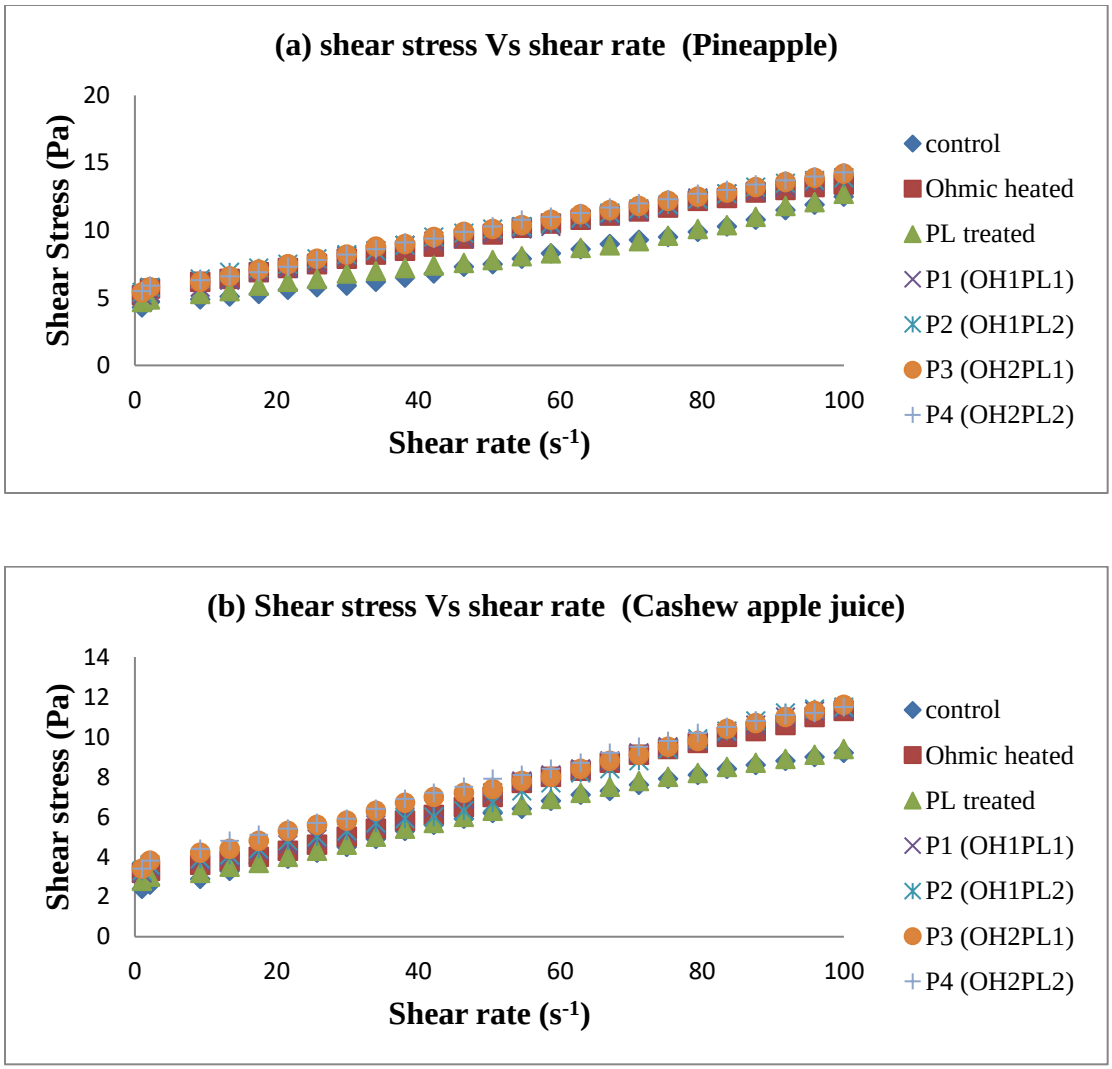


Fig. 4. 56 Effect of different treatments on shear stress of fruit juices

A plot of apparent viscosity versus shear rate is depicted in Fig. 4.57. (a) and (b) for pineapple and cashew apple juice. It may be inferred from the figures that viscosity decreased as shear rate increased. In general this behaviour is known as shear-thinning flow behaviour and the fluids with such behaviour are called pseudoplastic fluids (Rao, 2012). Similar results were also observed by Obot-Essien and Ufort-Usoh, (2016) in pineapple juice. The shear thinning behaviour had been noticed in passion fruit pulp by Moraes *et al.* (2011) and in mango pulps by Ahmed *et al.* (2004).

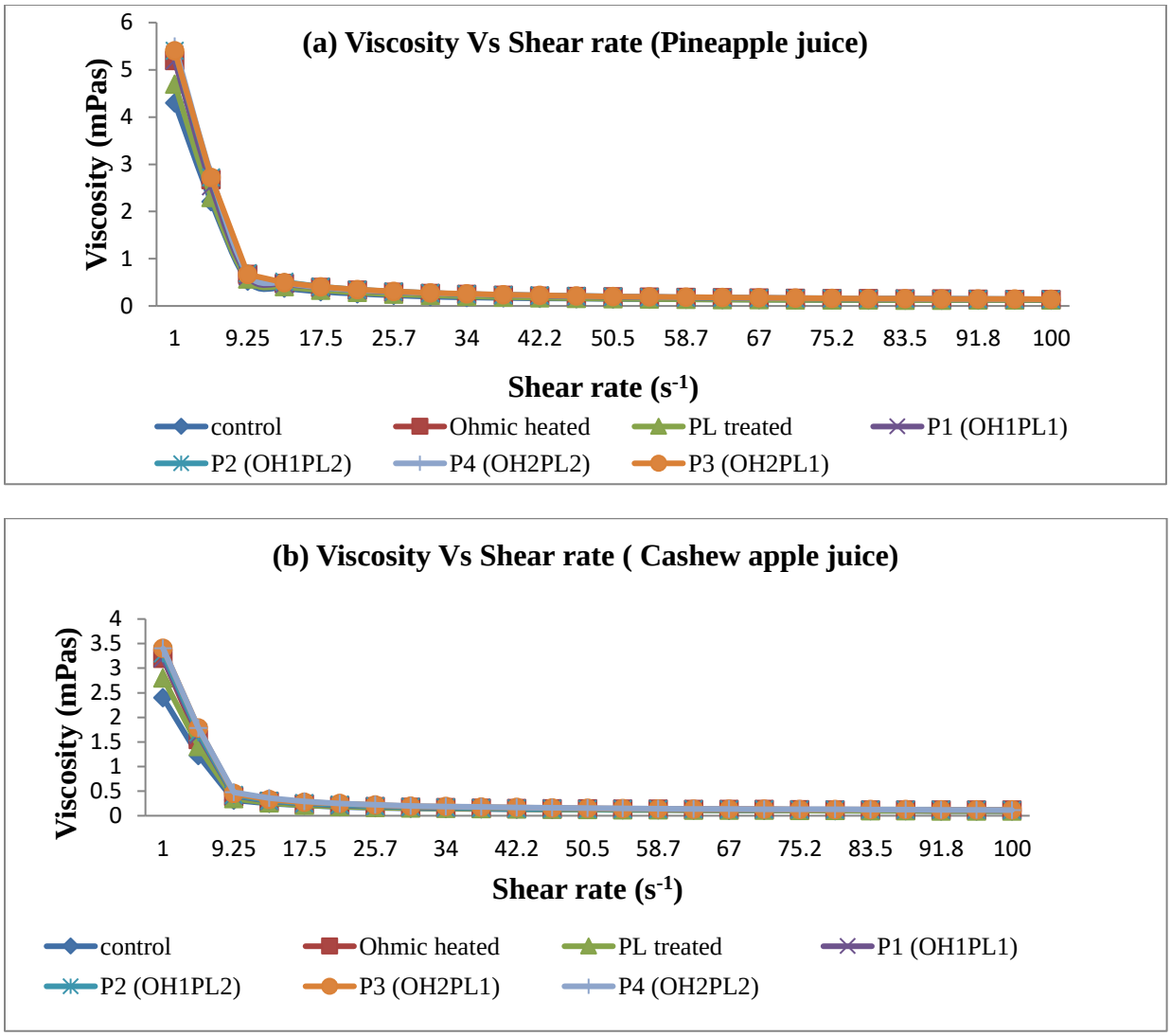


Fig. 4.57 Effect of different treatments on viscosity of fruit juices

Apparent viscosity of fruit juices, initially, decreased significantly with increasing shear rate until a shear rate of 9.25 s^{-1} in pineapple and cashew apple juice. Similar trend was observed for all other treatments too. At high shear rate, the viscosity reached some asymptotic value. According to Rao, (2012) three stage viscous response was observed when a shear-thinning fluid is sheared over a wide range of shear rates. The first stage at low shear rates, the viscosity of shear thinning fluid behaves like Newtonian fluid. This is then transited to pseudoplastic with further increase in shear rate as second stage. During

the last stage, at high shear rates, the relationship between the viscosity and shear stress was found to be limiting and constant infinite shear viscous.

It could also be observed from the Fig. 4.57 that among the treated samples ohmic heated and ohmic assited PL treated fruit juices of pineapple and cashew apple showed slight increase in viscosity variation pattern from control. Similar variation in viscosity pattern was observed in heated watermelon and carrot juice compared to the unprocessed juice (Vandresen *et al.,* 2009; Aguilo-Aguayo *et al.,* 2010). This could be due to the increase in particle size of the juices as a result of the coagulation of the colloidal materials present in the juices during the above ambient condition of ohmic heating (Yeom *et al.,* 2000; Vandresen *et al.,* 2009). Moreover the 'swelling' of the particles and the penetration of water between the cellulose chains during ohmic process could also impose viscosity increase to the heated juice samples (Cheftel, 1992; Vandresen *et al.,* 2009).

Data from the experiment were fitted into rheological models, the Herschel Bulkley and Ostwald (Power law) model, to obtain the rheological parameters of the pineapple and cashew apple juices and are listed in Table 4.9. Herschel Bulkley and Ostwald models best explains non Newtonian behavior. In this study these models exhibited very close fit to the experimental values of rheological parameters studied under various treatments for both the juices. Also, it may be concluded that the pineapple and cashew apple juices under all treatments were non Newtonian in nature.

From Table 4.9, the coefficient of determination (R^2) was found to be in the range of 0.83 and 0.89 for the fresh, ohmic heated, PL treated and ohmic assisted PL treated pineapple juice samples. The values for the flow behaviour index (n) of the pineapple juice ranged from 0.632 to 0.767. The flow behaviour index of fruit juices was lesser than one, indicating their non-Newtonian behaviour.

Table 4.9 Herchel-Bulkley and Ostwald parameters for pineapple and cashew apple juice

Herchel-Bulkley parameters of pineapple juice			
Treatments	Consistency Index (K), Pa·s	Flowindex (n)	R^2
Fresh	0.00356	0.767	0.87
Ohmic treated	0.00382	0.732	0.82
PL treated	0.00361	0.756	0.85
$P_1(OH_1PL_1)$	0.00384	0.715	0.89
$P_2(OH_1PL_2)$	0.00384	0.692	0.86
$P_3(OH_2PL_1)$	0.00383	0.632	0.85
$P_4(OH_2PL_2)$	0.00386	0.654	0.83
Ostwald model parameters of cashew apple juice			
Treatments	Consistency Index (K), Pa·s	Flowindex (n)	R^2
Fresh	0.032	0.876	0.89
Ohmic treated	0.053	0.863	0.86
PL treated	0.045	0.872	0.84
$C_1(OH_1PL_1)$	0.054	0.851	0.87
$C_2(OH_1PL_2)$	0.056	0.848	0.85
$C_3(OH_2PL_1)$	0.055	0.836	0.87
$C_4(OH_2PL_2)$	0.055	0.799	0.86

The consistency index (K) is a rheological parameter that reflects the viscosity of the fluid. The consistency index (K) obtained for fresh, ohmic treated and ohmic assisted PL treated pineapple juices are respectively 0.00356, 0.00382, 0.00384 Pa·s. This increase in the consistency index might be attributed to the decrease in viscosity values of pineapple juice in tune with the increase in heat sensitivity of the respective treatments.

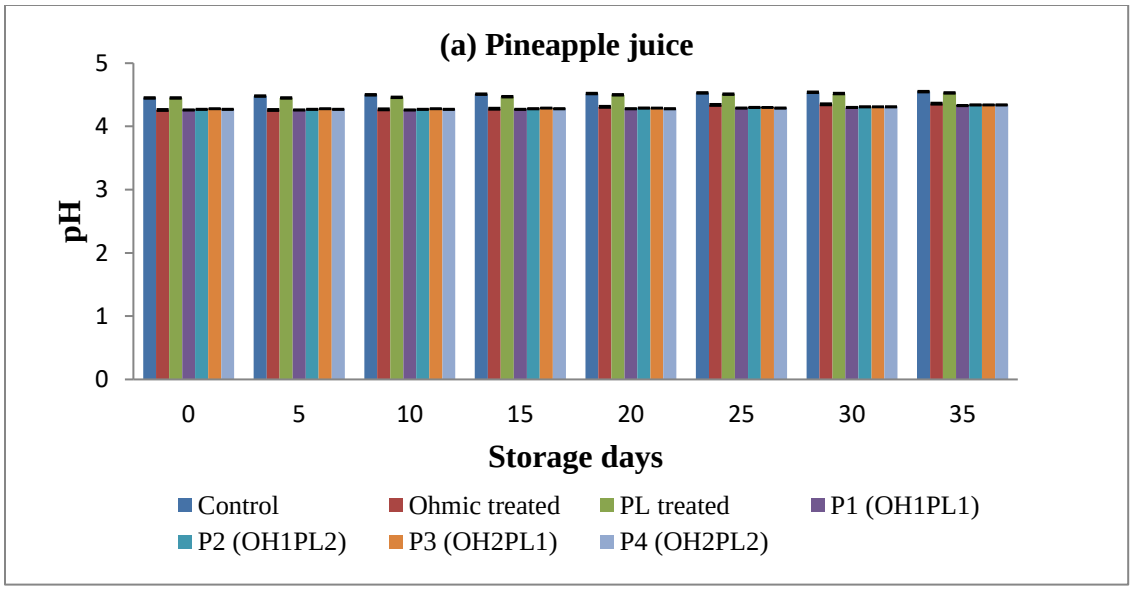
In cashew apple juice, the experimental data were fit into the Ostwald model and Herschel Bulkley model. The R^2 values for the Ostwald model were found to be higher than the Herschel-Bulkley. Ostwald model is a two-parameter model that is widely used to characterise flow behaviour of liquid food. Values of the shear stress and shear rate were fitted into Equation (3) using the rheoplus/32 V3.61 software to obtain the Ostwald parameters (Table 4.9). The results showed a good fitting to power law with higher R^2 values of approximately 0.87. The flow behavior indexes of the samples were found to be

between 0.799 and 0.876 indicating shear thinning behaviour (pseudoplastic nature) of the cashew apple juice. The flow behavior index was found to be below 1 (n<1) for both models. Silva *et al.* (2014) also reported flow behavior index (n) values of less than one in samples of cashew, mango, and acerola pulps. No significant differences in n values were observed between PL treated and fresh samples of both the juices. Similarly Shamsudin *et al.* (2013) also observed no significant differences in apparent viscosities among the UV-irradiated and untreated juice.

4.5 STORAGE STUDIES

The effect of different process conditions of ohmic heating, PL treatment, and ohmic assisted PL treatment on the quality characteristics of pineapple and cashew apple juices were discussed in previous sections. The shelf life of the optimally treated fruit juices were determined to access its keeping quality in the long run. In order to assess the effectiveness of the developed system towards microbial stability and quality retention during the storage of pineapple and cashew apple juices, the ohmic heated, pulsed light treated and optimally treated juices as mentioned in section (3.5) were analysed and compared with fresh juices as control. The physicochemical and microbiological characteristics of fresh and treated pineapple and cashew apple juices stored at refrigerated conditions (4±2°C) were analysed at five days interval for a storage period of 35 days. The prelmnary studies revealed that the maximum qualty retention period of combined treated fruit juices was ranged from 25 to 35 days of storage. The results are tabulated under Appendix A.30 and A.31 ANOVA tables are presented in Appendix A.32.

The variation in pH of the fruit juices under different treatment conditions during storage are presented in Fig. 4.58 (a) and (b).



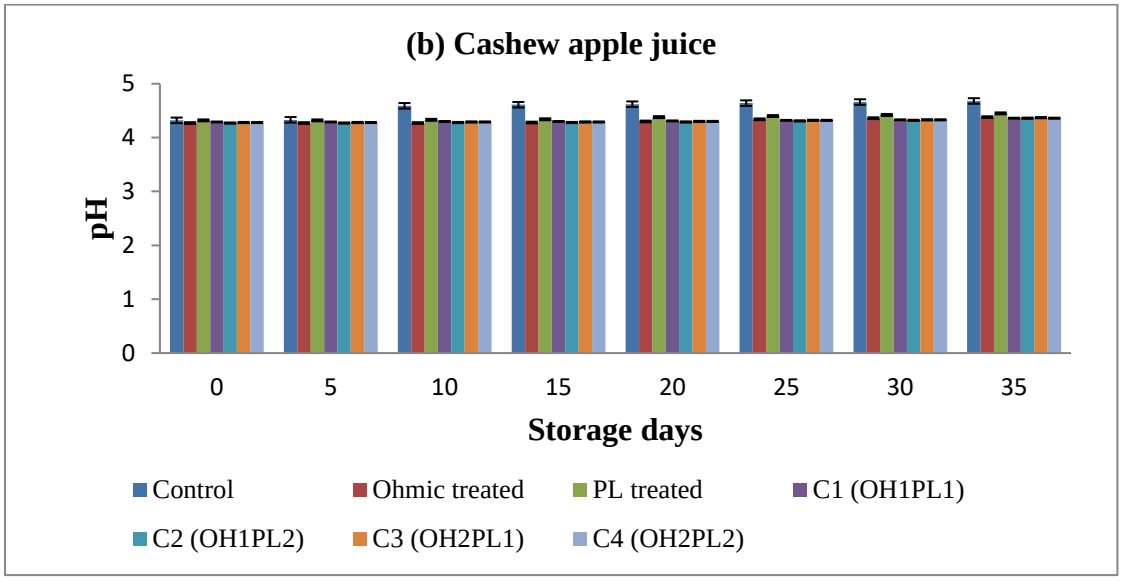


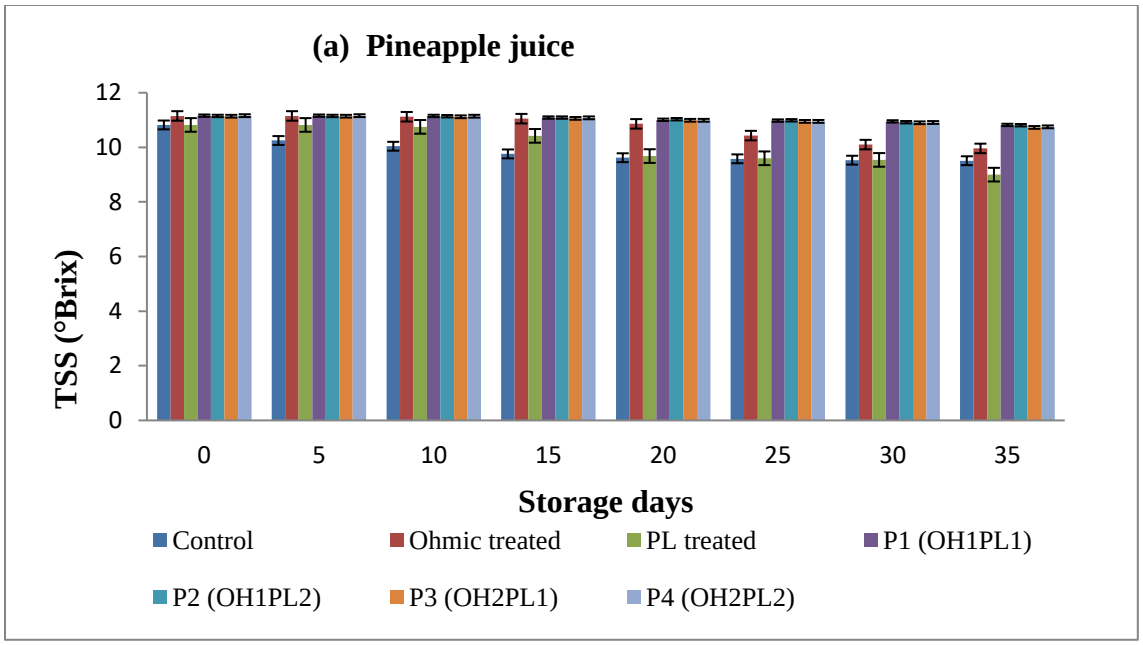
Fig. 4.58 Effect of different treatments on pH of fruit juices during storage

From the figures it could be revealed that the fresh pineapple and cashew apple juice had an initial pH of 4.45 and 4.32 respectively. In the case of control samples it was found that pH of juices suddenly increased after 5th day of storage and showed values

of 4.55 and 4.68 after 35 days of storage respectively. A higher increase in pH was observed after 10^{th} day of storage in PL processed pineapple juice, whereas, PL treated cashew apple juice showed a higher increase after 15^{th} day of storage. A significant variation in pH values was observed after the 20^{th} and 25^{th} day of storage in ohmic heated pineapple and cashew apple juice respectively. The combined treated pineapple and cashew apple juice samples exhibited a negligible gradual increase in pH until 25 and 30 days of storage at refrigerated condition and thereafter pH was found to exhibit sudden increase. It was found that among all the treatments under study, the optimally treated sample (P_1) and (C_1) showed minimum variation in pH from the initial pH values.

In the case of optimally treated pineapple juice, the initial pH values was 4.26 which varied to 4.3 at the end of 25 storage days under refrigerated condition. The initial pH of optimally treated cashew apple juice was 4.29 which increased to 4.33 at the end of 30 days under refrigerated storage. Though this variation is in statistically significant ($p<0.01$) this was found to be the minimum variation among all other treatments. Also, the variation is within the pH values of 4.5 showing acid pH range of fruit juices in which the fresh pineapple juice also falls. In case of optimally treated cashew apple juice, the initial pH value was 4.29 which varied to 4.33 at the end of 30 storage days under refrigerated condition. The variation in pH during storage could be attributed to the growth of microorganisms and associated conversion of total soluble solids to acids.

The effect of different treatment conditions on the total soluble solid content of pineapple and cashew apple juice during storage is presented in the Fig. 4.59 (a) and (b).



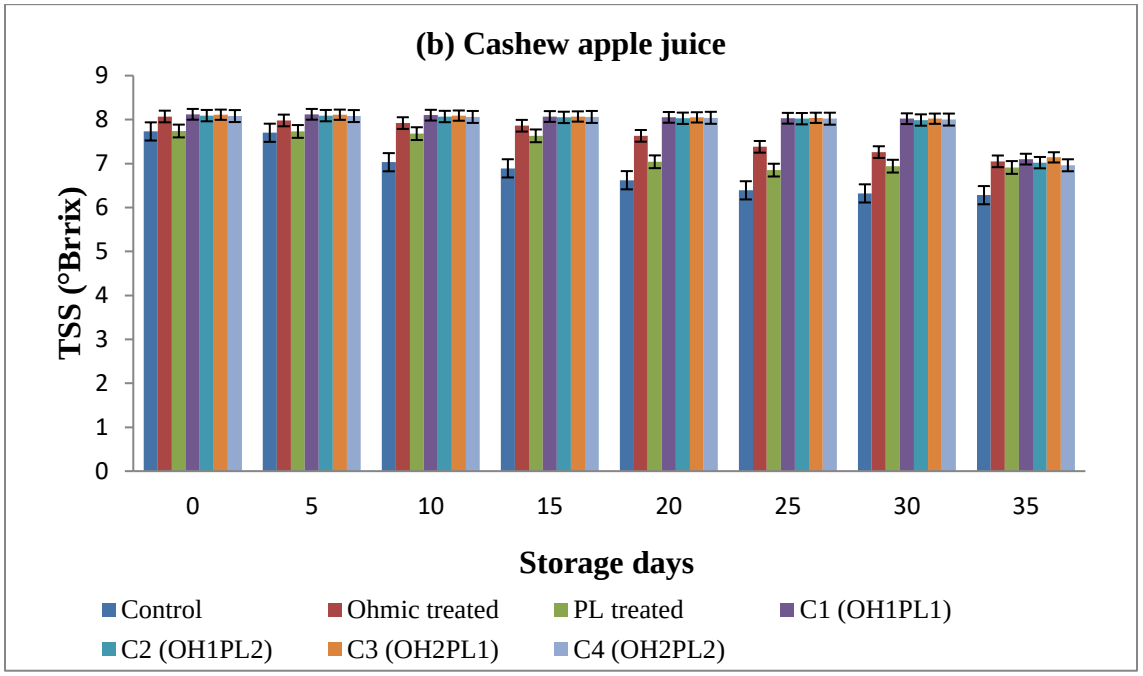


Fig. 4.59 Effect of different treatments on TSS of fruit juices during storage

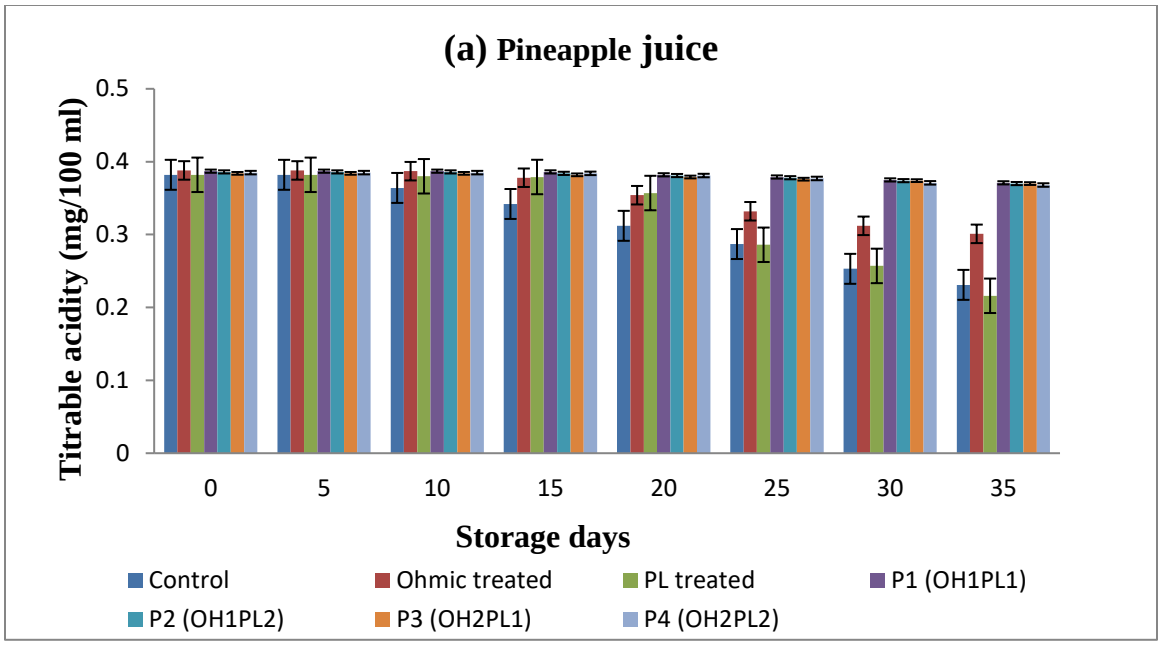
It was found that the fresh pineapple and cashew apple juice had an initial total soluble solid content of 10.82 and 7.73°Brix respectively. During the storage, the TSS content reduced to 9.51 and 6.28°Brix following an irregular pattern for pineapple and cashew apple juice respectively, at the end of 35 days of storage period. The TSS content of fruit juices registered a significant change ($p < 0.01$) during the storage period.

The ohmic heated pineapple juice showed an increased reduction in TSS after 20 days of storage whereas that of cashew apple juice the increased variation was recorded after 25 days of storage. The PL treated pineapple and cashew apple juice also observed a significant reduction in TSS after 10^{th} and 15^{th} days of storage, respectively, which might be due to the spoilage of juice with the action of microbiological population.

All the combined treated fruit juices showed gradual reduction in TSS with increase in storage period. Among all the combined treated samples, the optimally treated samples showed minimum variation of 1.8% and 1.2% respectively for pineapple (P_1) and cashew apple (C_1) respectively. In the case of optimally treated pineapple juice, the initial TSS value was 11.16°Brix which varied to 0.98°Brix at the end of 25 storage days under refrigerated condition. The initial pH of optimally treated cashew apple juice was 8.12°Brix which increased to 8.02°Brix at the end of 30 days under refrigerated storage. Though this variation is statistically significant ($p < 0.01$) this was found to be the minimum variation among all other treatments.

The change in TSS might be due to the growth of microorganisms during the storage period, which would have converted the total soluble solids into acid through fermentation (Rivas *et al.*, 2006). The microbial fermentation is a biochemical process of breaking down the glucose molecules to acids and other products (Rosen and Gothard, 2010). Chia *et al*. (2012) reported similar changes in TSS in UV treated pineapple juice. Teja *et al.* (2017) also reported minor changes in soluble solids of UV-treated apple cider during storage.

The effect of various treatment conditions on titrable acidity of pineapple and cashew apple juice during storage is presented in Fig. 4.60 (a) and (b).



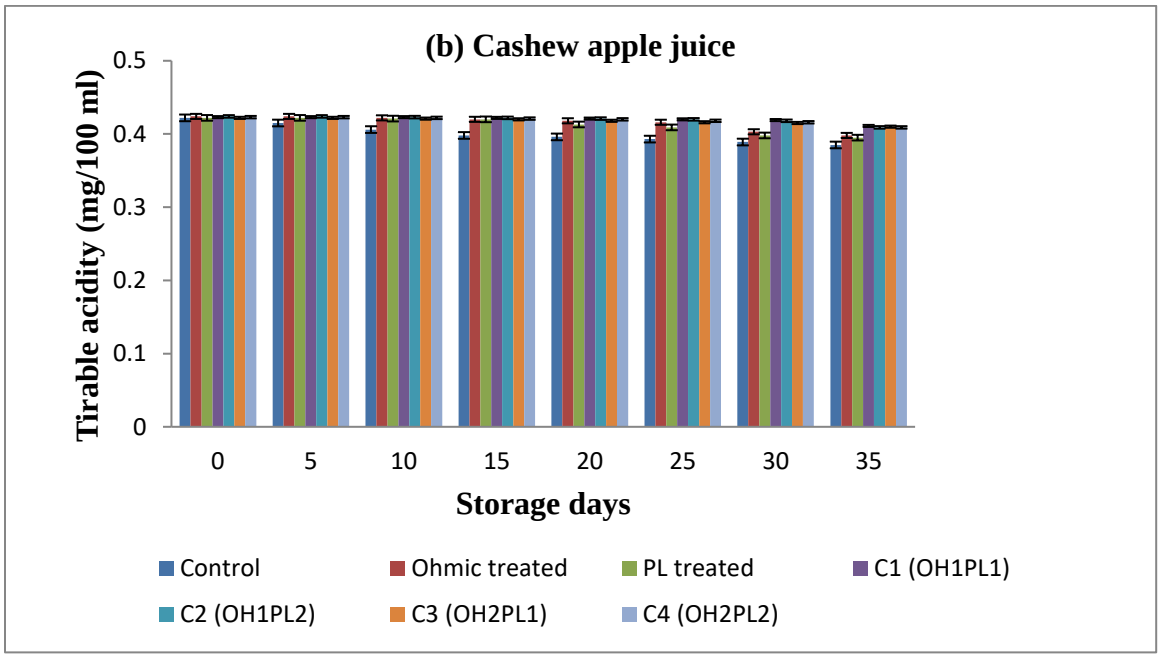


Fig. 4.60 Effect of different treatments on titrable acidity of fruit juices during storage

From the Fig. 4.60 (a) it could be derived that the fresh pineapple juice had an initial titrable acidity value of 0.382 mg/100 ml. It was found that titrable acidity of fresh pineapple juice suddenly decreased after 5th day of storage and showed a value of 0.18 mg/100 ml after 30 days of storage. A higher reduction in titrable acidity values was observed after 10th day of storage in PL processed pineapple juice. A significant variation in titrable acidity values was observed after the 20th day of storage in ohmic heated pineapple juice. The combined treated samples exhibited a negligible gradual decrease in titrable acidity until 30 days storage at refrigerated condition and thereafter titrable acidity was found to exhibit sudden decrease. It was found that among all the treatments under study, the optimally treated sample (P_1) showed minimum variation in titrable acidity values from the initial values. The optimally treated samples of pineapple juice resulted in only a 3.1% reduction in titrable acidity.

From the Fig. 4.60 (a) it could be seen that the fresh cashew apple juice had an initial titrable acidity value of 0.422 mg/100 ml. In the case of control samples it was found that titrable acidity suddenly increased after 5th day of storage and showed a value of 0.385 mg/100ml after 35 days of storage. A higher increase in titrable acidity values were observed after 15th day of storage in PL processed cashew apple juice. A significant variation in titrable acidity values were observed after the 25th day of storage in ohmic heated cashew apple juice. The combined treated samples exhibited a negligible gradual increase in titrable acidity until 30 days of storage at refrigerated condition and thereafter titrable acidity was found to exhibit sudden increase. It was found that among all the treatments under study, the optimally treated sample (C_1) showed minimum variation in titrable acidity values from the initial values. The reduction in titrable acidity during storage might be due to the release of acids by decomposition, hydrolysis, oxidation, or fermentation, which modifies the hydrogen ion concentration and consequently, food acidity (Talasila *et al.,* 2011). The titrable acidity may also vary due to different chemical changes such as breakdown of pectic substances, oxidation of reducing sugars and degradation of polysaccharides etc. during storage (Ibarz *et al.*, 2000).

The effect of different treatment conditions on the ascorbic acid content of pineapple and cashew apple juice during storage is presented in the Fig. 4.61 (a) and (b). The ascorbic acid values showed a significant reduction ($p < 0.01$) in all samples throughout the storage period.

It was found that the fresh pineapple and cashew apple juice had an initial ascorbic acid content of 28.96 and 162.96 mg/100 ml respectively. During the storage, the ascorbic acid content reduced to 14.32 and 98.01 mg/100 ml following an irregular pattern for pineapple and cashew apple juice respectively, at the end of 35 days of storage period and the reduction process were found to continue even further with storage.

The ohmic heated pineapple juice showed an increased reduction in ascorbic acid after 20 days of storage, whereas that of cashew apple juices, the increased variation was recorded after 25 days of storage. The PL treated pineapple and cashew apple juice also observed a significant reduction in ascorbic acid content after 10^{th} and 15^{th} day of storage respectively. All the combined treated fruit juices showed gradual reduction in ascorbic acid content with increase in storage period. Among all the combined treated samples, the optimally treated samples showed minimum variation of 9.8% and 5.9% respectively for pineapple (P_1) and cashew apple (C_1) juice respectively.

In the case of optimally treated pineapple juice, the initial ascorbic acid content was 27.13 mg/100ml which got reduced to 24.82 mg/100ml at the end of 25 storage days under refrigerated condition. The initial ascorbic acid content of optimally treated cashew apple juice was 150.02 mg/100 ml which was found reduced to 141.92 mg/100ml at the end of 30 days under refrigerated storage. Though this variation is statistically significant ($p < 0.01$) this was found to be the minimum variation among all other treatments. According to Campos *et al.* (2002), the cashew apple juice could be considered a potential source of vitamine C even after considerable loss of the same after usual thermal treatments. In this case only 5.9% reduction in ascorbic acid was reported at the end of 30 days of storage for the optimally treated sample.

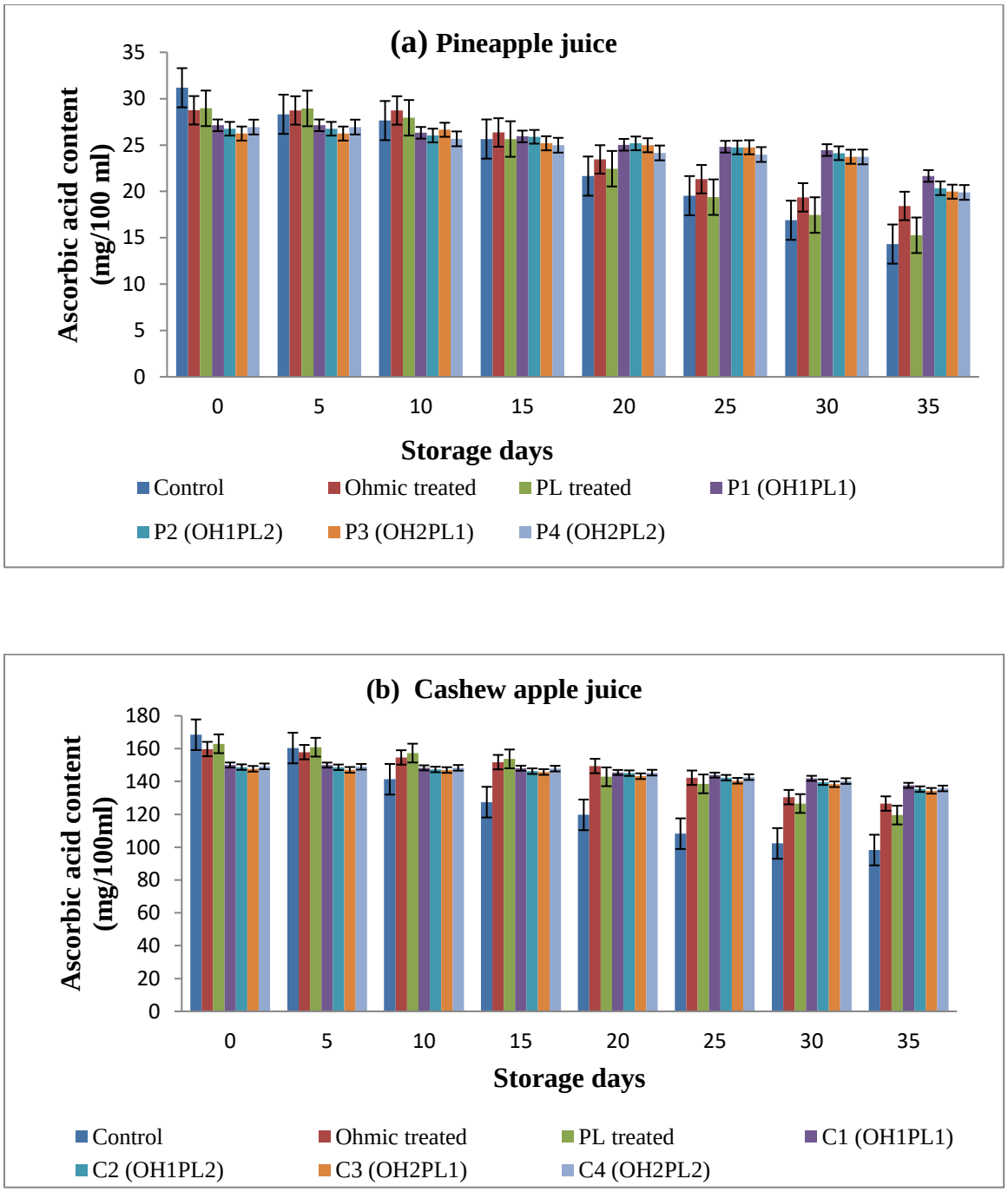


Fig. 4.61 Effect of different treatments on ascorbic acid content of fruit juices during storage

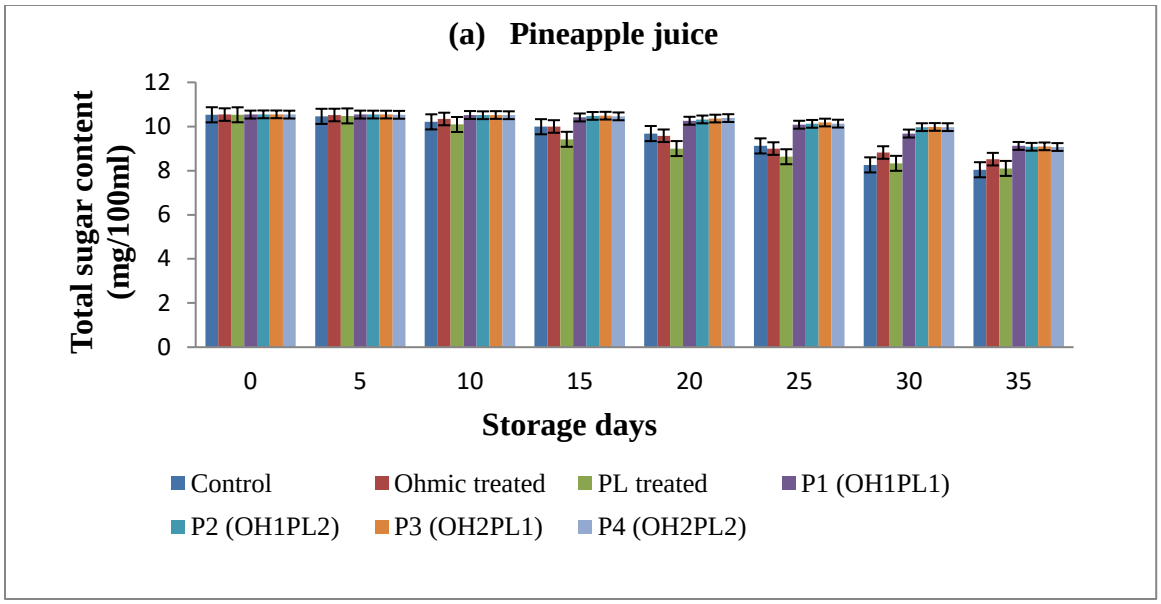
The reduction in ascorbic acid could be attributed to their oxidation due to the degradation caused by exposure to oxygen, light or heat or it could also be due to

enzymes such as peroxidase and ascorbate oxidase (Davey *et al*., 2000; Odriozola-Serrano *et al*., 2008). The storage temperature and packaging systems also affect the vitamin degradation. Though the variations are significant statistically, the real time magnitude of reduction is remains very small when compared to thermally treated fruit juices.

