

STUDY ON PACKAGING AND STORAGE OF TENDER COCONUT WATER

By

B. HEMALATHA

(Registration No-2050221007)

B. Tech. (Agril. Engg.)



**DEPARTMENT OF PROCESSING AND FOOD ENGINEERING
COLLEGE OF AGRICULTURAL ENGINEERING AND TECHNOLOGY
JUNAGADH AGRICULTURAL UNIVERSITY
JUNAGADH-362001**

OCTOBER - 2023

B. HEMALATHA

PROCESSING & FOOD ENGG.

OCTOBER - 2023

STUDY ON PACKAGING AND STORAGE OF TENDER COCONUT WATER

By

B. HEMALATHA

(Registration No-2050221007)

B. Tech. (Agril. Engg.)



**DEPARTMENT OF PROCESSING AND FOOD ENGINEERING
COLLEGE OF AGRICULTURAL ENGINEERING AND TECHNOLOGY
JUNAGADH AGRICULTURAL UNIVERSITY
JUNAGADH-362001**

OCTOBER - 2023

**STUDY ON PACKAGING AND STORAGE OF
TENDER COCONUT WATER**

**A THESIS SUBMITTED TO
JUNAGADH AGRICULTURAL UNIVERSITY
IN PARTIAL FULFILMENT OF THE REQUIREMENTS
FOR THE AWARD OF THE DEGREE OF**

**MASTER OF TECHNOLOGY
(Agricultural Engineering)**

**IN
PROCESSING AND FOOD ENGINEERING**

BY

B. HEMALATHA

(Registration No-2050221007)

B. Tech. (Agril. Engg.)



**DEPARTMENT OF PROCESSING AND FOOD ENGINEERING
COLLEGE OF AGRICULTURAL ENGINEERING AND TECHNOLOGY
JUNAGADH AGRICULTURAL UNIVERSITY
JUNAGADH-362001**

OCTOBER-2023

DEDICATION

*To my mother and my uncle
The reason for what I become today.
Thanks for your great support and continuous care.*

*To my respected guide
Who is the continuous Source of
Inspiration and constant encouragement for me.*

B. HEMALATHA

**DEPARTMENT OF PROCESSING AND FOOD ENGINEERING
COLLEGE OF AGRICULTURAL ENGINEERING AND
TECHNOLOGYJUNAGADH AGRICULTURAL UNIVERSITY
JUNAGADH – 362001**

Name of Student

B. Hemalatha

Major Advisor

Dr. V. K. Chandegara

**STUDY ON PACKAGING AND STORAGE OF TENDER
COCONUT WATER**

ABSTRACT

Keywords: Tender Coconut Water, Ozonation, Ultrasonication, glass bottle, PET bottle

Tender coconut water (TCW) is incredible healthy drink and the best one to hydrate the body. The sweet, clear, aqueous part inside the immature green coconut is referred as tender coconut water (TCW). TCW starts to deteriorate once it is exposed to the air and stored at ambient temperature due to microbial contamination as well as oxidation reactions, which cause changes in its sensory properties. Preserving the TCW with its wholesome natural property remains a challenge. Therefore nonthermal processing techniques treated tender coconut water (ozonation and ultrasonication) and packed in 200ml glass and PET bottles under refrigerated condition ($4\pm 2^{\circ}\text{C}$), and withdrawn a weekly interval to checked the quality and stability of TCW.

The effect of ozonation treatment time (10,20,30 min), ultrasonication treatment time (10,20,30 min), combination treatment time (ozonation and ultrasonication) and packaging materials (glass bottle, PET bottle) on different characteristics of tender coconut water *viz.* biochemical, enzyme activities and sensory characteristics of tender coconut water were studied. The Factorial Completely Randomized Design (FCRD) was used in designing the experiment and analyzing the results.

From the study, ozonation treated TCW was found at 20min treatment time and packed in glass bottle under refrigerated condition, which gave the experimental values of TSS 4.89°Brix, TA 0.08%, pH 4.94, EC 5.10 mS/cm, TDS 4.14ppm, TS 4.13 %, TPC 2.37 (mg GAE/ml), Total plate count 2.485log (CFU/ml), Yeast and Mould count,

2.253log (CFU/ml), POD and PPO 0.038 and 0.020 (Δ O.D./min/ml) and Overall acceptability 6.70 for after 4th week of storage period.

Ultrasonication treated TCW was found at 20min treatment time and packed in glass and PET bottle under refrigerated condition, which gave the experimental values of TSS 5.17 and 4.93°Brix, TA 0.08 and 0.10 %, pH 4.87 and 4.68, EC 5.24 and 5.15 mS/cm, TDS 4.03 and 4.62 ppm, TS 4.37 and 4.63 %, TPC 2.43 and 1.77 (mg GAE/ml), Total plate count 1.718 and 2.418log (CFU/ml), Yeast and Mould count 2.579 and 1.384log (CFU/ml), POD 0.014 and 0.021, PPO 0.017 and 0.021(Δ O.D./min/ml) and Overall acceptability 8.13 for after 4th week of storage period.

Ozonation and Ultrasonication treated TCW was found at 20min treatment time and packed in glass and PET bottle under refrigerated condition, which gave the experimental values of TSS 5.17 and 5.01°Brix, TA 0.08 and 0.09 %, pH 4.75 and 4.72, EC 5.82 and 5.93 mS/cm, TDS 4.60 and 4.68 ppm, TS 3.55 and 3.60 %, TPC 1.80 and 2.07 (mg GAE/ml), Total plate count 2.472 and 1.681log (CFU/ml), Yeast and Mould count 2.019 and 1.726log (CFU/ml), POD 0.014 and 0.016, PPO 0.010 and 0.014 (Δ O.D./min/ml) and Overall acceptability 7.33 and 7.50 for after 4th week of storage period.

It was recommended to tender coconut water treated with 20min ozonation treatment stored in glass bottle upto 4th week of storage period and 20min ultrasonication treatment stored in glass and PET bottle at 4 \pm 2°C upto 4th week of storage period. Combination treatment 20min ozonation and 20 min ultrasonication treatment stored in glass bottle and PET bottle at 4 \pm 2°C upto 4th week of storage period.

**COLLEGE OF AGRICULTURAL ENGINEERING AND TECHNOLOGY
JUNAGADH AGRICULTURAL UNIVERSITY
JUNAGADH**

CERTIFICATE – I

This is to certify that the thesis/project work report entitled “**Study on Packaging and Storage of Tender Coconut Water**” submitted by **Miss B. HEMALATHA (Reg. No. 2050221007)** in partial fulfilment of the requirements for the award of the degree of **Master of Technology (Agricultural Engineering)** in the subject of **PROCESSING AND FOOD ENGINEERING** to the Junagadh Agricultural University is a record of bonafide research work carried out by her under my guidance and supervision and the thesis has not previously formed the basis for the award of any degree or other similar title. The candidate had fulfilled all prescribe requirements. The assistance and help received during the course of investigation have been fully acknowledged. She has successfully completed the comprehensive / preliminary examination held on **June 12, 2023** as required under the regulation for post-graduate studies. She has submitted Kachcha bound thesis on **August 01, 2023**.

Place: Junagadh

Date: 01/ 08/2023

(V. K. Chandegara)

Major Guide

Associate Professor

Dept. of Processing and Food Engineering

College of Agricultural Engg. and Tech.

Junagadh Agricultural University

Junagadh.

**COLLEGE OF AGRICULTURAL ENGINEERING AND TECHNOLOGY
JUNAGADH AGRICULTURAL UNIVERSITY
JUNAGADH**

CERTIFICATE-II

Date: 09/10/2023

This is to certify that the thesis entitled “**STUDY ON PACKAGING AND STORAGE OF TENDER COCONUT WATER**” submitted by **Miss B. HEMALATHA (Reg. No. 2050221007)** to Junagadh Agricultural University, Junagadh in partial fulfillment of the requirements for award of the degree of **MASTER OF TECHNOLOGY (AGRICULTURAL ENGINEERING)** in the subject of **PROCESSING AND FOOD ENGINEERING** after recommendation by the external examiners were defended by the candidate before the following members of the examination committee. The performance of the candidate in the oral examination was satisfactory. We, therefore, forward with recommendation.

(V. K. Chandegara)
Major Guide and
Associate Professor
Dept. of Processing & Food Engg.
College of Agril. Engg. & Tech.
Junagadh Agricultural University
Junagadh

(P. J. Rathod)
Minor Guide and
Assistant Professor
Dep. of Biochemistry
College of Agriculture
Junagadh Agricultural University
Junagadh

(M. N. Dabhi)
Professor and Head,
Dept. of Processing & Food Engg.
College of Agril. Engg. & Tech.
Junagadh Agricultural University
Junagadh

(P. M. Chauhan)
Principal and Dean
College of Agril. Engg. & Tech.
Junagadh Agricultural University
Junagadh

Approved By

(R. B. Madariya)
Director of Research and Dean, P. G. Studies
Junagadh Agricultural University
Junagadh

ACKNOWLEDGEMENT

This thesis is the culmination of my journey of M.Tech. which was just like climbing a high peak step accompanied with encouragement, hardship, support and trust. When I found myself at top experiencing the feeling of fulfillment, I realized though only my name appears on the cover of this dissertation, a great many people including my family members, well-wishers, my friends, colleagues and various institutions have contributed to accomplish this huge work.

*This work would not have been possible without the encouragement and able guidance of **Dr. V. K. Chandegara**, Associate Professor, Department of Processing and Food Engineering, CAET, JAU, Junagadh. I have no words to express my heartfelt thanks for his meticulous guidance, writing interest, constant encouragement and suggestions during the course of work. His feedback and editorial comments were also invaluable for the writing of this thesis.*

*I place with deep sense of gratitude and respect with my heartfelt thanks to my Minor Guide **Dr. P. J. Rathod**, Assistant Professor, Department of Biotechnology and Biochemistry, College of Agriculture, JAU, Junagadh for his valuable suggestions, able guidance and encouragement to complete my course work.*

*I acknowledge with thanks to **Dr. V. P. Chovatia**, Hon'ble Vice-Chancellor, **Dr. R. B. Madariya**, Director of Research and Dean P.G. Studies and **Dr. P. M. Chauhan**, Principal and Dean, CAET, JAU, Junagadh for providing necessary support and encouragement during execution of different research activities.*

*I take this opportunity to thank all the members of my advisory committee, **Dr. M. N. Dabhi**, Professor and Head, Department of Processing and Food Engineering, CAET, JAU and **Dr. V. P. Sangani**, Assistant Professor, Department of PFE, College of Agricultural Engineering and Technology, JAU, Junagadh for their valuable and timely guidance and suggestions during the course of studies and keenly reviewing the manuscript of the thesis.*