The variation in the total sugar content of fresh and treated pineapple and cashew apple juices during storage are depicted in Fig. 4.62 (a) and (b). The storage period were found to show a significant effect on the total sugar of pineapple and cashew apple juice ($p < 0.01$).

It was found that the fresh pineapple and cashew apple juice had an initial total sugar content of 10.54 and 8.04 mg/100 ml respectively. During the storage, the total sugar content got reduced to 9.97 and 7.86 mg/100 ml at the end of 35 days of storage following an irregular pattern for pineapple and cashew apple juice respectively.

A higher reduction in total sugar was observed after 10^{th} day of storage in PL processed pineapple juice, whereas, PL treated cashew apple juice showed a higher reduction after 15^{th} day of storage. A significant variation in total sugar content was observed after the 20^{th} and 25^{th} day of storage in ohmic heated pineapple juice and cashew apple juice. The combined treated samples exhibited a negligible gradual reduction in total sugar content until 30 days storage at refrigerated condition and thereafter total sugar content was found to exhibit higher reduction. It was found that among all the treatments under study, the optimally treated pineapple juice (P_1) and cashew apple juice (C_1) showed minimum variation in total sugar from initial total sugar values.



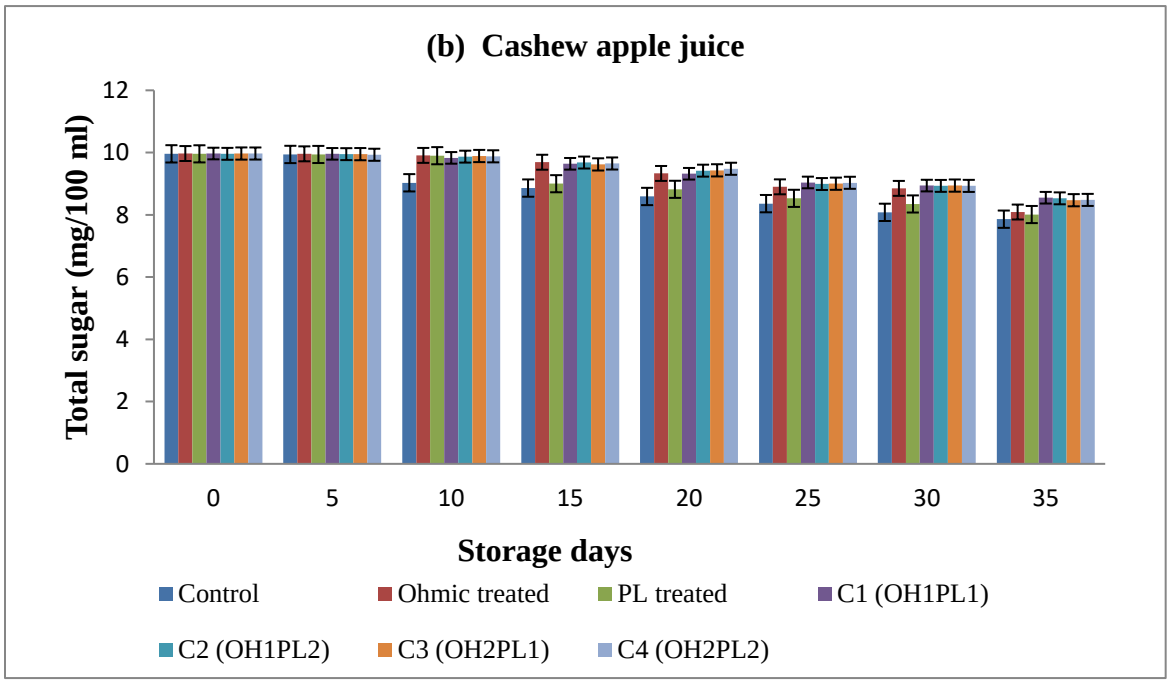


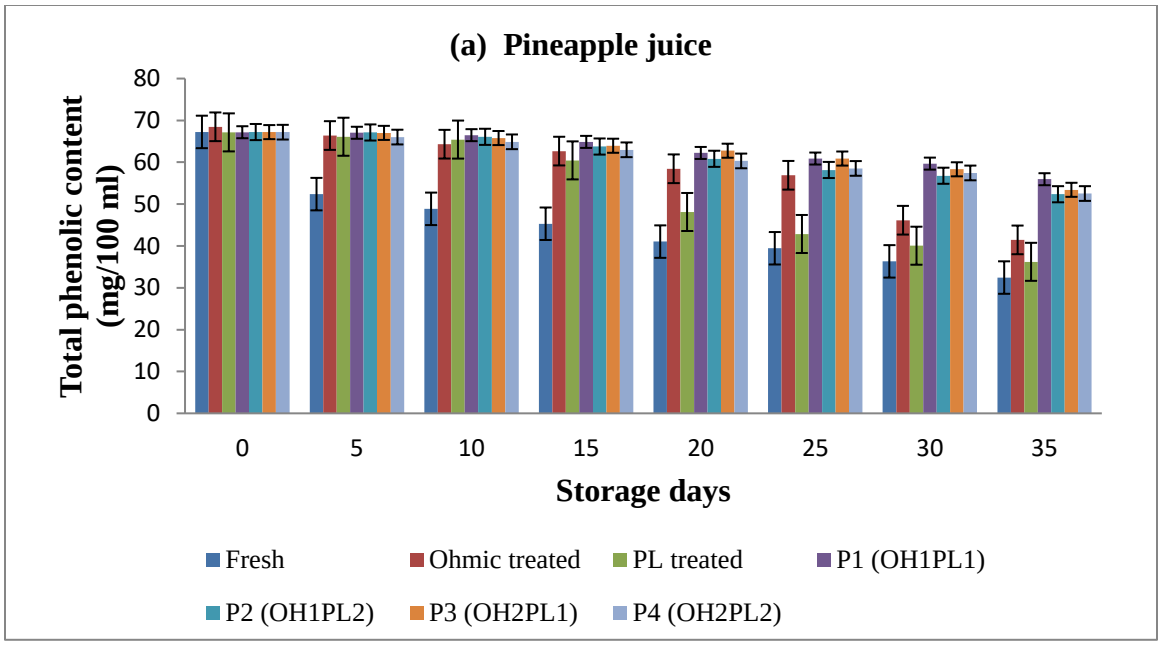
Fig. 4.62 Effect of different treatments on total sugar of fruit juices during storage

In the case of optimally treated pineapple juice, the initial total sugar content was 10.54 mg/100 ml which varied to 10.08 mg/100 ml at the end of 25 storage days under refrigerated condition. The initial total sugar content of optimally treated cashew apple juice was 9.97 mg/100 ml which was found reduced to 8.94 mg/100 ml at the end of 30 days under refrigerated storage condition. Though this variation is statistically significant ($p < 0.01$) this was found to be the minimum variation among all other treatments.

A similar reduction was also reported in cashew apple juice processed with chemical preservatives (Shaik, 2012; Talasila *et al.,* 2011). Kausar *et al.* (2012) also noted a decrease in reducing sugars with increased storage days for cucumber-melon functional drink. Bottle gourd-basil leaf juice reported a 70% reduction in reducing sugars during 6 months of storage when stored at a temperature of 4 ±2°C (Majumdar *et al.,* 2011). The decrease in sugar content of juices might be attributed to the non enzymatic reactions which include pigment destruction, caramelisation, and condensation between amino acids and reducing sugars that takes place during storage (Shaik, 2012; Talasila *et al.,* 2011). The decrease in sugars during the storage could also be due to the microbial fermentation wherein the sugars get converted to acids and the acidic conditions leads to an inversion of sucrose.

The effect of various processing treatments on the total phenolic content of pineapple and cashew apple juice during storage is presented in Fig. 4.63 (a) and (b). A significant decrease ($p < 0.01$) of total phenolic content was observed in all treated samples.

It may be seen from the figures that the total phenolic content of fresh pineapple and cashew apple juice samples recorded an initial phenolic content of 67.32 mg/100ml and 168.46 mg/100 ml respectively. In the case of fresh samples, it was found that the phenolic content of juices suddenly decreased after 5^{th} day of storage and showed a reduction of 51.17% and 42% after 35 days of storage respectively.



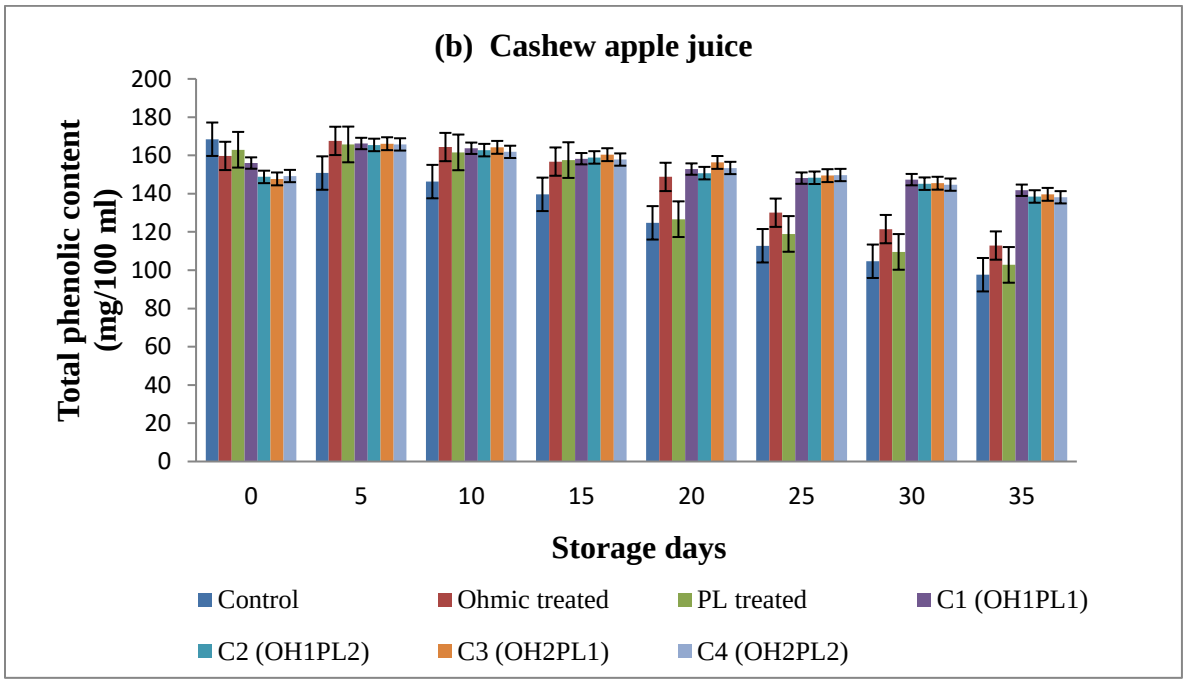


Fig. 4.63 Effect of different treatments on phenolic content of fruit juices during storage

A gradual reduction in phenolic content up to 10^{th} and 15^{th} day of storage period was observed in both PL treated pineapple and cashew apple juice. After that an increase in the reduction of phenolic content was noted in both juice samples.

A 8.4% and 18.5% reduction in phenolic content was observed up to the 20^{th} and 25^{th} day of storage in ohmic heated pineapple juice and cashew apple juice respectively. The combined treated samples of pineapple and cashew apple juice exhibited a negligible gradual reduction in phenolic content until 25 and 30 days storage at refrigerated condition and thereafter, phenolic content was found to decrease at a faster rate. It was found that among all the treatments under study, the optimally treated samples P_1 and C_1 showed a minimum reduction from the initial phenolic content. Even though reductions in phenolic contents were found in both juices, the variations did not exhibit any considerable and objectionable changes in the organoleptic qualities of fruit juices.

The variation in phenolic content during storage could be attributed to the product degradation caused by formation of haze, browning reactions, and sediment formation (Macheix *et al*., 1990). The lowest percent reduction during the storage observed in ohmic assisted PL treatment could be due to the inactivation of peroxidase enzyme which causes degradation of phenolic compounds (Odriozola-Serrano *et al.*, 2008).

The variation in tannin content of fresh clarified and treated clarified cashew apple juice during storage is presented in Fig. 4.64. A significant reduction ($P<0.01$) in tannin content was observed in fresh and treated cashew apple juice during storage period.

Fresh cashew apple juice had an initial tannin content of 0.57%. In the case of control sample it was found that tannin content was reduced to 0.32% after 30 days of storage. A higher reduction in tannin content was observed after 15^{th} day of storage in PL processed cashew apple juice whereas, the highest variation was observed after the 25^{th} day of storage in ohmic heated cashew apple juice. The combined treated samples exhibited a negligible gradual reduction in tannin content until 30 days storage at refrigerated condition and thereafter noticed a higher reduction in tannin content. It was

found that among all the treatments under study, the optimally treated sample (C_1) showed minimum variation in tannin content from the initial tannin content.

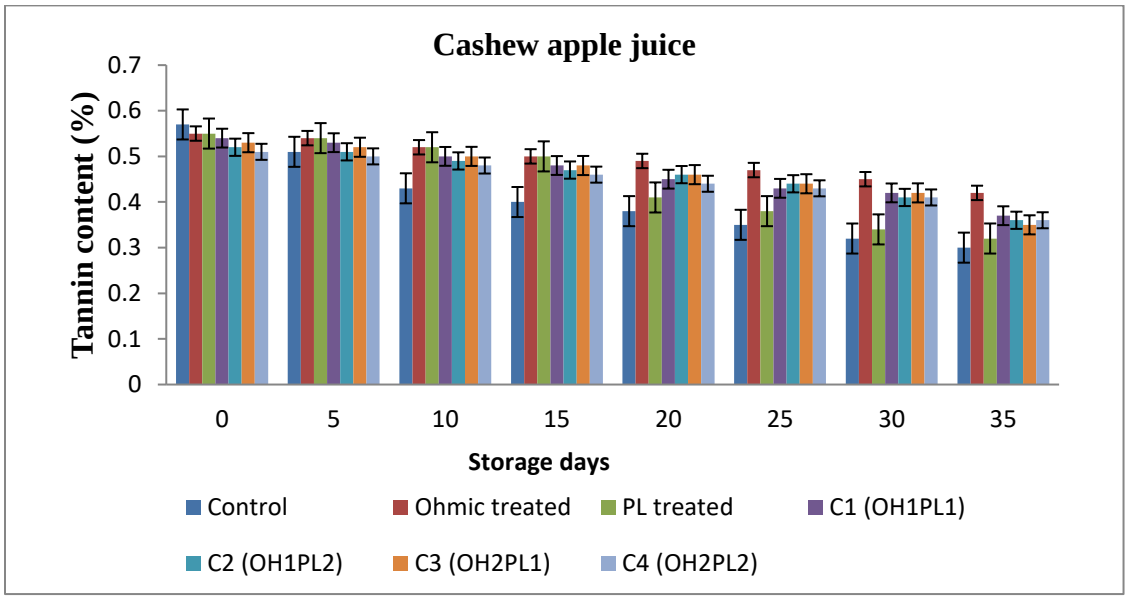


Fig. 4.64 Effect of different treatments on tannin content of cashew apple juice during storage

The highest reduction was found in fresh juice followed by PL treated, ohmic heated and combined treated juice samples in the decreasing order. A similar reduction in tannin content during storage is reported in cashew apple juice treated with chemical preservatives and combined physiochemical methods (Shaik, 2012; Talashila *et al.*, 2011). This could be due to the increase in the number of microorganisms during storage period. The microorganisms present in the juices produce certain enzymes, which might have degraded the tannins present in juices (Shiv-Kumar, 2015). Campos *et al.*, (2002) pointed out that the decrease in tannin content could also be due to the reaction of phenolic compounds leading to darkening of cashew juice.

The effect of different treatment conditions on the total colour difference of pineapple and cashew apple juice during storage is presented in the Fig. 4.65 (a) and (b). The total colour difference values showed an increasing trend with the progress of storage days for the pineapple and cashew apple juice.

The fresh juice samples of pineapple and cashew apple exhibited a high increase in colour difference from the 5th day of storage and arrived at a final value of 2.6 and 2.3 respectively. As per the colour difference classification of Cserhalmi *et al.* (2006), this value falls under the category of noticeable range. The PL treated pineapple and cashew apple juice also showed a noticeable change in total colour difference after 10th and 15th day of storage respectively. Ohmic assisted PL treated fruit juices fell in the category of slightly noticeable levels (Cserhalmi *et al.,* 2006). Ohmic heated pineapple juice recorded a colour difference value of 1.24 after 20 days of storage, and that of cashew apple juice recorded a colour dififference value of 1.5 after 25 days of storage. After 30 days of storage a visible change in colour was found in both the ohmic heated fruit juices. All ohmic assisted PL treated samples sustained total colour difference value less than one throughout the storage period, implying only slightly visual colour change even after 30 days of storage.

In the case of optimally treated pineapple and cashew apple juice, the colour difference increased with the progression of storage days, and finally recorded a total colour difference of 0.83 and 0.91 in pineapple and cashew apple juice at the end of 30 storage days under refrigerated condition. Though this variation is statistically significant ($p < 0.01$) this was found to be the minimum variation among all other treatments. Also, the variation is within the total colour difference of one, revealing only slightly visible colour change in fruit juices as stated by Choi *et al.* (2010).

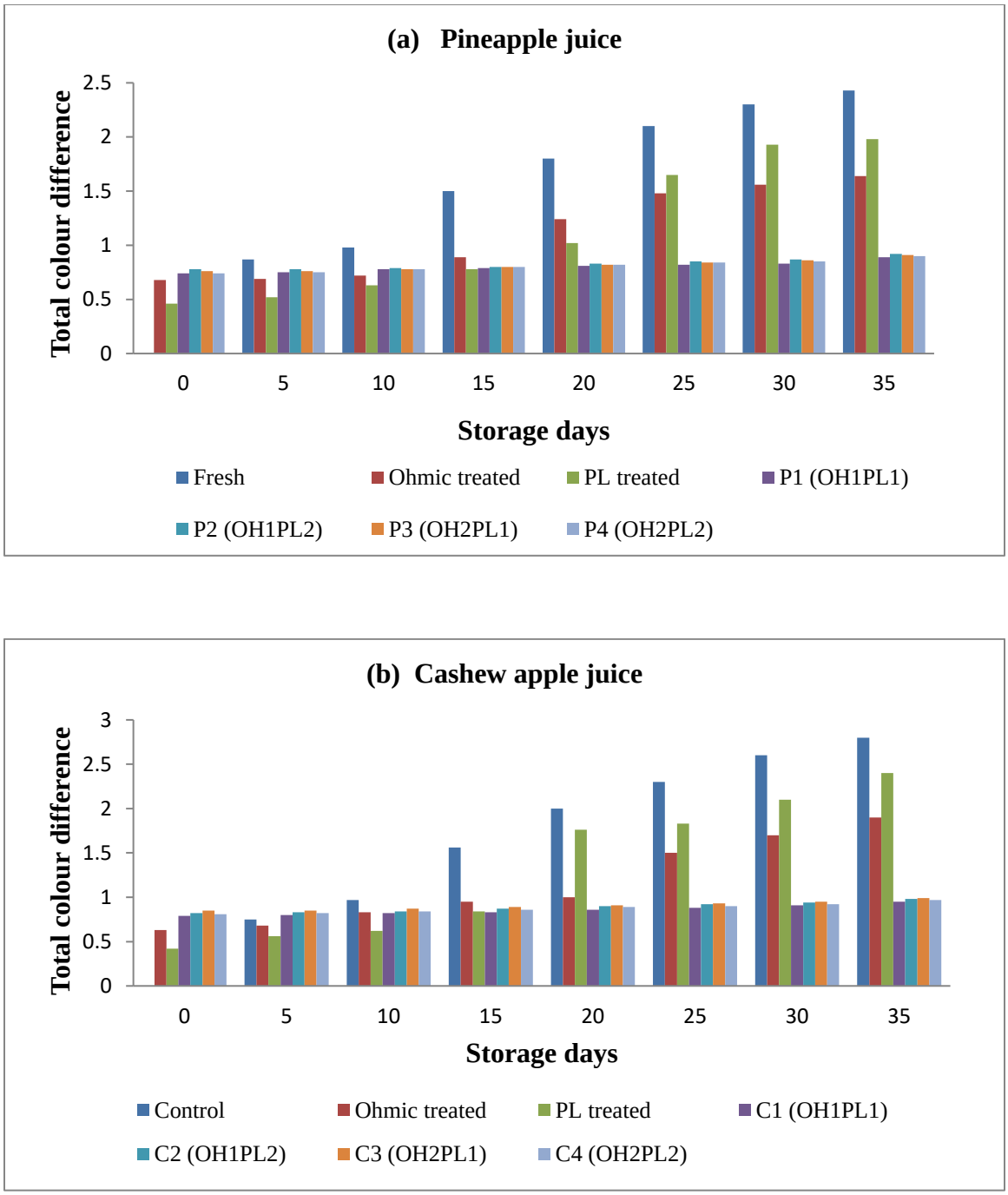


Fig. 4.65 Effect of different treatments on total colour difference of fruit juices during storage

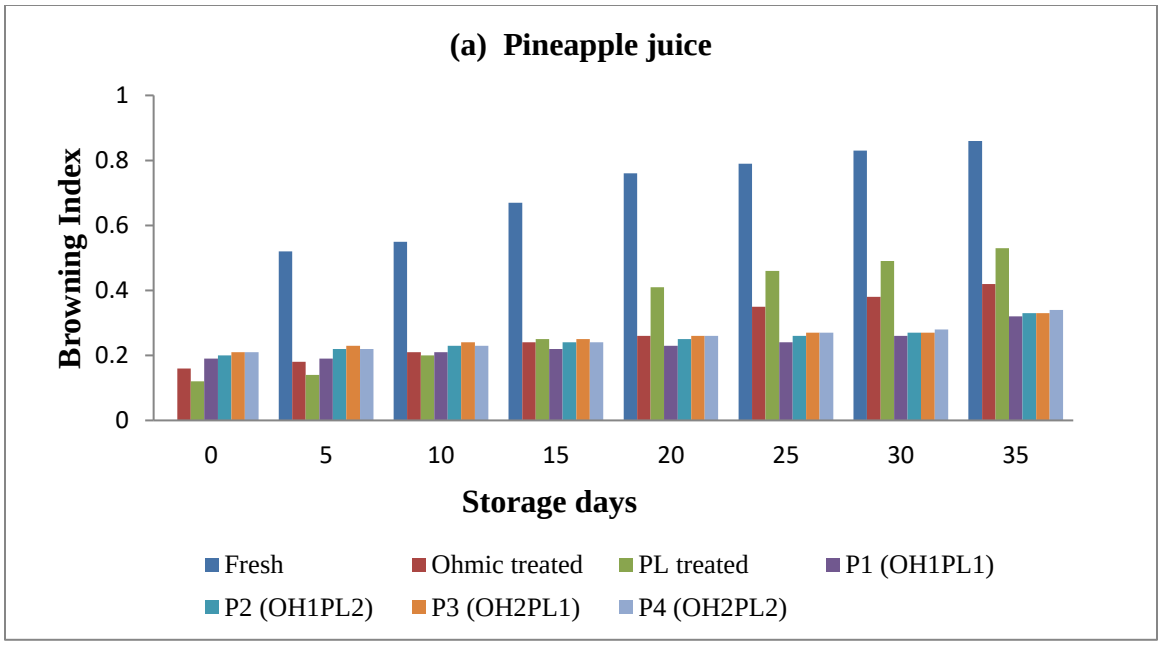
The deterioration of colour in fruit juices might be due to the non enzymatic Millard browning reaction in thermal processed samples (Moyer and Aitken, 1980). But during

storage, the colour change could be caused by the heat, air, and light conditions of storage. These key factors may cause oxidation of carotenoids and alterations in peroxide rings based on the storage period (Esteve and Frigola, 2007).

The variation in browning index of the fruit juices under different treatment conditions during storage is presented in Fig. 4.66 (a) and (b). A significant variation in the browning index ($p>0.001$) was observed in control and treated juice samples during storage period.

All samples showed an increase in the browning index with the increase in storage days. The control sample showed a higher increase from the initial zero value to 0.52 and 0.5 at 5^{th} day of storage of pineapple and cashew apple juice samples respectively. In the case of PL treated fruit juices, at 10^{th} and 15^{th} days of storage, objectionable variation in browning index was observed. The combined ohmic assisted PL treated pineapple and cashew apple juices had an initial browning index value of 0.19 and 0.2 respectively that showed a sudden increase after the 25^{th} and 30^{th} day of storage. The lowest variation in browning index was observed in ohmic assisted PL treated fruit juices, followed by ohmic heated further followed by PL treated samples.

A significant variation in the browning index ($p > 0.001$) was observed in control and treated juice samples during storage period. The increasing trend could be due to the oxidation of ascorbic acid as well as the breakdown of colouring pigments caused by PL and ohmic treatments. The variation in browning index observed in treated fruit juice sample might be due to non-enzymatic browning through several biochemical reactions such as Maillard condensation, pigment destruction, and caramelisation of sugar (Ibarz *et al.*, 2000).



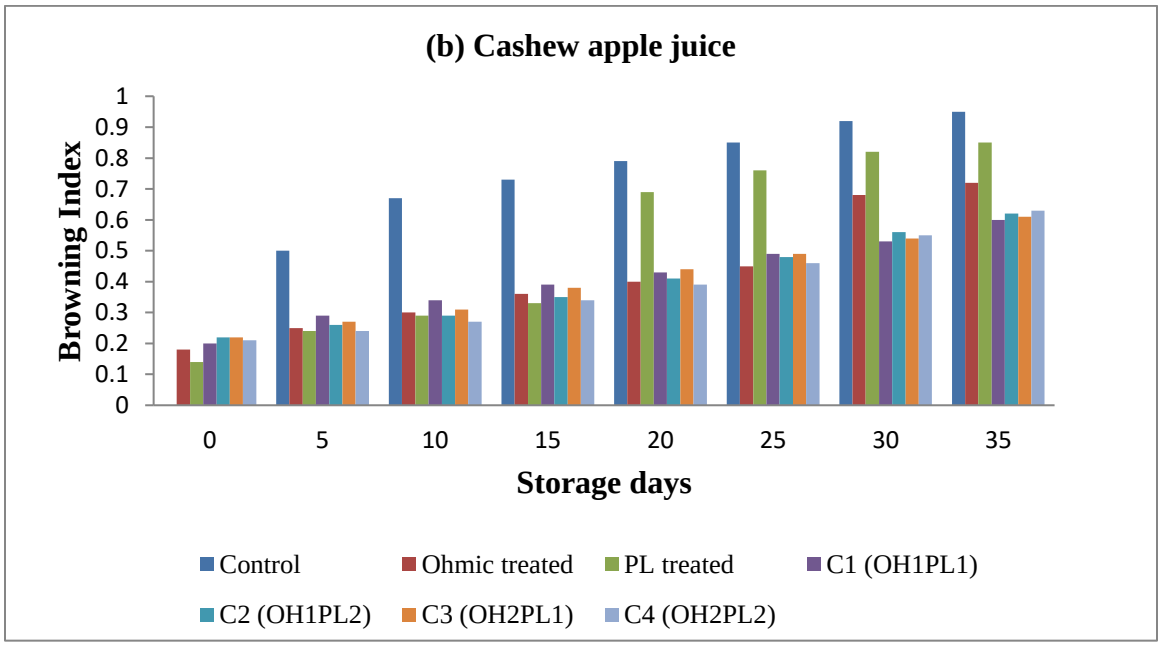


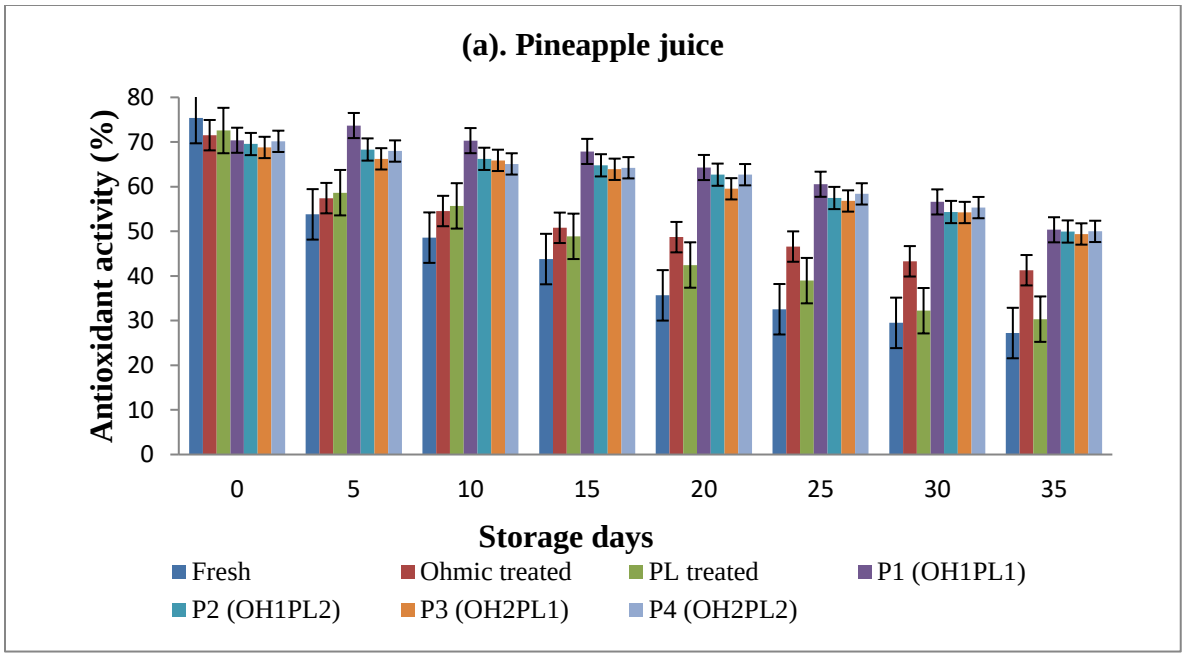
Fig. 4.66 Effect of different treatments on browning index of fruit juices during storage

The effect of different treatment conditions on the antioxidant activity of pineapple and cashew apple juice during storage is presented in the Fig. 4.67 (a) and (b). All samples of pineapple and cashew apple juice exhibited a significant decrease in antioxidant activity upon progress of storage period ($p < 0.01$).

It was found that the fresh pineapple and cashew apple juice had an initial antioxidant activity of 75.36% and 82.66% respectively. During the storage, the antioxidant activity reduced to 27.21% and 35.54% following an irregular pattern for pineapple and cashew apple juice respectively, at the end of 35 days of storage period.

The ohmic heated pineapple juice showed an increased reduction in antioxidant activity after 20 days of storage, whereas that of cashew apple juice a higher variation was recorded after 25 days of storage. The PL treated pineapple and cashew apple juice also observed a significant reduction in antioxidant activity after 10^{th} and 15^{th} day of storage, respectively.

All the combined treated fruit juices showed gradual reduction in antioxidant activity with increase in storage period. Among all the combined treated samples, the optimally treated samples showed minimum variation of 14% and 10.63% respectively for pineapple (P_1) and cashew apple (C_1) juice at 25^{th} and 30^{th} day of storage. In the case of optimally treated pineapple juice, the initial antioxidant activity was 70.39%, which varied to 60.54% at the end of 25 storage days under refrigerated condition. The initial antioxidant activity of optimally treated cashew apple juice was 75.85%, which increased to 67.78% at the end of 30 days under refrigerated storage. Though this variation is statistically significant ($p < 0.01$) this was found to be the minimum variation among all other treatments.



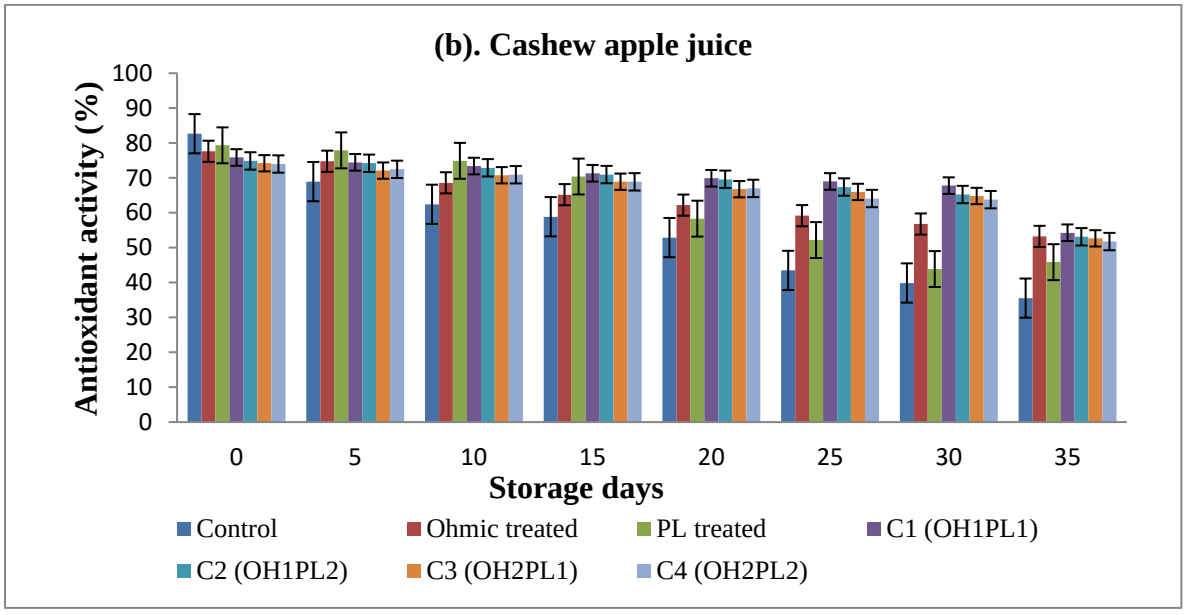


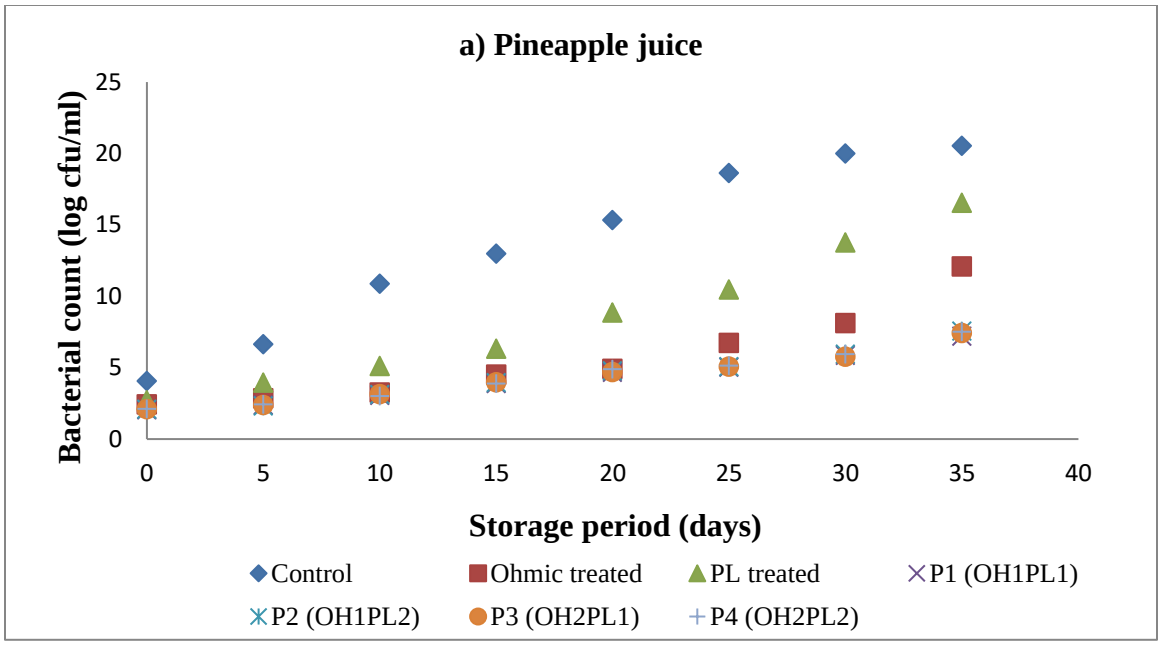
Fig. 4.67 Effect of different treatments on antioxidant activity of fruit juices during storage

4.5.1 Variation in Microbiological Properties during Storage of Fruit Juices

Microbiological studies of treated stored pineapple and cashew apple juices were carried out as mentioned in section 3.7.1. Gradual growth of microorganisms was observed during storage in all samples of pineapple and cashew apple juice. However, some treatments retarded the microbial growth higher than others. Generally, yeast and mould growth were found to be slow during the storage. The results are tabulated in Appendix A.33 and ANOVA tables are presented in Appendix A.34.

The variation in bacterial count of pineapple and cashew apple juice during storage is portrayed in Fig. 4.68 (a) and (b). From the figures it may be clearly inferred that the growth of microorganisms in fruit juices showed a significant ($p<0.05$) variation with respect to storage period. The combined treatment was found to be the most effective preservation delaying microbial growth in both the juices followed by ohmic treatment alone and further followed by PL treatment.

The fresh and treated juice samples exhibited an increase in bacterial count at different growth rates from initial values throughout the storage period. The fresh samples showed the highest growth rate of microorganism and found to reach values of 19.99 and 24.8 log cfu/ml at 30^{th} day of storage for both juices. This could be due to the higher initial microbial population which has not undergone any destruction process in its life path. A total bacterial count of only 5.85 and 5.89 log cfu/ml was seen in optimally processed ohmic assisted PL treated pineapple and cashew apple juice at 25^{th} and 30^{th} day of storage period respectively. The ohmic treated pineapple and cashew apple juice exhibited a growth of 4.91 and 5.48 log cfu/ml at 20^{th} and 25^{th} day of storage, whereas that of PL treated samples were 5.13 and 5.58 log cfu/ml after 10^{th} and 15^{th} day of storage respectively.