*I also express my sincere thanks and respect to **Dr. N. K. Dhamsaniya**, Principal, polytechnic in Agro-Processing, Junagadh Agricultural University, Junagadh, **Dr. P. R. Davara**, **Dr. S. P. Cholera**, **Prof. B. M. Devani** and **prof. A. M. Joshi** and all other staff members of the Department of PFE, CAET, JAU, Junagadh, and **Dr. H. P. Gajera**, Professor and Head, Department of Biochemistry, College of Agriculture, JAU, Junagadh for their constructive suggestions. I am also thankful to **Dr. Kavita**, Senior Research Fellow, **Heena**, Lab Ass., Department of Biochemistry, and **Prof. Jyoti** Department of Seed Science and Technology, College of Agriculture, JAU, Junagadh and **Dr. Urvesh Patel**, **Dr. B. R. Humbal**, **Dr. J. M. Chauhan** Department of VPT, COVS, Kamdhenu University, Junagadh for helping in my research work providing necessary facilities for conducting the research work.*

*I take this opportunity to express special thanks to my friends **Prachi**, **Arya**, **Narsimha**, **abi**, **dhaya**, **Vyas**, **Kuldeep**, **Pradeep** and my senior **Nirav Joshi**, **madhavan**, **rajesh** and my juniors **vidhya**, **poornina**, **vigneshwari**, **divi** for their moral support.*

*I wish my thanks to **Mitesh Dave**, **Pratik Pandiya** and **Vipulbhai** for academic support. I would like thank **Ravi Rathod**, **Chandreshbhai**, **Kishorbhai**, **Dakibhai** and **Parbatbhai** for helping in my research work.*

*I cannot eschew to express my whole hearted sense of reverence to my beloved parents **B. Pakkiya Lakshmi**, **P. Nallathambi perumal**, **Muruganandham** and **Jeya peruma** and my brothers **Manoj kumar**, **Vignesh kumar**, **Jana**, **ajith**, **sujith** and my sisters **Gayathiri**, **Kalarani** and special thanks to my baby **Sowjana adithi**.*

Last but not least, a million thanks to God, the almighty who made me to do this task and made every job a success for me.

...any omission in this small manuscript does not mean lack of gratitude.

*Place: Junagadh
Date: 01/08/2023*

(B. Hemalatha)

CONTENTS

CHAPTER NO.	TITLE	PAGE NO.
I	INTRODUCTION	1-5
	1.1 Practical Utility of Research Problem	4
	1.2 Objectives	5
II	REVIEW OF LITERATURE	6-22
	2.1 Composition of Tender Coconut Water	6
	2.2 Non - Thermal methods for preservation of tender coconut water	8
	2.2.1 Ozonation methods for preservation of tender coconut water	9
	2.2.2 Ultrasound methods for preservation of tender coconut water	11
	2.3 Packaging and Storage of Tender Coconut Water	15
	2.4 Quality Evaluation of Stored Tender Coconut Water	18
III	MATERIALS AND METHODS	23-38
	3.1 Experimental Location	23
	3.2 Raw Materials	23
	3.3 Process Variables	23
	3.3.1 Independent process variables	23
	3.3.2 Dependent process variables	24
	3.3.2.1 Physical properties	24
	3.3.2.2 Biochemical properties	24
	3.3.2.3 Enzyme activity	24
	3.3.2.4 Sensory analysis	24
	3.3.3 Experimental design	25
	3.3.3.1 Treatment details	25
	3.3.4 Different treatment combinations of variables	26
	3.4 Experimental Procedure	27
	3.4.1 Processing of tender coconut water	27
	3.5 Physical properties of fresh tender coconut	27
	3.5.1 Fruit weight	27
	3.5.2 Size	27
	3.5.3 Sphericity	28

3.5.4	Volume	28
3.5.5	Quantity of water	28
3.6	Biochemical properties	28
3.6.1	Titratable acidity	28
3.6.2	Total soluble solids (TSS)	29
3.6.3	pH	29
3.6.4	Total sugar (TS)	29
3.6.5	Electric conductivity (EC)	30
3.6.6	Total Dissolved Solids (TDS)	30
3.6.7	Total Phenol Content (TPC)	30
3.6.8	Enzyme activity	31
3.6.8.1	Assay of polyphenol oxidase (PPO)	31
3.6.8.2	Assay of peroxidase (POD)	31
3.6.9	Microbial analysis	33
3.6.9.1	Preparation of dilutions	33
3.6.9.2	Total yeast and mould count	33
3.6.9.3	Total plate count	33
3.7	Sensory analysis	35
3.8	Storage analysis	35
3.9	Statistical analysis	37
IV	RESULTS AND DISCUSSION	39-111
4.1	Physical properties of fresh coconut	39
4.2	Effect of different process parameters on treated tender coconut water	41
4.2.1	Effect of different treatment time and different packaging materials on the total soluble solids (TSS) of treated tender coconut water	41
4.2.1.1	Effect of ozonation treatment time on TSS for tender coconut water	41
4.2.1.2	Effect of ultrasonication treatment time on TSS for tender coconut water	43
4.2.1.3	Combined effect of ozonation and ultrasonication treatment time on TSS for tender coconut water	45
4.2.2	Effect of different treatment time and different packaging materials on the titratable acidity (TA) of treated tender coconut water	47
4.2.2.1	Effect of ozonation treatment time on TA for tender coconut water	48
4.2.2.2	Effect of ultrasonication treatment time on TA for tender coconut water	50
4.2.2.3	Combined effect of ozonation and ultrasonication treatment time on TA for tender coconut water	52

4.2.3	Effect of different treatment time and different packaging materials on the pH of treated tender coconut water	54
4.2.3.1	Effect of ozonation treatment time on pH for tender coconut water	54
4.2.3.2	Effect of ultrasonication treatment time on pH for tender coconut water	56
4.2.3.3	Combined effect of ozonation and ultrasonication treatment time on pH for tender coconut water	58
4.2.4	Effect of different treatment time and different packaging materials on the Electric conductivity (EC) of treated tender coconut water	60
4.2.4.1	Effect of ozonation treatment time on EC for tender coconut water	60
4.2.4.2	Effect of ultrasonication treatment time on EC for tender coconut water	62
4.2.4.3	Combined effect of ozonation and ultrasonication treatment time on EC for tender coconut water	64
4.2.5	Effect of different treatment time and different packaging materials on the Total Dissolved Solids (TDS) of treated tender coconut water	67
4.2.5.1	Effect of ozonation treatment time on TDS for tender coconut water	67
4.2.5.2	Effect of ultrasonication treatment time on TDS for tender coconut water	69
4.2.5.3	Combined effect of ozonation and ultrasonication treatment time on TDS for tender coconut water	71
4.2.6	Effect of different treatment time and different packaging materials on the Total sugar (TS) of treated tender coconut water	73
4.2.6.1	Effect of ozonation treatment time on TS for tender coconut water	74
4.2.6.2	Effect of ultrasonication treatment time on TS for tender coconut water	76
4.2.6.3	Combined effect of ozonation and ultrasonication treatment time on TS for tender coconut water	78
4.2.7	Effect of different treatment time and different packaging materials on the Total Phenol Content (TPC) of treated tender coconut water	80
4.2.7.1	Effect of ozonation treatment time on TPC for tender coconut water	80
4.2.7.2	Effect of ultrasonication treatment time on TPC for tender coconut water	82
4.2.7.3	Combined effect of ozonation and ultrasonication treatment time on TPC for tender coconut water	84

4.2.8	Effect of different treatment time and different packaging materials on the Peroxidase (POD) of treated tender coconut water	87
4.2.8.1	Effect of ozonation treatment time on POD for tender coconut water	87
4.2.8.2	Effect of ultrasonication treatment time on POD for tender coconut water	89
4.2.8.3	Combined effect of ozonation and ultrasonication treatment time on POD for tender coconut water	91
4.2.9	Effect of different treatment time and different packaging materials on the Polyphenoloxidase (PPO) of treated tender coconut water	93
4.2.9.1	Effect of ozonation treatment time on PPO for tender coconut water	94
4.2.9.2	Effect of ultrasonication treatment time on PPO for tender coconut water	96
4.2.9.3	Combined effect of ozonation and ultrasonication treatment time on PPO for tender coconut water	98
4.2.10	Microbiological parameters of treated tender coconut water	100
4.2.10.1	Total plate count	100
4.2.10.2	Yeast and mould count	101
4.3	Sensory analysis of treated tender coconut water	102
4.3.1	Effect of different treatment time and different packaging materials on the Overall acceptability of treated tender coconut water	102
4.3.1.1	Effect of ozonation treatment time on Overall acceptability for tender coconut water	102
4.3.1.2	Effect of ultrasonication treatment time on Overall acceptability for tender coconut water	104
4.3.1.3	Combined effect of ozonation and ultrasonication treatment time on Overall acceptability for tender coconut water	106
4.4	Comparison of different treatment time and different packaging materials for shelf life extension of tender coconut water	108
4.5	Recommendation of pretreatment for prolonging shelf life of tender coconut water	111
V	SUMMARY AND CONCLUSIONS	112-116
	BIBLIOGRAPHY	117-125

LIST OF TABLES

Table No.	Title	Page No.
2.1	Biochemical composition of coconut water	7
3.1	Details of Treatment combinations	25
4.1	Variation in weight, length, width, thickness, size, volume, sphericity and water quantity of tender coconut	40
4.2	Effect of different treatment time and different packaging materials on the total soluble solids (TSS) of ozonation treated tender coconut water	42
4.3	Effect of different treatment time and different packaging materials on the total soluble solids (TSS) of ultrasonication treated tender coconut water	44
4.4	Effect of different treatment time and different packaging materials on the total soluble solids (TSS) of combined treated tender coconut water	46
4.5	Effect of different treatment time and different packaging materials on the titratable acidity (TA) of ozonation treated tender coconut water	48
4.6	Effect of different treatment time and different packaging materials on the titratable acidity (TA) of ultrasonication treated tender coconut water	50
4.7	Effect of different treatment time and different packaging materials on the titratable acidity (TA) of combined treated tender coconut water	52
4.8	Effect of different treatment time and different packaging materials on the pH ozonation treated tender coconut water	55
4.9	Effect of different treatment time and different packaging materials on the pH of ultrasonication treated tender coconut water	57
4.10	Effect of different treatment time and different packaging materials on the pH of combined treated tender coconut water	58

4.11	Effect of different treatment time and different packaging materials on the Electrical Conductivity (EC) of ozonation treated tender coconut water	61
4.12	Effect of different treatment time and different packaging materials on the Electrical Conductivity (EC) of ultrasonication treated tender coconut water	63
4.13	Effect of different treatment time and different packaging materials on the Electrical Conductivity (EC) of combined treated tender coconut water	65
4.14	Effect of different treatment time and different packaging materials on the Total Dissolved Solids (TDS) of ozonation treated tender coconut water	68
4.15	Effect of different treatment time and different packaging materials on the total dissolved solids (TDS) of ultrasonication treated tender coconut water	70
4.16	Effect of different treatment time and different packaging materials on the total dissolved solids (TDS) of combined treated tender coconut water	72
4.17	Effect of different treatment time and different packaging materials on the total sugar (TS) of ozonation treated tender coconut water	75
4.18	Effect of different treatment time and different packaging materials on the total sugar (TS) of ultrasonication treated tender coconut water	76
4.19	Effect of different treatment time and different packaging materials on the total sugar (TS) of combined treated tender coconut water	78
4.20	Effect of different treatment time and different packaging materials on the total phenol content (TPC) of ozonation treated tender coconut water	81
4.21	Effect of different treatment time and different packaging materials on the total phenol content (TPC) of ultrasonication treated tender coconut water	84
4.22	Effect of different treatment time and different packaging materials on the total phenol content (TPC) of combined treated tender coconut water	85
4.23	Effect of different treatment time and different packaging materials on the peroxidase activity (POD) of ozonation treated tender coconut water	88

4.24	Effect of different treatment time and different packaging materials on the peroxidase activity (POD) of ultrasonication treated tender coconut water	90
4.25	Effect of different treatment time and different packaging materials on the peroxidase activity (POD) of combined treated tender coconut water	92
4.26	Effect of different treatment time and different packaging materials on the poly phenol oxidase (PPO) of ozonation treated tender coconut water	94
4.27	Effect of different treatment time and different packaging materials on the poly phenol oxidase (PPO) of ultrasonication treated tender coconut water	96
4.28	Effect of different treatment time and different packaging materials on the Poly Phenol Oxidase (PPO) activity of combined treated tender coconut water	99
4.29	Effect of different treatment time and different packaging materials on the microbial parameters treated tender coconut water	101
4.30	Effect of different treatment time and different packaging materials on the overall acceptability of ozonation treated tender coconut water	103
4.31	Effect of different treatment time and different packaging materials on the overall acceptability of ultrasonication treated tender coconut water	105
4.32	Effect of different treatment time and different packaging materials on the overall acceptability of combined treated tender coconut water	107
4.33	Comparsion of different treatment time and different packaging materials for shelflife extension of tender coconut waters	110

LIST OF FIGURES

Fig. No.	Title	Page No.
3.1	Process Flow Chart of experiment	38
4.1	Effect of different treatment time and different packaging materials on the total soluble solids (TSS) of ozonation treated tender coconut water	42
4.2	Effect of different treatment time and different packaging materials on the total soluble solids (TSS) of ultrasonication treated tender coconut water	44
4.3	Effect of different treatment time and different packaging materials on the total soluble solids (TSS) of combined treated tender coconut water	47
4.4	Effect of different treatment time and different packaging materials on the titratable acidity (TA) of ozonation treated tender coconut water	49
4.5	Effect of different treatment time and different packaging materials on the titratable acidity (TA) of ultrasonication treated tender coconut water	51
4.6	Effect of different treatment time and different packaging materials on the titratable acidity (TA) of combined treated tender coconut water	53
4.7	Effect of different treatment time and different packaging materials on the pH ozonation treated tender coconut water	55
4.8	Effect of different treatment time and different packaging materials on the pH of ultrasonication treated tender coconut water	57
4.9	Effect of different treatment time and different packaging materials on the pH of combined treated tender coconut water	59
4.10	Effect of different treatment time and different packaging materials on the Electrical Conductivity (EC) of ozonation treated tender coconut water	62

4.11	Effect of different treatment time and different packaging materials on the Electrical Conductivity (EC) of ultrasonication treated tender coconut water	64
4.12	Effect of different treatment time and different packaging materials on the Electrical Conductivity (EC) of combined treated tender coconut water	66
4.13	Effect of different treatment time and different packaging materials on the Total Dissolved Solids (TDS) of ozonation treated tender coconut water	68
4.14	Effect of different treatment time and different packaging materials on the total dissolved solids (TDS) of ultrasonication treated tender coconut water	70
4.15	Effect of different treatment time and different packaging materials on the total dissolved solids (TDS) of combined processed tender coconut water	73
4.16	Effect of different treatment time and different packaging materials on the total sugar (TS) of ozonation treated tender coconut water	75
4.17	Effect of different treatment time and different packaging materials on the total sugar (TS) of ultrasonication treated tender coconut water	77
4.18	Effect of different treatment time and different packaging materials on the total sugar (TS) of combined treated tender coconut water	79
4.19	Effect of different treatment time and different packaging materials on the total phenol content (TPC) of ozonation treated tender coconut water	82
4.20	Effect of different treatment time and different packaging materials on the total phenol content (TPC) of ultrasonication treated tender coconut water	84
4.21	Effect of different treatment time and different packaging materials on the total phenol content (TPC) of combined treated tender coconut water	86
4.22	Effect of different treatment time and different packaging materials on the peroxidase activity (POD) of ozonation treated tender coconut water	88
4.23	Effect of different treatment time and different packaging materials on the peroxidase activity (POD) of ultrasonication treated tender coconut water	90

4.24	Effect of different treatment time and different packaging materials on the peroxidase activity (POD) of combined treated tender coconut water	93
4.25	Effect of different treatment time and different packaging materials on the polphenol oxidase (PPO) of ozonation treated tender coconut water	95
4.26	Effect of different treatment time and different packaging materials on the poly phenol oxidase (PPO) of ultrasonication treated tender coconut water	97
4.27	Effect of different treatment time and different packaging materials on the Poly Phenol Oxidase PPO activity of combined treated tender coconut water	100
4.28	Effect of different treatment time and different packaging materials on the overall acceptability of ozonation treated tender coconut water	103
4.29	Effect of different treatment time and different packaging materials on the overall acceptability of ultrasonication treated tender coconut water	105
4.30	Effect of different treatment time and different packaging materials on the overall acceptability of combined treated tender coconut water	108

LIST OF PLATES

Plate No.	Title	page No.
1.1	Coconut tree , Coconut fruit and Tender coconut water	2
3.1	Ozonation and Ultrasonication treatment on tender coconut water	27
3.2	Biochemical properties of treated tender coconut water	32
3.3	Microbial analysis of treated tender coconut water	34
3.4	Sensory and Storage analysis of treated tender coconut water	36

LIST OF ABBREVIATIONS/NOMENCLATURE

Abbreviation	Meaning
mS	Millisiemen
cm	Centimetre
cm ³	Centimetre cube
g	gram
h	Hour
kHz	kilohertz
kg	Kilogram
mg	Milligram
mg/ml	Milligram per millimeter
min	Minute
ml	Millilitre
mm	Millimetre
Mpa	Mega pascal
N	Newton
ppm	Parts per millions
s	Second
W	Watt
<i>viz</i>	Namely
Anon.	Anonymous
ACP	Atmospheric Cold Plasma
AI	Acid Invertase
ANOVA	Analysis of Variance
AOAC	Association of Official Analytical Chemists
AOP	Advanced oxidation processes
ATCC	American Type Culture Collection
B. Tech	Bachelor of Technology
CD	Critical Difference
CFU	Colony Forming Unit
CV	Coefficient of Variance

EC	Electrical Conductivity
<i>et al.</i>	et alia (and other)
<i>etc.</i>	Etcetera (and the rest)
FCRD	Factorial Completely Randomized Design
Fig.	Figure
GAE	Gallic acid equivalent
GC	Green Coconut
G.F.	Graph factor
Govt.	Government
H ₂ O ₂	Hydrngen peroxide
HPP	High Pressure Process
HTST	High Temperature Short Time
ICAR	Indian Council of Agricultural Research
JAU	Junagadh Agricultural University
K	Potassium
LDPE	Low Density Polyethylene
MC	Mature Coconut
M. Tech	Master of Technology
NaOH	Sodium hydroxide
Na ₂ CO ₃	Sodium carbonate
NCTC	National Collection of Type Cultures
NMR	Nuclear magnetic resonance
No.	Number
NS	Non-Significant
NTU	Nephelometric turbidity units
O ₃	Ozone
O.D	Optical Density
p.	Page
PET	Polyethylene Terephthalate
pH	Potential of hydrogen ion
PME	Pectinmethylesterase
POD	Peroxidase
PPO	Polyphenoloxidase

R ²	Regression co-efficient
RH	Realtive Humidity
S.Em.±	Standard Error of Mean
Sr. no.	Serial number
TA	Titratable Acidity
TCW	Tender Coconut Water
TDS	Total Dissolved Soids
TFC	Total Flavonoid Content
TPC	Total Phenolic Content
TS	Total Sugars
TSS	Total soluble solids
US	Ultrasonication
UV	Ultra-Violet

LIST OF SYMBOLS

Symbol	Meaning
‘ ’	Single inverted comma
“ ”	Double inverted comma
’	Apostrophe
.	Full stop
,	Comma
:	Colon
;	Semicolon
—	Dash
-	Hyphen
/	Slash or per
&	And
%	Percent
@	At the rate of
()	Round bracket
[]	Square or box bracket
+	Plus
±	Plus-minus
=	Is equal to
-	Minus or subtract
*	Asterisk
μ	Micro
°C	Degree Celsius
°Brix	Degree Brix

CHAPTER I

INTRODUCTION

Coconut [*Cocos nucifera* L.] is an important multipurpose perennial horticultural crop in the world, providing food for millions of people, especially in the tropical and subtropical regions and with its many uses it is often called the “tree of life”. Coconut (*C. nucifera*) belongs to the family of the *Arecaceae* (*Palmae*), the subfamily *Cocoideae*. There are mainly two distinct groups of coconut i.e. tall and the dwarf. The tall varieties grow slow and bear fruits 6 to 10 years after planting. The dwarf varieties are fast growing and bear early i.e. takes 4 to 5 years (DebMandal and Mandal, 2011).

Extensively grown in the tropical and fragile coastal areas of the world. Coconut palm or tree is called as “KalpaVriksha” in Sanskrit, which means “the trees which provide all the necessities of life”. Coconut is a single-seeded drupe, its shape varies from spherical to oval, contains outer thick fibrous husk, inner hard shell, and endosperm filled with mildly sweet water. Coconut kernel, shell, husk, trunk, yarn, sap drink (neera), tender water, milk, oil and leaves are some of the products which have a strong economical potential in the global market (Naik *et al.*, 2020).

The major coconut producing countries include India, Indonesia, the Philippines, Brazil, Sri Lanka, Papua New Guinea, Vietnam, Mexico, Thailand and Malaysia. India is the largest coconut producing country in the world and accounted for about 31.45% of the world’s total production during 2021-22, with a production of 19,247 million nuts. The crop contributes around Rs. 307,498 million (US\$ 3.88 billion) to the country’s gross domestic product (GDP). The coconut palm provides food security and livelihood opportunities to more than 12 million people in India. It is also a fiber yielding crop for more than 15,000 coir-based industries, providing employment to nearly 6 lakh people. The productivity of coconut at a national level for 2021-22 was at 9,123 nuts per hectare and is one of the highest in the world. India’s coconut production is majorly situated in Kerala, Karnataka and Tamil Nadu and Andhra Pradesh, accounting for 89.13% of the coconut area and 90.04% of the coconut production in the country. Other coconut-producing states in the country are West Bengal, Orissa and Gujarat (IBEF, 2022).



Coconut tree



Coconut fruit



Tender Coconut water

Plate 1.1 Coconut tree , Coconut fruit and Tender coconut water

The edible part of the coconut fruit (coconut meat and coconut water) is the endosperm tissue. Endosperm tissues undergo one of three main modes of development, which are the nuclear, cellular and helobial modes and the development of coconut endosperm belongs to the nuclear mode. Initially, the endosperm is a liquid containing free nuclei generated by a process, in which the primary endosperm nucleus undergoes several cycles of division without cytokinesis (the process in which the cytoplasm of a single eukaryotic cell is divided to form two daughter cells). Cytokinesis then occurs, progressing from the periphery towards the centre, thus forming the cellular endosperm layer (Lopes and Larkins, 1993).