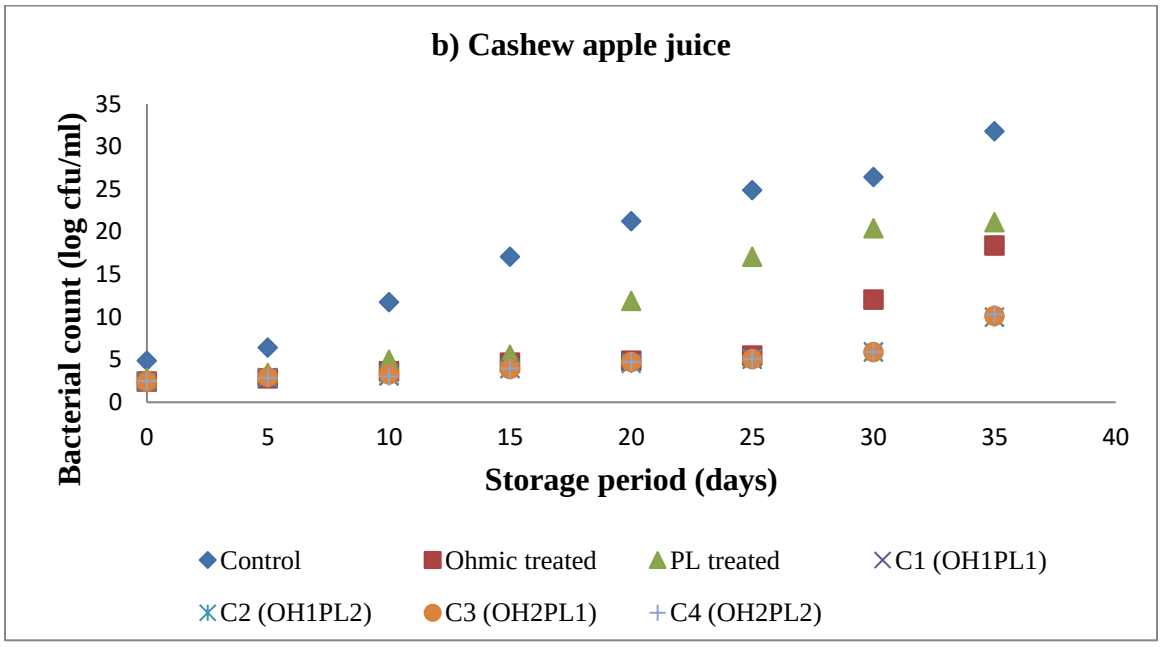
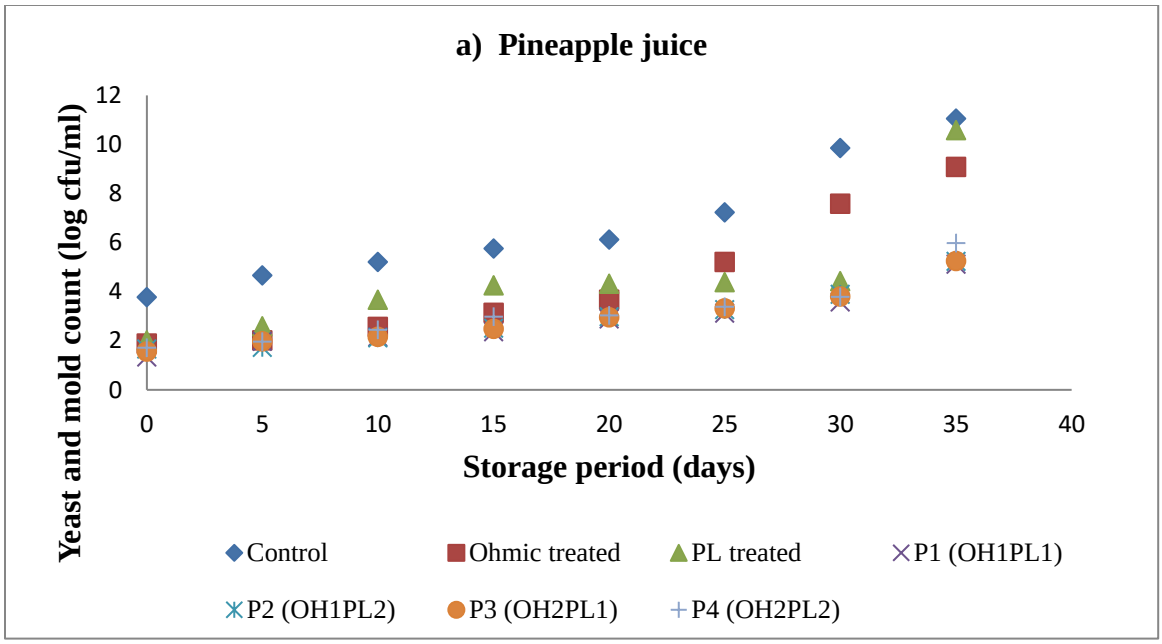


Fig. 4.68 Effect of different treatments on bacterial count of fruit juices during storage

As per FSSAI (2018) norms, the acceptable bacterial count in fruit juices should be limited to 6.0 log cfu/ml. Hence, the control samples were found not safe for consumption from the 5^{th} day of storage for both the juices. The PL treated pineapple juice retained an acceptable bacterial count limit up to 10^{th} day of storage, whereas cashew apple juice retained safe bacterial limit up to 15^{th} day of storage respectively. This might be due to the lower initial count of bacteria in the PL treated cashew apple juice compared to that of pineapple juice. The ohmic heated fruit juice samples resulted in a bacterial count below the acceptable limit up to 20^{th} day of storage. The optimally treated ohmic assisted PL processed fruit juices were found to be safe for consumption up to 25^{th} and 30^{th} days of storage in pineapple and cashew apple juice respectively. The rate of microbial growth in untreated liquid foods was observed to be higher than that of all treated samples. In ohmic assisted PL treated pineapple juice, total bacterial count was well within the limit of 6 log cfu/ml up to 25 days of storage whereas cashew apple juice remained in acceptable bacterial limit up to 30 days of storage.

Similar to bacterial count the yeast and mould population also increased during storage period at different growth rates for fresh and treated samples. The effect different treatment conditions on yeast and mould count in pineapple and cashew apple juice during storage are presented in the Fig. 4.69 (a) and (b).

The control and treated juice samples exhibited an increase in yeast and mould count at different growth rates from initial values throughout the storage period. The control samples showed the highest growth rates of microorganism and found to reach values of 11.06 and 14.46 log cfu/ml at 5^{th} day of storage for both juices. This might be due to the higher initial microbial population which has not undergone any destruction process in its life path. The minimum growth of 5.12 and 5.27 log cfu/ml was observed in optimally processed ohmic assisted PL treated pineapple and cashew apple juice at 25^{th} and 30^{th} day of storage respectively.



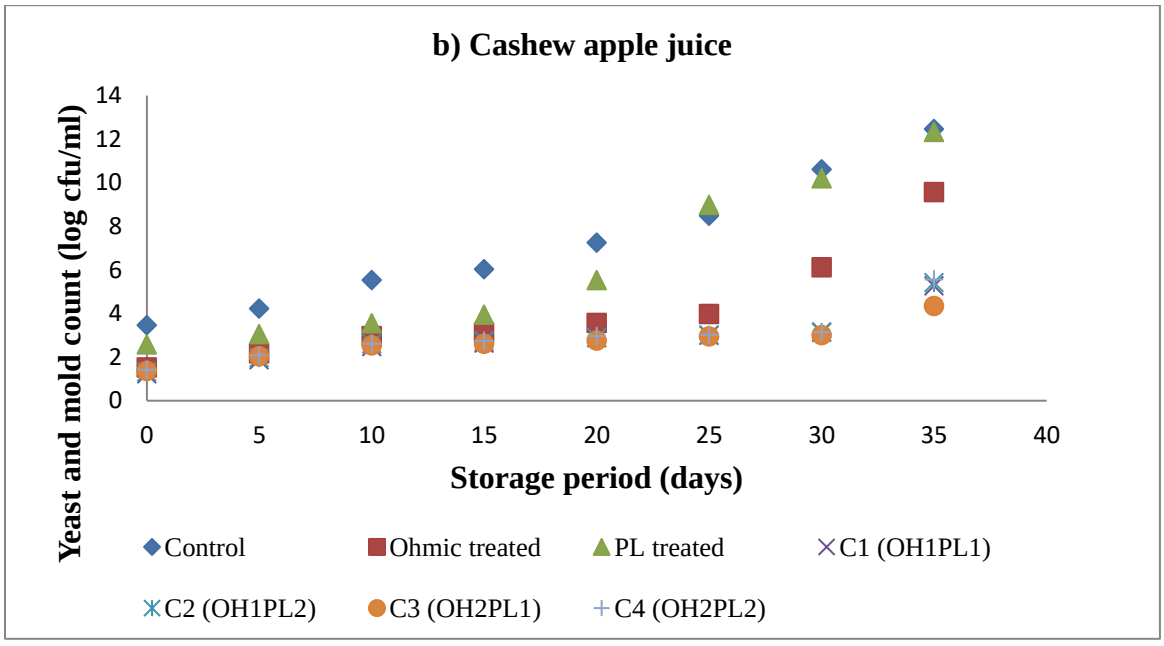


Fig. 4.69 Effect of different treatments on yeast and mould count of fruit juices during storage

The ohmic treated pineapple and cashew apple juice exhibited a growth of 3.67 and 3.98 log cfu/ml, at 20^{th} and 25^{th} days of storage, whereas that of PL treated samples were 3.67 and 3.96 log cfu/ml after 10^{th} and 15^{th} day of storage respectively.

The ohmic assisted PL treatment showed a gradual increase in yeast and mould count throughout the storage days, but does not exceed the permissible limit. The shelf life of the fruit juices were evaluated microbiologically based on FSSAI Microbiological Standards for fruits and vegetables and their products. Accordingly, the acceptable limit of yeast and mould count in fruit juices is $\leq$ 4.00 log cfu/ml. The PL treated pineapple juice samples attained safe limits up to the 10^{th} day of storage and cashew apple juice up to the 15^{th} day of storage. Ohmic heated and ohmic assisted PL treated fruit juice samples were found to be safe for consumtion up to 25^{th} and 30^{th} days of storage.

The microbial count of fresh fruit juices showed a significant increase after 5^{th} day of storage. The microorganisms already present in fruit juices showed a sudden increase due to the proliferation and growth, consuming the nutrients present in the juice samples. It is reported that most of the spoilage in refrigerated citrus juices is due to the presence of fermentative yeast, *Saccharomyces cerevisiae* (Alwazeer *et al*., 2002). The emergence of moulds affects flavour and results in the production of filamentous structures and enzymes such as amylases, proteases, and pectinases which ultimately causes juice spoilage (Swanson, 1989).

It was found that microbial population increased with increase in number of days of storage of the treated fruit juices under refrigerated conditions. This could be due to the germination of the survivor spore forming microbes present within the juice during the storage period. Once the microbes had started to germinate, it would multiply quickly making use of the nutrients available in the juices. Somboonsilp *et al.* (2011) also observed an increase in microbial load of ohmic heated orange, pineapple and guava juices during the storage under ambient as well as refrigerated storage conditions. A similar trend was also observed in the microbial count of thermally processed and UV treated pineapple juice samples (Chia *et al.*, 2012).

4.6. SENSORY EVALUATION

The pineapple and cashew apple juice samples treated under different process conditions and fresh samples stored at 4°C (control) were subjected to sensory evaluation. The sensory evaluation was carried out based on the nine-point Hedonic scale (Appendix.A.35) and fuzzy logic comprehensive model. Before each sensory evaluation of the stored samples, it was ensured that the samples were safe from microbial contamination through microbiological analysis. The scale factors *viz.* excellent (EX), good (GD), medium (MD), fair (FR) and not satisfactory (NS) were assigned to the quality factors *viz*.**,** colour and appearance, taste, flavour, and overall acceptability for all the samples as given in chapter III section 3.12.1 Fuzzy membership function (FMF) and normalised fuzzy membership function (NFMF) were then calculated and presented in the Appendix A. 36 and 37. The judgment membership functions were then compared with the weightage value of quality attributes and the minimum values were selected to assign the quality ranking.

The fresh pineapple juice (control), the PL treated pineapple juices at 10^{th} day of storage, ohmic heated pineapple juice at 20^{th} day of storage and ohmic assisted PL treated juice at 25^{th} day of storage were kept for sensory analysis based on the results

of the microbial characteristic studies as described in section. 4.5.1. Similarly, in cashew apple juice the fresh juice (control), PL treated juices at 15^{th} day of storage, ohmic heated juice samples at 25^{th} day of storage and ohmic assisted PL treated juices at 30^{th} day of storage were selected for analysis. Good sensory scores were obtained in the 9 - point Hedonic scale for the fresh followed by ohmic assisted PL treated pineapple (P_1) and cashew apple (C_1) juices at 25^{th} and 30^{th} days of storage. The sensory panel preferences for the ohmic heated alone and only PL treated juice samples were less than that of combined treated samples.

The fuzzy logic ranking was carried out based on the treatment conditions and presented in Table 4.10. The ranking order of pineapple juice was C= P1>P2=P3=P4>OH=PL. Similar ranking was also found in cashew apple juice i.e. C= C1> C4> C2=C3 >OH=PL. The fuzzy logic ranking for all the treatments is presented in

the Appendix-A.38. Based on the quality ranking, control samples and P1 samples scored highest (0.25) in pineapple juice, whereas, for cashew apple juices, control samples score highest (0.26) value followed by C1 (0.25). It is observed from the table that the sample C (control) gets the first rank based on the score obtained for flavour, colour and appearance for both fruit juices. Sample P1 and C1 scored 0.26 and 0.25 based on its colour and appearance and flavour.

Referring to Table 4.10 and 4.11, it may be derived that ohmic assisted PL treated pineapple juice samples at the optimised process condition scored 0.25, 0.25, 0.24, and 0.23 for colour and appearance, flavour, taste and overall acceptability respectively, whereas the corresponding weightage of these quality attributes were 0.27, 0.26, 0.24, and 0.23. It may be observed that, the sensory panels scored scores were on par with that of control for the optimally treated pineapple juice (P1) for all the quality attributes analysed. It may also be visualised that, only in colour and appearance, and flavour the sensory scores were slightly less than the weightages attributed to that quality. Similarly the ohmic assisted PL treated cashew apple juice samples scored 0.25, 0.24, 0.24, and 0.23 for colour and appearance, flavour, taste, and overall acceptability respectively. Therefore, it could very well be established that the ohmic assisted PL treated samples exhibited sensory characteristic on par with that of control and better than PL treated and ohmic heated samples.

The individual ohmic heated and PL treated juice samples resulted in higher variation in biochemical properties during storage and showed lower storage qualities compared to ohmic assisted PL treated samples. Therefore, it could be concluded from the results that combined treated pineapple and cashew apple juices at optimised process conditions would be acceptable by the consumer since these samples showed greater closeness to fresh pineapple and cashew apple juice in terms of sensory qualities.

Table 4.10 Quality ranking of pineapple juice treated at different conditions

Sensory attributes	Weightage	C	OH	PL	P1	P2	P3	P4
Colour and appearance	0.27	0.25	0.20	0.20	0.25	0.24	0.24	0.24
Flavour	0.26	0.25	0.20	0.21	0.25	0.24	0.24	0.24
Taste	0.24	0.24	0.21	0.20	0.24	0.24	0.24	0.24
Overall acceptability	0.23	0.23	0.20	0.21	0.23	0.22	0.23	0.23
		0.25	0.21	0.21	0.25	0.24	0.24	0.24
Quality ranking		I	IV	IV	II	III	III	III

Table 4.11 Quality ranking of cashew apple juice treated at different conditions

Sensory attributes	Weightage	C	OH	PL	C1	C2	C3	C4
Colour and appearance	0.27	0.26	0.20	0.19	0.25	0.23	0.23	0.24
Flavour	0.26	0.25	0.20	0.20	0.24	0.23	0.23	0.24
Taste	0.24	0.24	0.18	0.17	0.24	0.22	0.22	0.23
Overall acceptability	0.23	0.23	0.18	0.17	0.23	0.23	0.23	0.23
		0.26	0.20	0.20	0.25	0.23	0.23	0.24
Quality ranking		I	V	V	II	IV	IV	III

The fresh fruit juices are generally considered as a highly perishable commodity and also very difficult to store and market, preserving its authentic qualities. The thermal processing technologies and chemical preservation methods effectively extend the shelf life of the product for several months compromising the nutritional qualities.

In this study, during the process optimisation stage, it was established that the ohmic assisted PL treatment of pineapple and cashew apple juice could retain the nutritional qualities and effectively bring down the microbial population to safe levels for human consumption. Further, it was researched to find the optimum shelf life of the optimally ohmic assisted PL treated pineapple and cashew apple juice and compared the same with that of fresh and separately ohmic and PL treated juice. The variations in phisico-chemical properties during storage at refrigerated condition were analysed. From these analysis, it could be derived that, in general though there is an overall reduction in the magnitude of physico-chemical quality parameters listed, the intensity of such reductions were much severe in fresh samples and less severe in separately treated and least severe in optimally treated samples throughout the storage period under test as detailed earlier. The microbial analysis of the control and treated fruit juices at regular intervals of storage at refrigerated condition revealed that the optimally treated pineapple juice retained the microbial population under safe limit up to 25 days, whereas that of cashew apple juice up to 30 days. When applied separately, the PL treated juices showed very slight variation in quality attributes at first day of storage, but showed a gradual reduction in qualities up to 10^{th} day of storage in pineapple juice and 15^{th} day of storage in cashew apple juice and thereafter the microbial count exceeded the permissible limit.

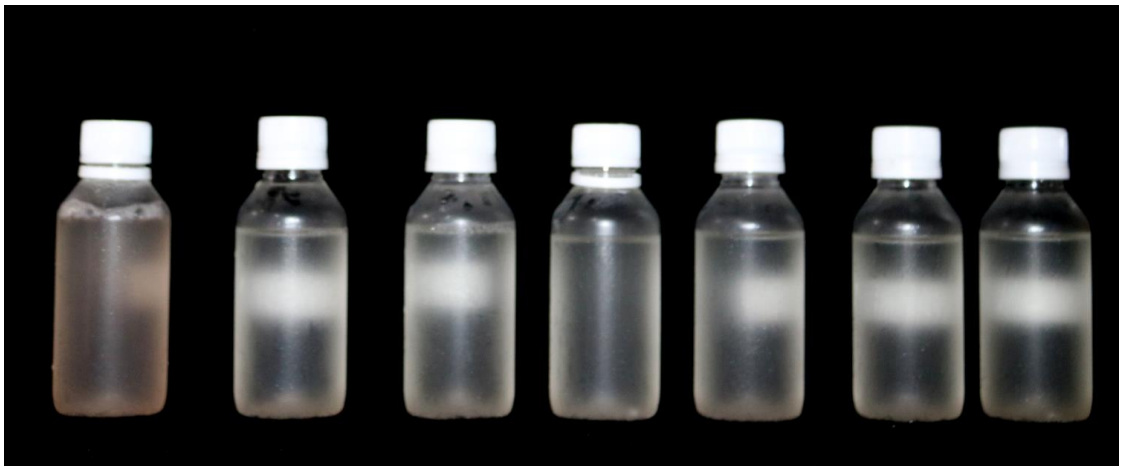
The sensory analysis proved that the panel scores on colour and appearance, flavour, taste and overall acceptability of the ohmic assisted PL treated fruit juices at optimised processing conditions were on par with that of control until 25 days in pineapple juice, and until 30 days in cashew apple juice under refrigerated condition. The pineapple and cashew apple juice treated at different treatment conditions stored at 25^{th} and 30^{th} day of refrigerated storage is shown in Plate 4.1 and 4.2 respectively.

C OH PL P1 P2 P3 P4

C- Control, OH- Ohmic heated, PL- Pulsed light treated

Combined ohmic and PL treated – P1 (OH_1PL_1**), P2 (**OH_1PL_2**), P3 (**OH_2PL_1**), P4 (**OH_2PL_2**)**

Plate 4.1 Pineapple juice treated at different treatment conditions stored at 25th days of storage



C OH PL C1 C2 C3 C4

C- Control, OH- Ohmic heated, PL- Pulsed light treated

Combined ohmic and PL treated – C1 (OH_1PL_1**), C2 (**OH_1PL_2**), C3 (**OH_2PL_1**), C4 (**OH_2PL_2**)**

Plate 4.2 Cashew apple juice treated at different treatment conditions stored at 30th days of storage

4.6 MICROBIAL INACTIVATION CHARACTERESTICS

The microbial inactivation studies of *Escherichia coli* MTCC 433 and *Listeria monocytogenes* MTCC 839 were carried out during ohmic assisted PL treatment of pineapple and cashew apple juices as discussed in section 3.13 of chapter 3 and the results are presented in Table.4.12.

Table 4.12 Log reductions of *Escherichia coli* and *Listeria monocytogenes* strains in optimally treated fruit juices

Bacterial strain	Microbial reduction (log cfu/ml)	
	Pineapple juice	Cashew apple juice
Escherichia coli MTCC 433	5.23 ± 0.73	5.46 ± 0.14
Listeria monocytogenes MTCC 839	5.21 ± 0.19	5.38 ± 0.25

From the table it could be clearly inferred that ohmic assisted PL treated pineapple juice showed a reduction of 5.23±0.73 and 5.21±0.19 log cfu/ml in *Escherichia coli* and *Listeria monocytogenes* strains respectively whereas, the highest reduction of 5.46±0.14 log cfu/ml in *Escherichia coli* and 5.38±0.25 log cfu/ml in *Listeria monocytogenes* were obtained in ohmic assisted PL treated cashew apple juice. The plate 4.3 and 4.4 portraits the enumerated plates after ohmic assisted PL treatment of pineapple and cashew apple juice inoculated with 10^8 cfu/ml *Escherichia coli* and *Listeria monocytogenes* strains. The plates did not show any colonies growing in the medium at 10^2 dilutions. The initial microbial populations of both the strains were 10^8 cfu/ml in both samples before the treatment and which was reduced to less than10^2 cfu/ml in all combined treated samples. The optimally processed PL treated samples of pineapple and cashew apple showed more than 5 log reductions in *Escherichia coli* and *Listeria monocytogenes* species. FDA HACCP regulation stipulates that a minimum 5 log reduction from the initial population in most resistant organism mandatory in treated fruit juices (FDA, 1998). All combined treated juice samples have met the minimum requirement as specified by the FDA.

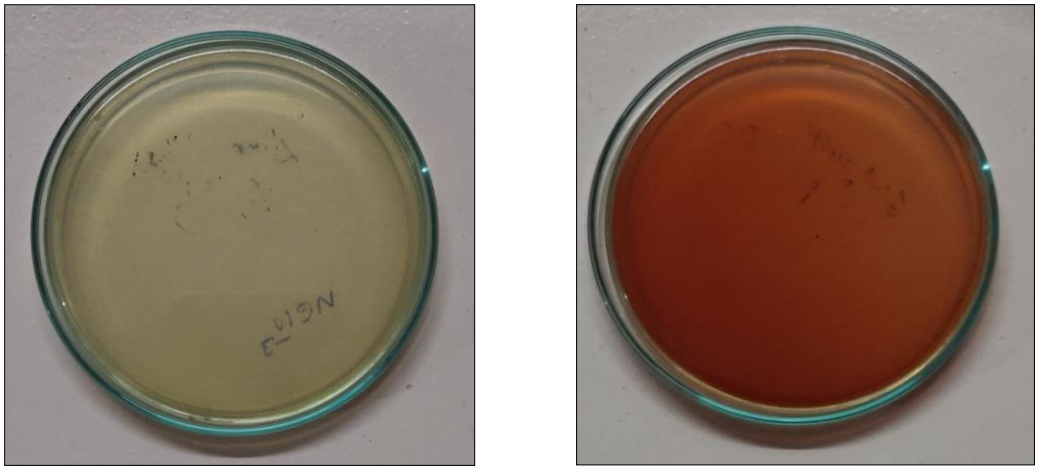


Plate 4.3 Enumeration of *Listeria monocytogenes* and *Escherichia coli* strains after optimal ohmic assisted PL treatment of pineapple juice

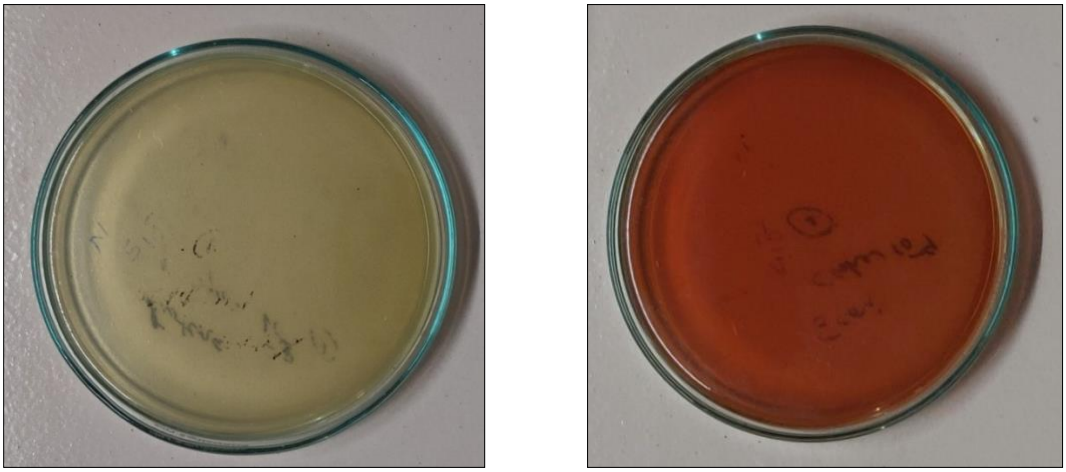


Plate.4.4 Enumeration of *Listeria monocytogenes* (a) and *Escherichia coli* (b) strains after optimal ohmic assisted PL treatment of cashew apple juice

The inoculation of the standard pathogenic microorganism could be attributed to the combined effect of ohmic and pulsed light process. It could be derived that the optimal voltage gradient, temperature and treatment time established at ohmic heating and the optimal PL dosage, sample-source distance and flow rate arrived during pulsed light treatment could have combindly produced mild thermal and the electroporation

effects (ohmic heating) and photochemical photo-themal and photo physical effects (PL treatment) causing reduction of microbial population to safe levels.

Mazzotta *et al.* (2001) reported that *Escherichia coli* O157:H7 showed the highest heat resistance than *Listeria* and *salmonella* species in the ohmic heating process. Whereas, *Listeria* species was found to be most resistant microorganism for pulsed light treatment. Thus the combination of ohmic and PL technologies ensure a better inactivation of *Listeria monocytogens* and *Escherichia coli* species at lower dosage levels of both technologies. Artíguez *et al.* (2015) found that combination of thermal treatments and PL process could assist to lessen the intensity of the different operating parameters such as processing time, temperature, and PL dosage than those required for inactivation when applied alone.

Artiguez *et al.* (2015) reported that heating followed by PL treatment showed efficient inactivation of *B. subtilis* spores than PL followed by thermal treatments. This could be due to the greater sub lethal damages induced by thermal treatment compared to PL treatment, results in increased sensitivity to the subsequent treatment. In our study also mild thermal ohmic heating followed by PL treatment could have resulted in sub-lethel injury to microorganisms.

The ohmic heating of buffer peptone water at 60°C for 30 s resulted in 3.59 and 3.40 log reduction in in *Escherichia coli O157: H7,* and *Listeria monocytogenes*, respectively (Park and Kang, 2013). Pulsed light irradiation at a dosage of 3 J/cm^2 was efficient to reduce *Listeria monocytogenes*, and *Escherichia coli 0157:H7* for 2.24 and 2.29 log cfu/g respectively on the surface of dry fermented salami (Rajkovic *et al.*, 2017). In a continues flow PL apparatus, the treatment at a fluence rate of 4 J/cm^2 resulted in a 4.00 and 2.90 Log-cycles for *Escherichia coli* and 2.98 and 0.93 Log-cycles for *Listeria innocua* microbial reductions in apple and orange juices respectively (Pataro *et al.*, 2011). From the research findings mentioned above, it could be clearly inferred that when applied separately, the ohmic and PL treatments could not achieve the recommended 5 log reduction in bacterial population whereas, when combined, they could achieve safe microbial limits at comparatively lower dosage levels.

Among the juice samples cashew juice samples showed a higher reduction in both *Escherichia coli* and *Listeria monocytogenesis* species. This might be due to the higher electrical conductivity of cashew apple juice, a property that helps in increasing the heating rate during ohmic heating of juice samples leading to higher reduction rates of micro-organisms. Lee *et al.* (2012) reported that tomato juice had a higher reduction in *Escherichia coli* and *Listeria monocytogenes* and higher heating rates than orange juice at all temperatures due to higher electrical conductivity. The efficiency of PL treatment increases with increase in transparency of product treated. Liquids such as drinking water have a higher transparency as compared to fruit juice with limited transparency due to absorption properties. The pineapple juices have a limited transparency, whereas the clarified cashew apple juice is clear and free of suspended particles and more transparent to light pulses (Pataro *et al.,* 2011). This property of cashew apple juice provides a better reduction in *Escherichia* and *Listeria.* Similar results were also reported in PL treatment for inactivation of *Escherichia coli* in liquids with varying transparency such as triptych soy broth (TSB), butter field's phosphate buffer (BPB), apple cider and apple juice. The PL susceptibility showed a decreasing trend from BPB to TSB and to apple juice and apple cider (Sauer and Moraru, 2009). Choi *et al*. (2010) reported that significantly higher effective inactivation of *Listeria monocytogenes* in a light coloured thin infant beverage than acoloured viscous infant meal. Pataro *et al.* (2011) also observed that the PL resistance of both *Escherichia coli* and *Listeria innocua* was greater in orange juice than in apple juice resulting in higher inactivation in apple juice.

.

Summary & Conclusion

CHAPTER V

SUMMARY AND CONCLUSION

Fruit juices are enjoyed by people due to their good taste and flavour apart from its usual thirst quenching ability and are consumed in fresh and processed form. Also, they supply vital components to enervate the physiological condition of human beings both physically and mentally. Seasonality of the fruit production restricts availability of fresh fruit juices throughout the year. So there has been a long run for development of suitable technologies in food business for making minimally processed fruit juices available to the consumer shelf throughout the year. Recently, juice industries have been facing various challenges due to the pathogenic bacteria developing resistance to stress. The microorganisms adapt themselves to hostile environments which were earlier detrimental to them (Wood and Moellering, 2003). Thus fruit juices need to be processed to inactivate pathogenic microorganisms to improve the shelf life adopting novel technologies ensuring safety as well as quality. Pineapple is one of the important fruit crop and cashew nut is an important plantation crop cultivated all over India. Due to lack of appropriate processing and storage facilities, high post harvest loss and distress sales are being reported in pineapple. The cashew trees are cultivated mainly for procuring nuts whereas the cashew apples are being wasted in the field after harvest of nuts. The processing and value addition of cashew apples employing suitable technologies are inevitable for making use of this highly nutrient rich fruit.

Ohmic heating is the process of heating food materials sandwiched between two electrodes. The food materials offer resistance to the flow of current and hence heat the food volumetrically. The pulsed light (PL) is a non thermal processing technology in which the food materials are exposed to high intense light pulses from the lamp to inactivate the microorganisms. Though Pulsed light system efficiently inactivate microorganism, the penetration depth is limited due to shadow effect resulting in uneven microbial destruction. Therefore, pulsed light treatment could be effectively combined with other such technologies resulting in a hybrid effect in which the weakness of one technology could be the strength of the other.

The present investigation was carried out to develop an ohmic heating assisted flow through pulsed light (PL) system and to evaluate the system for preservation of pineapple and cashew apple juice. The process operating parameters of the individual and combined process would be analysed for their effectiveness in delivering pineapple and cashew apple juice of better nutritional and sensory qualities ensuring microbial safety after processing and during storage.

The processing of juice occur in a two-step process: first, during ohmic heating, the mild thermal and electroporation generate cell pores or destroy cell membranes, and second, PL could penetrate deeply through the cell pores and disrupt the microbial DNA, preventing replication of microorganisms while retaining overall qualities of the juice.

The developed system consists of two sections *i.e.* ohmic heating part and pulsed light portion. The experimental ohmic heating system consists of a feed tank, ohmic heating chamber, varying power source, volt and ampere meter and temperature measuring system. The system can process 300ml of juice per batch. The ohmic heating chamber consists of a Teflon cylinder with inner and outer diameter of 70 mm and 80 mm respectively and having a length 150 mm. Both sides of the cylinder were fixed with stainless steel (SS 304) electrodes of 2 mm thickness with same inner diameter of chamber and covered with Teflon end caps. The effectiveness of the system was studied varying different operating parameters such as voltage gradient, process temperature, and holding time. A temperature detection probe was installed in the middle of the chamber to measure the system temperature during operation. A variable transformer was used to supply power to the ohmic heating chamber and also to vary the voltage gradient. A digital type Voltmeter and ammeter were used to monitor potential difference and changes in current during the process respectively

Pulsed light system consists of a xenon flash lamp, contactor, relay, resistors, rectifier, capacitors, micro processors, trigger transformer, thyristor and timer relay etc. An electronic circuit with these components was assembled to produce pulsed light. The xenon flash lamp comprises of a sealed tube with a thickness of 3 mm, bore diameter of 8

mm and arc length of 70 mm made of clear fused quartz (CFQ) filled with xenon gas. This was used to produce the broad spectrum flash of wavelength from 100 to 1100 nm.

The PL system can be controlled by varying three operating parameters *viz.* PL dosage, sample-source distance, and flow rate using a microcontroller. The microcontroller stores the functions as program in its memory and the output signals are displayed in the LCD display board. The three knobs provided in display panel is employed for varying the distance from the xenon lamp, flow rate and pulsed dosage. The stepper motor driver is used for controlling bipolar stepper motor employed for moving the Xenon lamp assembly by 5 cm, 10 cm, and 15 cm so as to change the distance between the lamp and the quartz tube. The pulsed light treatment chamber consists of the pulsed light lamp, mechanical assembly of backward and forward movement of PL lamp, quartz tube of 15 mm diameter and 70 mm length, circulation motor and associated electrical and electronic connecting terminals and circuits. The fruit juices are pumped using a booster pump though the quartz tube. A flow sensor is attached to the pipe routing to control the flow rate though the quartz tube.

The whole research work is divided in to four major sections *viz.* development of an ohmic heating assisted pulsed light treatment system, evaluation of the system for individual ohmic and pulsed light treatment as well as ohmic assisted pulsed light treatment, characterisation, storage and sensory studies of juices treated at optimised conditions and inoculation studies of *Escherichia coli* and *Listeria monocytogens* species in the pineapple and cashew apple juice.

Experiments were framed using Box-Bekhen Response Surface Methodology in the Design expert software version 7.0 to optimise the voltage gradient (10, 12.5 and 15 V/cm), process temperature (50, 55 and 60°C), and holding time (1, 3 and 5 min) combinations of ohmic heating process with respect to the microbial and biochemical quality of the fruit juices under study. Similarly, process parameters of pulsed light treatment such as pulsed light dose (10, 12.5 and 15 J/cm^2), sample- source distance (5, 10 and 15 cm) and flow rate (150, 200 and 300 ml/min) were optimised for the effective pasteurisation with respect to the physico-chemical and microbial quality of the fruit

juices. The physico-chemical parameters such as pH, TSS, titrable acidity, ascorbic acid, total sugars, total phenols, tannin content, total colour difference and microbiological properties *viz*. bacterial log reduction and yeast and mould reduction were fixed as the dependent parameters for the optimisation process of ohmic heating as well as PL process for both juices.

Fresh fruit juices were fed through the developed system wherein it first flow through the ohmic heating chamber and further to the pulsed light treatment chamber. While the juice reached ohmic heating system, it is exposed to electric fields and the temperature is monitored with thermocouples. Each fruit juice is subjected to ohmic heating at different voltage gradient and residence time as per the experimental design. Further the juice was subjected to pulsed light treatment at different flow rates, pulsed light dosages, and distances from the light source. The quality characteristics of the treated juices were then studied in detail. Based on the result the operating parameters were standardised.

The optimised process parameters of ohmic heating and PL treatment were combined in ohmic assisted PL treatment to obtain better quality product. For better and rigorous comparison, values of highest desirability next to that of optimised condition were also selected for conducting combined treatment studies. The combined treated pineapple and cashew apple juices were also analysed for the quality parameters such as pH, TSS, titrable acidity, ascorbic acid, total sugars, total phenols, tannin content and antioxidant activities, total colour difference, and rheological properties. The results were then compared with that of fresh, optimised ohmic and optimised PL treated samples.

The storage studies of both fruit juices treated at optimised process conditions of PL, ohmic and ohmic assisted PL treatment and other relevant combined treated samples as per the experimental design were conducted for 35 days of storage under refrigerated condition (4±2°C). The bacterial inactivation studies were conducted to evaluate the effect of combined treatments on microbial death kinetics using surrogate microbial strains of *Escherichia coli MTCC 433* and *Listeria monocytogenes 839*.

During the ohmic heating studies of both fruit juices, the process parameters showed a significant effect on the pH, TSS, ascorbic acid, total colour difference, and microbial reduction. The microbial reduction of fruit juices increased with increase in voltage gradient process temperature and holding time. The pH and ascorbic acid content were found to reduce during ohmic heating whereas total colour difference and TSS were found to increase.

The highest bacterial log reduction of 4.03 and highest yeast and mould reduction of 3.04 were observed in the treatment at voltage gradients of 15 V/cm, process temperature of 60°C with 3 min of holding time in pineapple juice. In cashew apple juice the highest bacterial log reduction of 4.26 and yeast and mould reduction of 3.17 was observed in ohmic heating treatment at voltage gradient of 12.5 V/cm and process temperature of 60°C with 5 min of holding time. The statistical analysis of the data revealed that in ohmic treatment studies a voltage gradient of 14.02 V/cm, holding time of 2.31 min and treatment temperature of 55.26°C were the optimum operating process conditions with respect to the quality of pineapple juice whereas, the optimum condition for cashew apple juice were a voltage gradient of 14.53 V/cm, process temperature of 55.25°C with holding time of 2.77 min.

PL process parameters showed a significant effect on ascorbic acid content, total colour difference, and microbial reduction of pineapple and cashew apple juice samples. The microbial reduction, total colour difference and reduction in ascorbic acid content increased with increase in PL dosage and decreased with increase in sample source distance and flow rate. The highest bacterial log reduction of 3.39 and highest yeast and mould reduction of 2.98 were observed in the samples treated at a PL dosage of 20 J/cm^2, flow rate of 150 ml/min with 5 cm of sample-source distance. In cashew apple juice, the highest bacterial log reduction of 3.56 and yeast and mould reduction of 2.94 was observed at a PL treatment dosage of 32 J/cm^2, flow rate of 225 ml/min, with a sample-source distance of 5 cm. The optimum process conditions for pineapple juice were found at a PL treatment with a PL dosage of 13.69 J/cm^2, sample-source distance of 10.26 cm

and flow rate of 165.06 ml/min whereas, that for cashew apple juice a PL dosage of 12.49 J/cm^2, sample-source distance of 8.63 cm with a flow rate of 164.01 ml/min.

Results of the experiments conducted with ohmic assisted PL treated samples at optimised process parameters arrived at their respective individual treatment s revealed a better inactivation of bacteria and yeast and mould population along with considerable retention of quality attributes. A bacterial reduction of 5.13 and 5.19 log cfu/ml and yeast and mould reduction of 4.86 and 4.95 log cfu/ml were observed in optimally treated pineapple and cashew apple juice respectively. The variation in ascorbic acid content and total colour difference were found to be higher in ohmic assisted PL treated samples when compared to individual ohmic and PL treatment.

The rheological properties of ohmic assisted PL treated pineapple and cashew apple juice did not show any variation from that of fresh sample. Both fluids showed a typical non Newtonian behavior and fell in the category of shear thinning behavior. The rheological models were fitted to experimental values of pineapple and cashew apple juice. The rheological behaviour of ohmic assisted PL treated pineapple juice showed a good fitting to Herschel Buckley model with an R^2 value of 0.89 and that of cashew apple juice showed a good fitting to power-law model with an R^2 value of 0.87.

The physicochemical characters of the juices were found to be varying throughout the storage period, wheares the magnitude of variation was less severe in ohmic assisted PL treatment compared to other treatments. Therefore, it was concluded that the quality attributes of fresh juice was observed to be suddenly varied from 5th day of storage for both fruit juices. The ohmic assisted PL treated samples were safe to consume up to 25th and 30th day of storage for pineapple and cashew apple juice respectively. The PL treated juice samples could maintain a safe microbial limit for consumption only up to 10th and 15th day of storage whereas, ohmic assisted samples could maintain the safe limits up to 20th and 25th days of storage for pineapple and cashew apple juice respectively.

The sensory evaluation was conducted for the treated fruit juices based on the 9-point Hedonic scale and fuzzy logic comprehensive model. The fuzzy logic ranking order of pineapple juice was found to be C= P1>P2=P3=P4>OH=PL. The cashew apple juice

also showed a similar ranking i.e. C= C1> C4> C2=C3 >OH=PL. Based on the quality ranking, the sensory panel scores of optimally treated pineapple juice (0.25) was on par with that of control (0.25) whereas that of cashew apple juice (0.25) was very close to that of control (0.26).

The inactivation studies conducted to study the influence of optimised combined treatment process parameters revealed a higher reduction of 5.46±0.14 log cfu/ml in *Escherichia coli* and 5.38±0.25 log cfu/ml reduction in *Listeria monocytogens* in ohmic assisted PL treated cashew apple juice.On the other hand, ohmic assisted PL treated pineapple juice showed a reduction of 5.23±0.73 and 5.21±0.19 log cfu/ml in *Escherichia* and *Listeria* strains respectively. It was concluded that ohmic assisted PL treated pineapple and cashew apple juice had met the minimum 5 log reduction in pathogenic microorganism as stipulated by FDA regulation.