Coconut water (CW), also called coconut juice, is a sweet refreshing drink taken directly from the inner part of coconut fruits. It differs from coconut milk, which is the oily white liquid extracted from the grated fresh kernel. The coconut water consumed as a beverage usually comes from immature coconut fruits. Due to its unique characteristics, coconut water is considered as a natural functional drink. Its sugar content and mineral composition make it an ideal rehydrating and refreshing drink after physical exercise. Previously considered as a simple tropical refreshment or occasionally as a medicine, coconut water is progressively becoming a natural healthy drink (Prades *et al.*, 2012).

Coconut water is served directly as a beverage to quench thirst, while coconut milk is usually used as a food ingredient in various traditional cooking recipes. The main components of coconut milk are water (50%), fat and protein, whereas coconut water contains mainly water (94%) (Seow and Gwee, 1997).

It is also believed that coconut water could be used as an important alternative for oral rehydration and even so for intravenous hydration of patients in remote regions. Coconut water may also offer protection against myocardial infarction. Interestingly, a study has shown that regular consumption of coconut water is effective in bringing about the control of hypertension. Furthermore, micronutrients (nutrients needed in small quantities) such as inorganic ions and vitamins in coconut water lay a vital role in aiding the human body antioxidant system (Alleyne *et al.*, 2005).

Tender coconut water (TCW), Coconut water (CW), and Green coconut water (GCW) have been synonymously used in numerous studies to denote the liquid endosperm of coconut. All the three terms differ in certain aspects, mostly indicating their maturity and variety. TCW refers to the water when the nut reaches 7–9 months of maturity, and the liquid at this time tastes the sweetest and is also termed as young

coconut water (Tetra Pack., 2019). The water from a nut that is 10–13 months old is the mature coconut water or referred to as CW, whereas the water derived from a variety called green dwarf or green coconut is called GCW (Dwiloka *et al.*, 2020).

Coconut water has been used for long as a therapeutic food around the world. Due to its unique chemical composition, it is more effective in relieving problems like heat stroke and severe dehydration caused by vomiting and diarrhea. It helps to relieve stomach pain, vomiting and prevents hot rashes, smallpox, measles, and chickenpox. It is useful for kidney disorders like kidney stone, and helps to clear the urinary tract infections. It has similar electrolytic balance as of the blood, and therefore, it can also be used as a blood plasma substitute. It is readily absorbed by the body and helps to maintain body pH level. TCW consumption helps in maintaining the body fluid level and absorption of drugs quickly (Rajashri *et al.*, 2020).

Coconut water's rich enzyme systems include very effective and selective reductase, polyphenol oxidase (PPO) and peroxidase (POD). These are involved in its development of a brownish colour when it is exposed to air for a long time. Based on its content and properties, coconut water has been used in the treatment of child and adult diarrhoea, and gastroenteritis as well as for urinary stone dissolution, short-term intravenous hydration and protecting against gastrointestinal tract infections. It is also a rich source of essential amino acids (lysine, histidine, tyrosine and tryptophan), fatty acids, glucose, fructose, cellulose, sucrose, and organic acids such as tartaric, citric and malic acids (Awua *et al.*, 2011).

1.1 PRACTICAL UTILITY

Demand for TCW is on the rise nowadays throughout the world, due to its unique therapeutic and nutritional properties. Although the water in the nut remains sterile, as soon as it is cracked open, enzymatic processes and microbiological contamination begin. According to reports, TCW is a favourable medium for microorganism development. TCW has a short shelf life due to its low acidity, which makes it susceptible to quality degradation from microbial development, the generation of rancid off-flavors, and a fall in pH. As the polyphenol oxidase (PPO) and peroxidase (POD) enzymes have optimal pH ranges around 6.0 and 5.5, respectively, and are endogenously present in fresh TCW, the intrinsic pH of TCW (pH 5.2) also favours the activity of these enzymes. The activity of these browning enzymes may speed up the oxidation cycle, which shortens the shelf life

of TCW by causing it to turn yellow, brown, or pinkish when stored at ambient temperature or in a refrigerator.

The development of novel food items with improved quality without compromising safety is made possible by the nonthermal processing of a variety of foodstuffs, giving customers better options and giving the food sector new chances. Nonthermal technologies are developing quickly and having a big, positive impact on the food processing industry. A number of nonthermal preservation techniques, including high pressure processing (HPP), ultrasound, ozonation, pulsed electric field (PEF) and ultra-violet (UV) treatment, can preserve food's nutritional and sensory properties while also extending its shelf life. These techniques have the potential to completely or partially replace the currently used thermal preservation techniques.

TCW is heat-sensitive because heating it changes its flavour and other quality parameters. As a result, thermal treatments like pasteurisation, sterilisation, etc., which can inactivate enzyme activity and reduce microbes, have a negative impact on the quality of TCW by causing the degradation of heat-sensitive vitamins and minerals, volatile flavour compounds, alteration or denaturation of protein, and loss of other nutrients. Thermal treatment is adverse for TCW preservation due to the changes in organoleptic qualities, including colour, flavour, TSS, turbidity, and rheological characteristics. Consequently, it is crucial to look for non-thermal substitutes that do not negatively affect or do so in a way that is less harmful to the nutritional and sensory qualities of TCW than thermal methods. Hence, research is required to ascertain the effects of non-thermal treatments like ozonation and ultrasound techniques on the storage life and quality of final produce of various types of packaging.

Thus by considering the above aspects, study has been undertaken with the following specific objectives:

1.2 OBJECTIVES

1. To standardize the process parameters for the tender coconut water preservation by ozonation and ultrasonication
2. To study the shelf life of tender coconut water packed in different packaging material
3. To study quality parameters during storage of tender coconut water

CHAPTER – II

REVIEW OF LITERATURE

The previous work done by scientist and researchers on the biochemical composition of tender coconut water, non-thermal methods of preservation for tender coconut water, packaging, and storage of tender coconut water during has been critically reviewed and presented in this chapter. Also quality evaluation in terms of nutrition and sensory also described to ascertain area needs research for shelf life extension of tender coconut water.

2.1 Composition of Tender Coconut Water

Coconut water is a clear and colorless liquid found inside young green coconuts. It has a mild sweetness and is low in calories (17.4 kcal/100 g). Due to its mineral-rich composition, it has been suggested as a natural alternative to sports drinks. The beneficial bioactive compounds found in coconut water include vitamin C, vitamin B, potassium, sodium, magnesium, calcium, arginine, alanine, lysine, and glutamic acid (Cappelletti *et al.*, 2015).

The cellular endosperm in young green coconuts is translucent and jelly-like, but as it matures, it hardens to become white flesh known as coconut meat. Unlike other plant endosperms, the cellularization process in coconuts doesn't completely fill the embryo sac cavity, leaving it solution-filled, known as coconut water. This coconut water originates from the cytoplasm. Nutrients from coconut water are obtained from the seed apoplasm and transported symplasmically into the endosperm through plasmodesmata, the connections between cytoplasm of adjacent cells (Patrick *et al.*, 2001). Coconut water is a natural and nutritious beverage widely consumed due to its refreshing taste and health benefits. It is believed to have various positive effects on health, often influenced by cultural and traditional practices (Sandhya *et al.*, 2008).

Table 2.1 Biochemical composition of coconut water (Jean *et al.*, 2009)

Proximates	(g/100 g)	Proximates	(mg/100 g)
Water	94.99	Inorganic ions	(mg/100 g)
Dry	5.01	Calcium, Ca	24
Energy value	19 kcal (79 kJ)	Iron, Fe	0.29
Protein	0.72	Magnesium, Mg	25
Total lipid (fat)	0.2	Phosphorus, P	20
Ash	0.39	Potassium, K	250
Carbohydrate, by difference	3.71	Sodium, Na	105
Fiber, total dietary	1.1	Zinc, Zn	0.1
Sugars	(g/100 g)	Copper, Cu	0.04
Sucrose	1.02	Manganese, Mn	0.142
Glucose	44.9	Selenium, Se	0.001
Fructose	43.9		
Vitamins		Chemical property	
Vitamin C, total ascorbic acid	2.4	pH	4.6 to 5.6
Thiamin (B1)	0.03		
Riboflavin (B2)	0.057		
Niacin (B3)	0.08		
Pantothenic acid (B5)	0.043		
Pyridoxine (B6)	0.032		
Folate, total	0.03		
Folic acid	0		
Folate, food	0.003		

Living organisms possess well-developed antioxidant systems that help counteract the harmful effects of oxidizing species. Micronutrients play crucial roles in this aspect. They can directly neutralize free radicals by donating electrons, or they can function indirectly as part of metalloid enzymes, such as selenium in glutathione peroxidase or zinc and copper in superoxide dismutase, to catalyze the removal of oxidizing species

(Shenkin *et al.*, 2006). Coconut water contains various components, including sugars, sugar alcohols, lipids, amino acids, nitrogenous compounds, organic acids, and enzymes, each contributing different functional roles in both plant and human systems due to their distinct chemical properties. These components play significant roles in the overall nutritional and health benefits of coconut water.

2.2 Non-Thermal Methods for Preservation of Foods

Kim *et al.* (1999) observed that ozone (O₃) is a strong antimicrobial with various applications in the food industry. It acts as a practical disinfectant, guaranteeing food safety due to its high reactivity and ability to decompose harmlessly to oxygen (O₂). Ozone effectively inactivates microflora on different food items, outperforming conventional disinfectants against resistant microorganisms like viruses and amoebic cysts. It also extends the shelf life of certain foods while maintaining their sensory qualities, leaving no noticeable hazardous residues after treatment.

Khadre *et al.* (2001) emphasized ozone's efficacy as an antibacterial agent in both gaseous and liquid forms. Ozone acts rapidly by reacting with intracellular enzymes, DNA material, and cell components, effectively inactivating microorganisms, including spores and viruses. When combined with specific initiators like UV or H₂O₂, ozone forms advanced oxidation processes (AOPs) capable of combating even the most resistant pathogens. Ozone is produced on-site, decomposes quickly, and leaves no residues when applied to food. Its versatility allows for the decontamination of produce, equipment, food-contact surfaces, and the processing environment.

Patil *et al.* (2010) studied ozone inactivation of acid-stressed *Listeria monocytogenes* and *Listeria innocua* in orange juice using a bubble column. Inoculated with different *Listeria* strains, the orange juice underwent direct ozone treatment at 0.098mg/min/ml for varying time periods (0-8 minutes). Results showed that ozone treatment required longer inactivation times for mild acid-stressed and mild acid stress habituated *L. monocytogenes* cells compared to non-acid stressed cells. Acid-stressed cells habituated in orange juice also exhibited longer inactivation times during ozonation compared to control and mild acid-stressed cells. Overall, gaseous ozone treatment of

orange juice reduced the *Listeria* population by 5 log cycles over a time range of 5 to 9 minutes.

Porto *et al.* (2020) investigated the impact of plasma and ozone processing on the quality of coconut water. Various ozone doses and temperatures were used for ozone processing, and multiple frequencies and voltages were employed for atmospheric cold plasma processing (ACP). The study confirmed that different ozone loads (0.075, 0.225, and 0.370 mg/mL) were absorbed by the samples, as indicated by comparing inlet and outlet ozone concentrations in the glass reactor. The amount of ozone absorbed depended on several factors, including the flow rate of the inlet gas, the ozone content in the gas, the size of gas bubbles, and their distribution in the reactor. Chemometric analysis and NMR spectroscopy were utilized to identify the principal substances present. Interestingly, the pH levels, total soluble solids, titratable acidity, and color of coconut water remained unaffected by both processing procedures. Furthermore, the concentration of phenolic compounds was not influenced by any of the ozone treatments, which effectively led to the complete inactivation of POD (peroxidase) activity.

2.2.1 Ozonation Methods for Preservation of Tender Coconut Water

Torres *et al.* (2011) investigated the effect of ozone processing on apple juice. They subjected apple juice samples to different ozone concentrations (ranging from 1% to 4.8% w/w) and processing times (ranging from 0 to 10 minutes). During ozonation, significant reductions were observed in color values (L, a, and b), rheological properties, and phenolic content of the juice. Using second-order polynomial regression modeling, they found that ozone concentration and processing time had significant effects on these changes. The predicted models were highly significant ($p < 0.05$) with low standard error and high coefficients of determination (R^2).

Tazeen *et al.* (2016) studied the impact of ozonation on physicochemical properties of tender coconut water (*Cocos nucifera* L.). They varied ozone concentration (50-100 $\mu\text{g/mL}$) and processing time (2-12 minutes) and stored the samples at 4°C (0-12 days). Ozone treatment is a non-thermal technique gaining popularity in the food industry for preservation. The optimal treatment (T7) resulted in minimal changes in pH (5.00), TSS (4.98 °B), and titratable acidity (0.316 g/100ml) of tender coconut water on the

zeroth day of storage. However, during ozone processing, some food products may undergo changes in their quality attributes, potentially leading to a sudden loss of nutritive value. Careful monitoring is necessary to minimize such losses and ensure the food remains fresh and stable for an extended period. Hence, the investigation aimed to assess the effect of ozone treatment on the physicochemical properties of tender coconut water before and after ozonation.

Pandiselvam *et al.* (2017) highlighted ozone as a triatomic form of activated oxygen with natural antibacterial properties and high oxidation potential, making it favorable for the food industry. Ozone is utilized to preserve fruits, vegetables, and fruit juices by inactivating bacteria. It is also a promising alternative to conventional sanitizers for preserving fruits and vegetables and managing insect pests that are resistant to phosphine in stored goods. Even at low concentrations, ozone exhibits effectiveness against various microorganisms, fungi, mycotoxins, and insect pests.

Jaramillo *et al.* (2018) studied the inactivation kinetics of peroxidase (POD) and polyphenol oxidase (PPO) in peach juice treated with gaseous ozone. Peach juice was exposed to ozone (0.11 and 0.20 mg O₃ min⁻¹ mL⁻¹) in a bubble column for up to 12 minutes at 20±1 °C. Enzyme activities were significantly reduced due to the treatments, with the extent of inactivation increasing with higher ozone levels and longer exposure times. After 12 minutes of treatment, POD activity reductions ranged from 99.5% to 99.8%, and PPO reductions ranged from 93.9% to 97.3%, depending on the ozone concentration used. The researchers successfully fitted the inactivation curves with first-order and Weibull models.

Rajashri *et al.* (2020) found a strong correlation between ozone concentration and exposure time with the reduction of enzyme activities. Combining ozone with a bio-preservative (nisin) effectively reduced microbial population (*Escherichia coli*) and enzyme activities while preserving nutritional compounds in tender coconut water (TCW). The use of non-thermal technology followed by bio-preservative treatment proved effective, as the bio-preservative's impact was enhanced by improved cell penetration from the preceding non-thermal treatments. They suggested using ozone gas dosages of 20 mg/L at flow rates of 1 L/min for up to 10 minutes, followed by the addition of nisin, as a potential method to extend the shelf-life of TCW up to 3 weeks.

Iqbal *et al.* (2022) studied the impact of ozonation on Kinnow juice. Ozonation at different times (5, 10, and 15 minutes) significantly influenced the juice's nutritional characteristics, increasing acidity, total soluble content, total phenolics content, flavonoids content, and antioxidant potential ($p \leq .05$). Longer ozone exposure (15 minutes) reduced microbial activity. OZ-15 showed the highest total soluble content (14.70 ± 0.10 °Brix), titrable acidity ($0.179 \pm 0.10\%$), total flavonoid content (244.44 ± 3.70 mg equivalent of catechin/100 mL), and total antioxidant activity (305.95 ± 2.70 µg/g equivalent of ascorbic acid of juice). It also had the lowest total mold counts (1.49 ± 0.02 colony forming unit/mL) and plate counts (1.53 ± 0.03 colony forming unit/mL). The study suggested ozonation as a potential method to enhance Kinnow juice properties and shelf life.

2.2.2 Ultrasound Methods for Preservation of Tender Coconut Water

De *et al.* (1999) studied the use of high-power ultrasound in food technology to inactivate peroxidase. They combined high power ultrasound with temperature (80°C) to treat peroxidase type VI from horseradish suspended in water. Ultrasound frequencies of 20, 40, and 60 kHz with actual powers ranging from 0 to 120 W were used. The experiments were conducted using a laboratory scale plant in continuous and batch mode. In continuous experiments, 46 ml of suspension was circulated in a 20 ml sanitation chamber, while in the batch mode, two sets of experiments treated 40 or 80 ml of suspensions in a 100 ml sanitation chamber. The study showed that the decimal reduction time of peroxidase at 80°C (D80) decreased from 65 to approximately 10 minutes with the application of ultrasounds.

Piyasena *et al.* (2003) explored ultrasound for the inactivation of microbes as an alternative method for pasteurization and sterilization in the food industry. They highlighted the increased demand for food processing methods that preserve nutritional content and overall food quality. While sonication (ultrasound processing) alone may not be sufficient to eradicate bacteria from food, combining ultrasound with pressure and/or heat shows promise. Thermosonic, manosonic, and manothermosonic treatments are effective and energy-efficient ways to inactivate germs. Ultrasonic irradiation, especially

higher-power ultrasound at lower frequencies (20 to 100 kHz), can induce cavitation and be utilized to inactivate microbes in food processing.

Zenkar *et al.* (2003) investigated the effects of continuous flow ultrasound thermal treatment for bacterial decontamination in model suspensions and various liquid food systems, including milk, fruit, and vegetable juices. They found that ultrasound-assisted thermal treatment was more efficient in bacterial decontamination compared to traditional indirect heating methods. During continuous flow operations at 26 liters/h with an isothermal process at 60.8°C, they achieved a $64.0 \pm 21.8\%$ reduction in D-value for *E. coli* and a $41.6 \pm 12.8\%$ reduction for *L. acidophilus*. This corresponded to a significant reduction in process time (at least 1.7-fold for *E. coli* and 1.4-fold for *L. acidophilus*) and process temperature (1.07-fold for *E. coli* and 1.03-fold for *L. acidophilus*).

Valero *et al.* (2007) evaluated the effects of ultrasonic treatments in orange juice processing. The selected batch ultrasonic treatment achieved a limited microbial inactivation of 61.08 log CFU ml⁻¹. However, microbial growth was observed during storage at refrigeration (5°C) and mild abusive (12°C) temperatures. The presence of pulp increased micro-organisms' resistance to ultrasound. Continuous ultrasonic treatments at high flow rates showed negligible reductions in microbial counts. Importantly, ultrasound did not adversely affect juice quality attributes. To prevent food-borne pathogen development in orange juice, combining ultrasound with more potent processing methods and reducing initial microbial concentration is necessary.

O'Donnell *et al.* (2010) studied the effect of ultrasound on enzymes in fruit juices and dairy products. Ultrasonic processing is now a viable alternative to traditional thermal methods for pasteurization and sterilization of food products. Ultrasound, when combined with modest heat treatment and/or pressure, has shown potential for inactivating both pathogens and enzymes. The effectiveness of sonication depends on various factors. At low power levels, ultrasound accelerates enzymatic reactions like hydrolysis, while at higher power levels, it inactivates spoilage enzymes like Pectin methyl esterase (PME) and Poly phenol oxidase (PPO).

Chemat and Khan (2011) highlighted the applications of ultrasound in food technology, including processing, preservation, and extraction. Ultrasound significantly accelerates various food processes, leading to faster completion times, higher

reproducibility, reduced costs, simplified manipulation, and higher purity of the final product. It also eliminates the need for post-treatment of waste water and reduces the time and energy required for conventional processes. Ultrasound has been successfully applied in freezing, cutting, drying, tempering, bleaching, sterilization, and extraction in the food industry. The advantages of using ultrasound for food processing include more effective mixing, faster energy and mass transfer, reduced thermal and concentration gradients, selective extraction, reduced equipment size, faster process extraction control, quicker start-up, increased production, and elimination of process steps. The effects of ultrasound on food processes are believed to be influenced by cavitation phenomena and mass transfer enhancement.

Rojas *et al.* (2016a) assessed ultrasound technology (US) for inactivating/sensitizing coconut water peroxidase (POD). US reduced enzyme activity by 27% after 30 minutes (286 W/L, 20 kHz), showing high resistance. Thermal inactivation was described by the Weibull model under non-isothermal conditions, and US pre-treatment sensitized the enzyme to heat. US also resulted in more uniform heat resistance. The study suggests US as a good technology for sensitizing enzymes before thermal processing, potentially reducing the undesirable effects of long processing times and high temperatures.

Rojas *et al.* (2016b) explored ultrasound technology as an emerging method for enzyme inactivation, focusing on peroxidase (POD) in coconut water. The enzymatic activity initially increased and then decreased, indicating complex behavior influenced by the applied ultrasonic energy and its mechanical effects on the product. The study highlights the potential of ultrasound for enzyme inactivation, with direct effects on the enzymatic intermediate states and structural conformation changes.

Zou and Jiang (2016) studied the effect of ultrasound treatment on carrot juice. Ultrasound treatment was performed at 40 kHz and 0.5 W/cm² for 20, 40, or 60 minutes. pH remained unchanged during treatment. Electrical conductivity, viscosity, and color values increased with longer treatment time. Total soluble solids, total sugars, total carotenoids, and ascorbic acid contents significantly improved. The study suggests that ultrasound treatment has the potential to enhance the quality and safety of carrot juice processing.

Ribeiro *et al.* (2017) investigated thermosonication to reduce enzymatic activity in coconut water and optimize the operating parameters (amplitude and time) and specific acoustic energy needed for complete enzymatic inactivation. Ultrasound, when used in combination with heat treatment, showed additional effects on inactivating polyphenoloxidase (PPO) and peroxidase (POD) enzymes. PPO and POD were significantly inhibited between 500 and 550 mW/mL, and at least 655.80 mW/mL of specific acoustic energy was required for complete inhibition. The study suggests that thermosonication, with its additive effect on heat treatment, is recommended for treating coconut water to inhibit PPO and POD enzymes effectively.