Conclusions

- An ohmic assisted pulsed light treatment system for fruit juices was developed and fabricated and the effectiveness of this system towards preservation of pineapple and cashew apple juice were analysed in detail
- A voltage gradient of 14.02 V/cm process temperatures of 55.60°C and holding time of 2.31 min was found to be optimal conditions for ohmic heating treatment of pineapple juice. A PL dosage of 13.69 J/cm^2, sample-source distance of 10.26 cm and flow rate of 165.06 ml/min were determined to be the optimal treatment of PL treatment of pineapple juice
- A voltage gradient of 12.57 V/cm process temperatures of 58.81°C and holding time of 1.52 min was found to be optimal conditions for ohmic heating treatment of cashew apple juice. A PL dosage of 12.66 J/cm^2, Sample-source distance of 8.83 cm and flow rate of 163.45 ml/min were determined to be the optimal treatment of PL treatment of cashew apple juice
- The optimised values of pineapple and cashew apple juice under ohmic and PL treatments were selected as optimal parameters for combined treatments

- The ohmic assisted PL treatment were found to be capable of retaining the quality parameters of the fresh fruit juices and showed fresh like characteristics during sensory analysis
- The ohmic assisted PL treatment at optimised conditions could maintain a storage life of 25 days for pineapple juice and 30 days for cashew apple juice at refrigerated conditions (4±2°C) retaining nutritional and sensory characterestics and ensuring microbial safety
- A 5 log cfu/ml reductions in *Escherichia* and *Listeria* strains could be achieved in optimally treated pineapple and cashew apple juice

Scope for further studies

- The developed ohmic assisted PL system can be evaluated for other different fruit juices, tender coconut water, and other refreshing drinks
- Development of ohmic assisted PL treatment for semi solid or concentrated products could be taken up
- The combinations of pulsed light and/ or ohmic heating system with other such non thermal/ mild thermal technologies could be taken up for various pumpable and semi solid foods.

References

REFERENCES

Abedelmaksoud, T. G., Mohsen, S. M., Duedahl-olesen, L., and Feyissa, A. H. 2018.Effect of ohmic heating parameters on inactivation of enzymes and quality of not-from-concentrate mango juice.*Asian J. Sci. Res*. 11(3): 383-392.

Abhilasha, P. and Pal, U. S. 2018.Effect of ohmic heating on quality and storability of sugarcane juice.*Int. J. Curr. Microbiol. App. Sci.*7(1): 2856–2868.

Achinewhu, S.C. and Hart, A.D. 1994. Effect of processing and storage on the ascorbic acid (vitamin C) content of some pineapple varieties grown in the rivers state of Nigeria.*Plant FoodHum.Nutr.*46: 335-337.

Adou, M., Tetchi, F.A., Gbane, M., Niaba, P.V.K., and Amani, N.G. 2011.Mineral composition of the cashew apple juice (*AnacardiumOccidentale* L.) of Yamoussoukro, Cote d'Ivoire.*Pak J. Nutr.* 10(12): 1109-1114.

Adou, M., Kouassi, D.A., Tetchi, F.A., and Amani, N.G. 2012.Phenolic profile of cashew (*Anacardium Ocidentale* L.) of Yamoussoukro and Korhogo, *J. Appl. Biosci.*49: 3331-3338.

Adzahan, N. M., Ling, L.P., Hashim, N., Shamsudin, R., Chew S.C., and Sobhi, B. 2011. Pineapple juice production using ultraviolet pasteurisation.*J. Agribus. Mark.* 4: 38-50.

Agostini-Costa, T.S., Lima, M.V., Lima, A., Aguiar, M.J., de Lima, J.B., and Paiva, J. 2002.Taninoempedúnculos de caju: efeito de algumasvariaçõesgenéticas e climáticas. *B. CEPPA.Curitiba.*20(2): 267-278.

Aguilar, K., Ibarz, R., Garvín, A., and Ibarz, A. 2016.Effect of UV-C irradiation on enzymatic activities and the physicochemical properties of nectarine juices from different varieties.*LWT - Food Sci. Technol.* 65: 969-977.

Aguilo-Aguayo, I., Soliva-Fortuny, R., and Martin-Belloso, O. 2010. Color and viscosity of watermelon juice treated by high-intensity pulsed electric fields or heat. *Innov.Food Sci. Emerg. Technol.*11: 299–305.

Ahmed, J. 2004. Rheological behaviour and colour changes of ginger paste during storage. *Int. J. Food Sci. Technol.* 39: 325–330.

Akbarpour, V., Hemmati, K., and Sharifani, M. 2009. Physical and chemical properties of pomegranate (*Punica granatum* L.) fruit in maturation stage. *Am. Eurasian J. Agri. Environ. Sci.* 6(4): 411– 416.

Akinwale, T.O. and Aladesua, O.O. 1999.Comparative study of the physico chemical properties and the effect of different techniques on the quality of cashew juice from Brazilian and local varieties.*Niger. J. Tree Crop Res.* 3: 60-66.

Akinwale, T.O. 2000. Cashew apple juice: its use in fortifying the nutritional quality of some tropical fruits. *Euro. Food Res. Technol.* 211: 205-211.

Allen, O. N. 1953. *Experiments in soil bacteriology*. Burgess Co. Minneapolis.Minn, 69-70p.

Alwazeer, D., Cachon, R., and Divics, C. 2002. Behaviour of *Lactobacillus plantarum* and *Saccharomyces cerevisiae* in fresh and thermally processed orange juice.*J. Food Prot*. 65(10): 1586-1589.

Amiali, M., Ngadi, M., Raghavan, V.G.S., and Nguyen, D.H. 2006. Electrical conductivities of liquid egg product and fruit juices exposed to high pulsed electric fields. *Int. J. Food Prop*. 9: 533–540.

An, H. J. and King, J. M. 2007. Thermal characteristics of ohmically heated rice starch and rice flours.*J. Food Sci*. 72(1): C84-C88.

Anderson, A. K. and Finkelsten, R. 1919.A Study of the electro-pure process of treating milk.*J. Dairy Sci*. 2: 374-406.

Anderson, J.G., Rowan, N.J., MacGregor, S.J., Fouracre, R.A., and Farish, O. 2000.Inactivation of food-borne enteropathogenic bacteria and spoilage fungi using pulsed-light.*IEEE Trans. Plasma. Sci.* 28: 83-88.

AOAC [Association of Official Analytical Chemistry]. 2000. *Official Methods of Analysis* (17th Ed.). Association of Official Analytica Chemistry, Gaithersburg, Washington. DC, 320p.

Aramwit, P., Bang, N., and Srichana, T. 2010. The properties and stability of anthocyanins in mulberry fruits.*Food Res. Int*. 43:1093−109.

Artes, F. and Allende, A. 2005.Processing lines and alternative preservation techniquesto prolong the shelf-life of minimally processed leafy vegetables.*Eur. J.Hortic. Sci.*70: 231-245.

Artiguez, M.L., Lasagabaster, A., and Marañón, I.M. 2011. Factors affecting microbial inactivation by pulsed light in a continuous flow-through unit for liquid products treatment.*Proced. Food Sci.* 1: 786-791.

Artiguez, M. L. and Maranon, I. M. 2015. Inactivation of *Bacillus subtilis* spores by combined pulsed light and thermal treatments. *Int. J. Food Microbiol*. 214: 31- 37.

Assiry, A., Sastry, S. K., and Samaranayake, C. 2003.Degradation kinetics of ascorbic acid during ohmic heating with stainless steel electrodes. *J. Appl. Electrochem*. 33(2): 187-196.

Assiry, A.M., Sastry, S.K., and Samaranayake, C. 2006. Influence of temperature, electrical conductivity, power and pH on ascorbic acid degradation kinetics during heating using stainless steel electrodes. *Bioelectrochem.* 68: 7-13.

Assiry, A.M.,Gaily, M.H.,Alsamee, M., and Sarifudin, A.2010.Electrical conductivity of sea water during ohmic heating.*Desalination*. 260: 9-17.

Attri, B. L. 2009. Effect of initial sugar concentration on the physico-chemical characteristics and sensory qualities of cashew apple wine.*Nat. Prod. Rad.* 8: 374-379.Available:https://patentimages.storage.googleapis.com/pdfs/US4464336.pdf [04 March 2018].

Awua, A. K., Doe, E. D., and Agyare, R. 2011. *Exploring the influence of sterilisation and storage on some physicochemical properties of coconut* (*Cocos nucifera* L.) *water* [BMC research notes]. 4: 451p.

Azevedo, D.C.S. and Rodrigues, A. 2000.Obtainment ofhigh-fructose solutions from cashew (*Anacardiumoccidentale*) apple juice by simulated moving-bed chromatography.*Sep.Sci Technol*. 35: 2561-2581.

Azoubel, P.M., Cipriani, D.C., El-Aouar, A.A., Antonio, G.C., and Murr, F.E.X. 2005.Effect of concentration on the physical properties of cashew juice.*J. Food Engg.* 66: 413–417.

Bachmann, R. 1975. Sterilization by intense ultraviolet radiation.*Brown Boveri Rev*. 62: 206–209.

Bank, H. L., John, J., Schmehl, M.K., and Dratch, R.J. 1990.Bactericidal effectiveness of modulated UV light.*Appl. Environ. Microbiol.* 56: 3888-3889.

Barbosa-Canovas, G.V., Pothakamury, U.R., Palou, E., and Swanson, B.G. 1998. *Nonthermal preservation of foods*.Marcel Dekker, Inc, New York, 235p.

Barros, S.T.D., Andrade, C.M.G., Mendes, E.S., and Peres, L. 2003.Study of fouling mechanism in pineapple juice clarification by ultrafiltration.*J. Membr. Sci.* 215: 213–224.

Bartolome, A.P., Ruperez, P., and Fuster, C. 1995. Pineapple fruit: morphological characteristics, chemical composition and sensory analysis of red spanish and smooth cayenne cultivars. *Food Chem.* 53: 75-79.

Bates, R.P., Morris, J. R., and Crandall, P.G. 2001. Principles and practices of small and medium scale fruit juice processing. *FAO Agricultural Services Bulletin.* 146: 135-149.Available: http://ucce.ucdavis.edu/files/datastore/234-2085.pdf [12 June 2019].

Berney, M., Weilenmann, H. U., Simonetti, A., and Egli, T. 2006.Efficacy of solar disinfection of *Escherichia coli*, *Shigella flexneri*, *Salmonella typhimurium* and *Vibrio cholera*.*J. Appl. Microbiol.* 104: 828–836.

Beveridge, T., Franz, K.Y., and Harrison, J.E. 1986. Clarified natural apple juice: production and storage stability of juice and concentrate. *J. Food Sci.* 51(2): 411-414.

Bevilacqua, A., Campaniello, D., Sinigaglia, M., and Corbo, M.R. 2015.Combination of ultrasound and antimicrobial compounds towards *Pichia* spp. and *Wickerhamomycesanomalus* in pineapple juice.*LWT - Food Sci. Technol.* 64(2): 616-622.

Bharate, S. S. and Bharate, S. B. 2014.Non-enzymatic browning in citrus juice: chemical markers, their detection, and ways to improve product quality. *J Food Sci Technol*. 51(10): 2271-2288.

Bhat, R., Ameran, S., Han, C.V., Karim, A.A., and Liong, M.T. 2011. Quality attributes of starfruit (*Averrhoacarambola* L.) juice treated with ultraviolet radiation. *Food Chem*. 127: 641-644.

Bhat, R. 2016. Impact of ultraviolet radiation treatments on the quality of freshly prepared tomato (*Solanum lycopersicum*) juice.*Food Chem*. 213: 635–640.

Bintsis, T., Tzanetaki, E.L.,and Robinson, R.K. 2000.Existing and potential applicationsof ultraviolet light in the food industry - A critical review.*J. Sci. Food Agric*. 80: 637-645.

Birmpa, A., Vantarakis, A., Paparrodopoulos, S., Whyte, P., and Lyng, J. 2014.Efficacy of three light technologies for reducing microbial populations in liquid suspensions. *Biomed. Res. Int.*1: 13-17.

Boldaji, M., Borghei, A. M., Beheshti, B., and Hosseini, S. E. 2014. The process of producing tomato paste by ohmic heating method. *J. Food Sci. Technol*. 52(6): 3598–3606.

Bolton, J.R. and Linden, K. G. 2003.Standardisation of methods for fluence (UV dose) determination in bench-scale UV experiments.*J. Environ. Eng.* 129: 209- 215.

Bordignon-Luiz, Marilde, G., Gris, C., Eliana, F., Leila.2007. Colour stability of anthocyanins from Isabel grapes (*Vitis labrusca* L.) in model systems. *LWT - Food Sci. Technol.* 40: 594-599.

Bozkurt, H. and Icier, F. 2012.The change of apparent viscosity of liquid whole egg during ohmic and conventional heating.*J. Food Process. Eng.* 35: 120-133.

Brochier, B. and Domeneghini, G. 2016. Influence of moderate electric field on inactivation kinetics of peroxidase and polyphenol oxidase and on phenolic compounds of sugarcane juice treated by ohmic heating. *LWT-Food Sci. Technol*. 74: 396-403.

Brochier, B. and Domeneghini, G. 2018. Effect of ohmic heating parameters on peroxidase inactivation, phenolic compounds degradation and colour changes of sugarcane juice.*Food Bioprod.Process*.111: 62–71.

Buzrul, S., Alpas, H., Largeteau, A., and Demazeau, G. 2008.Inactivation of *Escherichia coli* and *Listeria innocua* in kiwifruit and pineapple juices by high hydrostatic pressure.*Int. J. Food Microbiol.*124: 275-278.

Cacace, D. and Palmieri, L. 2014. Chapter 13 - High-intensity Pulsed Light Technology. *Emerging Technologies for Food Processing* (Second Edi). Elsevier Ltd. Available: https://doi.org/10.1016/B978-0-12-411479-1.00013-9 [23, August 2018].

Caminiti, I., Palgan, I., Muñoz, A., Noci, F., Whyte, P., Morgan, D., Cronin, D., and Lyng, J. 2010. The effect of ultraviolet light on microbial inactivation and quality attributes of apple juice. *Food Bioprocess Technology*. Available: http://dx.doi.org/10.1007/s11947-010-0365-x. [29 March 2019].

Caminiti, I. M., Palgan, I., Noci, F., Munoz, A., Whyte, P., and Cronin, D. A. 2011. The effect of pulsed electric fields (PEF) in combination with high intensity light pulses (HILP) on *Escherichia coli* inactivation and quality attributes in apple juice. *Innov. Food Sci. Emerg. Technol*. 12 (2): 118-123.

Caminiti, I. M., Noci, F., Morgan, D. J., Cronin, D. A., and Lyng, J. G. 2012. The effect of pulsed electric fields, ultraviolet light or high intensity light pulses

in combination with manothermosonication on selected physico-chemical and sensory attributes of an orange and carrot juice blend. *Food Bioprod.Process*. 90: 442-448.

Campos, D.C.P., Santos, A.S., Wolkoff, D.B., Matta, V.M., Cabral, L.M.C., and Couri, S. 2002. Cashew apple juice stabilization by microfiltration.*Desalination*. 148: 61-65.

Carneiro, L., Sa, I.D.S., Gomes, F.D.S., Matta, V.M., and Cabral, L.M.C., 2002. Cold sterilization and clarification of pineapple juice by tangential microfiltration. *Desalination.*148: 93-98.

Carvalho, J.M., Maia, G.A., de Figueiredo, R.W., de Brito, E.S., and Rodrigues, S. 2007. Storage stability of a stimulant coconut-cashew apple juice beverage.*J. Food Process. Pres.* 31: 178-189.

Cashew Hand Book- Global Perspective. 2014. Available: http://www.cashewinfo.com/cashewhandbook2014.pdf [28 December 2019].

Castro, A.J. and Barbosa-Canovas, G.V.1993.Microbial inactivation of foods by pulsed electric fields.*J. Food Process. Preserv*. 17:47–73.

Castro, I., Teixeira, J. A., Salengke, S., Sastry, S. K., and Vicente, A. A. 2003.The in- fluence of field strength, sugar and solid content on electrical conductivity of strawberry products.*J. Food Process Eng.* 26:17-29.

Castro, I., Macedo, B., Teixeira, J. A., and Vicente, A. A. 2004a.The effect of electric field on important food processing enzymes comparison of inactivation kinetics under conventional and ohmic heating.*J .Food Sci.* 69(9):696-701.

Castro, S. M., Van Loey, A., Saraiva, J. A., Smout, C., and Hendrickx, M. 2004b. Activity and process stability of purified green pepper (*Capsicum annuum*) pectin methylesterase.*J. Agric. Food Chem.* 52: 5724-5729.

Castro, S. M., Van-Loey, A., Saraiva, J., Smout, C., and Hendrickx, M. 2006.Inactivation of pepper (*Capsicum annum*) pectin methylesterase by combined high pressure and temperature treatments. *J. Food Eng*. 75: 50-58.

Cavalcante, A.A.M., Rubensam, G., Erdtmann, B., Brendel, M., and Henriques, J.A.P. 2005. Cashew (*Anacardiumoccidentale*) apple juice lowers mutagenicity of aflatoxin B1 in *S. typhimurium* TA102. *Genet.Mol Biol.* 28: 328-333.

Chaine, A., Levy, C., Lacour, B., Riedel, C., and Carlin, F. 2012. Decontamination of sugar syrup by pulsed light. *J. Food Prot*. 75: 913-917.

Chakraborty, I. and Athmaselvi, K. A. 2014.Changes in physicochemical properties of guava juice during ohmic heating. *J. ready eat food*.1(4): 152-157.

Chang, J.C., Ossoff, S.F., Lobe, D.C., Dorfman, M.H., Dumais, C. M., and Qualls, R.G. 1985.UV inactivation of pathogenic and indicator microorganisms. *Appl. Environ. Microbiol.* 49(6): 1361–1365.

Chanprasartsuk, O., Prakichaiwattana, C., and Sanguandeekul, R. 2013. Comparison ofmethods of identification of yeasts isolated during spontaneous fermentation of freshly crushed pineapple juices. *J. Agric. Sci. Technol.* 15: 1479-1490.

Chattopadhyay, N., Hore, J.K., and Sen, S.K. 1992.Extension of storage life of sweet orange (*C. sinensis Osbeck*) cv. *Jaffa. Indian J. Plant Physiol.* 35: 245-25.

Cheftel, J.C. and Cheftel, H. 1992.*Introduction a la Bioquimicay Technologia de los Alimentos*:*Vol. 1*. Zagoroza, Spain, 321p.

Chew, S.C., Ghazali, H.M., Martín-belloso, O., and Adzahan, N.M. 2014. Effects of combining ultraviolet and mild heat treatments on enzymatic activities and total phenolic contents in pineapple juice. *Innov. Food Sci. and Emerg.Technol.*26: 511-516.

Chia, S. L., Rosnah, S., Noranizan, M. A., and Wan-Ramli, W. D. 2012. The effect of storage on the quality attributes of ultraviolet-irradiated and thermally pasteurised pineapple juices. *Int. Food Res. J*. 19(3): 1001–1010.

Cho, H. Y., Yousef, A. E., and Sastry, S. K. 1999. Kinetics of inactivation of *Bacillus subtilis* spores by continuous or intermittent ohmic and conventional heating. *Biotechnol.Bioeng*. 62: 368-372.

Chobotova, K.,Vernallis, A.B., and Majid, F.A.2010. Bromelain's activity and potential as an anti-cancer agent: Current evidence and perspectives. *Cancer Lett*. 290(2): 148-56.

Choi, M. S., Cheigh, C.I., Jeong, E.A., Shin J.K., and Chung, M.S. 2010. Non-thermal sterilisation of *Listeria monocytogenes* in infant foods by intense pulsed-light treatment. *J. Food Eng*. 97: 504–509.

Christensen, J. and Linden, K.G. 2003. How particles affect UV light in the UV disinfection of unfiltered drinking water. *J. Am. Water Works Assoc.* 95(4): 179–189.

Chutintrasria, B. and Noomhorm, A. 2007. Colour degradation kinetics of pineapple puree during thermal processing. *LWT-Food Sci. Technol*. 40: 300–306.

Cordenunsi, Saura-Calixto, B., Diaz-Rubio, F., Zuleta, E.M., Tiné, A., Buckeridge, M., Silva, M., Carpio, G., Giuntini, C., Menezes, E., Lajolo, E., and Franco, 2010. Carbohydrate composition of ripe pineapple (cv. perola) and the glycemic response in humans. *Cienc.Tecnol.Aliment*.30(10).

Costa, M.C.O., Maia, G.A., Figueiredo, R.W., Souza Filho, M.M., and Brasil, I.M. 2003. Storage stability of cashew apple juice preserved by hot fill and aseptic processes.*Cienc.Tecnol.Aliment. Campinas.* 23: 106-109.

Cousin, J. F., Desailly, F., Goullieux, A., and Pain, J. P. 2001.Process for treating vegetables and fruit before cooking. United States Patent No. US6821540B2. Available: https://patents.google.com/patent/US6821540B2/en [13May 2019].

Crisosto, C. H., Seguel, X., and Michailides, T. 1998. Comparing pulsated ultraviolet lightand postharvest fungicide for peach fruit decay control. *ActaHortic.*465: 471-479.

Cserhalmi, Z., Sass-Kiss, Á., Tóth-Markus, M., and Lechner, N.2006. Study of pulsed electric field treated citrus juices. *Innov.Food Sci. Emerg. Technol*. 7: 49–54.

Damasceno, L.F., Fernandes, F.A.N., Magalhaes, M.M.A., and Brito, E.S. 2008.Evaluation and optimization of non-enzymatic browning of “cajuina” during thermal treatment.*Braz. J. Chem. Eng.*25(2): 313-320.

Darvishi, H., Hosainpour, A., Nargesi, F., Khoshtaghaza, M. H., and Torang, H. 2011. Ohmic processing: Temperature dependent electrical conductivities of lemon juice. *Mod. Appl. Sci*. 5(1): 209-216.

Darvishi, H., Hosainpour, A., and Nargesi, F. 2012. Ohmic heating behaviour and electrical conductivity of tomato paste. *J. Nutr. Food Sci*. 2: 9.

Darvishi, H., Khostaghaza, M.H., and Najafi, G. 2013. Ohmic heating of pomegranate juice: Electrical conductivity and pH change. *J. Saudi Soc. Agric. Sci*. 12(2): 101–108.

Das, I. and Arora, A. 2017. Post-harvest processing technology for cashew apple: A review. *J. Food Eng.*194: 87-98.

Davey, M. W., Van-Montagu, M., Inze, D., Sanmartin, M., Kanellis, A., Smimoff, N., Benzie, L.J.J., Strain, J.J., Favell, D., and Fletcher, J. 2000. Plant L-ascorbic: chemistry, function, metabolism, bioavailable and effects of processing. *J. Sci. Food Agric*. 80(7): 825-860.

Dedehou, S.C.A.E., Dossou, J., Ahohuendo, B., Saidou, A., Ahanchede, A., and Soumanou, M.M. 2015.Optimization of cashew (*Anacardiumoccidentale* L.) apple juice's clarification process by using cassava and rice starch. *J. Appl. Biosci.* 95: 8989-9002.

Dhakal, S., Balasubramaniam, V.M., Ayvaz, H., and Rodriguez-Saona, L.E. 2018. Kinetic modeling of ascorbic acid degradation of pineapple juice subjected to combined pressure-thermal treatment. *J. Food Eng.*224: 62-70.

Doyle, M.P., Beauchat, L.R. and Montville T.J. (eds). 1997. *Food Microbiology: Fundamentals and Frontiers*(5th Ed.). ASM Press, Washington D.C, pp. 497-519.

Dunn, J. E., Clark, R. W., Asmus, J. F., Pearlman, J. S., Boyer, K., Painchaud, F. and Hofmann, G.A. 1988. United StatesPatent No. 4,871,559. Washington, DC:

U.S. Patent and Trademark Office. Available: http://www.patentbuddy.com/ Patent/4871559[21 May 2018].

Dunn, J. E., Clark,R. W., Asmus,J. F., Pearlman,J. S., Boyer,K., andPainchaud, F. 1991.Methods for preservation of food- stuffs. United States Patent No. 5034235.Available:https://patents.google.com/patent/US5034235A/en [23May 2018].

Dunn, J., Ott, T., and Clark, W. 1995.Pulsed-light treatment of food and packaging.*Food Technol.* 49: 95–98.

Dunn, J., Bushnell, A., Ott, T., and Clark, W. 1997.Pulsed white light food processing.*Cereal Foods World*. 42: 510-515.

Echeverria, E. and Valich, J. 1989. Enzymes of sugar and acid metabolism in stored 'Valencia' oranges. *J. Am. Soc. Horticult. Sci. (USA).* 114: 445–449.

EFSA [European Food Safety Authority]. 2015. The European Union summary report on trends and sources of zoonoses, zoonotic agents and food-borne outbreaks in 2013.European Centre for Disease Prevention and Control, *EFSA J.* 13(1): 162.

Emelike, N.J.T., Hart, A.D., and Ebere, C.O. 2015. Influence of drying techniques on the properties, physicochemical and mineral composition of beetroot juice. *IOSR J. Environ. Sci., Toxicol. Food Technol.* 9(12-2): 20-26.

Eribo, B. and Ashenafi, M. 2003. Behavior of *Escherichia coli* O157:H7 in tomato and processed tomato products. *Food Res. Int.* 36:823–830.

Esteve, M.J.and Frigola, A. 2007. Refrigerated fruit juices: quality and safety issues. In: Taylor, S.L. (ed.),*Advances in food and nutrition research*. USA: Elsevier Inc., pp. 104-132.

Falguera, V., Pagan, J., and Ibarz, A. 2011.Effect of UV irradiation on enzymatic activities and physicochemical properties of apple juices from different varieties.*LWT- Food Sci. Technol*. 44(1): 115-119.

Farkas, J. 1997. *Analysis and optimum design of metal structures*, CRC Press, 436p.

FDA [Food and Drug Administration]. 1998. *Hazard Analysis Critical Control Point (HACCP): Procedures for the safe and sanitary processing and importing of juice. Department of Health and Human Services.*63 FR 20450[on- line].Available: https://www.federalregister.gov/documents/2001/01/19/01- 1291/ha zard-analysis-and-critical-control-point-haacp-procedures-for-the- safe-and-sanitary-processing [09 August 2017].

FDA [US Food and Drug Administration]. 2004. *FDA Guidance to Industry, 2004: Recommendations to Processors of Apple Juice or Cider on the Use of Ozone for Pathogen Reduction purposes* [on-line]. Available: https://www.fda.gov/ regulatory-information/search-fda-guidance documents/guidance-industry-reco mmendations-processors-apple-juice-or-cider-use-ozone-pathogen-reduction [08 August 2018].

Federal register. 1999. 'Pulsed light treatment of food', Fed Regist, 66, 338829–338830 [on-line] Available:https://www.accessdata.fda.gov/scripts/cdrh/cfdocs/cfcfr/CFRSearch.cfm?fr=179.41 [28 May 2018].

Fernando, M. B., Magnolia, L. S., Jose, Z. M., and Eduardo, M. S. 2000. Preparation and properties of pre-gelatinized cassava (manihot esculenta. crantz) and jicama (pachyrhizus erosus) starches using ohmic heating.*Cereal Chem*. 39(3): 275-283.

Ferrario, Guerrero, M., Alzamora, S., and Stella. 2013. Study of pulsed light-induced damage on *Saccharomyces cerevisiae* in apple juice by flow cytometry and transmission Electron Microscopy. *Food Bioprocess Technol.* 6(6): 312-321.

Ferrario, M., Maris, S. and Guerrero, S. 2015. Study of the inactivation of spoilage microorganisms in apple juice by pulsed light and ultrasound. *Food Microbiol*. 46: 635-642.

Ferrario, M. and Guerrero, S. 2016. Effect of a combined flow- through pulsed light system combined with ultrasound on microbial survivability, colour and sensory shelf life of apple juice. *Innov. Food Sci. Emerg. Technol*. 34: 214-224.

Ferrario, M.I. and Guerrero, S.N. 2018. Inactivation of *Alicyclobacillusacidoterrestris* ATCC 49025 spores in apple juice by pulsed light influence. *Rev. Argentina Microbiol.* 50(1): 3-11.

Ferreira, V. F., Nicoletti, C. D., Ferreira, P. G., Futuro, D. O., and Silva, F.C. 2016. Strategies for increasing the solubility and bioavailability of anticancer compounds: β-Lapachone and other Naphthoquinones. *Curr. Pharm. Des.* 22: 5899–5914.

Figueiredo, R.W., Lajolo, F.M., Alves, R.E., and Filgueiras, H.A.C. 2002. Physical-chemical changes in early dwarf cashew pseudo fruits during development and maturation. *Food Chem.* 77: 343–347.

Filgueiras, H.A.C., Alves, R.E., Masca, J.L., and Menezes, J.B. 1999. Cashew apple for fresh consumption: Res. on harvest and postharvest handling technology in Brazil. *Acta Hort.* 485: 155-160.

Fine, F. and Gervais, P. 2004. Efficiency of pulsed UV light for microbial decontamination of food powders. *J. Food Prot.* 67: 787–792.

Foley, D., Pickett, M., Varon, K., Lee, J. J., Min D. B., and Caporaso. F. 2002. Pasteurisation of fresh orange juice using gamma irradiation: microbiological, flavour, and sensory analyses. *J. Food Sci.* 67(4): 1495-1501.

Fontoin, F., Saucier, C., Teissedre, P.L., and Glories, Y. 2008.Effect of pH, ethanol and acidity on astringency and bitterness of grape seed tannin oligomers in model wine solution.*Food Qual. Prefer.* 19: 286-291.

Frazier, W. C., Westoff, D. C., and Vanitha, K. N. 1971.*Food Microbiology* (5th Ed.).McGraw-Hill,New York, 256p.

Fryer, P. J. and Li, Z. 1993. Electrical resistance heating of foods.*Trends Food Sci. Technol*. 4: 364-369.

FSSAI [Food Safety and Standards Authority of India].2018. Revised microbiological standards for Fruits and Vegetables and their products, Gazette Notification dtd 04/04/2018, Available:https://archive.fssai.gov.in/dam/jcr:e5d90e3a47954ff285d1ec355bbf612c/Gazette_Notification_Fruits_Vegetables_04_04_2018.pdf[27 July 2019].

Funes, G. J., Gomez, P. L., Resnik, S. L., and Alzamora, S. M. 2013.Application of pulsed light to patulin reduction in Mcllavine buffer and apple products. *Food Control*. 30(2): 405-410.

Fustier, P., St-Germain, F., Lamarche, F., and Mondor, M., 2011. Non-enzymatic browning and ascorbic acid degradation of orange juice subjected to electroreduction and electro-oxidation treatments. *Innov.Food Sci. Emerg. Technol.* 12: 491–498.

Gachovska, T.K., Kumar, S., Thippareddi, H., Subbiah, J., and Williams, F. 2008.Ultraviolet and pulsed electric field treatments have additive effect on inactivation of *E. coli* in apple juice. *J. Food Sci.* 73: 412–417.

Ghasemi, Z., Macgregor, S., Anderson, J., and Lamont, Y. 2003. Development of an integrated solid-state generator for light in- activation of food-related pathogenic bacteria.*Meas. Sci. Tech- nol.* 14: 26–32.

Gomez-Lopez, V.M., Devlieghere, F., Bonduelle, V., and Debe- vere, J. 2005a. Intense light pulses decontamination of minimally processed vegetables and their shelf-life. *Int. J. Food Microbiol.* 103: 79-89.

Gomez-Lopez, V.M., Devlieghere, F., Bonduelle, V., and Debe- vere, J. 2005b.Factors affecting the inactivation of microorganisms by intense light pulses.*J. Appl. Microbiol.*99: 460-470.

Gomez-Lopez, V. M., Ragaert, P., Debevere, J. and Devlieghere, V. M. 2007. Pulsed light for food decontamination: a review. *Trends Food Sci. Tech***.** 18: 464-473.

Gomez-Lopez, N., Vega-Sanchez, R., Castillo-Castrejon, M., Romero, R., Cubeiro-Arreola, K., and Vadillo-Ortega, F. 2013.Evidence for a role for the adaptive immune response in human term parturition.*Am. J. Reprod. Immunol.* 69: 212– 230.

Gould, W.G.1996. Methods for preservation and extension of shelf life.*Int. J. Food Microbiol.* 33: 51-54.

Gouma, M., Gayan, E., Raso, J., Condon, S., and Alvarez, I. 2015. Inactivation of spoilage yeasts in apple juice by UV-C light and in combination with mild heat. *Innov. Food Sci. Emerg. Technol.* 32: 146-155.

Grapperhaus, M., Schaefer, R. B., and Linden, K. 2005.Modelling of a new UV test cell for evaluation of lamp intensity effects on water treatment. In: Whistler, B.C. (ed.). *CDROM Proceedings of the 3rd International Congress on Ultraviolet Technologies*.International Ultraviolet Association, PO Box 1110, Ayr, ON, Canada, pp. 35-55.

Gregory, S. and Ortwerth, B. 2000.The non-oxidative degradation of ascorbic acid at physiological conditions.*Biochimic.biophysic.acta*. 1501: 12-24.

Guerrant, N.B., Adams Dutcher, R., Tabor, F. S., and Rasmussen, R. 1936. The vitamin content of canned pineapple juice. *J. Nutr*. 11(4): 383-390.

Guerrero-Beltrán, J. A. and Barbosa-Canovas, G.V. 2005.Reduction of *Saccharomyces cerevisiae*, *Escherichia coli* and *Listeria innocua* in apple juice by ultraviolet light. *J. Food Process Eng*. 28(5): 437–452.

Guerrouj, K., Sanchez-Rubio, M., Taboada-Rodriguez, A., Cava-Roda, R. M., and Marın-Iniesta, F. 2016. Sonication at mild temperatures enhances bioactive

compounds and microbiological quality of orange juice. *Food Bioprod. Process.* 99: 20–28.

Guillou, S. and Murr, N. E. 2002.Inactivation of *Saccharomyces cerevisiae* in solution by low-amperage electric treatment. *J. Appl. Microbiol*. 92: 860-865.

Hale, L. P., Greer, P. K., Trinh, C.T., and James, C. L. 2005. Proteinase activity and stability of natural bromelain preparations. *Int. Immunopharmacol*. 5(4): 783-793.

Hariharan, P.V. and Gerutti, P.A. 1977.Formation of products of the 5-6 dihydroxydihydrotymine type by ultraviolet light in HeLa cells. *Biochem.* 16(12): 2791-2795.

Haro-Maza, J. and Guerrero-Beltran, J. 2016. Ultraviolet-C Light Effect on the Physicochemical and Antioxidant Properties of Blackberry, Blueberry, and Raspberry Nectars, *J. Food Res.* 5(5): 11-22.

Helena, E., Rosenthal, A., Calado, V., Saraiva, J., and Mendo, S. 2009. Byssochlamysnivea inactivation in pineapple juice and nectar using high pressure cycles. *J. Food Eng.* 95(4): 664-669.

Hillegas, S.L. and Demirci, A. 2003.Inactivation of *Clostridium sporogenes* in clover honey by pulsed UV-light treatment. *Agric. Eng. Int.: CIGR J. Sci. Res. Dev*.5: 3-9.

Hiramoto, T. 1984. Method of sterilisation. United States Patent No. 4464336.Available: https://patents.google.com/patent/US4464336A/en [28 July 2017]

Holck, A., Liland, K., Dromtorp, S., Carlehog, M., and McLeod, A. 2018. Comparison of UV-C and pulsed UV light treatments for reduction of *Salmonella,Listeria monocytogenes,* and *Enterohemorrhagic Escherichia coli* on eggs. *J. Food Prot.* 81: 6-16.

Honorato, T.L. and Rodrigues, S. 2010. Dextransucrase stability in cashew apple juice. *Food Bioprocess Technol.* 3: 105-110.

Hounhouigan, M.H., Linnemann, A.R., Soumanou, M.M., and Van Boekel, M.A.J.S. 2014.Effect of processing on the quality of pineapple juice. *Food Rev. Int.* 30: 112-133.

Huang, Y., Ye, M., Cao, X., and Chen, H. 2017. Pulsed light inactivation of *murine norovirus*, *Tulane virus*, *Escherichia coli* O157:H7 and *Salmonella* in suspension and on berry surfaces. *Food Microbiol.* 61: 1-4.

Huffman, D.E., Slifko, T.R., Salisbury, K., and Rose, J.B. 2000. Inactivation of bacteria, virus and *Cryptosporidium* by a point-of-use device using pulsed broad white light. *Water Res.* 34: 2491-2498.

Hwang, H. J., Cheigh, C. I., and Chung, M. S. 2015. Relationship between optical properties of beverages and microbial inactivation by intense pulsed light.*Innov. Food Sci. Emerg. Technol*. 31: 91-96.

Ibarz, A., Pagan, J., and Garza, S. 1999. Kinetic models for color changes in pear puree during heating at relatively high temperatures. *J. Food Eng*. 39(4): 415– 422.

Ibarz, Albert, Pagán, J., and Salvador, G. 2000. Kinetic models of non-enzymatic browning in apple puree. *J. Sci. Food Agric*. 80: 1162 - 1168.

Icier, F. 2003.Experimental investigation and mathematical modelling of ohmic heating of foods.PhD thesis. Ege University, Izmir, 86p.

Icier, F. and Ilicali, C. 2005.The use of tylose as a food analog in ohmic heating studies. *J. Food Eng.* 69(1): 67-77.

Icier, F. and Bozkurt, H. 2009. Ohmic heating of liquid whole egg: rheological behaviour and fluid dynamics. *Food Bioprocess Technol.* 4: 1253-1263.

Icier, F., Yıldız, H and Baysal, T. 2008. Polyphenoloxidase deactivation kinetics during ohmic heating of grape juice. *J. Food Eng.* 85(3):410-417.

Icier, F., Yildiz, H. and Baysal, T. 2010. Peroxidase inactivation and colour changes during ohmic blanching of pea puree. *J. Food Eng*. 74: 424-429.

Inyang, U.E. and Abah, U.J. 1997. Chemical composition and organoleptic evaluation of juice from steamed cashew apple blended with orange juice. *Plant Food Hum.Nutr.* 50: 295-300.

IUPAC [International Union of Pure and Applied Chemistry]. 1996. *Pure and Applied Chemistry*. International Union of Pure and Applied Chemistry, 12: pp. 2223-2286.

Jagtian, D. 1980. *Fruit Preservation*. Vikas Publishing House, New Delhi,

275p. Jay, J.J. 1996.*Modern food microbiology*. Chapman & Hall, New York,

193p.

Jayaleshmy, V.G. and John, P.S. 2004. 'Sago' – A natural product for cashew apple juice clarification. *J. Trop. Agric.* 42(1-2): 67-68.

Jean, M., Anvoh, K. Y., and Zoro, A. 2007. Production and characterization of juice from mucilage of cocoa beans and its transformation to marmalade. *Pakistan J. Nutr*. 8(2): 129-133.

Jones, F. 1897. Apparatus for electrically treating liquids. United States Patent No. 592735. USA. Available: https://patentimages.storage.googleapis.com/0a/98/ba/3e3c8157d881fc/US592735.pdf [10 January 2018].

Jordan, S.L., Pascual, C., Bracey, E., and Mackey, B.M. 2001. Inactivation and injury of pressure-resistant strains of *Escherichia coli* O157 and *Listeria monocytogenes*in fruit juices. *J.Appl. Microbiol*. 91:463–469.

Jun, S., Irudayraj, J., Demirci, A., and Geiser, D. 2003. Pulsed UV-light treatment of corn meal for inactivation of *Aspergillus niger.Int. J. Food Sci. Technol*. 38: 883-888.

Kabasakalis, V., Siopidou, D., and Moshatou, E. 2000.Ascorbic acid content of commercial fruit juices and its rate of loss upon storage. *Food Chem*. 70(3):325–328.

Kabuo, N.O., Ojukwu, M., Omeire, G.C., and Ibeabuchi, J.C. 2015. Extraction and preservation of cashew juice using Sorbic and Benzoic acids. *Am. J. Food Sci. Technol.*3(2): 48–54.

Kasahara, I., Kaiser, S., Aguilar, F., and Marillanca, M. 2004. Pasteurisation of apple juice by pulsed ultraviolet light: processing conditions and effect in the product

characteristics. In:*Proceedings of the International Conference Engineering and Food*, Montpellier, France, pp. 36-48.

Kausar, H., Saeed, S., Mushtaq, A.M., and Salam, A. 2012. Studies on the development and storage stability of cucumber-melon functional drink. *J. Agric. Res.* 50(2): 239-248.

Kaushal, N. and Muchomba, F. 2013. How consumer price subsidies affect Nutrition. NEBR. *Working Paper*, pp. 194-204.

Keklik, N.M. and Demirci, A. 2014.Applications and modelling aspects of UV and pulsed UV-light for food decontamination. In: Boziaris I.S.(ed.), *Novel FoodPreservation and Microbial Assessment Techniques*. New York: CRC Press, pp. 67–77.

Kennedy, J. F., Rivera, Z. S., Lloyd, L. L., Warner, F. P., and Jumel, K. 1990. Studies on non-enzymic browning in orange juice using a model system based on freshly squeezed orange juice. *J Sci. Food Agric.* 52:85–95.

Keyser, M., Muller, I.A., Cilliers, F.P., Nel, W., and Gouws, P.A. 2008.Ultraviolet radiation as a non-thermal treatment for the inactivation of microorganisms in fruit juice. *Innov.Food Sci. Emerg. Technol.* 9: 348-354.

Khalid, N., Hafiz, A.R.S., and Iftikhar, A. 2016.*Pineapple Juice,Handbook of Functional Beverages and Human Health*. CRC Press, 489p.