Wu *et al.* (2021) investigated the impact of high-intensity ultrasound (US) treatment on sugar metabolism enzymes and quality of coconut water. The US-treated coconut water showed higher total soluble solids and sugar/acid ratio, as well as lower pH and conductivity compared to the control group. Sucrose, fructose, and glucose contents were also higher in the US-treated samples. Enzyme activities, including acid invertase (AI), were decreased by US treatment, with AI being the most affected. Circular dichroism and fluorescence spectra revealed structural changes in AI due to US intensity and time. Molecular docking and dynamics suggested that US treatment prevented sucrose breakdown by inhibiting the recognition and binding of sucrose and AI molecules.

Jacob *et al.* (2023) studied the effect of packaging materials and storage temperature on ultrasonicated mature coconut water (MCW) during storage. MCW was treated with ultrasound for 10 minutes at 60% amplitude with a pulse cycle of 5s on and 5s off. The study evaluated the impact of different packaging materials (glass bottles, PET bottles, and stand-up pouches) and storage temperatures (atmospheric and cold storage) on microbial growth and quality parameters (pH, Total Soluble Solids, total sugar, reducing sugar, and non-reducing sugar) during storage. The microbial load increased significantly ($p < 0.05$) during storage of both ultrasonicated and control samples. Glass bottles were the most effective packaging, followed by PET bottles and stand-up pouches, in maintaining quality parameters and microbial load.

2.3 Packaging and Storage of Tender Coconut Water

Maciel *et al.* (1992) studied the storage conditions for preserving green coconut water. Coconut water overall acceptance scores were not affected by the storage system for two weeks. After three weeks, higher acceptability scores were observed for coconut water stored wrapped at 12°C and 22°C compared to 4°C. In the fourth week, coconut water stored wrapped at 22°C and unwrapped at 4°C received the highest acceptability scores. By the fifth week, the acceptability of coconuts was fair for both storage systems at 4°C and 12°C.

Soares and Hotchkiss (1999) studied the detrimental impact of oxygen on orange juice quality, including the degradation of ascorbic acid, increased browning, and microbial growth. To mitigate oxygen exposure, traditional juice packaging methods use high barrier materials like glass or foil laminates in brickpacks. Additional measures include nitrogen flushing or blending PET with aromatic polyamides to improve gas barrier properties. These methods aim to preserve the freshness and quality of the juice during storage.

Moyssiadi *et al.* (2004) studied the mechanical properties, clearness, UV resistance and good oxygen barrier properties of PET (polyethylene terephthalate) for usage in food packaging increased for liquid foods such as milk or oil and advised to use PET as a good packaging material.

Perez-Vicente *et al.* (2004) reported the packaging material selection as well as influences of processing in the quality of foods like color changes and nutrient composition during storage as a result of contact with oxygen and light transmission through them. Paperboard cartons with low density polyethylene (LDPE) coating or glass containers are commonly used packaging materials for juices and reported higher loss of aroma compounds from citrus juices in contact with LDPE than with other packaging.

Reddy *et al.* (2007) developed a nonthermal sterilization process for green coconut water using two-stage filtration with low ash filter paper and cellulose nitrate membrane. The water after the second stage was sterile with no visible microbial growth. However, flavor and overall acceptability decreased by 9% and 11%, respectively. The water remained sterile even after 1 month in aseptically packed conditions, but overall

acceptability further decreased by 6%. Filtration led to reductions in nutrients such as fat, ash, total sugar, reducing sugar, and protein by 40.0%, 43.9%, 23.4%, 29.2%, and 13.3%, respectively. Additionally, potassium, magnesium, calcium, iron, and copper were removed by 10.15%, 16.14%, 19.04%, 20.85%, and 22.21%, respectively. The removal of these nutrients resulted in increased surface tension and decreased viscosity of the coconut water.

Vila and Ampuero (2007) examined the role of packaging in positioning orange juice. They focused on the marketing mix and how packaging plays a crucial role in positioning a product. The study revisited the functions of packaging and its graphic elements, such as color, shape, typography, and image. The aim was to identify associations between various positioning strategies and the graphic design variables used to define orange juice packaging. Designer and consumer opinions were taken into account to understand the impact of packaging on product positioning.

El-Sheikha *et al.* (2009) assessed the quality of pasteurized physalis juice during storage in glass bottles and flexible laminated packs at 5°C. The fresh juice had a pH of 3.5, titratable acidity of 1.43%, and contained polyphenols (76.6mg/100mL) and vitamin C (38.8mg/100mL). It was rich in carotenoids (70µg/mL). Throughout the 6-month storage, the carotenoids, polyphenols, and ascorbic acid contents gradually reduced in both packaging types, with a more significant reduction observed in juice stored in flexible laminated packs.

Nwafo and Ikenebomeh (2009) studied the effects of different packaging materials on the microbiological, physico-chemical, and organoleptic quality of zobo drink during storage at room temperature ($29 \pm 2^\circ\text{C}$) for 6 months. The zobo drink was aseptically dispensed into glass bottles, plastic bottles, and polythene sachets after preparation at 45°C. Quality changes were assessed monthly. The results showed varying degrees of changes in quality among the different packaging materials, with more significant changes observed in the polythene sachet packaged samples. The packaged samples had lower pH and total soluble solids (TSS) values compared to the fresh sample, but higher carbohydrate, protein, and vitamin C contents. Glass bottles were identified as the best packaging material for storing zobo drink at ambient temperature.

Revi *et al.* (2014) investigated the impact of different packaging materials on the enological parameters and volatile compounds of dry white wine during a 6-month storage period at 20°C. They compared dark colored glass bottles with two commercial bag-in-box (BIB) pouches, one lined with low-density polyethylene (LDPE) and the other with ethylene vinyl acetate (EVA). The study monitored various parameters, including titratable acidity, volatile acidity, pH, total SO₂, free SO₂, color, volatile compounds, and sensory attributes. The results indicated that the BIB packaging materials influenced the titratable acidity, total and free SO₂, and color of the wine. A significant portion of the wine's aroma compounds was absorbed by the plastic materials or lost due to leakage from the valve fitment. Among the two plastics, the LDPE-lined pouch exhibited higher aroma sorption compared to EVA. On the other hand, wine packaged in glass retained the highest amount of its aroma compounds. Sensory evaluation showed that the white wine packaged in both plastics remained of acceptable quality for 3 months, while the wine in glass bottles retained its quality for at least 6 months.

Pimental *et al.* (2015) studied the effect of supplementing clarified apple juice with probiotic *Lactobacillus paracasei ssp.* and *Oligofructose* on physicochemical characteristics, probiotic viability, and acceptability during refrigerated storage in plastic or glass packages at 4°C for 28 days. Probiotic juices had similar chemical properties to unsupplemented juices regarding density, acceptability, and purchase intent, but they were more acidic, turbid, and red in color. *Oligofructose* supplementation improved probiotic survival during storage without affecting physicochemical properties, acceptability, or storage stability. *Oligofructose* was stable in apple juice storage and could be used as a sugar substitute, as products with *Oligofructose* showed similar properties and consumer acceptance as those with sucrose. Glass packaging was preferred to maintain probiotic viability. Both plastic and glass packaging had no significant impact on the physicochemical properties or acceptability of the juices.

Ekasari and Widyarti (2019) compared the physicochemical properties of natural coconut water with packaged coconut water. They highlighted coconut water as a healthy beverage and a solvent that can enhance the solubility of a solution during heating processes. Natural coconut water is prone to changes in the open air and needs to be processed into packaged coconut water. However, the packaging process often introduces additional ingredients that may alter the taste and nutritional content of coconut water.

The study reported that all samples of packaged coconut water had pH values ranging from 5.42 to 5.49, turbidity ranging from 19.48 to 69.63 NTU, and conductivity ranging from 15.48 to 19.88 mS. These values provide essential information for understanding the physicochemical characteristics of packaged coconut water and its suitability for consumption and various applications.

Tan and Easa (2021) conducted a study to investigate the impact of storage temperature and time on the physicochemical and microbiological properties of green (GC) and mature coconut (MC) water. They observed that at temperatures above 25°C, total soluble solids in coconut water decreased slightly, and pH decreased while titratable acidity increased. These changes were more pronounced in MC water than in GC water and correlated positively with storage temperature. Both GC and MC water could be stored at temperatures between 25°C and 35°C for up to 12 and 6 hours, respectively, before becoming acidified and microbiologically unsafe. However, at 4°C, both types of coconut water remained microbiologically safe for 24 hours, indicating that lower temperatures are more suitable for preserving the microbiological quality of coconut water during storage.

2.4 Quality Evaluation of stored Tender Coconut Water

Campos *et al.* (1996) investigated the chemical composition and enzyme activity of green coconut water and its impact on flavor quality. They found that both poly phenol oxidase and peroxidase enzymes exhibited optimal activity at pH 6.0 and 5.5, and at temperatures of 25°C and 35°C, respectively. The most effective methods for enzyme inactivation were heating at 90°C for 550 seconds and adding ascorbic acid. Ascorbic acid addition did not affect sensory qualities, but prolonged heat treatment at 90°C negatively impacted the flavor. However, flavor quality remained unaffected by heat treatment with the addition of potassium metabisulfite, ascorbic acid, or both. This study highlights the importance of enzyme inactivation techniques in preserving the flavor quality of green coconut water.

Tan *et al.* (2014) analysed composition, physicochemical properties and thermal inactivation kinetics of polyphenol oxidase and peroxidase from coconut water and they obtain from immature, mature and over mature coconut that turbidity of the coconut water

increased with maturity, the pH tended to rise, with inverse changes in acidity and soluble solids ($^{\circ}$ Brix, the total sugar content) tended to increase but fall off with maturities above 12 months, with a general decrease in Brix values from around 7 months onwards.

Jackson *et al.* (2004) studied the chemical composition changes in coconut water during fruit maturation. They observed that the volume of nut water increased significantly from 233 to 504 ml, with the highest quantity at 9 months of maturity. Additionally, fat, protein, soluble solids, acidity, and turbidity steadily increased with maturity, while pH and ash showed variations during maturation. The study also highlighted the significant impact of fruit variety and maturity stage on the chemical composition of coconut water. These findings provide valuable insights into the changes occurring in coconut water as the fruit develops, which can be essential for understanding its nutritional and sensory characteristics.

Kailaku *et al.* (2005) studied on carbohydrates-electrolytes characteristics of coconut water from different varieties and its potential as natural isotonic drink and found that Total sugar % in dwarf was 6.01 ± 0.06 and for tall 5.57 ± 0.02 , Sucrose % in dwarf 0.63 ± 0.01 and for tall 0.50 ± 0.00 , fructose % in dwarf 2.67 ± 0.04 and for tall 2.56 ± 0.04 , glucose % in dwarf 2.71 ± 0.01 and for tall 2.52 ± 0.01 , potassium (mg/kg) in dwarf 1497.40 ± 43.73 and for tall 1567.96 ± 128.77 , sodium (mg/kg) in dwarf 30.30 ± 6.18 and for tall 37.33 ± 6.18 , magnesium (mg/kg) in dwarf 95.4 ± 3.88 and for tall 217.73 ± 119.42 content found.