Khalil, M.I., Motallib, M.A., Anisuzzaman, A., Sathi, Z.S., Hye, M., and Shahjahan, M. 2001. Biochemical analysis of different brands of unifloral honey available at the northern region of Bangladesh. *J Med. Sci.* 1: 385– 388.

Kim, J. and Pyun, Y. 1995. Extraction of soy milk using ohmic heating.In: *Proceedings of the ninth congress on food science and technology*, Budapest, Hungary, pp. 287-296.

Kim, S. S. and Kang, D. H. 2015. Comparison of pH effects on ohmic heating and conventional heating for inactivation of *Escherichia coli* O157 : H7, *Salmonella enterica SerovarTyphimurium* and *Listeria monocytogenes* in orange juice. *LWT-Food Sci. Technol.*, 64 (2): 860-866.

Kim, S. S., Jo, Y. and Kang, D. H. 2019. Combined inhibitory effect of milk fat and lactose for inactivation of foodborne pathogens by ohmic heating.*LWT-Food Sci. Technol.* 86: 159-165.

Koutchma, T., Keller, S., Chirtel, S. and Parisi, B. 2004. Ultraviolet disinfection of juice products in laminar and turbulent flow reactors.*Innov.Food Sci. Emerg. Technol.* 5: 179-189.

Koutchma, T. 2009. Advances in ultraviolet light technology for non-thermal processingof liquid foods. *Food Bioprocess Technol*. 2: 138-155.

Koutchma, T., Forney L., and Moraru, C. 2009.*Ultraviolet light in food technology: Principles and applications.* Boca Raton: CRC, 462p.

Krebbers, B., Matser, A. M., Hoogerwerf, S. W., Moezelaar, R., and Tomassen, M. M. M. 2019. Combined high-pressure and thermal treatments for processing of tomato puree : Evaluation of microbial inactivation and quality parameters, *Innov. Food Sci. Emerg. Technol.*8(4):1763-1781Available: https://doi.org/10.1016/S1466-8564(03)00045-6 [06 Nov 2019].

Krebbers, B., Matser, A. M., Hoogerwerf, S. W., Moezelaar, R., Tomassen, M. M. M, Van Den-Berg, R. W. 2003. Combined high-pressure and thermal treatments for processing of tomato puree: evaluation of microbial inactivation and quality parameters. *Innov. Food Sci. Emerg. Technol.* 4(4): 377-385.

Krishnamurthy, K. 2006. Decontamination of milk and water by pulsed uv-light and infrared heating Agricultural and Biological Engineering, Department of

Agricultural and Biological Engineering, PhD thesis, The Pennsylvania State University, 86p.

Krishnamurthy, K., Demirci, A., and Irudayaraj, J.M. 2007. Inactivation of *Staphylococcus aureus* in milk using flow-through pulsed UV-light treatment system. *J. Food Sci.* 72(7): 233-239.

Kulshrestha, S. A. and Sastry, S. K. 2010. Changes in permeability of moderate electric field (MEF) treated vegetable tissue over time. *Innov. Food Sci. Emerg. Technol.* 11(1): 78–83.

Kwaw, E., Ma, Y., Tchabo, W., Apaliya, M. T., Sackey, A. S., Wu, M., and Ma, Y. 2018. Effect of pulsed light treatment on the phytochemical, volatile and sensorial attributes of lactic-acid-fermented mulberry juice. *Int. J. Food Prop*. 21(1): 228-243.

Lagnika, C., Adjovi, Y.C.S., Lagnika, L., and Gogohounga, F.O. 2017.Effect of combining ultrasound and mild heat treatment on physicochemical, nutritional quality and microbiological properties of pineapple juice. *Food Nutr. Sci.* 8: 227-241.

Lakkakula, N. R., Lima, M. and Walker, T. 2004.Rice bran stabilization and rice bran oil extraction using ohmic heating. *Bioresour. Technol*. 92: 157-161.

Lamsal, B.P. and Jindal, V.K. 2014. Variation in Electrical Conductivity of Selected Fruit Juices During Continuous Ohmic Heating. *Int. J. Appl. Sci. Technol.* 7(1): 47-56.

Laorko, A., Tongchitpakdee, S., and Youravong, W. 2013. Storage quality of pineapple juice non-thermally pasteurized and clarified by microfiltration. *J. Food Eng.*116(2): 554-561.

Lasagabster, A., Arboleya, J.C., and Martinez, D.M. 2011. Pulsed light technology for surface decontamination of eggs, Impact on *Salmonella* inactivation and egg quality.*Innov. Food Sci. Emerg. Technol.* 12(2): 124-128.

Lavinas, F.C., Miguel, M.A.L., Lopes, M.L.M., and Valente Mesquita, V.L. 2008.Effect of high hydrostatic pressure on cashew apple (*Anacardium occidentale,* L.) juice preservation. *J. Food Sci.* 73(6): 273-277.

Lebovka, N. I., Shynkaryk, M., and Vorobiev, E. 2007.Moderate electric field treatment of sugarbeet tissues. *Biosyst. Eng*. 96: 47-56.

Lebovka, N., Praporscic, I. and Vorobiev, E. 2004.Effect of moderate thermal and pulsed electric field treatment on textural properties of carrots, potatoes and apples. *Innov.Food Sci. Emerg. Technol*. 5: 9-16.

Lee, S. H. and Yoon, S. 1999. Effect of ohmic heating on the structure and permeability of the cell membrane of *Saccharomyces cerevisae*.IFT Annual Meeting 1999. IFT, Chicago, pp. 135-146

Lee, H.S. and Coates, G.A. 2003.Effect of thermal pasteurization on Valencia orange juice color and pigments. *LWT- Food Sci. Technol*. 36: 153-156.

Lee, J. Y., Kim, S.S., and Kang, D.H. 2015. Effect of pH for inactivation of *Escherichia coli* O157:H7, *Salmonella Typhimurium*and *Listeria monocytogenes* in orange juice by ohmic heating. *Lebensm-Wiss. Technol*. 62:83–88.

Lee, S. H. 2014. Hybrid combination of emerging food processing technologies: microwave and pulsed ohmic heating, PhD (Mol. Biosci. and Eng.) thesis, University of Hawaii, Manoa, 171p.

Lee, S. H., Yamada, K., and Jun, S. 2013. Ultraviolet radiation assisted with ohmic current for microbial inactivation in apple juice. *T. ASABE*, *56*(3): 1085–1091.

.Lee, S. Y., Sagong, H. G., Ryu, S., and Kang, D. H. 2012. Effect of continuous ohmic heating to inactivate *Escherichia coli* O157:H7, *Salmonella Typhimurium* and *Listeria monocytogenes* in orange juice and tomato juice. *J. Appl. Microbiol*. 112: 723-731.

Leizerson, S. and Shimoni, E. 2005a.Effect of ultrahigh-temperature continuous ohmic heating treatment on fresh orange juice. *J. Agric. Food Chem.* 53(9): 3519-3524.

Leizerson, S. and Shimoni, E. 2005b. Stability and sensory shelf life of orange juice pasteurised by continuous ohmic heating. *J. Agr. Food Chem*. 53(10): 4012-4018.

Levy, C., Aubert, X., Lacour, B., and Carlin, F. 2012. Relevant factors affecting microbial surface decontamination by pulsed light. *Int. J. Food Microbiol.* 152: 168-174.

Li, B. and Sun, D.W. 2002. Novel methods for rapid freezing and thawing of foods-a review. *J. Food Eng*. 54: 175-182.

Li, S.Q., Zhang, H.Q., Jin, T.Z., Turek, E.J., and Lau, M.H. 2005. Elimination of *Lactobacillus plantarum* and achievement of shelf stable model salad dressing by pilot scale pulsed electric fields combined with mild heat. *Innov. Food Sci. Emerg. Technol.* 6(2): 125-133.

Lima M., Heskitt, B. F., Burianek, L. L., Nokes, S. E., and Sastry, S. K. 1999. Ascorbic acid degradation kinetics during conventional and ohmic heating. *J. Food Process Eng.* 23(5):421-434.

Lima, M. and Sastry, S. K. 1999. The effects of ohmic heating frequency on hot air drying rate and juice yield. *J. Food Eng*. 41: 115-119.

Loghavi, L., Sastry, S.K., and Yousef, A.E. 2008.Effect of moderate electric field frequency on growth kinetics and metabolic activity of *Lactobacillus acidophilus*. *Biotechnol.Prog*. 24: 148-153.

Lojewska, Z., Farkas, D. L., Ehrenberg, B., and Loew, L. M. 1989.Analysis of the effect of medium and membrane conductance on the amplitude and kinetics of membrane potentials induced by externally applied electric fields. *Biophys. J*. 56: 121-128.

Lowor, S.T. and Agyente-Badu, C.K. 2009.Mineral and proximate composition of cashew apple (*AnacardiumOccidentale L.*) juice from northern savannah,

forest and coastal savannah regions in Ghana.*Am. J. Food Technol.* 4: 154-161.

Luzuriaga, D.A., Roberts, J.S., and Balaban, M.O. 1996.Electrical conductivity of frozen shrimp and flounder at different temperatures and volt- age levels. *J Agric Food Proc Technol.* 3(5): 41–63.

Mac-Gregor, S.J., Rowan, N.J., McIlvaney, L., Anderson, J.G.,Fouracre, R.A., and Farish, O.1998. Light inactivation of food-related pathogenic bacteria using a pulsed powersource. *Lett. Appl. Microbiol.* 27: 67-70.

Macheix, J.J., Fleuriet, J.A., and Billot, J. 1990. *Fruit phenolics*. Boca Raton, FY: CRC Press, 364p.

Mafeti, N.A., Ramos, A.Y.V., Nicolau, A.I., Martin-Belloso, O., and Soliva-Fortuny, R. 2014. Influence of process paramters on the pulsed light inactivation of *Penicillin expansum* in apple juice. *Food Control*. 41: 27-31.

Majumdar, T.K., Wadikar, C.R, Vasudish, C.R., Premavalli,K.S., and Bawe, R.S. 2011. Effect of storage on physic-chemical, microbiological and sensory qualities of bottlegourd- basil leaves juice, *Am. J. Food Technol*. 6(3):226-234.

Makroo, H. A., Rastogi, N. K., and Srivastava, B. 2016. Enzyme inactivation of tomato juice by ohmic heating and its effects on physico-chemical characteristics of concentrated tomato paste. *J. Food Process. Eng*. 40(3): 1-10.

Makroo, H. A. and Srivastava, B. 2017.Effects of ohmic heating on different liquid food materials. In: *Trends and innovations in food processing technology*, 95-115 February 2017, Tezpur University [on-line]. Available: https://www.researchgate.net/publication/316038217 [10 December 2019].

Mansor, A., Shamsudin, R., Adzahan, N.M., and Hamidon, M.N. 2014.Efficacy of Ultraviolet Radiation as Non-thermal Treatment for the Inactivation of *Salmonella typhimurium* TISTR 292 in Pineapple Fruit Juice.*Agric. Agric.l Sci. Proc.* 2: 173-180.

Marc, A., Kouassi, Didier, T., Amani, F.A., and Guessan, N. 2012.Phenolic profile of cashew apple juice (*Anacardium occidentale* L.) from Yamoussoukro and Korhogo (Côte d'Ivoire). *J. Appl. Biosci.* 49: 3331-3338.

Marquenie, D., Geeraerd, A.H., Lammertyn, J., Soontjens, C., Van Impe, J.F., andMichiels, C.W. 2003.Combinations of pulsed white light and UV-C or mild heat treatment to inactivate conidia of *Botrytis cinerea* and *Moniliafructigena.Int. J. Food Microbiol.* 85: 185-196.

Marra, F., Zell, M., Lyng, J. G., Morgan, D. J., and Cronin, D.A. 2009.Analysis of heat transfer during ohmic processing of a solid food. *J. Food Eng*. 91: 56-63.

Martinez-Bustos, F., Lopez-Soto, M., Zazueta-Morales, J. J.,and Morales-Sanchez, E. 2005.Preparation and properties of pre-gelatinized cassava (*Manihot esculenta Crantz*) and jicama (*Pachyrhizus erosus*) starches using ohmic heating. *Agrociencia*. 39(3): 275-283.

Masamba, K.G. and Mndalira, K. 2016. Vitamin C stability in pineapple, guava and baobab juices under different storage conditions using different levels of sodium benzoate and metabisulphite. *Afr. J.Biotechnol.* 12(2): 186-191.

Masschelein, W. J. 2002. *Ultraviolet light in water and wastewater sanitation*. Boca Raton: CRC Press, 254p.

Matthews R.F. 1994. Frozen concentrated orange juice from florida oranges.Univ. Fla. *IFAS Extension; Gainsville,* FL, USA. pp. 1–4.

Mazzotta, A. S. 2001. Thermal inactivation of stationary-phase and acid-adapted *Escherichia coli* O157 : H7, *Salmonella* , and *Listeria monocytogenes* in fruit Juices. *J. Food Prot*. 64(3): 315–320.

McDonald, K. F., Curry, R. D., Clevenger, T. E., Brazos, B. J., Unklesbay, K., and Eisenstark, A. 2000. The development of photosensitized pulsed and continuous ultraviolet light decontamination techniques for surfaces and solutions.*IEEE Trans. Plasma Sci*. 28: 89-95.

Meng.J., Doyle, M.P., Zhao, T., and Zhao, S. 2001.Enterohemorrhagic *Escherichia coli*. In: Doyle, M.P., Beuchat L.R., and Montville, T.J. (eds), *Food Microbiology: Fundamentals and Frontiers* (2nd Ed.). ASM Press, Washington, DC, pp. 193-214.

Mercali, G. D., Schwartz, S., Marczak, L. D. F., Tessaro, I. C., and Sastry, S. 2014. Ascorbic acid degradation and color changes in acerola pulp during ohmic heating: Effect of electric field frequency. *J. Food Eng*. 123(0): 1-7.

Meydav, S., Saguy, I., and Kopelman, I. J. 1977.Browning determination in citrus products. *J Agric. food Chem.* 25(3): 602–604.

Miao, Y., Chen, J.Y., and Noguchi, A. 2007.Studies in the ohmic thawing of frozen surimi. *Food Sci. Technol. Res*. 13(4): 296-300.

Michodjehoun-Mestres, L., Souquet, J.M., Fulcrand, H., Bouchut, C., Reynesa, M., and Brillouet, J. M. 2009.Monomeric phenols of cashew apple (*Anacardium occidentale* L.). *Food Chem.* 112:851–857.

Mizrahi, S. 1996. Leaching of soluble solids during blanching of vegetables by ohmic heating. *J. Food Eng*. 29: 153-166.

MOFPI [Ministry of Food Processing Industries]. 2015. *Annual report 2014-15* [on-line]. Available: https://mofpi.nic.in/sites/default/files/annualreport 201415eng. pdf0.pdf [06 June 2017].

Moraes, I.C., Fasolin, L.H., Cunha, R.L., and Menegalli, F.C. 2011. Dynamic and steady: shear rheological properties of xanthan and guar gums dispersed in yellow passion fruit pulp (*Passiflora edulis* f. flavicarpa), *Braz J. Chem Eng.* 28: 483–494.

Morgan, R. 1989. UV ‘green’ light disinfection. *Dairy Ind. Int.* 54(11): 33–35.

Morton, J.F. 1961. The Cashew's brighter future. *Econ. Bot.* 15: 57-78.

Moyer, J.C. and Aitken, H.C. 1980.Apple juice. In: Nelson, P.E. and Tressler, D.K. (eds), *Fruit and vegetable juice processing,*USA: AVI Publishing Co., Inc., pp. 212-267.

Muller, A., Stahl, M.R., Graef, V., Franz, C. M., and Huch, M. 2011. UV-C Treatment of Juices to inactivate microorganisms using Dean Vortex Technology. *J. Food Eng.*107: 268-275.

Munoz , A., Palgan, I., Noci, F., Morgan, D. J., Cronin, D. A., Whyte, P. and Lyng, J. G. 2011. Combinations of high intensity light pulses and thermosonication for the inactivation of *Escherichia coli* in orange juice. *Food Microbiol*. 28: 1200- 1204.

Munoz, A., Caminiti, I., Palgan, I., Pataro, G., Noci, F., Morgan, D. and Lyng, J. 2012.Effects on *Escherichia coli* inactivation and quality attributes in apple juice treated by combinations of pulsed light and thermosonication. *Food Res. Int*. 45: 299-305.

Murashita, S., Kawamura, S., and Koseki, S. 2017.Effects of ohmic heating, including electric field intensity and frequency, on thermal inactivation of *Bacillus subtilis* spores. *J. Food Prot.* 80: 164-168.

NACMCF [National Advisory Committee on Microbiological Criteria for Foods].2006. Requisite scientific parameters for establishing the equivalence of alternative methods of pasteurization. *J Food Prot.* 69(5): 1190–1216.

Nair, G. R., Divya, V. R., Prasannan, L., Habeeba, V., Prince, N. V., and Raghavan, G.S.V. 2012. Ohmic heating as a pre-treatment in solvent extraction of rice bran. *J. Food Sci. Technol.* 51(40): 2692-2698.

Nazir, A., Wani, S.M., Ganil, A., Masoodi, F.A., Haq, E., Mir, S.A., and Riyaz, U. 2013. Nutritional, antioxidant and antiproliferative properties of persimmon (*Diospyros kaki*) – a minor fruit of J&K. *Int. J. of adv. Res.* 1(7): 545-554.

Ngadi, M., Smith, J., and Cayouette, B. 2003. Kinetics of ultraviolet light inactivation of *Escherichia coli* O157:H7 in liquid foods. *J. Sci Food Agric.* 83: 1551-1555.

Ngozi, O.K., Moses Ojukwu, Gloria, C.O., and Ibeabuchi, J.C. 2015. Extraction and preservation of cashew juice using sorbic and benzoic acids. *Am. J. Food Technol.* 3(2): 48-54.

NHB [National Horticultural Board].2017. *Indian Horticulture Database 2015* [on-line]. Available:http://nhb.gov.in/areapro/NHB_Database_2015.pdf [23 July 2017].

Nicorescu, I., Nguyen, B., Moreau-Ferret, M., Agoulon, A., Chevalier, S., and Orange, N. 2013. Pulsed light inactivation of *Bacillus subtilis* vegetative cells in suspensions and spices. *Food Control*. 31: 151-157.

Nisperos-Carriedo, M.O., Shaw, P.E., and Baldwin, E.A. 1990.Changes in volatile flavor components of pineapple orange juice as influenced by the application of lipid and composite films. *J. Agric. Food Chem.* 38: 1382-1387.

Noci, F., Riener, J., Walkling-Ribeiro, M., Cronin, D.A., Morgan, D.J., and Lyng, J.G. 2008. Ultraviolet irradiationand pulsed electric fields (PEF) in a hurdle strategy for the preservation of fresh apple juice. *J. Food Eng*. 85: 141-146.

Nwachukwu, E. and Ezejiaku, F.C. 2014. Microbial and physicochemical characteristics of locally produced pineapple juice treated with garlic and ginger. *Int.J.Curr.Microbiol.App.Sci.*3(6): 895-901.

Obeta, J.A.N. and Ugwuanyi, J.O. 1997. Shelf life study of some Nigerian fruit juices inoculated with ascospores of Neosartorya spp. *Plant Foods Hum. Nutr.* 50: 325-331.

Obot-Essien.S and Ufort-Usoh, E. 2016.Effect of temperature on the rheological properties of pineapple juice (a*nanascomosus*). *Int. J. Latest Res. Sci. Technol.5*(2): 86-90.

Ochoa-velasco, C. E. and Quimica, D. I. 2012. Ultraviolet-C Light Effect on Pitaya (*Stenocereus griseus*) Juice. *J. Food Res*. 1(2): 60–70.

Odriozola-Serrano, I., Aguiló-Aguayo, I., Soliva-Fortuny, R., Gimeno-Añó, V., and Martín-Belloso, O. 2007. Lycopene, vitamin C, and antioxidant capacity of

tomato juice as affected by high- intensity pulsed electric fields critical parameters. *J. Agric. Food Chem*. 55(22): 9036-9042.

Odriozola-Serrano, I., Soliva-Fortuny, R., Gimeno-A~no´, V., and Marti´n-Belloso, O. 2008. Kinetic study of anthocyanins, vitamin C, and antioxidant capacity in strawberry juices treated by high-intensity pulsed electric fields. *J. Agric. Food Chem*. 56 (18): 8387–8393.

Oms-Oliu, G., Martín-Belloso, O., and Soliva-Fortuny, R. 2010.Pulsed light treatments for food preservation- A Review. *Food Bioprocess Technol.* 3: 13-23.

Orlowska, M., Koutchma, T., Grapperhaus, M., Gallagher, J., Schaefer, R. and Defelice, C. 2013.Continuous and pulsed ultraviolet light for non-thermal treatment of liquid foods. Part 1: effects on quality of fructose solution, apple juice and milk. *Food Bioproc.Tech*. 6(6): 1580-1592.

Ozer, N.P. and Demirci, A. 2006. Inactivation of *Escherichia coli* O157:H7 and *Listeria monocytogenes* inoculated on raw salmon fillets by pulsed UV-light treatment.*Int. J. Food Sci. Technol.* 41: 354-360.

Pai, S.T. and Zhang, Q. 1995.*In Introduction to high power pulse technology*.World Scientific Publishing, Singapore, 255p.

Pala, C.U. and Toklucu, A.K. 2013.Effects of UV-C light processing on some quality characteristics of grape juices. *Food Bioprocess Technol*. 6: 719-72.

Palaniappan,S.,Sastry, S.K., and Richter, E.R.1990.Effects of electricity on microorganisms: a review. *J. Food Process. Preserv.* 14: 393–414.

Palaniappan, S. and Sastry, S. K. 1991. Electrical conductivity of selected juices: influences of temperature, solids content, applied voltage, and particle size. *J. Food Process Eng*. 14:247-260.

Palaniappan, S., Sastry, S.K., and Richter, E.R. 1992.Effects of electro conductive heat treatment and electrical pretreatment on thermal death kinetics of selected microorganisms. *Biotechnol.Bioeng.* 39: 225–232.

Palaniappan, S. andSastry, S.K. 2002.Ohmic heating. In: Juneja V.K. and Sofos J.N. (eds), *Control of food borne microorganisms*. Marcel Dekker, New York, pp. 451-460.

Palgan, I., Caminiti, A., Munoz, A., Noci, F., Whyte, P., and Morgan, D. J. 2011.Effectiveness of height intensity light pulses (HILP) treatments for the control of *Escherichia coli* and *Listeria innocua* in apple juice, orange juice and milk. *Food Microbiol*. 28: 14-20.

Pandhare, G.R., Satwase, A.N., Jaju, R.H., and Awalgaonkar, G.S. 2018. Effect of natural preservatives on pineapple juice, *J. Pharmacognosy Phytochem*.*7*(4): 746–750.

Park, I. K. and Kang, D. H. 2013. Effect of electropermeabilization by ohmic heating for inactivation of *Escherichia coli* O157: H7, *Salmonella enterica serovar typhimurium* and *Listeria monocytogenes* in buffered peptone water and apple juice. *Appl. Environ. Microbiol*. 79 (23): 7122-7129.

Park, S. H., Balasubramaniam, V. M., Sastry, S. K., and Lee, J. Y. 2013. Pressure ohmic thermal sterilization: A feasible approach for the inactivation of *Bacillus amyloliquefaciens* and *Geobacillus stearothermophilus* spores. *Innov. Food Sci. Emerg. Technol.* 19: 115-123.

Park, I. K., Ha, J. W., and Kang, D. H. 2017. Investigation of optimum ohmic heating conditions for inactivation of *Escherichia coli* O157:H7, *Salmonella enterica serovar Typhimurium* and *Listeria monocytogenes* in apple juice. *BMC Microbiol*.17: 117.

Parmar, P.R., Tripathi, S., Tiwari, S., and Singh, R. 2016. Design, fabrication, and performance evaluation of ohmic heater, *Int.J. Res. Sci. Technol.* 5(6):59-69.

Paskeviciute, E., Buchovec, I., and Luksiene, Z. 2011. High-power pulsed light for decontamination of chicken from food pathogens: A study on antimicrobial efficiency and organoleptic properties. *J. Food Saf.* 31: 61-68.

Pataro, G., Munoz, A., Palgan, I., Noci, F., Ferrari, G., and Lyng, J. G. 2011. Bacterial inactivation in fruit juices using a continuous flow pulsed light (PL) system. *Food Res. Int.* 44: 1642-1648.

Paul, R. and Ghosh, U. 2012. Effect of thermal treatment on ascorbic acid content of pomegranate juice.*Indian. J. Biotechnol*.11:309-313.

Pereira, R. N. and Vicente, A. A. 2010.Environmental impact of novel thermal and non-thermal technologies in food processing. *Food Res. Int*. 43: 1936-1943.

Pevere, A., G. Guibad, E. van Hullenbusch, P. Lens and M. Baudu. 2006. Viscosity evolution of anaerobic granular sludgee. *Biochem. Engg. J.* 27: 315-322.

Piyapong, P. and Naruemon, L. 2017. Cashew Apple Juice: Contents and Effects on Health. *Nutri. Food Sci. Int J.* 4(1): 555-629.

Pollock, A. M., Singh, A. P., Ramaswamy, H. S., and Ngadi, M. 2017. Pulsed light destruction kinetics of L. *monocytogenes*. *LWT-Food Sci. Technol*. 84: 114-121 (in press).

Poojitha, P.and Athmaselvi, K. A. 2018. Influence of sucrose concentration on electric conductivity of banana pulp during ohmic heating. *Food Sci Technol Int*. 24 (8): 664-672.

Praporscic, I., Ghnimi, S., and Vorobiev, E. 2005. Enhancement of pressing of sugar beet cuts by combined ohmic heating and pulsed electric field treatment. *J. Food Process. Preserv.*29: 378-389.

Preetha, P., Varadharaju, N., Kennedy, Z.J., Malathi, D., and Shridar, B. 2016a. Non- thermal Inactivation of *Escherichia coli* in Pineapple juice by Pulsed Light Treatment. *Int. J. Food Ferment. Technol.*6(1): 57-65.

Preetha, P., Varadharaju, N., John, Z., Malathi, D., and Shridar, B. 2016b.Evaluation of non-thermal process for decontamination of orange juice using a pulsed light system.*Int. J. food ferment. technol*. 5 (2): 121 -128.

Prasertsri, Piyapong,and Leelayuwat, N. 2017. Cashew apple juice: Contents and effects on health. *Nutri. Food Sci.* 4 (1): 1-3. .

Prescott, S. C. 1927. The treatment of milk by an electrical method.*Am. J. Public Health* 17: 221-223.

Purvis, A.C. 1983. Effect of film thickness and storage temperature on water loss and internal quality of sealed packed grape fruit. *J. Am. Soc. Horti. Sci*. 108: 562- 566.

Qualls, R.G. and Johnson, J.D.1983.Bioassay and dose measurement in UV disinfection.*Appl. and Environ. Microbiol*. 45(3):872-877.

Queiroz, C., Moreira, C.F.F., Lavinas, F.C., Lopes, M.L.M., Fialho, E., and Valente- Mesquita, V.L. 2010.Effect of high hydrostatic pressure on phenolic compounds, ascorbic acid and antioxidant activity in cashew apple juice.*High. Pr. Res.* 30(4): 507-513.

Queiroz, C., da Silva, A.J.R., Lopes, M.L.M., Fialho, E., and Valente-Mesquita, V.L. 2011. Polyphenol oxidase activity, phenolic acid composition and browning in cashew apple (*Anacardium occidentale* L.) after processing.*Food Chem.* 125: 128-132.

Rajkovic, A., Tomasevic, I., De Meulenaer, B., andDevlieghere, F. 2017. The effect of pulsed UV light on *Escherichia coli* O157: H7, *Listeria monocytogenes, Salmonella Typhimurium, Staphylococcus aureus* and *Staphylococcal enterotoxin* A on sliced fermented salami and its chemical quality. *Food Control.*73: 829-837.

Ranganna, S. 1991. *Handbook of analysis and quality control for fruit and vegetable products* (1st Ed.). Tata McGraw-Hill Education, pp.920-933.

Ranganna, S. 1995. *Hand book of analysis and quality control for fruits and vegetable products* (2nd Ed.). Tata McGraw- Hill Publishing Company Limited. New Delhi-2, 321p.

Rao, D.G. 2012. *Fundamentals of Food Engineering* (Eastern Economy Edition). PHI Learning Private Limited, pp. 126-142.

Rattanathanalerk, M., Chiewchan, N., and Srichumpoung, W. 2005.Effect of thermal processing on the quality loss of pineapple juice. *J. Food Eng.* 66(2): 259-265.

Ravi-Shankar, Kaushik, U., and Shayeeb, A.B. 2014.Intense pulsed light in food technology-A review. *Int. J. Curr. Eng. Technol.* 4: 2314-2327.

Reznick, D. 1996. Ohmic heating of fluid foods: Ohmic heating for thermal processing of foods: Government, industry and academic perspectives. *Food Technol*. 50(5): 250-251.

Rivas, A., Rodrigo, D., Martınez, A., Barbosa-Canovas, G.V. and Ro- drigo, M. 2006.Effect of PEF and heat pasteurization on the physical–chemical characteristics of blended orange and carrot juice. *LWT- Food Sci. Technol.* 39: 1163–1170.

Roberts, P. and Hope, A. 2003. Virus inactivation by high intensity broad spectrum pulsed light. *J. Virol. Methods*, 110: 61-65.

Rocha-Andrade, Rodrigues-Sousa, Antonio, F., Pinto-Abreu, Ximenes, F.S. and Garruti, D.S. 2018.Changes in cashew apple Juice flavour after tangential microfiltration process. *Annals Nutr.Food Sci.* 2(4):1029

Rocha M. V. P., Oliveira A. H. S., Souza M. C. M., and Goncalaves L. R. B. 2006.Natural cashew apple juice as fermentation medium for biosurfactant production by *Acinetobactercalcoaceticus*.*World J. Microbiol.Biotechnol.* 26: 875-883.

Rodríguez, Ó., Gomes, W.F., Rodrigues, S., and Fernandes, F.A.N. 2017. Effect of indirect cold plasma treatment on cashew apple juice (*Anacardiumoccidentale*L.). *LWT-Food Sci. Technol.*84: 457-463.

Rosen, J. and Gothard, L.Q. 2010.Encyclopedia of physical science.Vol. 1. New York: Infobase Publishing. Sanchez-Moreno, pp. 2302-2315.

Rowan, N.J., Macgregor,S.J., Anderson,J.G., Fouracre, R.A., Mcilvaney, L., and Farish, O. 1999. Pulsed light Inactivation of Food- related Microorganisms. *Appl. Environ. Microbiol.* 65(3): 1312-1315.

Ryang, J. H., Kim, N. H., Lee, B. S., Kim, C. T., Lee, S. H., Hwang, I. G., and Rhee, M. S. 2015. Inactivation of Bacillus cereus spores in a tsuyu sauce using continuous ohmic heating with five sequential elbow type electrodes. *J. Appl. Microbiol*. 120: 175-184.

Sadasivam, S. and Manickam, C. 1992.*Biochemical methods for agricultural science* (1st Ed.).Wiley Eastern Limited, New Delhi, 532p.

Sangong, H. G., Park, S. H., Choi, Y. J., Ryu, S., and Kang, D. H. 2011. Inactivation of *Escherichia coli* O157: H7, *Salmonella typhimurium* and *Listeria monocytogenes* in orange and tomato juice using ohmic heating. *J. Food Prot*. 74(6): 899-904.

Saikiran, K. C.H.S., Lavanya, M., and Venkatachalapathy, N. 2016. Different pulsed light systems and their application in foods: a review. *Int. J Sci. Environ. Technol.* 5(3): 1463 – 1476

Sakr,M. and Liu, S. 2014.A comprehensive review on applications of ohmic heating (OH).*Renew. Sustain. Energy Rev*. 39: 262-269.

Salvador, J., García, G., Fernando, D., Hierro, E., Hospital, X.F., Espinosa, I., and Fernández, M. 2018.Characterization of damage on *Listeria innocua* surviving to pulsed light: Effect on growth , DNA and proteome. *Int. J. Food Microbiol.*284: 63-72.

Samaranayake, C.P. and Sastry, S.K. 2005.Electrodeand pH effects on electrochemical reactions during Ohmic heating. *J. Electroanal. Chem.* 577:125-35.

Samaranayake, C. P., Sastry, S. K., and Zhang, H. 2005. Pulsed ohmic heating-a novel technique for minimization of electrochemical reactions during processing. *J. Food Sci.* 70(8): 460-465

Sambrook, J., Fritch, E.F., and Maniatis, T. 1989. *Molecular cloning volume2*.Cold spring harbour laboratory press, New York, 623p.

Samprovalaki, K., Bakalis, S., and Fryer, P. J. 2007. Ohmic heating: models and measurements. In: Yanniotis, S. and Sunden, B. (eds), *Heat transfer in food*

processing,pp. 159-186, WIT press, Southampton [on-line]. Available: doi:10.2495/978-1-85312-932-2/05 [06 October 2018].

Sana, F., Nitya, S., Surendra, P.S., Alok, J., and Arvind, K. 2016.Fuzzy analysis of sensory data for ranking of beetroot candy. *Int. J. Food Eng.* 2(1): 26-30.

Sang-Soon, K., Sang-Hyun, P., Soo-Hwan, K., Dong-Hyun, K. 2019. Synergistic effect of ohmic heating and UV-C irradiation for inactivation of *Escherichia coli* O157:H7, *Salmonella Typhimurium* and *Listeria monocytogenes* in buffered peptone water and tomato juice. *Food Control*. 102: 69-75.

Saradhuldhat, Paull, P., and Robert, 2007. Pineapple organic acid metabolism and accumulation during fruit development. *Sci. Hortic.* 112: 297-303.

Sarang, S., Sastry, S. K., and Knipe, L. 2008. Electrical conductivity of fruits and meats during ohmic heating, *J. Food Eng.*87: 351–356.

Sastry, S. K. and Li, Q. 1996. Modeling the ohmic heating of foods. *Food Technol*. 50(5): 246-248.

Sastry, S. K. and Salengke, S.1998. Ohmic heating of solid-liquid mixtures: A comparison of mathematical models under worst-case heating conditions. *J. Food Process Eng*. 21: 441-458.

Sauer, A. and Moraru, C. 2009. Inactivation of *Escherichia coli* ATCC 25922 and *Escherichia coli* O157:H7 in apple juice and apple cider, using pulsed light treatment. *J. Food Prot.* 72: 937-944.

Sawamura, M., Takemoto, K., Matsuzaki, Y., Ukeda, H., and Kusunose, H. 1994.Identification of two degradation products from aqueous dehydroascorbic acid. *J Agric Food Chem*. 42:1200–1203.

Saxena J., Makroo, H. A., and Srivastava, B. 2016.Optimization of time-electric field combination for PPO inactivation in sugarcane juice by ohmic heating and its shelf life assessment. *LWT-Food Sci. Technol*. 71: 329-338.

Sean.Y. D. and Prince, M. V. 2015.Development and evaluation of anultraviolet radiation assisted with ohmic heating system for preservation of pineapple juice. *Trends Biosci*.8(16): 4221-4226.

Sean, Y. D., Prince, M. V., Sudheer, K. P. and Sariga, S. 2016. Storage studies of ultraviolet radiation assisted ohmic heating of pineapple juice. *Agr. Eng. Today*. 40(4): 29-35.

Setlow, R.B. and Carrier, W.L. 1966.Pyrimidine dimmers in ultraviolet-irradiated DNAs. *J. Mol. Biol.* 17: 237-254.

Shah, N.A.D., Shamsudin, R., Rahman, R.A.and Adzahan, N.M. 2016. *Fr*uit juice production using ultraviolet pasteurisation: a review. *Bev.* 2: 22.

Shaik, K. B. 2012. Storage stability of cashew apple juice-use of chemical preservatives, *Am.J. Food Technol*. 10(4): 117-123.

Shamsudin, R., Ramli, W., Daud, W., Takrif, M.S., Hassan, O., and Ilicali, C. 2009. Rheological properties of Josapine pineapple juice at different stages of maturity. *Int. J. Food Sci. Technol.* 757–762.

Shamsudin, R., Su, C., Mohd, N., Ramli, W., and Daud, W. 2013.Rheological properties of ultraviolet-irradiated and thermally pasteurized Yankee pineapple juice. *J. Food Eng.* 116(2): 548-553

Shamsudin, R., Noranizan, M.A., Yap, P.Y., and Mansor, A. 2014.Effect of repetitive ultraviolet irradiation on the physico-chemical properties and microbial stability of pineapple juice. *Innov. Food Sci. Emerg. Technol.* 10: 166-171.

. Sharifi-Yazdi, M.K., and Darghahi, H. 2006. Inactivation of pathogenic bacteria using pulsed UV-light and its application in water disinfection. *Acta Med Iran* 44:305–308

Sharma, R.R. and Demirci, A. 2003. Inactivation of *Escherichia coli* O157:H7 on inoculated alfalfa seeds with pulsed ultraviolet light and response surface modelling. *J. Food Sci*. 68: 1448-1453.

Shiv-Kumar, 2015. Role of enzymes in fruit juice processing and its quality enhancement, *Adv. Appl. Sci. Res*. 6(6): 114-124.

Silva, L. G. Maia, P. Sousa, R. Figueiredo, M. Afonso , M. Gonzaga, Gomes, C., and Figueiredo, E. 2014. Effect of heat treatment on rheological properties of mixed nectars based on cashew apple, mango and acerola pulps, *Acta Alimentaria.*43(1): 19–27.

Sivagurunathan, P., Sivasankari, S., and Muthukkaruppan, S.M.J. 2010. Characterisation of cashew apple (*Anacardium occidentale* L.) fruits collected from Ariyalur District. *Biosci. Res.* 1:101–107.

Singh, R. P. and Heldman, D. R. 2014.Introduction to food engineering (5^{th} Ed.).Academic Press Incorporation, Orlando, Florida, 892p.

Sitzmann, W. 1995. High-voltage pulsed techniques for food preservation, In Gould W (ed.*), New methods of food preservation*. Blackie Academic and Professional, London, United Kingdom, pp. 236–251.

Slieman, T.A. and Nicholson, W.L. 2000. Artificial and solar UV radiation induces strand breaks and cyclobutane dimers in *Bacillus subtilis* spore DNA. *Appl. Environ.Microbiol.* 66: 199-205.

Slongo, A. P. and de Aragao, G. M. F. 2008.Behaviour of *Neosartorya fisheriascopores* in pineapple juice. *Bol. Cent. Pesqui.Process.Aliment.* 26: 51- 60.

Sm, H., Mk, A., Massudi, R., and Elikaei, A. 2011. Pulsed UV laser light on *Escherichia coli* and *Saccharomyces cerevisiae* suspended in non-alcoholic beer, *Iranian J. Microbiol.* 3(1): 31–35.

Smith, K., Mittal, G.S., and Griffith, M.W. 2002.Pasteurization of milk using pulsed electric field and antimicrobials. *J. Food Sci.* 67: 2304-2308.

Somboonsilp, P., Tia, S., and Yoovidhya, T. 2011. Development of a small-scale ohmic pasteurization system for flavor retention in fruit juice production. In: *The 12th ASEAN Food Conference 2011* [On-line]. Available:

http://technology.kku.ac.th/foodtechnology/AFC2011/index.asp_files/Full_P ap er/Oral/OB_009.pdf [15 September 2019].

Somogyi, L. P., Ramaswamy H.S., and Y.H. Hui. 1996. Biology, principles and applications In: *Processing fruits: science and technology* (Vol. 1). Lancaster, Pa.:Technomic Publishing Co. Inc., pp. 95-113.

Sonenshein, A.L. 2003. Killing of Bacillus spores by high-intensity ultraviolet light. *Sterilization and decontamination using high- energy light*. Xenon Corporation, Woburn, Mass. pp. 15–19.

Stanel, J. and Zitny, R. 2010. Milk fouling at direct ohmic heating.*J. Food Eng*. 99: 437-444.

Stevens, C., Khan, V. A., Lu, J.Y., Wilson, C.L., Chalutz, E., and Droby, S. 1999. Induced resistance of sweet potato to *Fusarium* root rot by UV-C hormesis. *Crop Prot.* 18: 463-470.

Sun, H., Kawamura, S., Himoto, J., Itoh, K., Wada, T., and Kimura, T. 2008.Effects of ohmic heating on microbial counts and denaturation of proteins in milk. *Food Sci. Technol. Res.* 14: 117–123.

Supratomo, S., Laga, A., Tahir, M., Mochtar, A. A., and Salengk, S. 2019. Design and performance test of ohmic-assisted cocoa fermentation apparatus. *ARPN J. Eng. Appl. Sci*. 14(8): 1515-1523.

Swanson, H. L, 1989. Effects of Subvocal Suppression on Learning Disabled Readers' Sentence Comprehension [on-line], Available: https://journals.sagepub.com/doi/abs/10.2307/1510207[20 August 2020].