Vigliar *et al.* (2006) observed the biochemical profile of coconut water from coconut palms planted in an inland region and state that, Tender coconut water comprises of 95.5% water, 4% sugars, 0.1% fat, 0.02% calcium, 0.01% phosphorous, 0.5% iron, considerable amounts of amino acids, mineral salts, vitamin B complex, vitamin C and cytokines etc.

Kwiatkowski *et al.* (2008) conducted a study on the quality of coconut water from green dwarf fruit variety at different stages of development in Brazil. They found variations in liquid volume (280 to 350 ml per fruit), pH (4.73 to 5.74), acidity (8.0 to 11.0 g/l citric acid), total soluble solids content (5.23 to 6.13 $^{\circ}$ Brix), reducing sugars (33.5 to 49.3 g/l), non-reducing sugars (2.20 to 3.50 g/l), total sugars (37.0 to 51.5 g/l), and

electrical conductivity (14.58 to 15.00 $\mu\text{S}/\text{cm}$). The study also analyzed mineral elements, with potassium concentration being the highest (2400.96 to 2726.03 mg/l), followed by calcium concentration (145.36 to 197.21 mg/l). Other minerals appeared in smaller concentrations. Sensory analysis indicated that coconut water from fruits with 7-9 months of development was preferred as a reference, suggesting that this stage offers the optimal sensory characteristics of coconut water. This research provides valuable information on the quality attributes and mineral content of coconut water at different stages of fruit development, which can be useful for the coconut industry and consumers.

Chuku *et al.* (2014) evaluated the protein and mineral content of coconut water from different species and they found that the concentration of this mineral element studied lies within the ranges (ppm) with Ca (25-55), Na (40-85), Fe (0.542-1.625), Cu (0.000-0.018), S (10.35-30.00), P (0.01-0.35), Cl (2050.00-2565.00). And the value of protein of three different species ranged between 0.30-0.40, 0.10-0.20 and 0.49-0.55.

Tiwari *et al.* (2008) conducted experiment on the kinetics of freshly squeezed orange juice quality changes during ozone processing at various gas flow rates (0.25, 0.125, 0.0625, and 0.0312 L min^{-1}). ozone concentration ranges for these flow rates were 0.6-3.0, 1.0-4.8, 1.6-7.8, and 2.3-10.0% w/w, respectively. Samples were treated for 0 (control), 2, 4, 6, 8, and 10 min processing times. Treatments were performed at 20 (0.5°C). No significant changes in pH, $^\circ\text{Brix}$, TA, cloud value, and NEB ($p < 0.05$) were found. L^* , a^* , and b^* color values were significantly affected by gas flow rate, ozone concentration, and treatment time. The ascorbic acid content was found to decrease from 41.59 to 12.70 mg/100 mL after 10 min of treatment time.

Chattopadhyay *et al.* (2013) evaluated different coconut cultivars for tender nut water in West Bengal and found significant variations in water volume with different stages of nut maturity. The highest volume was observed at 7 months (268.0 ml) and 6 months (267.1 ml) of maturity. Across all stages of maturity, nut water volume varied significantly among cultivars, ranging from 27.9 ml to 198.1 ml. 'Jamaican Tall' exhibited the highest water volume (273.9 ml), followed by 'East Coast Tall' (260.7 ml) and 'Zanzibar' (245.5 ml), while 'FMS Big' had the lowest volume (198.1 ml). The interaction between stage of maturity and cultivars also had a significant effect on water volume. 'Jamaican Tall' showed the highest water volume (327.5 ml) during the 6th month of

maturity, followed by 'Zanzibar' (315.6 ml) and 'East Coast Tall' (305.6 ml) at the 7th month of maturity. This study provides valuable insights into the water volume variations of different coconut cultivars at various stages of maturity, which can be beneficial for cultivar selection and coconut water production in West Bengal.

Mahnot *et al.* (2014) investigated the impact of additives on the quality of tender coconut water processed using a nonthermal two-stage microfiltration technique. Citric acid (0.02 g/100 mL), ascorbic acid (0.18 g/100 mL), and L-cysteine (0.009 g/100 mL) were added to enhance the taste. After nitrogen flushing and refrigeration (4 °C), the coconut water was packed into glass and plastic bottles. Water packed in glass bottles showed superior quality in all aspects. Total soluble solids increased from 5.4 to 6 °Brix, pH from 5.7 to 5.8, and soluble sugar concentration from 1.9 g/100 mL to 3.1 g/100 mL. The free fatty acid content also increased from 0.064 mg KOH/g to 2.8 mg KOH/g after 46 days of storage. This study highlights the positive effects of additives on the quality of tender coconut water and the benefits of using glass bottles for packaging to maintain its quality during storage.

Priya *et al.* (2014) studied on tender coconut water nature's elixir to mankind and found that soluble solids (Brix) 5.08 at 7 months and 6.10 at 9 months coconut water, Ph 4.8 at 7 months and 5.29 at 9 months fruit, Glucose (g/100ml) 2.4 at 7 months fruit and 2.9 at 9 months, Fructose 2.1 at 7 months and 2.5 at 9 months, Sucrose 0.4 at 9 months, Total sugar 5.0 at 7 months and 6.3 at 9 months, potassium 198.7 at 7 months and 215.8 at 9 months, Calcium 14.5 at 7 months and 11.5 at 9 months, Magnesium 4.6 at 7 months and 5.1 at 9 months.

Sun *et al.* (2015) investigated the impact of sonication on fresh apple juice (cv. Red Fuji). Ultrasound power at 900 W and frequency between 20 to 25 kHz was used. Sonication was effective in inhibiting enzyme browning in the juice. However, it led to a decrease in the contents of total phenolic content (TPC), total flavonoid content (TFC), and chlorogenic acid, resulting in reduced antioxidant activity. The study highlights the trade-off between inhibiting browning and preserving the beneficial phytochemicals and antioxidant properties of fresh apple juice when using ultrasound technology.

Ma *et al.* (2019) reported the quality and shelf life of coconut water treated at 4°C under high pressure (HPP) and high temperature for a short period of time (HTST) were

compared. The microbiological safety of coconut water during refrigerated storage of 25 and 15 days, respectively, could be guaranteed by HPP of 500 MPa (5 min) and HTST of 72°C (15 s) treatments. After 15 days of storage, the HTST group had lost 51.54% of its amino acids and 32.37% of its protein but had retained 65.0% of its total sugars, 64.51% of its ascorbic acid, and 74.34% of its total phenols. After 25 days of storage, the HPP groups had more nutrient content, including 93.17% of total phenols, 76.76% of total protein, and 76.85% of total amino acids.

Mahnot *et al.* (2019) evaluated the shelf-life enhancement of refrigerated tender coconut water using nonthermal microfiltration and additives. The two-stage microfiltration process with 0.8 µm and 0.45 µm pore size filters reduced total sugars by 13.4%, protein by 13.0%, and reducing sugars by 21.5% without significantly affecting overall acceptability. The addition of 200 mg/L citric acid, 180 mg/L ascorbic acid, and 5% (w/v) orange honey further improved the stability of the final product. Analyses of microfiltered samples showed no evidence of hemolytic activity, indicating product safety. Moreover, the use of additives effectively reduced the activity of polyphenol oxidase and peroxidase, contributing to the preservation of the coconut water's quality during refrigerated storage.

CHAPTER III

MATERIALS AND METHODS

This chapter deals with the materials and methods employed for the processed tender coconut water by nonthermal treatment (Ozonation and Ultrasound) and storage stability in the two different packaging materials. Preliminary trials conducted for the selection of raw materials, procedure for the preparation of treated coconut water using Ozone gas flow rate, time, amplitude, frequency and pulse, determination of biochemical, Enzyme activity and sensory characteristics of processed tender coconut water and optimization of the process parameters for the preparation of better quality and nutritious coconut water through statistical design. The details of machineries, instruments and materials used in the experiments are explained here.

3.1 Experimental Location

The experiment was carried out at Department of Processing and Food Engineering, College of Agricultural Engineering and Technology, Junagadh Agricultural University, Junagadh during the year of 2022 – 2023.

3.2 Raw Materials

The tender coconuts was purchased from Instructional Farm, Jambuvadi, College of Horticulture, Junagadh Agricultural University, Junagadh, Gujarat, India. Coconut water samples were extracted from tender coconuts (*Cocos nucifera* L.) at the 7-9th maturation month. Coconuts were immersion in water for 20min. The extraction was done manually with the aid of a sterile stainless steel knife. After extraction, the samples were homogenized and then filtered on standard muslin cloth.

3.3 Process Variables

There are two types of process variables as independent and dependent variables described briefly in following subheadings.

3.3.1 Independent process variables

The effects of independent variables for ozonation treatment *viz.* Ozone gas flow rate and time, Ultrasound treatment *viz.* Time, amplitude, frequency and pulse.

3.3.2 Dependent variables

The following biochemical, enzyme activity and sensory properties were measured during experiment and the analysis is reported in Chapter-IV.

3.3.2.1 Physical properties

- i. Weight
- ii. Size
- iii. Sphericity
- iv. Volume
- v. Quantity of water

3.3.2.2 Biochemical properties

- i. Titratable Acidity
- ii. Total Soluble Solids
- iii. pH
- iv. Total Sugar
- v. Electrical conductivity
- vi. Total viable count
- vii. Yeast and mould count
- viii. Total phenol content
- ix. Total Dissolved Solids

3.3.2.3 Enzyme activity

- i. Peroxidase
- ii. Poly Phenol Oxidase

3.3.2.4 Sensory analysis

- i. Taste
- ii. Color
- iii. Flavor
- iv. Overall acceptability

3.3.3 Experimental design

The experiment was designed using Factorial Completely Randomized Design (FCRD) with three replications and had fifteen treatments viz., in first factor ozone treatment different time and second factor processed tender coconut water stored in two different packaging materials for ozone and ultrasound treatment. Combination treatment first factor ultrasound treatment three different time, second factor ozonation treatment three different time and third factor processed tender coconut water stored in two different packaging materials. The experiment is planned giving nonthermal treatment as ozonation and ultrasonication with different packaging material as PET and glass bottle stored at $4\pm 2^{\circ}\text{C}$ temperature.