Takeshita, K., Shibato, J., Sameshima, T.,Fukunaga, S.,Isobe, S., Arihara, K., and Itoh, M.2003. Damage of yeast cells induced by pulsed light irradiation. *Int. J. Food Microbiol.* 85(1-2): 151-158.

Talasila, U., Rao, R., and Beebi, K. 2011. Preservation and shelf life extension of cashew apple juice. *Int. J. Food Saf.*13: 275-280.

Talasila, U., Vechalapu, R.R., and Shaik, K.B. 2012.Clarification, preservation, and shelf life evaluation of cashew apple juice. *Food Sci. Biotechnol.* 21: 709-714.

Talasila, U., and Shaik, K. B. 2013. Quality, spoilage and preservation of cashew apple juice : a review. *J Food Sci Technol*. Available: https://doi.org/10.1007/s13197-013-0931-0[24 August 2017].

Tasnim, F., Hossain, A., Nusrath, S., Hossain, M.K., Lopa, D., and Formuzul, H.K.M. 2010. An Assessment of Microbiological Quality of Some Commercially Packed and Fresh Fruit Juice Available in Dhaka City: A comparative Study. Institute of Food Science and Technology, Bangladesh. *Malaysian J. Nutr.*16(3): 431-438.

Taussig, S.J., and Batkin, S. 1988. Bromelain, the enzyme complex of pineapple (*Ananascomosus*) and its clinical application- An update. *J. Ethnopharmacol.* 22(2): 191-203.

Teja, C. K., Sanganamoni, S., and Prabhakar, B. 2017. Effect of UV – C light treatment on physicochemical and bioactive compounds in apple and pineapple juices, 6(6): 2321–2333.

Thangalakshmi, S., Tadakod, M., Rani, S., and Singh, R. 2018. Effect of ohmic heating on quality parameters of sapota juice. *J Emerg. Technol. Innov. Res.* 5(8): 390-394.

Tikekar, R. V., Anantheswaran, R. C., and La-Borde, L. F. 2011a.Ascorbic acid degradation in a model apple juice system and in apple juice during ultraviolet processing and storage. *J Food Sci.* 76: 62-71.

Tikekar, R., Anantheswaran, R., Laborde, R., and Luke, F.L. 2011b. Ultraviolet-Induced Oxidation of Ascorbic Acid in a Model Juice System: Identification of Degradation Products. *J. Agric. Food chem.* 59: 8244-8

Tiwari, B. K., Muthukumarappan, K., O'Donnell, C.P., and Cullen, P.J. 2009. Inactivation kinetics of pectinmethylesterase and cloud retention in sonicated orange juice. *Innov. Food Sci. Emerg. Technol*. 10: 166-171.

Tran, M.T.T. and Farid, M. 2004. Ultraviolet treatment of orange juice. *Innov .Food Sci. Emerg. Technol*. 5:495-502.

Trevisan, M.T.S., Pfundstein, B., Haubner, R., Wurtele, G., Spiegelhalder, B., Bartsch, H., and Owen, R.W. 2006.Characterization of alkyl phenols in cashew (*Anacardium occidentale*) products and assay of their antioxidant capacity. *Food Chem. Toxicol.* 44:188–197.

Trevisan, M.T.S., Pfundstein, B., Haubner, R., Wurtele, G., Spiegelhalder, B., Bartsch, H., and Owen, R.W. 2006.Characterization of alkyl phenols in cashew (*Anacardiumoccidentale*) products and assay of their antioxidant capacity. *Food Chem. Toxicol.* 44: 188-197.

Tumpanuvatr, T. and Jittanit, W. 2012.The temperature prediction of some botanical beverages, concentrated juices and purees of orange and pineapple during ohmic heating. *J. Food Eng.* 113(2): 226-233.

Turtoi, M. and Nicolau, A. 2007.Intense light pulse treatment as alternative method for mouldspores destruction on paper–polyethylene packaging material. *J. Food Eng.* 83: 47-53.

Uemura, K. and Isobe, S. 2003. Developing a new apparatus for inactivating *Bacillus subtilis* spore in orange juice with a high electric field AC under pressurised conditions. *J. Food Eng*. 56:325–329.

Unluturk, S., Arastoopour, H., and Koutchma, T. 2004.Modelling of UV dose distribution in a thin-film UV reactor for processing of apple cider. *J. Food Eng.* 65: 125–136.

USA-FDA [United States of America, Food and Drug Administration]. 2000. *Center for Food Safety and Applied Nutrition Kinetics of micro- bial inactivation for alternative food processing technologies: ohmic and inductive heating*. Available: http://www.cfsan.fda.gov/wcomm/IFT-OHM.html. [09 August 2019].

Uslu, G., Demirci, A., and Regan, J. M. 2016. Disinfection of synthetic and real municipal wastewater effluent by flow-through pulsed UV-light treatment system, *J Water Process Eng.*10: 89–97.

Vandresen, S., Quadri, M.G.N., Souza, J.A.R.D., and Hortza, D., 2009.Temperature effect on the rheological behaviour of carrot juices. *J. Food Eng*. 92: 269–274.

Varghese, K. S., Pandey, M. C., Radhakrishna, K., and Bawa, A. S. 2014. Technology, applications and modelling of ohmic heating: A review. *J. Food Sci. Technol*. 51: 2304-2317.

Walkling-Ribeiro, M., Noci, F., Cronin, D.A., Riener, J., Lyng, J.G., and Morgan, D.J. 2008.Reduction of *Staphylococcus aureus* and quality changes in apple juice processed by ultraviolet irradiation, pre-heating and pulsed electric fields. *J. Food Eng*. 89(3): 267–273.

Wang,W.C. and Sastry, S.K. 2002. Effects of moderate electrothermal treatments on juice yield from cellular tissue. *Innov. Food Sci. Emerg. Technol.* 3: 371-377.

Weaver, J.C. and Chizmadzhev, Y.A. 1996. Theory of electroporation: a review. *Bioelectrochem Bioenergy* 41:135–160.

Wekhof, A. 2000. Disinfection with flash lamps.*PDA J. Pharma. Sci. Technol.* 54(3): 264-276.

Wekhof, A., F. J. Trompeter and O. Franken. 2001. Pulse UV disintegration (PUVD): a new sterilisation mechanism for packaging and broad medical-hospital applications. Proceedings of the first international conference on ultraviolet technologies.Washington D.C. USA.

Wekhof, A. 2003. Sterilization of packaged pharmaceutical solutions, packaging and surgical tools with pulsed UV light.*CDROM Proceedings of the 2nd International Congress on Ultraviolet Technologies*, July 2003, Vienna, Austria, International Ultraviolet Association. 295p.

Wood, M. J. and Moellering, J. 2003. Microbial resistance: Bacteria and more.

Clinical Infectious Diseases, 36: S2–S3.

Xenon Corp. 2005.AN-104 application note. Achieving faster cure time with pulsed ultraviolet [online]. Available:http://www.globalspec.com [09 august 2017].

Yang, W., Chung, S. Y., Ajayi, O., Krishnamurthy, K., Konan, K., and Goodrich-Schneider, R. 2010. Use of pulsed ultraviolet light to reduce the allergenic potency of soybean extracts. *Int. J. Food Eng.* 6(3): 1–2.

Yang, Y., Shen, Tian, S., You, Y., Guo, Z., and Yurong. 2019. Effect of thermal pasteurization and ultraviolet treatment on the quality parameters of not-from- concentrate apple juice from different varieties. *CyTA – J. Food.* 17: 189-198.

Yeom, H.W., Streaker, C.B., Zhang, Q.H., and Min, D.B. 2000. Effect of pulsed electric fields on the quality of orange juice and comparison with heat pasteurization. *J. Agric. Food Chem*. 48: 4597–4605.

Yildiz, H., Bozkurt, H. and Icier, F. 2010. Ohmic and conventional heating of pomegranate juice: effects on rheology, colour, and total phenolics. *Food Sci. Technol. Int.* 15(5): 503-512.

Yoon, S. W., Lee, C. Y. J., Kim, K. M., and Lee, C. H. 2002.Leakage of cellular material from *Saccharomycies cerevisiae* by ohmic heating. *J. Microbiol. Biotechn*. 12: 183-188.

Zapata , A. and Ramirez-Arcos, S. 2015. A comparative study of McFarland turbidity standards and the Densimat photometer to determine bacterial cell density, *Curr.Microbiol.* 70(6): 907-909.

Zareifard, M.R., Ramaswamy, R.S., Trigui, M., and Marcotte, M. 2003.Ohmic heating behaviour and electrical conductivity of two-phase food systems. *Innov. Food Sci. Emerging. Technol*. 4: 45-55.

Zell, M., Lyng, G.J., Cronin, D.A., and Morgan, D.J. 2010. Ohmic cooking of whole turkey meat: effect of rapid ohmic heating on selected product parameters. *J. Agric. Food Chem*. 120:724–729.

Zell, M., Lyng, J. G., Morgan, D. J., and Cronin, D. A. 2011.Minimising heat losses during batch ohmic heating of solid food. *Food Bioprod.Process.* 89: 128-134.

Zheng, H. and Lu, H. 2011. Use of kinetic, Weibull and PLSR models to predict the retention of ascorbic acid, total phenols and antioxidant activity during storage of pasteurized pineapple juice. *LWT - Food Sci. Technol.*44(5): 1273-1281.

Appendices

Appendix A.1

ANOVA for electrical conductivity during ohmic heating of pineapple juice at different voltage gradients

Voltage gradient	Model	Sum of Squares	df	Mean Square	F	Sig.
7.5V/cm	Regression	.093	1	.093	1.109E4	.000[a]
	Residual	.000	18	.000		
	Total	.093	19			
10 V/cm	Regression	.105	1	.105	3.603E3	.000[a]
	Residual	.001	18	.000		
	Total	.106	19			
12.5 V/cm	Regression	.120	1	.120	5.017E3	.000[a]
	Residual	.000	18	.000		
	Total	.120	19			
15 V/cm	Regression	.156	1	.156	933.891	.000[a]
	Residual	.003	18	.000		
	Total	.159	19			
17 V/cm	Regression	.164	1	.164	4.654E3	.000[a]
	Residual	.001	18	.000		
	Total	.165	19			

Correlation Coefficients of temperature during ohmic heating of pineapple juice

Dependent variable		Unstandardised Coefficients		Standardised Coefficients		
	Model	B	Std. Error	Beta	t	Sig.
7.5V/cm	(Constant)	-.031	.002		-13.017	.000
	temperature	.006	.000	.999	105.323	.000
10 V/cm	(Constant)	-.003	.004		-.581	.568
	temperature	.006	.000	.998	60.027	.000
12.5 V/cm	(Constant)	.026	.004		6.483	.000
	temperature	.007	.000	.998	70.832	.000
15 V/cm	(Constant)	.027	.011		2.540	.021
	temperature	.008	.000	.991	30.560	.000
17 V/cm	(Constant)	.076	.005		15.548	.000
	temperature	.008	.000	.998	68.222	.000

Appendix. A.2

ANOVA for electrical conductivity during ohmic heating of cashew apple juice at different voltage gradients

Voltage gradient	Model	Sum of Squares	df	Mean Square	F	Sig.
7.5V/cm	Regression	0.292	1	0.292	1.899E4	.000[a]
	Residual	0.000	18	0.000		
	Total	0.293	19			
10 V/cm	Regression	0.262	1	0.262	2.950E3	.000[a]
	Residual	0.002	18	0.000		
	Total	0.263	19			
12.5 V/cm	Regression	0.392	1	0.392	1.090E4	.000[a]
	Residual	0.001	18	0.000		
	Total	0.393	19			
15 V/cm	Regression	0.500	1	0.500	1.048E4	.000[a]
	Residual	0.001	18	0.000		
	Total	0.501	19			
17 V/cm	Regression	0.746	1	0.746	1.097E4	.000[a]
	Residual	0.001	18	0.000		
	Total	0.747	19			

Correlation Coefficients of temperature during ohmic heating of cashew apple juice

Dependent variable		Unstandardised Coefficients		Standardised Coefficients		
	Model	B	Std. Error	Beta	t	Sig.
7.5V/cm	(Constant)	0.151	0.003		46.572	.000
	temperature	0.010	0.000	1.000	137.804	.000
10 V/cm	(Constant)	0.216	0.008		27.764	.000
	temperature	0.010	0.000	0.997	54.311	.000
12.5 V/cm	(Constant)	0.203	0.005		40.898	.000
	temperature	0.012	0.000	0.999	104.397	.000
15 V/cm	(Constant)	0.207	0.006		36.324	.000
	temperature	0.014	0.000	0.999	102.348	.000
17 V/cm	(Constant)	0.169	0.007		24.804	.000
	temperature	0.017	0.000	0.999	104.720	.000

Appendix A. 3 (a)Variation of electrical conductivity of Pineapple juice with different voltage gradients

Temperature	Voltage gradient (V/cm)				
(°C)	7.5	10	12.5	15	17
22	0.105324	0.145684	0.173761	0.214065	0.26144
24	0.11256	0.151403	0.189908	0.220095	0.274168
26	0.12261	0.163443	0.201476	0.23919	0.283456
28	0.129846	0.177289	0.215454	0.248838	0.292744
30	0.143112	0.185115	0.228468	0.258687	0.308568
32	0.159996	0.196252	0.236903	0.269742	0.321124
34	0.169644	0.210098	0.25305	0.280194	0.332648
36	0.183714	0.224546	0.26992	0.291852	0.351912
38	0.192558	0.227556	0.28679	0.308736	0.371348
40	0.200598	0.238994	0.297153	0.318284	0.390096
42	0.216276	0.253141	0.306311	0.329841	0.403684
44	0.227934	0.268793	0.319325	0.342303	0.423636
46	0.23919	0.284445	0.329688	0.368433	0.441868
48	0.250848	0.299796	0.345112	0.385116	0.454424
50	0.264516	0.311234	0.359331	0.406623	0.473688
52	0.275772	0.325682	0.367766	0.423105	0.485212
54	0.286626	0.341635	0.381744	0.449637	0.499144
56	0.304716	0.357588	0.40006	0.462702	0.511184
58	0.315168	0.366317	0.427775	0.487425	0.538704
60	0.323208	0.37625	0.435969	0.506842	0.55212

Appendix A.3 (b)

Variation of electrical conductivity of cashew apple juice with different voltage gradients

Temperature	Voltage gradient (V/cm)				
(°C)	7.5	10	12.5	15	17
22	0.384312	0.44548	0.46031	0.51912	0.56244
24	0.400392	0.461734	0.48682	0.539926	0.576888
26	0.4221	0.479493	0.51574	0.56547	0.596324
28	0.443004	0.497252	0.542973	0.589984	0.634164
30	0.46029	0.512302	0.570447	0.613468	0.666156
32	0.484008	0.531265	0.595752	0.639218	0.697976
34	0.510138	0.558656	0.618888	0.66023	0.734354
36	0.529434	0.571298	0.646603	0.699164	0.765228
38	0.554358	0.588154	0.669498	0.721412	0.802208
40	0.567624	0.601097	0.686609	0.758698	0.8342
42	0.590538	0.62006	0.709022	0.786096	0.86516
44	0.613854	0.642334	0.73746	0.811022	0.905924
46	0.632346	0.664608	0.763729	0.84596	0.934648
48	0.658476	0.685979	0.789516	0.875294	0.9761
50	0.67737	0.701932	0.816749	0.900014	1.004308
52	0.703098	0.729022	0.84109	0.922674	1.03888
54	0.721188	0.751898	0.861816	0.940884	1.076376
56	0.732846	0.772968	0.880614	0.965934	1.116796
58	0.758172	0.805175	0.899412	0.997658	1.149476
60	0.77184	0.83377	0.91821	1.037004	1.16788

Appendix 4 (a)

ANOVA for Response Surface Quadratic Model for pH of pineapple juice during ohmic heating

Analysis of variance table [Partial sum of squares - Type III]						
Source	Sum of Squares	df	Mean Square	F Value	p-value Prob > F	
Model	1.0543	9	0.117152059	43.959	< 0.0001	significant
A-voltage gradient	0.525	1	0.5253125	197.11	< 0.0001	
B-Temperature	0.0002	1	0.0002	0.0750469	0.7920	
C-Time	0.292	1	0.2926125	109.79	< 0.0001	
AB	0.000625	1	0.000625	0.234	0.6430	
AC	0.0025	1	0.0025	0.9380863	0.3650	
BC	2.5E-05	1	2.5E-05	0.00938	0.9256	
A^2	0.1417	1	0.141777895	53.199	0.0002	
B^2	0.001	1	0.001077895	0.4044	0.5450	
C^2	0.075	1	0.0750	28.157	0.0011	
Residual	0.018655	7	0.002665			
Lack of Fit	0.009	3	0.003325	1.532	0.3361	not significant
Pure Error	0.00868	4	0.00217			
Cor Total	1.073	16				
Std. Dev.	0.0516		R-Squared		0.982	
Mean	11.15521		Adj R-Squared		0.960	
C.V. %	0.462		Pred R-Squared		0.838	
PRESS	0.1731		Adeq precision		22.60	

Appendix A. 4 (b)

ANOVA for Response Surface Quadratic Model for pH of cashew apple juices during ohmic heating

Analysis of variance table [Partial sum of squares - Type III]						
Source	Sum of Squares	df	Mean Square	F Value	p-value Prob > F	
Model	0.023	9	0.003	8.364	0.005	significant
A-voltage gradient	0.0128	1	0.0128	41.100	0.000	
B-Process temperature	5E-05	1	5E-05	0.160	0.7006	
C-Holding time	0.008	1	0.008	27.133	0.0012	
AB	0.0001	1	0.0001	0.321	0.589	
AC	0.0004	1	0.0004	1.284	0.294	
BC	0	1	0	0	1.000	
A^2	0.0015	1	0.001	4.627	0.069	
B^2	0	1	0.000	0.571	0.475	
C^2	5.15E-05	1	5.15E-05	0.165	0.696	
Residual	0.002	7	0.0003			
Lack of Fit	0.0007	3	0	0.631	0.632	not significant
Pure Error	0.00148	4	0.00037			
Cor Total	0.025	16				
Std. Dev.	0.018		R-Squared	0.915		
Mean	4.275		Adj R-Squared	0.806		
C.V. %	0.4135		Pred R-Squared	0.7736		
PRESS	0.0135125		Adeq Precision	10.713		

Appendix A. 5 (a)

ANOVA for Response Surface Quadratic Model for TSS of pineapple juices during ohmic heating

Analysis of variance table [Partial sum of squares - Type III]						
Source	Sum of Squares	df	Mean Square	F Value	p-value Prob > F	
Model	0.650693235	9	0.07229925	18.958	0.0004	significant
A-voltage gradient	0.32	1	0.32	83.910	< 0.0001	
B-Temperature	0.099	1	0.0990	25.963	0.0014	
C-Time	0.099	1	0.099	25.963	0.0014	
AB	0.021	1	0.021	5.5130	0.0512	
AC	0.021	1	0.021	5.5132	0.0512	
BC	0.0225	1	0.0225	5.899	0.0455	
A^2	0.0039	1	0.003	1.027	0.3446	
B^2	0.025	1	0.025	6.717	0.0359	
C^2	0.032	1	0.032	8.550	0.0222	
Residual	0.026695	7	0.0038			
Lack of Fit	0.009375	3	0.003125	0.721	0.5893	not significant
Pure Error	0.01732	4	0.00433			
Cor Total	0.677	16				
Std. Dev.	0.06175412		R-Squared		0.960	
Mean	8.01352941		Adj R-Squared		0.909	
C.V. %	0.77062325		Pred R-Squared		0.838	
PRESS	0.1770625		Adeq Precision		13.354	

Appendix A. 5(b)

ANOVA for Response Surface Quadratic Model for TSS of cashew apple juices during ohmic heating

Analysis of variance table [Partial sum of squares - Type III]						
Source	Sum of Squares	df	Mean Square	F Value	p-value Prob > F	
Model	0.646	9	0.071	25.412	0.0002	significant
A-voltage gradient	0.332	1	0.332	117.443	< 0.0001	
B-Process temperature	0.125	1	0.125	44.203	0.0003	
C-Holding time	0.082	1	0.082	29.001	0.001	
AB	0.021	1	0.021	7.434	0.029	
AC	0.0196	1	0.0196	6.931	0.033	
BC	0.009	1	0.009	3.191	0.117	
A^2	0.008	1	0.008	3.082	0.122	
B^2	0.0194	1	0.019	6.884	0.034	
C^2	0.024	1	0.024	8.487	0.022	
Residual	0.019	7	0.002			
Lack of Fit	0.002	3	0.0008	0.1908	0.897	not significant
Pure Error	0.0173	4	0.00433			
Cor Total	0.666	16				

Std. Dev.	0.0531	R-Squared	0.970
Mean	11.517	Adj R-Squared	0.932
C.V. %	0.461	Pred R-Squared	0.899
PRESS	0.066	Adeq Precision	16.120

Appendix A. 6 (a)

ANOVA for Response Surface Quadratic Model for titrable acidity of pineapple juice during ohmic heating

Analysis of variance table [Partial sum of squares - Type III]						
Source	Sum of Squares	df	Mean Square	F Value	p-value Prob > F	
Model	1.590	9	0.177	109.413	< 0.0001	significant
A-voltage gradient	1.140	1	1.140	705.913	< 0.0001	
B-Temperature	0.002	1	0.002	0.937	0.365	
C-Time	0.285	1	0.285	176.478	< 0.0001	
AB	0.002	1	0.002	1.254	0.300	
AC	0.004	1	0.004	2.616	0.150	
BC	0.003	1	0.003	1.548	0.254	
A^2	0.130	1	0.130	80.759	< 0.0001	
B^2	0.007	1	0.007	4.490	0.072	
C^2	0.025	1	0.025	15.258	0.006	
Residual	0.011	7	0.002			
Lack of Fit	0.006	3	0.002	1.521	0.338	not significant
Pure Error	0.005	4	0.001			
Cor Total	1.602	16				
Std. Dev.	0.040		R-Squared		0.993	
Mean	3.965		Adj R-Squared		0.984	
C.V. %	1.014		Pred R-Squared		0.935	
PRESS	0.105		Adeq Precision		36.743	

Appendix A. 6 (b)

ANOVA for Response Surface Quadratic Model for titrable acidity of cashew apple juice during ohmic heating

Analysis of variance table [Partial sum of squares - Type III]						
Source	Sum of Squares	df	Mean Square	F Value	p-value Prob > F	
Model	7E-06	9	8E-07	1.323	0.364	not significant
A-voltage gradient	1E-06	1	1E-06	1.944	0.206	
B-Process temperature	3E-06	1	3E-06	5.401	0.053	
C-Holding time	5E-07	1	5E-07	0.864	0.384	
AB	1E-06	1	1E-06	1.728	0.230	
AC	3E-07	1	3E-07	0.432	0.532	
BC	3E-07	1	3E-07	0.432	0.532	
A^2	9E-08	1	9E-08	0.164	0.698	
B^2	5E-07	1	5E-07	0.891	0.377	
C^2	4E-08	1	4E-08	0.073	0.795	
Residual	4E-06	7	6E-07			
Lack of Fit	1E-06	3	4E-07	0.595	0.651	not significant
Pure Error	3E-06	4	7E-07			
Cor Total	1E-05	16				
Std. Dev.	8E-04		R-Squared	0.6298		
Mean	4E-01		Adj R-Squared	0.1539		
C.V. %	8E-01		Pred R-Squared	-1.2278		
PRESS	2E-05		Adeq Precision	4.0711		

Appendix A.7 (a)

ANOVA for Response Surface Quadratic Model for ascorbic acid of pineapple juice during ohmic heating

Analysis of variance table [Partial sum of squares - Type III]						
Source	Sum of Squares	df	Mean Square	F Value	p-value Prob > F	
Model	1.601	9	0.178	5.122	0.021	significant
A-voltage gradient	0.218	1	0.218	6.270	0.041	
B-Temperature	0.485	1	0.485	13.966	0.007	
C-Time	0.401	1	0.401	11.531	0.012	
AB	0.194	1	0.194	5.574	0.050	
AC	0.000	1	0.000	0.012	0.918	
BC	0.003	1	0.003	0.087	0.777	
A^2	0.078	1	0.078	2.234	0.179	
B^2	0.165	1	0.165	4.764	0.065	
C^2	0.062	1	0.062	1.797	0.222	
Residual	0.243	7	0.035			
Lack of Fit	0.201	3	0.067	6.364	0.053	not significant
Pure Error	0.042	4	0.011			
Cor Total	1.844	16				
Std. Dev.	0.186		R-Squared	0.868		
Mean	27.904		Adj R-Squared	0.799		
C.V. %	0.668		Pred R-Squared	0.780		
PRESS	3.282		Adeq Precision	7.136		

Appendix A.7(b)

ANOVA for Response Surface Quadratic Model for Ascorbic acid of cashew apple juice during ohmic heating

Analysis of variance table [Partial sum of squares - Type III]						
Source	Sum of Squares	df	Mean Square	F Value	p-value Prob > F	
Model	181.314	9	20.146	10.033	0.003	significant
A-voltage gradient	25.134	1	25.134	12.517	0.010	
B-Process temperature	108.708	1	108.708	54.135	0.000	
C-Holding time	38.413	1	38.413	19.129	0.003	
AB	1.988	1	1.988	0.990	0.353	
AC	4.285	1	4.285	2.134	0.188	
BC	2.356	1	2.356	1.173	0.315	
A^2	0.006	1	0.006	0.003	0.958	
B^2	0.285	1	0.285	0.142	0.717	
C^2	0.160	1	0.160	0.080	0.786	
Residual	14.056	7	2.008			
Lack of Fit	9.312	3	3.104	2.617	0.188	not significant
Pure Error	4.744	4	1.186			
Cor Total	195.371	16				
Std. Dev.	1.417		R-Squared		0.928	
Mean	160.991		Adj R-Squared		0.836	
C.V. %	0.880		Pred R-Squared		0.824	
PRESS	156.408		Adeq Precision		11.571	

Appendix A. 8 (a)

ANOVA for Response Surface Quadratic Model for total sugar content of pineapple juice during ohmic heating

Analysis of variance table [Partial sum of squares - Type III]						
Source	Sum of Squares	df	Mean Square	F Value	p-value Prob > F	
Model	0.0013	9	0.00014	0.925	0.554	not significant
A-voltage gradient	0.0001	1	5.00E-05	0.326	0.586	
B-Temperature	0.0003	1	0.00031	2.035	0.197	
C-Time	0.0000	1	1.25E-05	0.081	0.784	
AB	0.0000	1	0	0.000	1.000	
AC	0.0004	1	0.0004	2.605	0.151	
BC	0.0002	1	0.00023	1.465	0.265	
A^2	0.0001	1	5.92E-05	0.386	0.554	
B^2	0.0001	1	5.92E-05	0.386	0.554	
C^2	0.0002	1	0.00016	1.071	0.335	
Residual	0.0011	7	0.00015			
Lack of Fit	0.0003	3	9.17E-05	0.458	0.726	not significant
Pure Error	0.0008	4	0.0002			
Cor Total	0.0024	16				
Std. Dev.	0.012		R-Squared		0.543	
Mean	10.553		Adj R-Squared		-0.044	
C.V. %	0.117		Pred R-Squared		-1.401	
PRESS	0.006		Adeq Precision		3.288	

Appendix A. 8 (b)

ANOVA for Response Surface Quadratic Model for total sugar content of pineapple juice during ohmic heating

Analysis of variance table [Partial sum of squares - Type III]						
Source	Sum of Squares	df	Mean Square	F Value	p-value Prob > F	
B-Process temperature	1.25E-05	1	1.25E-05	0.052	0.827	
C-Holding time	1.25E-05	1	1.25E-05	0.052	0.827	
AB	0	1	0	0.000	1.000	
AC	0.0001	1	0.0001	0.413	0.541	
	2.50E-05	1	2.50E-05	0.103	0.757	
BC	2.50E-05	1	2.50E-05	0.103	0.757	
A^2	8.53E-05	1	8.53E-05	0.352	0.572	
B^2	0.00046	1	0.00046	1.917	0.209	
C^2	0.00021	1	0.00021	0.852	0.387	
Residual	0.0017	7	0.00024			
Lack of Fit	0.00038	3	0.00013	0.379	0.774	not significant
Pure Error	0.00132	4	0.00033			
Cor Total	0.00259	16				
Std. Dev.	0.016		R-Squared		0.345	
Mean	9.956		Adj R-Squared		-0.497	
C.V. %	0.156		Pred R-Squared		-2.115	
Press	12.34		Adeq Precision		4.32	

Appendix A. 9 (a)

ANOVA for Response Surface Quadratic Model for total phenolic content of pineapple juice during ohmic heating

Analysis of variance table [Partial sum of squares - Type III]						
Source	Sum of Squares	df	Mean Square	F Value	p-value Prob > F	
Model	13.578	9	1.509	7.990	0.006	significant
A-voltage gradient	3.213	1	3.213	17.016	0.004	
B-Temperature	4.047	1	4.047	21.432	0.002	
C-Time	1.022	1	1.022	5.415	0.053	
AB	0.336	1	0.336	1.782	0.224	
AC	0.837	1	0.837	4.434	0.073	
BC	1.156	1	1.156	6.120	0.043	
A^2	0.755	1	0.755	3.999	0.086	
B^2	0.070	1	0.070	0.368	0.563	
C^2	1.924	1	1.924	10.190	0.015	
Residual	1.322	7	0.189			
Lack of Fit	0.809	3	0.270	2.106	0.242	not significant
Pure Error	0.512	4	0.128			
Cor Total	14.900	16				
Std. Dev.	0.435		R-Squared	0.911		
Mean	166.864		Adj R-Squared	0.797		
C.V. %	0.260		Pred R-Squared	0.077		
PRESS	13.750		Adeq Precision	8.750		

Appendix A.9(b)

ANOVA Response Surface Quadratic Model for Total phenolic content of cashew apple juice during ohmic heating

Analysis of variance table [Partial sum of squares - Type III]						
Source	Sum of Squares	df	Mean Square	F Value	p-value Prob > F	
Model	0.0014	9	0.0002	0.319	0.943	not significant
A-voltage gradient	0.0000	1	0.0000	0.002	0.963	
B-Process temperature	0.0008	1	0.0008	1.635	0.242	
C-Holding time	0.0002	1	0.0002	0.472	0.514	
AB	0.0001	1	0.0001	0.131	0.728	
AC	0.0001	1	0.0001	0.270	0.619	
BC	0.0000	1	0.0000	0.008	0.931	
A^2	0.0001	1	0.0001	0.222	0.652	
B^2	0.0001	1	0.0001	0.116	0.743	
C^2	0.0000	1	0.0000	0.032	0.863	
Residual	0.0034	7	0.0005			
Lack of Fit	0.0025	3	0.0008	3.379	0.135	not significant
Pure Error	0.0010	4	0.0002			
Cor Total	0.0048	16				
Std. Dev.	0.022		R-Squared		0.291	
Mean	0.520		Adj R-Squared		-0.621	
C.V. %	4.251		Pred R-Squared		-7.450	
PRESS	0.041		Adeq Precision		2.063	

Appendix A. 10

ANOVA Response Surface Quadratic Model for tannin content of cashew apple juice during ohmic heating

Analysis of variance table [Partial sum of squares - Type III]						
Source	Sum of Squares	df	Mean Square	F Value	p-value Prob > F	
Model	13.578	9	1.509	7.990	0.006	significant
A-voltage gradient	3.213	1	3.213	17.016	0.004	
B-Process temperature	4.047	1	4.047	21.432	0.002	
C-Holding time	1.022	1	1.022	5.415	0.053	
AB	0.336	1	0.336	1.782	0.224	
AC	0.837	1	0.837	4.434	0.073	
BC	1.156	1	1.156	6.120	0.043	
$A^{\wedge 2}$	0.755	1	0.755	3.999	0.086	
$B^{\wedge 2}$	0.070	1	0.070	0.368	0.563	
$C^{\wedge 2}$	1.924	1	1.924	10.190	0.015	
Residual	1.322	7	0.189			
Lack of Fit	0.809	3	0.270	2.106	0.242	not significant
Pure Error	0.512	4	0.128			
Cor Total	14.900	16				
Std. Dev.	0.435		R-Squared		0.911	
Mean	166.864		Adj R-Squared		0.797	
C.V. %	0.260		Pred R-Squared		0.077	
PRESS	13.750		Adeq Precision		8.750	

Appendix 11 Colour values of ohmic heated pineapple and cashew apple juices

	Pineapple juice			Cashew apple juice		
Treatment	**L * values**	**a* values**	**b* values**	**L * values**	**a* values**	**b* values**
T_1	39.33	-3.46	22.77	1.35	0.48	-1.34
T_2	39.17	-3.31	22.58	1.3	0.59	-1.5
T_3	38.94	-3.26	22.51	1.09	0.62	-1.61
T_4	39.2	-3.35	22.63	1.48	0.33	-1.23
T_5	39.18	-3.33	22.6	1.18	0.6	-1.58
T_6	38.88	-3.2	22.46	1.43	0.38	-1.28
T_7	39.15	-3.29	22.56	1.29	0.55	-1.38
T_8	39.26	-3.41	22.68	1.38	0.45	-1.31
T_9	39.36	-3.49	22.78	1.33	0.51	-1.34
T_{10}	39.24	-3.4	22.67	1.31	0.53	-1.36
T_{11}	39.19	-3.34	22.62	1.11	0.61	-1.6
T_{12}	39.39	-3.53	22.82	1.22	0.56	-1.47
T_{13}	39.22	-3.38	22.65	1.27	0.56	-1.4
T_{14}	39.27	-3.42	22.69	1.46	0.36	-1.26
T_{15}	39.32	-3.46	22.76	1.41	0.41	-1.3
T_{16}	38.9	-3.22	22.5	1.26	0.57	-1.43
T_{17}	38.88	-3.2	22.46	1.24	0.58	-1.44

Appendix A.12 (a)

ANOVA for Response Surface Quadratic Model for total colour differences of pineapple juice during ohmic heating

Analysis of variance table [Partial sum of squares - Type III]						
Source	Sum of Squares	df	Mean Square	F Value	p-value Prob > F	
Model	0.624	9	0.069	25.150	0.000	significant
A-voltage gradient	0.211	1	0.211	76.639	< 0.0001	
B-Temperature	0.183	1	0.183	66.395	< 0.0001	
C-Time	0.104	1	0.104	37.553	0.001	
AB	0.053	1	0.053	19.192	0.003	
AC	0.026	1	0.026	9.287	0.019	
BC	0.021	1	0.021	7.628	0.028	
A^2	0.022	1	0.022	7.864	0.026	
B^2	0.001	1	0.001	0.310	0.595	
C^2	0.002	1	0.002	0.898	0.375	
Residual	0.019	7	0.003			
Lack of Fit	0.015	3	0.005	4.358	0.095	not significant
Pure Error	0.005	4	0.001			
Cor Total	0.643	16				
Std. Dev.	0.053		R-Squared		0.970	
Mean	0.814		Adj R-Squared		0.931	
C.V. %	6.449		Pred R-Squared		0.751	
PRESS	0.243		Adeq Precision		15.770	

Appendix A. 12 (b)

ANOVA for Response Surface Quadratic Model for total Colour difference of cashew apple juice during ohmic heating

Analysis of variance table [Partial sum of squares - Type III]						
Source	Sum of Squares	df	Mean Square	F Value	p-value Prob > F	
Model	0.502	9	0.056	37.637	< 0.0001	significant
A-voltage gradient	0.157	1	0.157	105.844	< 0.0001	
B-Process temperature	0.168	1	0.168	113.539	< 0.0001	
C-Holding time	0.092	1	0.092	62.406	< 0.0001	
AB	0.026	1	0.026	17.281	0.004	
AC	0.008	1	0.008	5.468	0.052	
BC	0.004	1	0.004	2.430	0.163	
A^2	0.029	1	0.029	19.580	0.003	
B^2	0.008	1	0.008	5.255	0.056	
C^2	0.006	1	0.006	4.104	0.082	
Residual	0.010	7	0.001			
Lack of Fit	0.006	3	0.002	2.023	0.253	not significant
Pure Error	0.004	4	0.001			
Cor Total	0.512	16				
Std. Dev.	0.038			R-Squared	0.980	
Mean	0.579			Adj R-Squared	0.954	
C.V. %	6.650			Pred R-Squared	0.792	
PRESS	0.106			Adeq Precision	19.309	

Appendix A. 13 (a)

ANOVA for Response Surface Quadratic Model for bacterial log reduction pineapple juice during ohmic heating

Analysis of variance table [Partial sum of squares - Type III]						
Source	Sum of Squares	df	Mean Square	F Value	p-value Prob > F	
Model	7.880	9	0.876	70.90	< 0.0001	significant
A-voltage gradient	2.940	1	2.940	238.10	< 0.0001	
B-Temperature	2.060	1	2.060	166.85	< 0.0001	
C-Time	0.656	1	0.656	53.08	0.000	
AB	0.087	1	0.087	7.05	0.033	
AC	0.212	1	0.212	17.13	0.004	
BC	0.106	1	0.106	8.55	0.022	
A^2	0.952	1	0.952	77.09	< 0.0001	
B^2	0.141	1	0.141	11.42	0.012	
C^2	0.562	1	0.562	45.55	0.000	
Residual	0.086	7	0.012			
Lack of Fit	0.071	3	0.024	6.00	0.0581	not significant
Pure Error	0.016	4	0.004			
Cor Total	7.966	16				
Std. Dev.	0.111		R-Squared		0.989	
Mean	3.034		Adj R-Squared		0.975	
C.V. %	3.663		Pred R-Squared		0.855	
PRESS	1.156		Adeq Precision		26.135	

Appendix A.13 (b)

ANOVA for Response Surface Quadratic Model for bacterial log reduction of cashew apple juice during ohmic heating

Analysis of variance table [Partial sum of squares - Type III]						
Source	Sum of Squares	df	Mean Square	F Value	p-value Prob > F	
Model	10.911	9	1.212	17.654	0.001	significant
A-voltage gradient	2.475	1	2.475	36.045	0.001	
B-Process temperature	4.220	1	4.220	61.444	0.000	
C-Holding time	1.345	1	1.345	19.583	0.003	
AB	0.009	1	0.009	0.131	0.728	
AC	0.078	1	0.078	1.142	0.321	
BC	0.012	1	0.012	0.176	0.687	
A^2	1.530	1	1.530	22.276	0.002	
B^2	0.280	1	0.280	4.073	0.083	
C^2	0.709	1	0.709	10.319	0.015	
Residual	0.481	7	0.069			
Lack of Fit	0.397	3	0.132	6.326	0.053	not significant
Pure Error	0.084	4	0.021			
Cor Total	11.392	16				
Std. Dev.	0.262		R-Squared		0.958	
Mean	3.250		Adj R-Squared		0.904	
C.V. %	8.063		Pred R-Squared		0.831	
PRESS	6.483		Adeq Precision		13.030	

Appendix A. 14 (a)

ANOVA for Response Surface Quadratic Model for yeast and mould reduction of pineapple juice during ohmic heating

Analysis of variance table [Partial sum of squares - Type III]						
Source	Sum of Squares	df	Mean Square	F Value	p-value Prob > F	
Model	5.775	9	0.642	42.443	< 0.0001	significant
A-voltage gradient	2.205	1	2.205	145.861	< 0.0001	
B-Temperature	1.638	1	1.638	108.357	< 0.0001	
C-Time	0.414	1	0.414	27.389	0.001	
AB	0.073	1	0.073	4.822	0.064	
AC	0.116	1	0.116	7.647	0.028	
BC	0.048	1	0.048	3.202	0.117	
A^2	0.594	1	0.594	39.272	0.000	
B^2	0.145	1	0.145	9.584	0.017	
C^2	0.419	1	0.419	27.725	0.001	
Residual	0.106	7	0.015			
Lack of Fit	0.087	3	0.029	5.970	0.059	not significant
Pure Error	0.019	4	0.005			
Cor Total	5.880	16				
Std. Dev.	0.123		R-Squared		0.982	
Mean	2.194		Adj R-Squared		0.959	
C.V. %	5.605		Pred R-Squared		0.790	
PRESS	1.414		Adeq Precision		20.732	

Appendix A.14 (b)

ANOVA for Response Surface Quadratic Model for Yeast and mould log reduction of cashew apple juice during ohmic heating

Analysis of variance table [Partial sum of squares - Type III]						
Source	Sum of Squares	df	Mean Square	F Value	p-value Prob > F	
Model	4.307	9	0.479	25.698	0.000	significant
A-voltage gradient	0.994	1	0.994	53.382	0.000	
B-Process temperature	1.531	1	1.531	82.231	< 0.0001	
C-Holding time	0.781	1	0.781	41.954	0.000	
AB	0.068	1	0.068	3.630	0.098	
AC	0.160	1	0.160	8.592	0.022	
BC	0.116	1	0.116	6.208	0.042	
A^2	0.348	1	0.348	18.690	0.004	
B^2	0.019	1	0.019	1.030	0.344	
C^2	0.238	1	0.238	12.754	0.009	
Residual	0.130	7	0.019			
Lack of Fit	0.026	3	0.009	0.328	0.807	not significant
Pure Error	0.105	4	0.026			
Cor Total	4.437	16				
Std. Dev.	0.136		R-Squared		0.971	
Mean	2.271		Adj R-Squared		0.933	
C.V. %	6.008		Pred R-Squared		0.870	
PRESS	0.575		Adeq Precision		15.574	

Appendix A.15 (a)

ANOVA for Response Surface Quadratic Model for pH of pineapple juice during PL treatment

Analysis of variance table [Partial sum of squares - Type III]						
Source	Sum of Squares	df	Mean Square	F Value	p-value Prob > F	
Model	0.00028	9	3.09E-05	1.494	0.305	not significant
A-PL dosage	1.3E-05	1	1.3E-05	0.603	0.463	
B-Distance from the light source	0.00011	1	0.00011	5.431	0.053	
C-Flow rate	5.00E-05	1	5.00E-05	2.414	0.164	
AB	0	1	0	0.000	1.000	
AC	2.50E-05	1	2.50E-05	1.207	0.308	
BC	2.50E-05	1	2.50E-05	1.207	0.308	
A^2	3.79E-05	1	3.79E-05	1.829	0.218	
B^2	1.68E-05	1	1.68E-05	0.813	0.397	
C^2	1.05E-06	1	1.05E-06	0.051	0.828	
Residual	0.00015	7	2.07E-05			
Lack of Fit	2.50E-05	3	8.33E-06	0.278	0.840	not significant
Pure Error	0.00012	4	3.00E-05			
Cor Total	0.00042	16				
Std. Dev.	0.005		R-Squared		0.658	
Mean	4.455		Adj R-Squared		0.217	
C.V. %	0.102		Pred R-Squared		-0.387	
PRESS	0.001		Adeq Precision		3.581	

Appendix A.15 (b)

ANOVA for Response Surface Quadratic Model for pH of cashew apple juice during PL treatment

Analysis of variance table [Partial sum of squares - Type III]						
Source	Sum of Squares	df	Mean Square	F Value	p-value Prob > F	
Model	0.00039	9	4.32E-05	0.992	0.516	not significant
A-PL dosage	5.00E-05	1	5.00E-05	1.148	0.320	
B-Distance from the light source	1.3E-05	1	1.3E-05	0.287	0.609	
C-flow rate	1.25E-05	1	1.25E-05	0.287	0.609	
AB	2.50E-05	1	2.50E-05	0.574	0.474	
AC	2.50E-05	1	2.50E-05	0.574	0.474	
BC	0.0001	1	0.0001	2.295	0.174	
A^2	6.74E-05	1	6.74E-05	1.546	0.254	
B^2	5.16E-05	1	5.16E-05	1.184	0.313	
C^2	5.16E-05	1	5.16E-05	1.184	0.313	
Residual	0.00031	7	4.36E-05			
Lack of Fit	2.50E-05	3	8.33E-06	0.119	0.944	not significant
Pure Error	0.00028	4	7.00E-05			
Cor Total	0.00069	16				
Std. Dev.	0.007		R-Squared	0.561		
Mean	4.321		Adj R-Squared	-0.004		
C.V. %	0.153		Pred R-Squared	-0.207		
PRESS	0.001		Adeq Precision	4.197		

Appendix A.16 (a)

ANOVA for Response Surface Quadratic Model for TSS of pineapple juice during PL treatment

Analysis of variance table [Partial sum of squares - Type III]						
Source	Sum of Squares	df	Mean Square	F Value	p-value Prob > F	
Model	0.00044	9	4.88E-05	1.339	0.358	not significant
A-PL dosage	1.25E-05	1	1.25E-05	0.343	0.576	
B-Distance from the light source	0	1	0	0.000	1.000	
C-Flow rate	1.25E-05	1	1.25E-05	0.343	0.576	
AB	2.50E-05	1	2.50E-05	0.686	0.435	
AC	1.00E-04	1	1.00E-04	2.745	0.142	
BC	0.00023	1	0.00023	6.176	0.042	
A^2	5.16E-05	1	5.16E-05	1.416	0.273	
B^2	4.21E-06	1	4.21E-06	0.116	0.744	
C^2	9.47E-06	1	9.47E-06	0.260	0.626	
Residual	0.00026	7	3.64E-05			
Lack of Fit	0.00018	3	5.83E-05	2.917	0.164	not significant
Pure Error	8.00E-05	4	2.00E-05			
Cor Total	0.00069	16				
Std. Dev.	0.006		R-Squared	0.633		
Mean	10.809		Adj R-Squared	0.160		
C.V. %	0.056		Pred R-Squared	-3.214		
PRESS	0.003		Adeq Precision	3.780		

Appendix A. 16 (b)

ANOVA for Response Surface Quadratic Model for TSS of cashew apple juice during PL treatments

Analysis of variance table [Partial sum of squares - Type III]						
Source	Sum of Squares	df	Mean Square	F Value	p-value Prob > F	
Model	0.0009	9	0.0001	4.839	0.025	significant
A-PL dosage	0	1	0	0.000	1.000	
B-Distance from the light source	1.25E-05	1	1.25E-05	0.603	0.463	
C-flow rate	0.00061	1	0.00061	29.569	0.001	
AB	1.00E-04	1	1.00E-04	4.828	0.064	
AC	0	1	0	0.000	1.000	
BC	2.50E-05	1	2.50E-05	1.207	0.308	
A^2	1.29E-05	1	1.29E-05	0.623	0.456	
B^2	2.37E-06	1	2.37E-06	0.114	0.745	
C^2	0.00014	1	0.00014	6.721	0.036	
Residual	0.00015	7	2.07E-05			
Lack of Fit	2.50E-05	3	8.33E-06	0.278	0.840	not significant
Pure Error	0.00012	4	3.00E-05			
Cor Total	0.00105	16				
Std. Dev.	0.005		R-Squared	0.862		
Mean	11.238		Adj R-Squared	0.683		
C.V. %	0.040		Pred R-Squared	0.439		
PRESS	0.001		Adeq Precision	6.804		

Appendix A. 17 (a)

ANOVA for Response Surface Quadratic Model for Titrable acidity of pine apple juice during PL treatment

Analysis of variance table [Partial sum of squares - Type III]						
Source	Sum of Squares	df	Mean Square	F Value	p-value Prob > F	
Model	7.39E-06	9	8.21E-07	1.036	0.493	not significant
A-PL dosage	3.13E-06	1	3.13E-06	3.941	0.088	
B-Distance from the light source	1.13E-06	1	1.13E-06	1.419	0.272	
C-Flow rate	0	1	0	0.000	1.000	
AB	1.00E-06	1	1.00E-06	1.261	0.298	
AC	2.5E-07	1	2.5E-07	0.315	0.592	
BC	2.5E-07	1	2.5E-07	0.315	0.592	
A^2	9.47E-08	1	9.47E-08	0.119	0.740	
B^2	9.47E-08	1	9.47E-08	0.119	0.740	
C^2	1.52E-06	1	1.52E-06	1.912	0.209	
Residual	5.55E-06	7	7.93E-07			
Lack of Fit	2.75E-06	3	9.17E-07	1.310	0.387	not significant
Pure Error	2.8E-06	4	7E-07			
Cor Total	1.29E-05	16				
Std. Dev.	0.001		R-Squared	0.571		
Mean	0.382		Adj R-Squared	0.020		
C.V. %	0.233		Pred R-Squared	-2.738		
PRESS	0.000		Adeq Precision	3.478		

Appendix A. 17 (b)

ANOVA for Response Surface Quadratic Model for Titrable acidity of cashew apple juice during PL treatment.

Analysis of variance table [Partial sum of squares - Type III]						
Source	Sum of Squares	df	Mean Square	F Value	p-value Prob > F	
Model	8.42E-06	9	9.36E-07	1.083	0.469	not significant
A-PL dosage	1.25E-07	1	1.25E-07	0.145	0.715	
B-Distance from the light source	1.25E-07	1	1.25E-07	0.145	0.715	
C-flow rate	0	1	0	0.000	1.000	
AB	2.25E-06	1	2.25E-06	2.603	0.151	
AC	1E-06	1	1E-06	1.157	0.318	
BC	1E-06	1	1E-06	1.157	0.318	
A^2	1.16E-06	1	1.16E-06	1.343	0.285	
B^2	2.63E-09	1	2.63E-09	0.003	0.958	
C^2	2.53E-06	1	2.53E-06	2.926	0.131	
Residual	6.05E-06	7	8.64E-07			
Lack of Fit	1.25E-06	3	4.17E-07	0.347	0.794	not significant
Pure Error	4.80E-06	4	1.2E-06			
Cor Total	1.45E-05	16				
Std. Dev.	9E-04		R-Squared		0.582	
Mean	4E-01		Adj R-Squared		0.044	
C.V. %	2E-01		Pred R-Squared		-0.900	
PRESS	3E-05		Adeq Precision		3.331	

Appendix 18 (a)

ANOVA for Response Surface Quadratic Model for Ascorbic acid of pineapple juice during PL treatment

Analysis of variance table [Partial sum of squares - Type III]						
Source	Sum of Squares	df	Mean Square	F Value	p-value Prob > F	
Model	8.993	9	0.999	15.048	0.001	significant
A-PL dosage	3.075	1	3.075	46.311	0.000	
B-Distance from the light source	2.311	1	2.311	34.806	0.001	
C-Flow rate	0.858	1	0.858	12.922	0.009	
AB	0.008	1	0.008	0.122	0.737	
AC	0.230	1	0.230	3.470	0.105	
BC	0.360	1	0.360	5.421	0.053	
A^2	1.488	1	1.488	22.411	0.002	
B^2	0.107	1	0.107	1.613	0.245	
C^2	0.390	1	0.390	5.879	0.046	
Residual	0.465	7	0.066			
Lack of Fit	0.343	3	0.114	3.767	0.116	not significant
Pure Error	0.122	4	0.030			
Cor Total	9.458	16				

Std. Dev.	0.258	R-Squared	0.951
Mean	29.744	Adj R-Squared	0.888
C.V. %	0.866	Pred R-Squared	0.829
PRESS	5.683	Adeq Precision	11.713

Appendix A.18 (b)

ANOVA for Response Surface Quadratic Model for Ascorbic acid of cashew apple juice during PL treatment

Analysis of variance table [Partial sum of squares - Type III]						
Source	Sum of Squares	df	Mean Square	F Value	p-value Prob > F	
Model	183.759	9	20.418	5.053	0.022	significant
A-PL dosage	95.981	1	95.981	23.755	0.002	
B-Distance from the light source	36.937	1	36.937	9.142	0.019	
C-flow rate	16.820	1	16.820	4.163	0.081	
AB	5.290	1	5.290	1.309	0.290	
AC	4.264	1	4.264	1.055	0.338	
BC	2.205	1	2.205	0.546	0.484	
A^2	12.565	1	12.565	3.110	0.121	
B^2	4.316	1	4.316	1.068	0.336	
C^2	3.298	1	3.298	0.816	0.396	
Residual	28.283	7	4.040			
Lack of Fit	4.122	3	1.374	0.227	0.873	not significant
Pure Error	24.160	4	6.040			
Cor Total	212.042	16				
Std. Dev.	2.010		R-Squared	0.867		
Mean	162.166		Adj R-Squared	0.795		
C.V. %	1.240		Pred R-Squared	0.731		
PRESS	103.705		Adeq Precision	7.281		

Appendix A. 19 (a)

ANOVA for Response Surface Quadratic Model for total sugar of pine apple juice during PL treatment

Analysis of variance table [Partial sum of squares - Type III]						
Source	Sum of Squares	df	Mean Square	F Value	p-value Prob > F	
Model	0.00032	9	3.54E-05	0.120	0.998	not significant
A-PL dosage	5.00E-05	1	5.00E-05	0.169	0.693	
B-Distance from the light source	5.00E-05	1	5.00E-05	0.169	0.693	
C-Flow rate	5.00E-05	1	5.00E-05	0.169	0.693	
AB	1.00E-04	1	1.00E-04	0.338	0.579	
AC	0	1	0	0.000	1.000	
BC	0	1	0	0.000	1.000	
A^2	1.68E-05	1	1.68E-05	0.057	0.818	
B^2	3.79E-05	1	3.79E-05	0.128	0.731	
C^2	1.68E-05	1	1.68E-05	0.057	0.818	
Residual	0.00207	7	0.0003			
Lack of Fit	0.00135	3	0.00045	2.500	0.199	not significant
Pure Error	0.00072	4	0.00018			
Cor Total	0.00239	16				
Std. Dev.	0.017		R-Squared	0.133		
Mean	10.546		Adj R-Squared	-0.981		
C.V. %	0.163		Pred R-Squared	-8.515		
PRESS	0.023		Adeq Precision	1.137		

Appendix A. 19 (b)

ANOVA for Response Surface Quadratic Model for total sugar of cashew apple juice during PL treatments

Analysis of variance table [Partial sum of squares - Type III]						
Source	Sum of Squares	df	Mean Square	F Value	p-value Prob > F	
Model	0.0009	9	9.98E-05	0.662	0.724	not significant
A-PL dosage	5.00E-05	1	5.00E-05	0.332	0.583	
B-Distance from the light source	0.00031	1	0.00031	2.073	0.193	
C-flow rate	0.00011	1	0.00011	0.746	0.416	
AB	0.0001	1	0.0001	0.664	0.442	
AC	0.0001	1	0.0001	0.664	0.442	
BC	2.50E-05	1	2.50E-05	0.166	0.696	
A^2	0.00012	1	0.00012	0.770	0.409	
B^2	3.18E-05	1	3.18E-05	0.211	0.660	
C^2	3.18E-05	1	3.18E-05	0.211	0.660	
Residual	0.00106	7	0.00015			
Lack of Fit	0.00038	3	0.00013	0.735	0.583	not significant
Pure Error	0.00068	4	0.00017			
Cor Total	0.00195	16				
Std. Dev.	0.012		R-Squared		0.460	
Mean	9.957		Adj R-Squared		-0.235	
C.V. %	0.123		Pred R-Squared		-2.616	
PRESS	0.007		Adeq Precision		2.522	

Appendix A. 20 (a)

ANOVA for Response Surface Quadratic Model for total phenolic content of pineapple juice during PL treatment

Analysis of variance table [Partial sum of squares - Type III]						
Source	Sum of Squares	df	Mean Square	F Value	p-value Prob > F	
Model	1.135	9	0.126	3.478	0.057	not significant
A-PL dosage	0.076	1	0.076	2.098	0.191	
B-Distance from the light source	0.174	1	0.174	4.802	0.065	
C-Flow rate	0.072	1	0.072	1.992	0.201	
AB	0.141	1	0.141	3.880	0.090	
AC	0.004	1	0.004	0.117	0.743	
BC	0.133	1	0.133	3.675	0.097	
A^2	0.466	1	0.466	12.862	0.009	
B^2	0.040	1	0.040	1.110	0.327	
C^2	0.025	1	0.025	0.693	0.433	
Residual	0.254	7	0.036			
Lack of Fit	0.150	3	0.050	1.942	0.265	not significant
Pure Error	0.103	4	0.026			
Cor Total	1.388	16				
Std. Dev.	0.190		R-Squared	0.817		
Mean	66.712		Adj R-Squared	0.582		
C.V. %	0.285		Pred R-Squared	-0.850		
PRESS	2.569		Adeq Precision	6.044		

Appendix A. 20 (b)

ANOVA for Response Surface Quadratic Model for total phenolic content of cashew apple juice during PL treatment

Analysis of variance table [Partial sum of squares - Type III]						
Source	Sum of Squares	df	Mean Square	F Value	p-value Prob > F	
Model	4.256	9	0.473	3.440	0.059	not significant
A-PL dosage	0.054	1	0.054	0.396	0.549	
B-Distance from the light source	0.039	1	0.039	0.285	0.610	
C-flow rate	0.022	1	0.022	0.160	0.701	
AB	0.026	1	0.026	0.186	0.679	
AC	0.608	1	0.608	4.426	0.074	
BC	1.346	1	1.346	9.789	0.017	
A^2	0.701	1	0.701	5.099	0.059	
B^2	0.073	1	0.073	0.534	0.489	
C^2	1.476	1	1.476	10.735	0.014	
Residual	0.962	7	0.137			
Lack of Fit	0.957	3	0.319	259.431	< 0.0001	significant
Pure Error	0.005	4	0.001			
Cor Total	5.218	16				
Std. Dev.	0.371		R-Squared		0.816	
Mean	163.76		Adj R-Squared		0.579	
C.V. %	0.226		Pred R-Squared		-1.937	
PRESS	15.324		Adeq Precision		6.805	

Appendix A. 21

ANOVA for Response Surface Quadratic Model for total tannin content of cashew apple juice during PL treatment

Analysis of variance table [Partial sum of squares - Type III]						
Source	Sum of Squares	df	Mean Square	F Value	p-value Prob > F	
Model	0.00026	9	2.9E-05	2.549	0.115	not significant
A-PL dosage	7.75E-05	1	7.8E-05	6.733	0.036	
B-Distance from the light source	1.90E-06	1	1.9E-06	0.165	0.697	
C-flow rate	6.13E-06	1	6.1E-06	0.532	0.489	
AB	7.31E-05	1	7.3E-05	6.351	0.040	
AC	1.6E-05	1	1.6E-05	1.390	0.277	
BC	3E-05	1	3.0E-05	2.628	0.149	
A^2	2.95E-06	1	3.0E-06	0.257	0.628	
B^2	3.57E-05	1	3.6E-05	3.103	0.122	
C^2	1.92E-05	1	1.9E-05	1.671	0.237	
Residual	8.06E-05	7	1.2E-05			
Lack of Fit	7.38E-05	3	2.5E-05	14.466	0.013	significant
Pure Error	6.80E-06	4	1.7E-06			
Cor Total	0.00034	16				
Std. Dev.	0.003		R-Squared		0.766	
Mean	0.541		Adj R-Squared		0.466	
C.V. %	0.626		Pred R-Squared		2.456	
PRESS	0.001		Adeq Precision		5.678	

Appendix 22

Colour values of pineapple and cashew apple juice during PL treatment

Treatment	Pineapple juice			Cashew apple juice		
	L * values	a* values	b* values	L * values	a* values	b* values
T_1	39.33	-3.46	22.77	1.45	0.36	-1.25
T_2	39.17	-3.31	22.58	1.27	0.49	-1.5
T_3	38.94	-3.26	22.51	1.37	0.41	-1.32
T_4	39.2	-3.35	22.63	1.38	0.4	-1.31
T_5	39.18	-3.33	22.6	1.49	0.33	-1.12
T_6	38.88	-3.2	22.46	1.28	0.48	-1.48
T_7	39.15	-3.29	22.56	1.36	0.44	-1.4
T_8	39.26	-3.41	22.68	1.37	0.41	-1.32
T_9	39.36	-3.49	22.78	1.48	0.34	-1.19
T_{10}	39.24	-3.4	22.67	1.33	0.45	-1.39
T_{11}	39.19	-3.34	22.62	1.32	0.45	-1.41
T_{12}	39.39	-3.53	22.82	1.39	0.39	-1.31
T_{13}	39.22	-3.38	22.65	1.41	0.38	-1.29
T_{14}	39.27	-3.42	22.69	1.43	0.37	-1.27
T_{15}	39.32	-3.46	22.76	1.29	0.47	-1.44
T_{16}	38.9	-3.22	22.5	1.33	0.44	-1.36
T_{17}	38.88	-3.2	22.46	1.34	0.42	-1.34

Appendix A. 23 (a)

ANOVA for Response Surface Quadratic Model for total colour difference of pineapple juice during PL treatment

Analysis of variance table [Partial sum of squares - Type III]						
Source	Sum of Squares	df	Mean Square	F Value	p-value Prob > F	
Model	0.494	9	0.055	21.997	0.000	significant
A-PL dosage	0.171	1	0.171	68.543	< 0.0001	
B-Distance from the light source	0.074	1	0.074	29.687	0.00	
C-Flow rate	0.097	1	0.097	38.775	0.00	
AB	0.000	1	0.000	0.160	0.70	
AC	0.013	1	0.013	5.298	0.05	
BC	0.001	1	0.001	0.491	0.51	
A^2	0.046	1	0.046	18.595	0.00	
B^2	0.024	1	0.024	9.487	0.02	
C^2	0.053	1	0.053	21.346	0.00	
Residual	0.017	7	0.002			
Lack of Fit	0.012	3	0.004	3.327	0.138	not significant
Pure Error	0.005	4	0.001			
Cor Total	0.512	16				
Std. Dev.	0.050		R-Squared		0.966	
Mean	0.572		Adj R-Squared		0.922	
C.V. %	8.730		Pred R-Squared		0.795	
PRESS	0.207		Adeq Precision		13.37	

Appendix A. 23 (b)

ANOVA for Response Surface Quadratic Model for total colour difference of cashew apple juice during PL treatments

Analysis of variance table [Partial sum of squares - Type III]						
Source	Sum of Squares	df	Mean Square	F Value	p-value Prob > F	
Model	0.248	9	0.028	25.552	0.0002	significant
A-PL dosage	0.115	1	0.115	106.737	< 0.0001	
B-Distance from the light source	0.070	1	0.070	65.147	< 0.0001	
C-flow rate	0.023	1	0.023	21.415	0.002	
AB	0.000	1	0.000	0.023	0.883	
AC	0.006	1	0.006	5.212	0.056	
BC	0.002	1	0.002	1.482	0.263	
$A^{\wedge 2}$	0.018	1	0.018	16.994	0.004	
$B^{\wedge 2}$	0.006	1	0.006	5.783	0.047	
$C^{\wedge 2}$	0.005	1	0.005	4.378	0.075	
Residual	0.008	7	0.001			
Lack of Fit	0.006	3	0.002	5.473	0.067	not significant
Pure Error	0.001	4	0.000			
Cor Total	0.256	16				
Std. Dev.	0.033		R-Squared		0.970	
Mean	0.457		Adj R-Squared		0.932	
C.V. %	7.188		Pred R-Squared		0.811	
PRESS	0.100		Adeq Precision		17.16	

Appendix A. 24 (a)

ANOVA for Response Surface Quadratic Model for bacterial log reduction of pineapple juice during PL treatment

Analysis of variance table [Partial sum of squares - Type III]						
Source	Sum of Squares	df	Mean Square	F Value	p-value Prob > F	
Model	6.9231	9	0.769	30.81	< 0.0001	significant
A-PL dosage	2.6335	1	2.634	105.48	< 0.0001	
B-Distance from the light source	1.0225	1	1.022	40.95	0.00	
C-Flow rate	1.3530	1	1.353	54.19	0.00	
AB	0.3136	1	0.314	12.56	0.01	
AC	0.0380	1	0.038	1.52	0.26	
BC	0.1600	1	0.160	6.41	0.04	
A^2	0.6120	1	0.612	24.51	0.00	
B^2	0.4145	1	0.414	16.60	0.00	
C^2	0.2350	1	0.235	9.41	0.02	
Residual	0.1748	7	0.025			
Lack of Fit	0.0396	3	0.013	0.39	0.77	not significant
Pure Error	0.1352	4	0.034			
Cor Total	7.0978	16				
Std. Dev.	0.158		R-Squared	0.975		
Mean	2.441		Adj R-Squared	0.943		
C.V. %	6.471		Pred R-Squared	0.88		
PRESS	0.844		Adeq Precision	16.67		

Appendix A. 24 (b)

ANOVA for Response Surface Quadratic Model for Bacterial log reduction of cashew apple juice during PL treatments

Analysis of variance table [Partial sum of squares - Type III]						
Source	Sum of Squares	df	Mean Square	F Value	p-value Prob > F	
Model	8.054	9	0.895	50.938	< 0.0001	significant
A-PL dosage	3.125	1	3.125	177.889	< 0.0001	
B-Distance from the light source	0.911	1	0.911	51.872	0.0002	
C-flow rate	1.394	1	1.394	79.378	< 0.0001	
AB	0.308	1	0.308	17.534	0.0041	
AC	0.286	1	0.286	16.293	0.005	
BC	0.021	1	0.021	1.197	0.3102	
A^2	1.667	1	1.667	94.903	< 0.0001	
B^2	0.176	1	0.176	9.999	0.0159	
C^2	0.050	1	0.050	2.861	0.1346	
Residual	0.123	7	0.018			
Lack of Fit	0.032	3	0.011	0.470	0.719	not significant
Pure Error	0.091	4	0.023			
Cor Total	8.177	16				
Std. Dev.	0.133		R-Squared		0.985	
Mean	2.552		Adj R-Squared		0.966	
C.V. %	5.193		Pred R-Squared		0.920	
PRESS	0.655		Adeq Precision		20.560	

Appendix A. 25 (a)

ANOVA for Response Surface Quadratic Model for Yeast and mould log reduction of pineapple juice during PL treatment

Analysis of variance table [Partial sum of squares - Type III]						
Source	Sum of Squares	df	Mean Square	F Value	p-value Prob > F	
Model	6.277	9	0.697	29.543	< 0.0001	significant
A-PL dosage	2.761	1	2.761	116.96	< 0.0001	
B-Distance from the light source	0.656	1	0.656	27.767	0.001	
C-Flow rate	1.073	1	1.073	45.456	0.000	
AB	0.203	1	0.203	8.578	0.022	
AC	0.010	1	0.010	0.424	0.536	
BC	0.133	1	0.133	5.643	0.049	
A^2	0.609	1	0.609	25.788	0.001	
B^2	0.480	1	0.480	20.346	0.003	
C^2	0.209	1	0.209	8.849	0.021	
Residual	0.165	7	0.024			
Lack of Fit	0.118	3	0.039	3.327	0.138	not significant
Pure Error	0.047	4	0.012			
Cor Total	6.442	16				
Std. Dev.	0.154		R-Squared		0.974	
Mean	2.135		Adj R-Squared		0.941	
C.V. %	7.196		Pred R-Squared		0.896	
PRESS	1.961		Adeq Precision		16.187	

Appendix A. 25 (b)

ANOVA for Response Surface Quadratic Model for Yeast and mould reduction of cashew apple juice during PL treatments

Analysis of variance table [Partial sum of squares - Type III]						
Source	Sum of Squares	df	Mean Square	F Value	p-value Prob > F	
Model	7.060	9	0.784	84.669	< 0.0001	significant
A-PL dosage	1.815	1	1.815	195.846	< 0.0001	
B-Distance from the light source	0.952	1	0.952	102.774	< 0.0001	
C-flow rate	1.059	1	1.059	114.249	< 0.0001	
AB	0.276	1	0.276	29.749	0.001	
AC	0.130	1	0.130	13.988	0.0073	
BC	0.198	1	0.198	21.373	0.0024	
A^2	1.278	1	1.278	137.973	< 0.0001	
B^2	0.497	1	0.497	53.622	0.0002	
C^2	0.595	1	0.595	64.249	< 0.0001	
Residual	0.065	7	0.009			
Lack of Fit	0.045	3	0.015	2.973	0.16	not significant
Pure Error	0.020	4	0.005			
Cor Total	7.125	16				
Std. Dev.	0.096		R-Squared		0.991	
Mean	2.084		Adj R-Squared		0.979	
C.V. %	4.618		Pred R-Squared		0.895	
PRESS	0.748		Adeq Precision		24.365	

A.26 ANOVA for the Effect of ohmic assisted PL treatment on the physiochemical properties of pineapple juice

Treatment	pH	TSS (°Brix)	Titrable acidity (mg/100 ml)	Ascorbic acid (mg/100 ml)	Total sugar (mg/100 ml)	Total phenolic content (mg/100ml)	Total colour difference (ΔE)	Yellowness index
Fresh	4.32	10.82	0.382	31.18	10.53	67.25	0	82.18
Ohmic treated	4.27	11.15	0.388	28.75	10.54	68.48	0.68	81.92
PL treated	4.32	10.82	0.382	28.96	10.55	67.14	0.46	82.10
P1 (OH1PL1)	4.29	11.16	0.387	26.24	10.54	67.22	0.78	81.91
P2 (OH1PL2)	4.27	11.15	0.386	26.76	10.55	67.18	0.74	81.91
P3 (OH2PL1)	4.28	11.14	0.384	27.13	10.53	67.21	0.76	81.87
P4 (OH2PL2)	4.28	11.16	0.385	26.94	10.54	67.19	0.74	81.78

Treatment	Browning index	Antioxidant Activity	Calcium	Potassium	Sodium	Bacterial log reduction (log cfu/ml)	Yeast and mold log reduction (log cfu/ml)
Fresh	0	82.66	5.04	65.34	17.68	0	0
Ohmic treated	0.18	77.63	4.98	65.18	17.63	3.24	2.54
PL treated	0.14	79.34	5.02	65.32	17.66	2.42	2.16
P1 (OH1PL1)	0.2	75.85	4.97	64.98	17.64	5.13	4.95
P2 (OH1PL2)	0.21	74.86	4.98	65.03	17.62	5.11	4.92
P3 (OH2PL1)	0.22	74.19	4.96	64.95	17.59	5.04	4.89
P4 (OH2PL2)	0.21	73.98	4.99	64.93	17.6	4.96	4.9

A.27 ANOVA for the Effect of ohmic assisted PL treatment on the physiochemical properties of cashew apple juice

Treatment	pH	TSS (°Brix)	Titrable acidity (mg/100 ml)	Ascorbic acid (mg/100 ml)	Total sugar (mg/100 ml)	Total phenolic content (mg/100ml)	Tannin content	Total colour difference (ΔE)
Fresh	4.45	7.73	0.422	168.46	9.96	168.46	0.57	0
Ohmic treated	4.26	8.07	0.424	159.74	9.97	169.43	0.55	0.68
PL treated	4.45	7.74	0.422	162.96	9.96	167.12	0.55	0.46
C1 (OH1PL1)	4.27	8.12	0.423	150.02	9.97	168.14	0.54	0.78
C2 (OH1PL2)	4.26	8.09	0.424	148.76	9.96	167.89	0.52	0.74
C3 (OH2PL1)	4.28	8.11	0.422	147.72	9.97	168.26	0.53	0.76
C4 (OH2PL2)	4.27	8.08	0.423	149.23	9.97	167.6	0.51	0.74

Treatment	Yellowness index	Browning index	Antioxidant Activity	Calcium	Potassium	Sodium	Bacterial log reduction (log cfu/ml)	Yeast and mold log reduction (log cfu/ml)
Fresh	-86.08	0	75.36	10.53	123.78	19.34	0	0
Ohmic treated	-160.86	0.16	71.53	10.36	122.98	19.2	3.24	2.54
PL treated	-134.89	0.12	72.57	10.48	123.03	19.32	2.42	2.16
C1 (OH1PL1)	-192.45	0.19	70.39	10.27	122.69	19.21	5.13	4.95
C2 (OH1PL2)	-191.28	0.2	69.55	10.25	122.73	19.18	5.11	4.92
C3 (OH2PL1)	-196.85	0.21	68.78	10.18	122.56	19.22	5.04	4.89
C4 (OH2PL2)	-193.79	0.21	70.15	10.19	122.81	19.21	4.96	4.9

Appendix A.28

ANOVA for the effect of treatments on physiochemical and microbiological properties of pineapple juice

		Sum of Squares	df	Mean Square	F	Sig.
pH	Between Groups	.296	7	.042	91.510	.000
	Within Groups	.007	16	.000		
	Total	.304	23			
TSS	Between Groups	3.077	7	.440	462.744	.000
	Within Groups	.015	16	.001		
	Total	3.092	23			
Titrable acidity	Between Groups	.000	7	.000	11.071	.000
	Within Groups	.000	16	.000		
	Total	.000	23			
Ascorbic acid	Between Groups	303.092	7	43.299	31.097	.000
	Within Groups	22.278	16	1.392		
	Total	325.370	23			
Total sugar	Between Groups	.467	7	.067	121.247	.000
	Within Groups	.009	16	.001		
	Total	.476	23			
Phenolic content	Between Groups	18.769	7	2.681	37.245	.000
	Within Groups	1.152	16	.072		
	Total	19.921	23			
Total Colour Difference	Between Groups	4.225	7	.604	58.509	.000
	Within Groups	.165	16	.010		
	Total	4.390	23			

		Sum of Squares	df	Mean Square	F	Sig.
Browning Index	Between Groups	.148	7	.021	68.571	.000
	Within Groups	.005	16	.000		
	Total	.153	23			
Antioxidant Activity	Between Groups	493.262	7	70.466	23.112	.000
	Within Groups	48.782	16	3.049		
	Total	542.045	23			
Ca	Between Groups	.030	7	.004	.593	.752
	Within Groups	.115	16	.007		
	Total	.145	23			
K	Between Groups	7.937	7	1.134	.133	.994
	Within Groups	136.721	16	8.545		
	Total	144.658	23			
Na	Between Groups	9.224	7	1.318	.5914	.082
	Within Groups	3.565	16	2.23		
	Total	12.789	23			
Bacterial Reduction	Between Groups	77.098	7	11.014	616.884	.000
	Within Groups	.286	16	.018		
	Total	77.383	23			
Yeast and Mould Reduction	Between Groups	77.069	7	11.010	2.028E3	.000
	Within Groups	.087	16	.005		
	Total	77.156	23			

Appendix. A.29

ANOVA for the effect of treatments on physiochemical and microbiological properties of cashew apple juice

		Sum of Squares	df	Mean Square	F	Sig.
pH	Between Groups	.081	7	.012	31.189	.000
	Within Groups	.006	16	.000		
	Total	.087	23			
TSS	Between Groups	4.663	7	.666	1.615E3	.000
	Within Groups	.007	16	.000		
	Total	4.669	23			
Titrableacidity	Between Groups	.000	7	.000	1.973	.123
	Within Groups	.000	16	.000		
	Total	.000	23			
Ascorbicacid	Between Groups	4076.380	7	582.340	94.509	.000
	Within Groups	98.588	16	6.162		
	Total	4174.968	23			
Totalsugar	Between Groups	.875	7	.125	146.319	.000
	Within Groups	.014	16	.001		
	Total	.889	23			
Phenolic content	Between Groups	29.892	7	4.270	2.784	.043
	Within Groups	24.544	16	1.534		
	Total	54.436	23			
Tannin	Between Groups	.015	7	.002	4.806	.004
	Within Groups	.007	16	.000		
	Total	.022	23			

		Sum of Squares	df	Mean Square	F	Sig.
Total Colour Difference	Between Groups	5.601	7	.800	741.502	.000
	Within Groups	.017	16	.001		
	Total	5.619	23			
Browning Index	Between Groups	.156	7	0.022	95.633	0.000
	Within Groups	.004	16	0.000		
	Total	.160	23			
Antioxidant Activity	Between Groups	594.834	7	84.976	28.415	0.000
	Within Groups	47.849	16	2.991		
	Total	642.683	23			
Ca	Between Groups	.019	7	0.003	0.189	0.984
	Within Groups	.232	16	0.014		
	Total	.251	23			
K	Between Groups	.659	7	0.094	1.527	0.228
	Within Groups	.986	16	.062		
	Total	1.645	23			
Na	Between Groups	.020	7	0.003	0.003	1.000
	Within Groups	12.918	16	0.807		
	Total	12.938	23			
Bacterial Reduction	Between Groups	71.977	7	10.282	4.360E3	0.000
	Within Groups	.038	16	.002		
	Total	72.015	23			
Yeast and Mould Reduction	Between Groups	71.682	7	10.240	6.238E3	0.000
	Within Groups	.026	16	.002		
	Total	71.708	23			

Appendix. A.30 Changes in physicochemical properties during storage of pineapple juice

Parameter	Storage days	Treatments						
		Control	Ohmic treated	PL treated	P_1 (OH_1PL_1)	P_2 (OH_1PL_2)	P_3 (OH_2PL_1)	P_4 (OH_2PL_2)
pH	0	4.45	4.26	4.45	4.26	4.27	4.28	4.27
	5	4.48	4.26	4.45	4.26	4.27	4.28	4.27
	10	4.5	4.27	4.46	4.26	4.27	4.28	4.27
	15	4.51	4.28	4.47	4.27	4.28	4.29	4.28
	20	4.52	4.31	4.5	4.28	4.29	4.29	4.28
	25	4.53	4.34	4.51	4.29	4.3	4.3	4.29
	30	4.54	4.35	4.52	4.3	4.31	4.31	4.31
	35	4.55	4.36	4.53	4.33	4.34	4.34	4.34
TSS	0	10.82	11.15	10.82	11.16	11.15	11.14	11.16
	5	10.25	11.15	10.82	11.16	11.15	11.14	11.16
	10	10.04	11.12	10.75	11.15	11.14	11.12	11.14
	15	9.76	11.05	10.42	11.09	11.09	11.06	11.08
	20	9.62	10.86	9.68	11.01	11.03	10.99	10.99
	25	9.58	10.43	9.6	10.98	10.99	10.95	10.95
	30	9.53	10.1	9.54	10.95	10.92	10.9	10.91
	35	9.51	9.96	9	10.82	10.81	10.73	10.75
Titrable acidity	0	0.382	0.388	0.382	0.387	0.386	0.384	0.385
	5	0.382	0.388	0.382	0.387	0.386	0.384	0.385
	10	0.364	0.387	0.38	0.387	0.386	0.384	0.385
	15	0.342	0.378	0.379	0.386	0.384	0.382	0.384
	20	0.312	0.354	0.357	0.382	0.381	0.379	0.381
	25	0.287	0.332	0.286	0.379	0.378	0.376	0.377
	30	0.253	0.312	0.257	0.375	0.374	0.374	0.371
	35	0.231	0.301	0.216	0.371	0.37	0.37	0.368

Parameter	Storage days	Treatments						
		Control	Ohmic treated	PL treated	P_1 (OH_1PL_1)	P_2 (OH_1PL_2)	P_3 (OH_2PL_1)	P_4 (OH_2PL_2)
Ascorbic acid content	0	31.18	28.75	28.96	27.13	26.76	26.24	26.94
	5	28.32	28.73	28.95	27.13	26.76	26.24	26.94
	10	27.64	28.73	27.94	26.32	26.03	26.65	25.67
	15	25.65	26.36	25.65	25.93	25.89	25.19	24.98
	20	21.65	23.45	22.45	25.03	25.18	24.98	24.15
	25	19.54	21.32	19.38	24.82	24.74	24.76	23.98
	30	16.89	19.36	17.45	24.46	24.12	23.74	23.72
	35	14.32	18.43	15.27	21.67	20.34	19.97	19.89
Total Sugar	0	10.53	10.54	10.53	10.54	10.55	10.55	10.54
	5	10.46	10.52	10.48	10.54	10.54	10.54	10.53
	10	10.21	10.34	10.09	10.52	10.51	10.52	10.51
	15	9.99	10	9.42	10.41	10.48	10.49	10.46
	20	9.68	9.58	9.0	10.26	10.32	10.36	10.38
	25	9.12	9.0	8.63	10.08	10.12	10.18	10.13
	30	8.26	8.82	8.33	9.68	9.97	9.98	9.97
	35	8.04	8.52	8.1	9.12	9.08	9.1	9.07
Total phenolic content	0	67.25	68.48	67.14	67.18	67.22	67.21	67.19
	5	52.39	66.39	66.1	67.05	67.12	67.02	66.02
	10	48.87	64.32	65.42	66.48	66.08	65.79	64.89
	15	45.31	62.67	60.45	64.87	63.76	63.95	62.98
	20	41.04	58.45	48.12	62.23	60.83	62.76	60.32
	25	39.45	56.89	42.86	60.89	58.16	60.89	58.53
	30	36.32	46.15	40.06	59.68	56.78	58.32	57.45
	35	32.45	41.45	36.21	55.95	52.36	53.42	52.53
Total Colour difference	0	0	0.68	0.46	0.74	0.78	0.76	0.74
	5	0.87	0.69	0.52	0.75	0.78	0.76	0.75
	10	0.98	0.72	0.63	0.78	0.79	0.78	0.78
	15	1.5	0.89	0.78	0.79	0.8	0.8	0.8
	20	1.8	1.24	1.02	0.81	0.83	0.82	0.82
	25	2.1	1.48	1.65	0.82	0.85	0.84	0.84
	30	2.3	1.56	1.93	0.83	0.87	0.86	0.85
	35	2.43	1.64	1.98	0.89	0.92	0.91	0.9

Parameter	Storage days	Treatments						
		Control	Ohmic treated	PL treated	P_1 (OH_1PL_1)	P_2 (OH_1PL_2)	P_3 (OH_2PL_1)	P_4 (OH_2PL_2)
Browning Index	0	0	0.16	0.12	0.19	0.2	0.21	0.21
	5	0.52	0.18	0.14	0.19	0.22	0.23	0.22
	10	0.55	0.21	0.2	0.21	0.23	0.24	0.23
	15	0.67	0.24	0.25	0.22	0.24	0.25	0.24
	20	0.76	0.26	0.41	0.23	0.25	0.26	0.26
	25	0.79	0.35	0.46	0.24	0.26	0.27	0.27
	30	0.83	0.38	0.49	0.26	0.27	0.27	0.28
	35	0.86	0.42	0.53	0.32	0.33	0.33	0.34
Antioxidant Activity	0	75.36	71.53	72.57	70.39	69.55	68.78	70.15
	5	53.79	57.43	58.65	73.69	68.32	66.23	67.98
	10	48.58	54.54	55.69	70.31	66.23	65.89	65.09
	15	43.78	50.78	48.87	67.89	64.78	63.89	64.23
	20	35.65	48.69	42.45	64.29	62.68	59.52	62.68
	25	32.54	46.58	38.95	60.54	57.46	56.79	58.38
	30	29.49	43.28	32.21	56.58	54.32	54.21	55.31
	35	27.21	41.28	30.32	50.34	49.95	49.4	49.98

Appendix. A.31 Changes in physicochemical properties during storage of cashew apple juice

Parameter	Storage days	Treatments						
		Control	Ohmic treated	PL treated	C_1 (OH_1PL_1)	C_2 (OH_1PL_2)	C_3 (OH_2PL_1)	C_4 (OH_2PL_2)
pH	0	4.45	4.26	4.45	4.3	4.27	4.28	4.27
	5	4.48	4.26	4.45	4.3	4.27	4.28	4.27
	10	4.5	4.27	4.46	4.3	4.27	4.28	4.27
	15	4.51	4.28	4.47	4.3	4.28	4.29	4.28
	20	4.52	4.31	4.5	4.3	4.29	4.29	4.28
	25	4.53	4.34	4.51	4.3	4.3	4.3	4.29
	30	4.54	4.35	4.52	4.3	4.31	4.31	4.31
	35	4.55	4.36	4.53	4.3	4.34	4.34	4.34
TSS	0	10.82	11.15	10.82	11	11.15	11.14	11.16
	5	10.25	11.15	10.82	11	11.15	11.14	11.16
	10	10.04	11.12	10.75	11	11.14	11.12	11.14
	15	9.76	11.05	10.42	11	11.09	11.06	11.08
	20	9.62	10.86	9.68	11	11.03	10.99	10.99
	25	9.58	10.43	9.6	11	10.99	10.95	10.95
	30	9.53	10.1	9.54	11	10.92	10.9	10.91
	35	9.51	9.96	9	11	10.81	10.73	10.75
Titrable acidity	0	0.422	0.424	0.422	0.4	0.424	0.422	0.423
	5	0.415	0.424	0.422	0.4	0.424	0.422	0.423
	10	0.406	0.422	0.421	0.4	0.423	0.421	0.422
	15	0.398	0.42	0.42	0.4	0.422	0.42	0.421
	20	0.396	0.418	0.413	0.4	0.421	0.418	0.42
	25	0.393	0.416	0.409	0.4	0.42	0.416	0.418
	30	0.389	0.403	0.398	0.4	0.418	0.415	0.416
	35	0.385	0.398	0.395	0.4	0.409	0.41	0.409

Parameter	Storage days	Treatments						
		Control	Ohmic treated	PL treated	C_1 (OH_1PL_1)	C_2 (OH_1PL_2)	C_3 (OH_2PL_1)	C_4 (OH_2PL_2)
Ascorbic acid content	0	168.46	159.74	162.96	150	148.76	147.7	149.23
	5	160.38	157.87	160.87	150	148.65	147.1	148.95
	10	141.34	154.64	157.28	148	147.32	147	148.31
	15	127.42	151.78	153.75	148	146.29	145.8	147.84
	20	119.67	149.42	142.86	145	145.07	143.2	145.38
	25	108.21	142.28	138.54	144	142.31	140.5	142.67
	30	102.27	130.46	126.56	142	139.58	138.3	140.26
	35	98.24	126.56	119.54	138	135.34	134.3	135.76
Total sugar	0	9.96	9.97	9.96	9.97	9.96	9.97	9.97
	5	9.94	9.96	9.94	9.96	9.95	9.95	9.93
	10	9.03	9.91	9.9	9.83	9.87	9.89	9.88
	15	8.86	9.69	9	9.64	9.68	9.62	9.65
	20	8.59	9.33	8.82	9.32	9.42	9.43	9.48
	25	8.36	8.9	8.53	9.04	8.99	9	9.03
	30	8.08	8.85	8.35	8.94	8.93	8.94	8.93
	35	7.86	8.09	8.01	8.55	8.53	8.47	8.48
Total Phenolic content	0	168.46	159.74	162.96	156.02	148.76	147.72	149.23
	5	150.78	167.59	165.73	166.23	165.48	166.12	165.75
	10	146.32	164.38	161.58	163.74	162.76	164.21	161.86
	15	139.65	156.76	157.56	158.28	158.95	160.39	157.84
	20	124.76	148.76	126.67	152.86	150.73	156.32	153.43
	25	112.78	130.04	118.96	148.12	148.32	149.43	149.76
	30	104.67	121.48	109.56	147.34	145.23	145.46	144.72
	35	97.65	112.89	102.78	141.78	138.54	139.65	138.12
Tannin content	0	0.57	0.55	0.55	0.54	0.52	0.53	0.51
	5	0.51	0.54	0.54	0.53	0.51	0.52	0.5
	10	0.43	0.52	0.52	0.5	0.49	0.5	0.48
	15	0.4	0.5	0.5	0.48	0.47	0.48	0.46
	20	0.38	0.49	0.41	0.45	0.46	0.46	0.44
	25	0.35	0.47	0.38	0.43	0.44	0.44	0.43
	30	0.32	0.45	0.34	0.42	0.41	0.42	0.41
	35	0.3	0.42	0.32	0.37	0.36	0.35	0.36

Parameter	Storage days	Treatments						
		Control	Ohmic treated	PL treated	C_1 (OH_1PL_1)	C_2 (OH_1PL_2)	C_3 (OH_2PL_1)	C_4 (OH_2PL_2)
Total Colour Difference	0	0	0.63	0.42	0.79	0.82	0.85	0.81
	5	0.75	0.68	0.56	0.8	0.83	0.85	0.82
	10	0.97	0.83	0.62	0.82	0.84	0.87	0.84
	15	1.56	0.95	0.84	0.83	0.87	0.89	0.86
	20	2	1	1.76	0.86	0.9	0.91	0.89
	25	2.3	1.5	1.83	0.88	0.92	0.93	0.9
	30	2.6	1.7	2.1	0.91	0.94	0.95	0.92
	35	2.8	1.9	2.4	0.95	0.98	0.99	0.97
Browning Index	0	0	0.18	0.14	0.2	0.22	0.22	0.21
	5	0.5	0.25	0.24	0.29	0.26	0.27	0.24
	10	0.67	0.3	0.29	0.34	0.29	0.31	0.27
	15	0.73	0.36	0.33	0.39	0.35	0.38	0.34
	20	0.79	0.4	0.69	0.43	0.41	0.44	0.39
	25	0.85	0.45	0.76	0.49	0.48	0.49	0.46
	30	0.92	0.68	0.82	0.53	0.56	0.54	0.55
	35	0.95	0.72	0.85	0.6	0.62	0.61	0.63
Antioxidant Activity	0	82.66	77.63	79.34	75.85	74.86	74.19	73.98
	5	68.94	74.78	77.89	74.46	74.19	72.09	72.45
	10	62.43	68.59	74.89	73.39	72.89	70.76	70.9
	15	58.87	65.19	70.39	71.32	70.95	68.9	68.87
	20	52.89	62.19	58.32	69.89	69.6	66.76	66.98
	25	43.48	59.17	52.18	68.98	67.39	65.98	64.09
	30	39.87	56.78	43.87	67.78	65.23	64.82	63.76
	35	35.54	53.23	45.85	54.28	53.12	52.67	51.75

Appendix.A.32
ANOVA for linear model-Storage studies of pineapple and cashew apple juice treated under different conditions

ANOVA of pH values during storage of pineapple juice treated under different conditions

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	1.596[a]	55	.029	1.080	.360
Intercept	3180.546	1	3180.546	1.184E5	.000
treatment	1.467	6	.244	9.099	.000
days	.113	7	.016	.603	.752
treatment * days	.016	42	.000	.014	1.000
Error	3.009	112	.027		
Total	3185.151	168			
Corrected Total	4.605	167			

a. R Squared = 0.347 (Adjusted R Squared = .026)

ANOVA of pH values during storage of cashew apple juice treated under different conditions

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	1.868[a]	55	.034	1.261	.151
Intercept	3180.807	1	3180.807	1.180E5	.000
treatment	1.260	6	.210	7.795	.000
days	.321	7	.046	1.704	.115
treatment * days	.287	42	.007	.253	1.000
Error	3.018	112	.027		
Total	3185.694	168			
Corrected Total	4.886	167			

a. R Squared = .682 (Adjusted R Squared = .079)

ANOVA of TSS content during storage of pineapple juice treated under different conditions

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	56.708[a]	55	1.031	6.189	.000
Intercept	19178.507	1	19178.507	1.151E5	.000
treatment	35.137	6	5.856	35.152	.000
days	13.070	7	1.867	11.208	.000
treatment * days	8.501	42	.202	1.215	.210
Error	18.658	112	.167		
Total	19253.873	168			
Corrected Total	75.366	167			

a. R Squared = .752 (Adjusted R Squared = .731)

ANOVA of TSS content during storage of cashew apple juice treated under different conditions

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	49.530[a]	55	.901	10.312	.000
Intercept	9837.748	1	9837.748	1.127E5	.000
treatment	25.091	6	4.182	47.886	.000
days	17.800	7	2.543	29.118	.000
treatment * days	6.640	42	.158	1.810	.007
Error	9.781	112	.087		
Total	9897.059	168			
Corrected Total	59.311	167			

a. R Squared = 0.835 (Adjusted R Squared = 0.754)

ANOVA of titrable acidity during storage of pineapple juice treated under different conditions

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	.297[a]	55	.005	28.960	.000
Intercept	21.859	1	21.859	1.172E5	.000
treatment	.102	6	.017	91.048	.000
days	.101	7	.014	77.542	.000
treatment * days	.094	42	.002	11.994	.000
Error	.021	112	.000		
Total	22.177	168			
Corrected Total	.318	167			

a. R Squared = 0.934 (Adjusted R Squared = 0.902)

ANOVA of titrable acidity during storage of cashew apple juice treated under different conditions

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	.017[a]	55	.000	1.233	.175
Intercept	28.954	1	28.954	1.169E5	.000
treatment	.007	6	.001	4.775	.000
days	.007	7	.001	4.169	.000
treatment * days	.002	42	5.898E-5	.238	1.000
Error	.028	112	.000		
Total	28.998	168			
Corrected Total	.045	167			

a. R Squared = 0.377 (Adjusted R Squared = 0.071)

ANOVA of total sugar content during storage of pineapple juice treated under different conditions

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	94.272[a]	55	1.714	11.996	.000
Intercept	16454.990	1	16454.990	1.152E5	.000
treatment	20.559	6	3.427	23.982	.000
days	62.161	7	8.880	62.151	.000
treatment * days	11.552	42	.275	1.925	.003
Error	16.003	112	.143		
Total	16565.264	168			
Corrected Total	110.275	167			

a. R Squared = 0.855 (Adjusted R Squared = 0.784)

ANOVA of total sugar content during storage of cashew apple juice treated under different conditions

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	67.818[a]	55	1.233	9.827	.000
Intercept	14435.059	1	14435.059	1.150E5	.000
treatment	7.624	6	1.271	10.126	.000
days	56.366	7	8.052	64.172	.000
treatment * days	3.828	42	.091	.726	.880
Error	14.054	112	.125		
Total	14516.930	168			
Corrected Total	81.871	167			

a. R Squared = 0.828 (Adjusted R Squared = 0.744)

ANOVA of ascorbic acid content during storage of pineapple juice treated under different conditions

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	2240.786[a]	55	40.742	57.325	.000
Intercept	99546.274	1	99546.274	1.401E5	.000
treatment	96.039	6	16.007	22.522	.000
days	1646.035	7	235.148	330.861	.000
treatment * days	498.711	42	11.874	16.707	.000
Error	79.600	112	.711		
Total	101866.660	168			
Corrected Total	2320.386	167			

a. R Squared = 0.966 (Adjusted R Squared = 0.949)

ANOVA of ascorbic acid content during storage of cashew apple juice treated under different conditions

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	31132.646[a]	55	566.048	19.110	.000
Intercept	3413117.454	1	3413117.454	1.152E5	.000
treatment	5902.718	6	983.786	33.214	.000
days	15571.658	7	2224.523	75.103	.000
treatment * days	9658.270	42	229.959	7.764	.000
Error	3317.412	112	29.620		
Total	3447567.512	168			
Corrected Total	34450.058	167			

a. R Squared = .904 (Adjusted R Squared = .856)

ANOVA of total phenolic content during storage of pineapple juice treated under different conditions

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	15912.439[a]	55	289.317	59.503	.000
Intercept	562423.501	1	562423.501	1.157E5	.000
treatment	5978.653	6	996.442	204.934	.000
days	7834.133	7	1119.162	230.173	.000
treatment * days	2099.654	42	49.992	10.282	.000
Error	544.574	112	4.862		
Total	578880.515	168			
Corrected Total	16457.013	167			

a. R Squared = 0.967 (Adjusted R Squared = 0.951)

ANOVA of total phenolic content during storage of cashew apple juice treated under different conditions

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	55677.487[a]	55	1012.318	31.970	.000
Intercept	3615914.423	1	3615914.423	1.142E5	.000
treatment	12118.116	6	2019.686	63.783	.000
days	31453.138	7	4493.305	141.903	.000
treatment * days	12106.233	42	288.244	9.103	.000
Error	3546.450	112	31.665		
Total	3675138.361	168			
Corrected Total	59223.938	167			

a. R Squared = 0.940 (Adjusted R Squared = 0.911)

ANOVA of tannin content during storage of cashew apple juice					
Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	.758[a]	55	.014	46.061	.000
Intercept	34.644	1	34.644	1.157E5	.000
treatment	.095	6	.016	52.901	.000
days	.588	7	.084	280.744	.000
treatment * days	.075	42	.002	5.970	.000
Error	.034	112	.000		
Total	35.436	168			
Corrected Total	.792	167			

a. R Squared = 0.958 (Adjusted R Squared = .937)

ANOVA of total colour difference during storage of pineapple juice treated under different conditions

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	36.881[a]	55	.671	384.429	.000
Intercept	167.341	1	167.341	9.593E4	.000
treatment	9.935	6	1.656	949.275	.000
days	12.412	7	1.773	1.017E3	.000
treatment * days	14.534	42	.346	198.391	.000
Error	.195	112	.002		
Total	204.417	168			
Corrected Total	37.076	167			

a. R Squared = 0.995 (Adjusted R Squared = 0.992)

ANOVA of total colour difference during storage of cashew apple juice treated under different conditions

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	50.832[a]	55	.924	460.794	.000
Intercept	198.621	1	198.621	9.903E4	.000
treatment	12.343	6	2.057	1.026E3	.000
days	17.625	7	2.518	1.255E3	.000
treatment * days	20.864	42	.497	247.671	.000
Error	.225	112	.002		
Total	249.678	168			
Corrected Total	51.057	167			

a. R Squared = 0.996 (Adjusted R Squared = 0.993)

ANOVA of browning index during storage of pineapple juice treated under different conditions

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	5.300[a]	55	.096	554.237	.000
Intercept	16.878	1	16.878	9.708E4	.000
treatment	2.736	6	.456	2.623E3	.000
days	1.305	7	.186	1.072E3	.000
treatment * days	1.259	42	.030	172.376	.000
Error	.019	112	.000		
Total	22.198	168			
Corrected Total	5.319	167			

a. R Squared = 0.996 (Adjusted R Squared = 0.995)

ANOVA of browning index during storage of cashew apple juice treated under different conditions

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	7.851[a]	55	.143	384.964	.000
Intercept	35.328	1	35.328	9.527E4	.000
treatment	1.588	6	.265	713.613	.000
days	5.116	7	.731	1.971E3	.000
treatment * days	1.148	42	.027	73.686	.000
Error	.042	112	.000		
Total	43.221	168			
Corrected Total	7.893	167			

a. R Squared = 0.995 (Adjusted R Squared = 0.992)

ANOVA of antioxidant activity during storage of pineapple juice treated under different conditions

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	25692.169[a]	55	467.130	99.607	.000
Intercept	523510.104	1	523510.104	1.116E5	.000
treatment	9741.331	6	1623.555	346.194	.000
days	13134.647	7	1876.378	400.104	.000
treatment * days	2816.192	42	67.052	14.298	.000
Error	525.250	112	4.690		
Total	549727.523	168			
Corrected Total	26217.419	167			

a. R Squared = .980 (Adjusted R Squared = 0.970)

ANOVA of antioxidant activity during storage of cashew apple juice treated under different conditions

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	11719.023[a]	55	213.073	32.625	.000
Intercept	743816.059	1	743816.059	1.139E5	.000
treatment	720.066	6	120.011	18.376	.000
days	9155.185	7	1307.884	200.257	.000
treatment * days	1843.772	42	43.899	6.722	.000
Error	731.475	112	6.531		
Total	756266.558	168			
Corrected Total	12450.498	167			

a. R Squared = 0.941 (Adjusted R Squared = 0.912)

Appendix A.33 Changes in microbial properties of pineapple and cashew apple juice during storage

Parameter	Storage days	Treatments						
		Control	Ohmic treated	PL treated	(OH_1PL_1)	(OH_1PL_2)	(OH_2PL_1)	(OH_2PL_2)
Bacterial log reduction (Pineapple)	0	4.06	2.43	2.72	2.04	2.09	2.1	2.11
	5	6.65	2.86	3.95	2.38	2.32	2.38	2.42
	10	10.89	3.26	5.13	3.05	3.12	3.15	3.01
	15	12.99	4.52	6.34	3.88	3.96	3.97	3.89
	20	15.34	4.91	8.86	4.67	4.78	4.69	4.89
	25	18.64	6.74	10.47	5.02	5.08	5.07	5.14
	30	19.99	8.12	13.78	5.85	5.94	5.78	5.94
	35	20.54	12.08	16.54	7.21	7.56	7.43	7.52
Yeast and mold reduction (Pineapple)	0	3.78	1.89	2.02	1.34	1.67	1.56	1.72
	5	4.67	2.02	2.59	1.98	1.73	1.98	1.96
	10	5.21	2.56	3.67	2.16	2.12	2.16	2.45
	15	5.76	3.14	4.26	2.37	2.51	2.49	2.98
	20	6.12	3.67	4.32	2.89	2.98	2.96	3.04
	25	7.23	5.21	4.39	3.12	3.27	3.32	3.38
	30	9.85	7.58	4.43	3.58	3.89	3.81	3.79
	35	11.06	9.09	10.59	5.12	5.24	5.25	5.98
Bacterial log reduction (Cashew apple)	0	4.87	2.44	2.86	2.33	2.41	2.44	2.52
	5	6.43	2.83	3.45	2.78	2.93	2.98	2.89
	10	11.74	3.64	4.98	3.09	3.18	3.26	3.03
	15	17.08	4.67	5.58	3.98	3.95	3.92	3.99
	20	21.25	4.9	11.89	4.79	4.56	4.73	4.78
	25	24.89	5.48	17.08	5.05	5.09	5.08	5.12
	30	26.42	12.04	20.43	5.89	5.92	5.91	5.9
	35	31.79	18.38	21.12	9.98	9.99	10.12	10.32
Yeast and mold reduction (Cashew apple)	0	3.47	1.53	2.59	1.24	1.32	1.36	1.42
	5	4.23	2.18	3.05	1.88	1.97	2.04	2.09
	10	5.54	2.96	3.55	2.48	2.62	2.54	2.62
	15	6.04	3.12	3.95	2.64	2.75	2.62	2.74
	20	7.26	3.57	5.53	2.89	2.89	2.76	2.96
	25	8.48	3.98	8.98	3.01	3	2.95	3.02
	30	10.62	6.12	10.21	3.16	3.12	3.01	3.14
	35	12.46	9.57	12.34	5.27	5.42	4.34	5.56

Appendix.A.34
ANOVA for Microbial count during storage of pineapple and cashew apple juice treated under different conditions

ANOVA of bacterial log count during storage of pineapple juice treated under different conditions

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	3584.468[a]	55	65.172	710.257	.000
Intercept	6952.503	1	6952.503	7.577E4	.000
treatment	1787.968	6	297.995	3.248E3	.000
days	1348.573	7	192.653	2.100E3	.000
treatment * days	447.927	42	10.665	116.228	.000
Error	10.277	112	.092		
Total	10547.249	168			
Corrected Total	3594.745	167			

a. R Squared = 0.997 (Adjusted R Squared = 0.996)

ANOVA of bacterial log count during storage of cashew apple juice treated under different conditions

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	8065.158[a]	55	146.639	936.789	.000
Intercept	10331.396	1	10331.396	6.600E4	.000
treatment	3660.397	6	610.066	3.897E3	.000
days	2975.782	7	425.112	2.716E3	.000
treatment * days	1428.980	42	34.023	217.354	.000
Error	17.532	112	.157		
Total	18414.085	168			
Corrected Total	8082.689	167			

a. R Squared = 0.998 (Adjusted R Squared = 0.997)

ANOVA of yeast and mold log count during storage of pineapple juice treated under different conditions

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	846.601[a]	55	15.393	520.147	.000
Intercept	2590.736	1	2590.736	8.755E4	.000
treatment	290.769	6	48.461	1.638E3	.000
days	460.913	7	65.845	2.225E3	.000
treatment * days	94.919	42	2.260	76.368	.000
Error	3.314	112	.030		
Total	3440.651	168			
Corrected Total	849.915	167			

a. R Squared = 0.996 (Adjusted R Squared = 0.994)

ANOVA of yeast and mold log count during storage of cashew apple juice treated under different conditions

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Corrected Model	1245.879[a]	55	22.652	615.329	.000
Intercept	2887.407	1	2887.407	7.843E4	.000
treatment	506.797	6	84.466	2.294E3	.000
days	542.380	7	77.483	2.105E3	.000
treatment * days	196.702	42	4.683	127.220	.000
Error	4.123	112	.037		
Total	4137.409	168			
Corrected Total	1250.002	167			

a. R Squared = 0.997 (Adjusted R Squared = 0.995)

Appendix A.35 Sensory score card for sensory analysis

SENSORY SCORE CARD

Department of processing and Food Engineering
KCAET, Tavanur

Name of judge: **Date:**

You are requested to assess the product in terms of general acceptability on a 9 point hedonic scale

Characteristics	**Sample code**						
	A	B	C	D	E	F	G
Colour & Appearance							
Flavour							
Taste							
Overall acceptability							

Score system:

Dislike extremely: 1, Dislike very much: 2, Dislike moderately: 3, Dislike slightly: 4

Neither like nor dislike: 5, Like slightly: 6, Like moderately: 7, Like very much: 8

Like extremely: 9

Comments if any:
Signature

Appendix A. 36 Scale factor, fuzzy membership and normalized membership functions for quality attributes of

			Sample C			Sample OH			Sample PL			Sample P1			Sample P2			Sample P3			Sample P4		
Sensory attributes	**SF**	**SF**		**FMF**	**NFM**		**FMF**	**NFF**		**FMF**	**NFMF**		**FMF**	**NFF**		**FMF**	**NFMF**		**FMF**	**NFMF**		**FMF**	**NFM**
Colour and appearance	**EX**	1	13	0.65	0.65	0	0	0	0	0	0	7	0.35	0.35	5	0.25	0.25	6	0.3	0.3	6	0.3	0.3
	GD	0.9	7	0.35	0.31	7	0.35	0.31	6	0.3	0.27	13	0.65	0.58	15	0.75	0.67	14	0.7	0.63	13	0.65	0.58
	MD	0.7	0	0	0	13	0.65	0.45	14	0.7	0.49	0	0	0	0	0	0	0	0	0	1	0.05	0.03
	FR	0.4	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
	NS	0.1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Total			**20**	**1**	**0.96**	**20**	**1**	**0.77**	**20**	**1**	**0.76**	**20**	**1**	**0.93**	**20**	**1**	**0.925**	**20**	**1**	**0.93**	**20**	**1**	**0.92**
Flavour	**EX**	1	12	0.6	0.6	0	0	0	0	0	0	10	0.5	0.5	7	0.35	0.35	6	0.3	0.3	8	0.4	0.4
	GD	0.9	8	0.4	0.36	8	0.4	0.36	9	0.45	0.405	10	0.5	0.45	11	0.55	0.495	13	0.65	0.59	10	0.5	0.45
	MD	0.7	0	0	0	12	0.6	0.42	11	0.55	0.385	0	0	0	2	0.1	0.07	1	0.05	0.04	2	0.1	0.07
	FR	0.4	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
	NS	0.1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Total			**20**	**1**	**0.96**	**20**	**1**	**0.78**	**20**	**1**	**0.79**	**20**	**1**	**0.95**	**20**	**1**	**0.915**	**20**	**1**	**0.92**	**20**	**1**	**0.92**
Taste	**EX**	1	8	0.4	0.4	0	0	0	0	0	0	7	0.35	0.35	6	0.3	0.3	7	0.35	0.35	6	0.3	0.3
	GD	0.9	12	0.6	0.54	10	0.5	0.45	8	0.4	0.36	13	0.65	0.585	12	0.6	0.54	11	0.55	0.5	14	0.7	0.63
	MD	0.7	0	0	0	10	0.5	0.35	12	0.6	0.42	0	0	0	2	0.1	0.07	2	0.1	0.07	0	0	0
	FR	0.4	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
	NS	0.1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Total			**20**	**1**	**0.94**	**20**	**1**	**0.8**	**20**	**1**	**0.78**	**20**	**1**	**0.93**	**20**	**1**	**0.91**	**20**	**1**	**0.92**	**20**	**1**	**0.93**
Overall acceptability	**EX**	1	8	0.4	0.4	0	0	0	0	0	0	5	0.25	0.25	2	0.1	0.1	2	0.1	0.1	5	0.25	0.25
	GD	0.9	12	0.6	0.54	8	0.4	0.36	9	0.45	0.405	15	0.75	0.675	12	0.6	0.54	15	0.75	0.68	12	0.6	0.54
	MD	0.7	0	0	0	12	0.6	0.42	11	0.55	0.385	0	0	0	6	0.3	0.21	3	0.15	0.11	2	0.13	0.09
	FR	0.4	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
	NS	0.1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Total			**20**	**1**	**0.94**	**20**	**1**	**0.78**	**20**	**1**	**0.79**	**20**	**1**	**0.92**	**20**	**1**	**0.85**	**20**	**1**	**0.88**	**19**	**0.98**	**0.88**

SF: Scale factor; FMF: Fuzzy membership functions; NFMF: Normalized fuzzy membership functions; EX: Excellent; GD: Good: MD: Medium; FR: Fair; NS: Not satisfactory

pineapple juice samples treated at different conditions

Appendix A. 37 Scale factor, fuzzy membership and normalized membership functions for quality attributes of cashew apple juice samples treated at different conditions

			Sample C			Sample OH			Sample PL			Sample C1			Sample C2			Sample C3			Sample C4		
Sensory attributes	**SF**	**SF**		**FMF**	**NFMF**		**FMF**	**NFMF**		**FMF**	**NFMF**		**FMF**	**NFMF**		**FMF**	**NFMF**		**FMF**	**NFMF**		**FMF**	**NFMF**
Colour and appearance	**EX**	1.0	12	0.60	0.60	0	0	0	0	0	0	8	0.4	0.4	5	0.25	0.25	4	0.2	0.2	4	0.2	0.2
	GD	0.9	8	0.40	0.36	8	0.4	0.36	6	0.3	0.27	12	0.6	0.54	10	0.5	0.45	13	0.65	0.585	16	0.8	0.72
	MD	0.7	0	0.0	0.00	10	0.5	0.35	11	0.55	0.385	0	0	0	5	0.25	0.175	3	0.15	0.105	0	0	0
	FR	0.4	0	0.0	0.00	2	0.1	0.04	3	0.15	0.06	0	0	0	0	0	0	0	0	0	0	0	0
	NS	0.1	0	0.0	0.00	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Total			**20**	**1.0**	**0.96**	**20**	**1**	**0.75**	**20**	**1**	**0.715**	**20**	**1**	**0.94**	**20**	**1**	**0.875**	**20**	**1**	**0.89**	**20**	**1**	**0.92**
Flavour	**EX**	1	11	0.55	0.55	0	0	0	0	0	0	6	0.3	0.3	4	0.2	0.2	5	0.25	0.25	4	0.2	0.2
	GD	0.9	9	0.45	0.405	9	0.45	0.405	7	0.35	0.315	14	0.7	0.63	12	0.6	0.54	12	0.6	0.54	14	0.7	0.63
	MD	0.7	0	0	0	8	0.4	0.28	10	0.5	0.35	0	0	0	4	0.2	0.14	3	0.15	0.105	2	0.1	0.07
	FR	0.4	0	0	0	3	0.15	0.06	3	0.2	0.08	0	0	0	0	0	0	0	0	0	0	0	0
	NS	0.1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Total			**20**	**1**	**0.955**	**20**	**1**	**0.745**	**20**	**1.05**	**0.745**	**20**	**1**	**0.93**	**20**	**1**	**0.88**	**20**	**1**	**0.895**	**20**	**1**	**0.9**
Taste	**EX**	1	10	0.5	0.5	0	0	0	0	0	0	4	0.2	0.2	3	0.15	0.15	4	0.2	0.2	3	0.15	0.15
	GD	0.9	10	0.5	0.45	6	0.3	0.27	5	0.25	0.225	15	0.75	0.675	11	0.55	0.495	9	0.45	0.405	12	0.6	0.54
	MD	0.7	0	0	0	10	0.5	0.35	9	0.45	0.315	1	0.05	0.035	6	0.3	0.21	7	0.35	0.245	5	0.25	0.18
	FR	0.4	0	0	0	4	0.2	0.08	6	0.3	0.12	0	0	0	0	0	0	0	0	0	0	0	0
	NS	0.1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Total			**20**	**1**	**0.95**	**20**	**1**	**0.7**	**20**	**1**	**0.66**	**20**	**1**	**0.91**	**20**	**1**	**0.855**	**20**	**1**	**0.85**	**20**	**1**	**0.87**
Overall acceptability	**EX**	1	10	0.5	0.5	0	0	0	0	0	0	5	0.25	0.25	4	0.2	0.2	3	0.15	0.15	5	0.25	0.25
	GD	0.9	10	0.5	0.45	5	0.25	0.225	4	0.2	0.18	12	0.6	0.54	10	0.5	0.45	15	0.75	0.675	10	0.5	0.45
	MD	0.7	0	0	0	12	0.6	0.42	11	0.55	0.385	3	0.15	0.105	6	0.3	0.21	2	0.1	0.07	5	0.25	0.18
	FR	0.4	0	0	0	3	0.15	0.06	5	0.25	0.1	0	0	0	0	0	0	0	0	0	0	0	0
	NS	0.1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Total			**20**	**1**	**0.95**	**20**	**1**	**0.705**	**20**	**1**	**0.665**	**20**	1	**0.895**	**20**	**1**	**0.86**	**20**	**1**	**0.895**	**20**	**1**	**0.88**

SF: Scale factor; FMF: Fuzzy membership functions; NFMF: Normalized fuzzy membership functions; EX: Excellent; GD: Good: MD: Medium; FR: Fair; NS: Not satisfactory

Appendix A. 38 Fuzzy ranking of fruit juice samples

Pineapple juice	
Control	C&A=Flavour>Taste>O&A
Ohmic heated	Taste>=C&A=Flavour=O&A
PL treated	Flavour=O&A>Taste>O&A
P1	C&A=Flavour>Taste=O&A
P2	C&A=Flavour=O&A>Taste
P3	C&A=Flavour=O&A>Taste
P4	C&A=Flavour=O&A>Taste
Cashew apple juice	
Control	C&A>Flavour>Taste>O&A
Ohmic heated	C&A=Flavour>Taste=O&A
PL treated	Flavour>C&A>Taste>O&A
C1	C&A>Flavour=Taste>O&A
C2	C&A=Flavour=O&A>Taste
C3	C&A=Flavour=O&A>Taste
C4	C&A=Flavour>O&A=Taste

Appendix A.39

Cost economics of ohmic assisted PL treated pineapple and cashew apple juice

Cost of machineries		
Cost of ohmic heating system	=	Rs.16,000
Cost of Pulsed light system	=	Rs.79,000
Cost of juice extractor	=	Rs.6000 /-
Floor space 5 m^2	=	1,00,000
Cold storage	=	35000
Packing machine	=	25000
Miscellaneous item	=	Rs.10,000/-
Total cost	=	Rs.196600 /-

Assumptions

Life span (L)	=	3 years
Annual working hours (H)	=	275days (per day 8 hrs)
	=	2200 hours
Salvage value (S)	=	10% of initial cost
Interest on initial cost (i)	=	15% annually
Repair and maintenance	=	10% of initial cost
Insurance and taxes	=	2% of initial cost
Electricity charge	=	Rs.6.50/unit
Labour wages/person	=	Rs 400/day

1. Total fixed cost per year

i. Depreciation = $\frac{C-S}{L \times H} = \frac{196600 - 19660}{3 \times 2200}$ = Rs.26.80 /h

ii. Interest = $\frac{\frac{C+S}{2} \times \frac{i}{H}}{}$

$\frac{196600 + 19660}{2} \times \frac{15}{100 \times 2200}$

= Rs.7.37/h

iii. Insurance & taxes = 2% of initial cost

$\frac{2}{100 \times 2200} \times 196600$ = Rs.1.78/h

Total fixed cost = i + ii + iii = 34.77/h

Time consumed to process one litre juice = 15 min (maximum)

Total fixed cost for processing 1 litre juice = 34.77/4=8.69

2. Total variable cost per year

i. Repair & maintenance = 10% of initial cost $= \frac{10}{100} \times 196600$

= 8.93/h

Repair & maintenance for 1litre juice = 8.93/4=2.23/litre

ii. Electricity cost

a) Energy consumed by the ohmic heating system for one litre of juice = 0.56kwh

b) Energy consumed by PL system for 1 litre of juice = 0.64 kwh

Total energy consumption = 1.2 kwh

Cost of energy consumption/h = Rs. 6.50

Cost of energy consumption to process 1 litre juice	=	1.66*1.2=1.95/litre
iii. Labour cost (1 persons)	=	Rs. 400
Labour cost for 1h	=	Rs. 50
Labour cost for processing 1 litre juice	=	Rs. 12.5

iv. Cost of raw materials for production of fruit juices

Quantity of juice processed within 1 day	=	30 litre
Quantity of pineapple required for 1 litre juice	=	1.8kg
Cost of 1kg pineapple	=	Rs. 65
Total cost of pineapple for making1 litre juice	=	Rs. 117
Quantity of cashew apple required for producing 1litre juice	=	2kg
Cost of 1 kg cashew apple	=	Rs. 20
Total cost of cashew apple for making one litre juice	=	Rs. 40
Cost of packaging material Cost of one 250 ml PET bottle	=	Rs. 3.50
Cost of packaging material for 1 litre juice	=	Rs.14

Total variable cost for pineapple juice	=	2.23+1.93+12.5+117+14
	=	147.66
Total variable cost for pineapple juice	=	2.23+1.93+12.5+40+14
	=	70.66

Total cost for production of one liter of

pineapple fruit juice = Fixed cost + Variable cost

= 8.69+147.66

= 156.35

Total cost for production of one litter of cashew apple juice = 8.69+70.66

= Rs. 79.35

Market selling price of pineapple juice = Rs. 300/litre

Market selling price of cashew apple juice = Rs. 130/litre

Benefit cost ratio for pineapple juice $= \frac{300}{156.35}$

= 1.91

The benefit cost ratio for the production of 1litre of ohmic assisted PL treated pineapple juice was found to be **1.91.**

Benefit cost ratio for cashew apple juice $= \frac{130}{79.35}$

= 1.63

The benefit cost ratio for the production of 1litre of ohmic assisted PL treated cashew apple juice was found to be **1.63.**

Appendix A.40 Changes yellowness index of fruit juice during storage

Fruit juice	Storage days	Treatments						
		Control	Ohmic treated	PL treated	(OH_1PL_1)	(OH_1PL_2)	(OH_2PL_1)	(OH_2PL_2)
Pineapple	0	82.18	81.92	82.10	81.91	81.91	81.87	81.78
	5	80.27	81.89	82.08	81.92	81.90	81.87	81.68
	10	81.09	81.61	81.86	81.90	81.89	81.88	81.63
	15	80.79	81.26	81.81	81.74	81.87	81.86	81.59
	20	79.85	80.82	81.74	81.30	81.87	81.81	81.49
	25	78.36	80.76	81.70	81.00	81.86	81.75	81.38
	30	77.95	80.55	79.77	80.92	81.84	81.70	81.00
	35	73.74	78.20	79.44	80.75	81.77	81.60	80.77
Cashew apple	0	-86.08	-160.86	-134.8	-192.45	-191.28	-196.85	-193.79
	5	-180.9	-169.79	-185.3	-195.65	-192.50	-202.28	-194.29
	10	-228.8	-201.84	-190.4	-192.50	-195.36	-205.36	-196.36
	15	-253.6	-224.69	-192.50	-192.5	-200.00	-206.64	-200.00
	20	-450.7	-253.65	-348.2	-202.28	-206.64	-211.69	-204.81
	25	-596.7	-394.37	-362.4	-207.21	-211.69	-216.94	-206.64
	30	-923.6	-483.00	-782.6	-212.32	-214.95	-220.30	-209.19
	35	-1331.	-552.07	-1087.	-223.91	-221.63	-231.63	-218.19

DEVELOPMENT AND EVALUATION OF FLOW THROUGH OHMIC HEATING ASSISTED PULSED LIGHT TREATMENT SYSTEM FOR PRESERVATION OF FRUIT JUICE

by

ASHITHA G N

(2016-28-001)

ABSTRACT OF THESIS

Submitted in partial fulfillment of the requirement for the degree of

DOCTOR OF PHILOSOPHY

IN

AGRICULTURAL ENGINEERING

(Agricultural Processing and Food Engineering)

Kerala Agricultural University

DEPARTMENT OF PROCESSING AND FOOD ENGINEERING

KELAPPAJI COLLEGE OF AGRICULTURAL ENGINEERING AND TECHNOLOGY

TAVANUR, MALAPPURAM-679573

KERALA, INDIA

2020

ABSTRACT

Ohmic heating is a promising thermal processing technology having wide application in food processing. This causes instantaneous heat generation in the food product, creating a rapid and uniform heating that reduces thermal abuse, unlike the conventional thermal processing methods. Pulsed light (PL) treatment is one of the non-thermal processing technologies that have been proven to be effective against a number of food borne pathogens. But the practical application of the PL treatment on food products is limited due to its low penetration depth. These noval technologies could contribute to shorten processing times, energy savings, and highly balanced safe food; however, they alone still cannot guarantee food safety without damaging the food's quality. Therefore, a new concept combining ohmic heating with PL has been extensively evaluated. This combination technology would optimize each of the individual technology's strengths and reduce each of their individual weaknesses. The present study envisages development of an ohmic assisted PL treatment system for pineapple and cashew apple juices and evaluation of the developed system towards maintaining their quality characteristics, microbial safety, and storage stability.

In this study, an ohmic heating batch system and a flow through PL system was developed and fabricated to pasteurize the pineapple and cashew apple juice. The process parameters *viz*. Voltage gradient (10, 12.5 and 15V/cm), Process temperature (50, 55 and 60°C) and treatment time (1, 3 and 5 min) of ohmic heating and PL dosage (10, 12.5 and 15 J/cm^2), sample-source distance (5, 10 and 15 cm) and flow rate (150, 200 and 300 ml/min) of the PL system were optimized separately based on the physic-chemical and microbial quality characteristics of both fruit juices on responses using Response Surface Methodology (RSM). The optimized condition for ohmic heating of pineapple juice were obtained at a voltage gradient of 14.02 V/cm, holding time of 2.31 min and treatment temperature of 55.26°C and that of cashew apple juice were a voltage gradient of 14.53 V/cm, process temperature of 55.25°C with holding time of 2.77 min. The optimum process operating conditions for pineapple juice were found at a PL dosage of 13.69

J/cm^2, sample-source distance of 10.26 cm and flow rate of 165.06 ml/min and that for cashew apple juice were a PL dosage of 12.49 J/cm^2, sample-source distance of 8.63 cm at a flow rate of 164.01 ml/min.

The optimised process operating conditions of both treatments were selected for analysis of ohmic assisted PL treatment process. The optimally treated ohmic assisted PL treated pineapple and cashew apple juice samples were found to be superior to individual treatments as well as other combined ohmic and PL treatment combinations studied in terms of physico-chemical, microbiological and organoleptical characteristics. The ohmic assisted PL treated pineapple and cashew apple juice resulted in more than 5 log reductions in *Escherichia* and *Listeria* strains from initial count thus confirms to the recommendations by FDA safety standards for fruit juices. Storage studies of ohmic assisted PL processed samples at optimised process operating conditions revealed that, the process could provide a shelf life of 25 days and 30 days respectively for pineapple and cashew apple juice under refrigerated conditions of 4±2°C retaining their physico-chemical and sensory characteristics while keeping the microbial level at safe limits.