3.3.3.1 Treatment details

1. Ultrasound Treatment : 3 levels (First factor)

$U_1 = 10 \text{ min}$

$U_2 = 20 \text{ min}$

$U_3 = 30 \text{ min}$

Amplitude = 75%

Frequency = 20kHz

Pulse = 30sec

2. Ozonation Treatment : 3 levels (First factor)

$O_1 = 10 \text{ min}$

$O_2 = 20 \text{ min}$

$O_3 = 30 \text{ min}$

Ozone gas flow rate = 200 mg/h

3. Packaging Treatment : 2 levels (Second factor)

$S_1 = \text{Glass Bottle}$

$S_2 = \text{PET Bottle (Polyethylene Terephthalate)}$

Table 3.1 Details of Treatment combinations

Ultrasound + Packaging Treatment

S. No	Treatments		
1	U_1S_1	U_2S_1	U_3S_1
2	U_1S_2	U_2S_2	U_3S_2

Ultrasound + Ozone + Packaging Treatment

S. No	Treatments		
1	U ₁ O ₁ S ₁	U ₂ O ₁ S ₁	U ₃ O ₁ S ₁
2	U ₁ O ₁ S ₂	U ₂ O ₁ S ₂	U ₃ O ₁ S ₂
3	U ₁ O ₂ S ₁	U ₂ O ₂ S ₁	U ₃ O ₂ S ₁
4	U ₁ O ₂ S ₂	U ₂ O ₂ S ₂	U ₃ O ₂ S ₂
5	U ₁ O ₃ S ₁	U ₂ O ₃ S ₁	U ₃ O ₃ S ₁
6	U ₁ O ₃ S ₂	U ₂ O ₃ S ₂	U ₃ O ₃ S ₂

Ozone + Packaging Treatment

S. No	Treatments		
1	O ₁ S ₁	O ₂ S ₁	O ₃ S ₁
2	O ₁ S ₂	O ₂ S ₂	O ₃ S ₂

3.3.4 Different treatment combinations of variables

- a) Total number of treatment combinations

$$= (3*2) + (3*3*2) + (3*2)$$

$$= 30$$
- b) Storage period = 4 weeks

$$= 30*4$$

$$= 120 \text{ samples}$$
- c) Packaging materials = 2
 - i. 60 samples in Glass bottle
 - ii. 60 samples in PET bottle
- d) Quantity of Samples
 - i. 200ml of coconut water = Glass bottle/per
 - ii. 200ml of coconut water = PET bottle/per
- e) Total quantity of Bottles = 120 Bottles



Plate 3.1 Ozonation and Ultrasonication treatment on tender coconut water

3.4 Experimental Procedure

3.4.1 Processing of tender coconut water

The filtered tender coconut water was treated with ozonation and ultrasonication methods for different times and variables. Treated samples were packed in glass and PET bottles. Then the packed samples were refrigerated at $4\pm 2^{\circ}\text{C}$ and stored for 4 weeks.

3.5 Physical properties of fresh tender coconut:

Various physical parameters like fruit weight, size, sphericity, volume and quantity of water of fresh coconut fruit were determined as described here under. Ten representative samples of well graded and uniform size fresh coconut fruits were selected for determination of physical parameters.

3.5.1 Fruit weight

Weight of coconut fruit was measured with help of digital weight balance having an accuracy of 0.01g. The observations were recorded in kilogram.

3.5.2 Size

Size of the coconut fruit was measured with the help of digital Vernier Calipers, having a least count 0.01mm. Dimensions like length, width and thickness were recorded in millimeter. The size of the fresh coconut fruit was determined by the following equation.

$$\text{Size (mm)} D = (lbh)^{1/3}$$

Where, l = length of the coconut fruit (mm)

b = width of the coconut fruit (mm)

h = thickness of the coconut fruit (mm)

3.5.3 Sphericity

The sphericity was determined by using the equation given below with three dimensions as length, width and thickness of coconut.

$$\text{Sphericity } \varepsilon = [(lbh)^{1/3}]/l$$

Where, l = length of the coconut fruit (mm)

b = width of the coconut fruit (mm)

h = thickness of the coconut fruit (mm)

3.5.4 Volume

The volume of the coconut was calculated by using the equation given below

$$\text{Volume (m}^3\text{)} V = 4/3 \pi lbh$$

Where, l = length of the coconut fruit (mm)

b = width of the coconut fruit (mm)

h = thickness of the coconut fruit (mm)

3.5.5 Quantity of water

The quantity of water was measured with help of measuring cylinder having an accuracy of 0.01ml. The observations were recorded in milli litre.

3.6 Biochemical properties of tender coconut water

3.6.1 Titratable acidity

Titratable acidity was estimated as reported by Kannangara *et al.* (2015). A TCW sample of 10 mL was dissolved in 100 mL of water. 10 mL of sample was pipetted into a 100 mL conical flask, and a few drops of phenolphthalein indicator were added. Titration

was performed against 0.1 N NaOH until the pale pink end point was achieved. This method was continued until concordant readings were obtained. The titratable acidity was estimated in terms of citric acid equivalency using the formula shown below.

$$\text{Titratable acidity (\%)} = \frac{\text{Titre (ml)} \times 0.1\text{N NaOH (ml)} \times \text{Volume made up (ml)} \times \text{Equivalent weight of citric acid}}{\text{Weight of sample (ml)} \times \text{Aliquot taken} \times 1000} \times 100$$

3.6.2 Total soluble solids (TSS)

Total soluble solids (TSS) of processed tender coconut water were determined following procedure described by Ali *et al.* (2010). The total soluble solids are primarily sugars like sucrose, fructose, and glucose. Citric acid and minerals in the juice also contribute to the soluble solids. A digital refractometer was used to measure TSS. A drop of processed tender coconut water was placed on the prism glass of a digital refractometer to obtain the measurement of TSS. Between samples, the prism of the refractometer was washed with distilled water and dried with tissue paper before use. The refractometer was standardized against distilled water (0% TSS).

3.6.3 pH measurement

The pH is defined as the logarithm of the reciprocal of hydrogen ion concentration. It is of importance as a measure of the activity acidity which influences the flavour or palatability of a product and affects the processing requirements. The pH meter was standardized with double distilled water of PH 7.0 and standards at pH 4.0 and 9.2. Samples were poured into a beaker glass. The pH was measured using pH meter by dipping the electrodes of pH meter into the sample.

3.6.4 Total sugar

Total sugars were estimated by the modified method of Dubois *et al.* (1956). A sample of 0.1 ml was mixed with 10 ml of 2.5 N methanol. Then 0.1 ml aliquot was taken and added 0.9 ml distilled water to make a final volume of 1.0 ml. 1.0 ml of 5% phenol and 5.0 ml of 96% H₂SO₄ were added one by one. Then all samples were put in a water bath for 10-15 minutes. Spectrophotometer reading was taken at 490 nm wavelength.

$$\text{Total sugar (\%)} = \frac{\text{Sample O.D} \times \text{Standard O.D} \times \text{Total volume (ml)} \times \text{Dilution factor} \times 100}{\text{Aliquot taken (ml)} \times \text{Sample weight (ml)} \times 1000}$$

3.6.5 Electric conductivity

The conductivity meter was calibrated against standard buffer solutions. The samples were mixed well to homogenize and the conductivity was measured using the calibrated conductivity meter and expressed in mS/cm.

3.6.6 Total dissolved solids (TDS)

Total Dissolved Solids (TDS) is the total number of compounds dissolved in a liquid. These substances can include salts, minerals, metals, calcium, and other organic and inorganic components. TDS is defined as everything present in water that is not pure water and is not a suspended solid. TDS is most typically determined by measuring specific conductivity to detect the presence of ions in water (EC). After determining the EC, a conversion factor is applied (usually by the metre making the measurement) to calculate the TDS. The conversion factor will differ depending on the sample you are testing, conversion factors generally range from 0.4 – 1.0 (EC value 5 to 6.5 mS/cm conversion factor 0.79) (Anon., 2023). The TDS of the samples was calculated from their electrical conductivity (EC). TDS is generally measured in parts per million (ppm) but can also be measured in mg/L. Generally, good quality water will fall between 0 and 600 ppm and readings over 1200 ppm are often considered an unsatisfactory level of TDS.

3.6.7 Total phenol content

Phenol content was estimated by the method suggested by Mahayothee *et al.* (2015). 0.1 ml of sample was extracted in 10 ml of 80% ethanol. Centrifugation was done and supernatant was collected. Then 0.1, 0.2, of working standard into a series of test tubes was pipetted out and total volume of each was made up to 3 ml using distilled water. 3 ml of distilled water in a test tube was set for blank solution. 0.5 ml of Folin-Ciocalteu reagent was then added to each tube including blank. After three minutes, 2 ml of 20% Na₂CO₃ solution was added to each. The solution was mixed thoroughly and was placed in boiling water bath for one minute. After cooling to room temperature, colour was read at 650 nm using UV visible spectrophotometer (Thermo Scientific, GENESYS 50). At last, percentage of total phenol was calculated by preparing standard curve of catechol. Following formula was used.

$$\text{Total Phenol content (\%)} = \frac{\text{Graph factor } (\mu\text{g}) \times \text{Optical density} \times \text{Total volume (ml)}}{\text{Sample aliquot (ml)} \times \text{weight of sample (ml)} \times 1000}$$

3.6.8 Enzyme activity

3.6.8.1 Assay of polyphenol oxidase (PPO)

Processed tender coconut water 10ml of 100mM sodium phosphate buffer having pH 6.5. The homogenate was centrifuged at 10,000 rpm for 15 min at 4°C and the supernatant was used for enzyme assay.

The reaction mixture contained 2.9ml of catechol (10mM catechol in 10 mM phosphate buffer, pH 6.5) and reaction was initiated by the addition of 100 μ l of enzyme extract. The changes in the colour due to the oxidized catechol was read at 490nm for 15 minutes at an interval of 3min. Blank was carried out without substrate. The enzyme activity was expressed as Δ O.D/min/ml. (Malick and Singh 1980).

3.6.8.2 Assay of peroxidase (POD)

Processed tender coconut water (10ml) was homogenized in a pre-chilled mortar with 2ml of extraction buffer, containing 50 mM sodium phosphate buffer pH 7.0. The homogenates were centrifugal at 10,000 rpm for 15minutes and the supernatant was used for the assay of antioxidant enzymes *viz.* peroxidase and catalase.

The reaction mixture contained 2.99ml of 0.03% H₂O₂ in 0.1M phosphate buffer (pH 6.0) containing 0.01% orthodianisidine dye (freshly prepared, dissolved in methanol). The reaction was initiated by the addition of 10 μ l of enzyme extract. The change in colour of oxidized dye was read at 460nm up to 1 minute at the interval of 15 seconds. Blank was run without the addition of enzyme (Malick and Singh (1980)). The enzyme activity was expressed as Units Δ O.D/min/ml.



(A) Titratable acidity



(B) TSS measurement



(C) Electrical Conductivity



(D) pH measurement



(E) Total Sugar



(F) Total Phenol Content

Plate 3.2 Biochemical Properties of treated tender coconut water

3.6.9 Microbial analysis

3.6.9.1 Preparation of dilutions

The microbial analysis was carried out using the dilution plate count method. The processed tender coconut water was analysed to estimate the fungi and bacteria population. The potato dextrose agar (PDA) plates for the fungi count were prepared by adding 39g of PDA and 24g of agar –agar powder in one litre of water. The nutrient agar plate for the bacterial count was prepared using 13g of nutrient agar and 40g of agar-agar powder in one litre of water.

One ml processed tender coconut water was mixed with 9ml of sterile water in a volumetric flask to obtain 10^{-1} dilution. Using the serial dilution, 10^{-3} was obtained by transferring 0.1ml to the tube containing 9.9ml of sterile water. Similarly, 10^{-5} and 10^{-7} dilutions was obtained.

3.6.9.2 Total yeast and mould count (CFU/ml)

One ml of suitable dilution from each sample prepared as described in a precious section was used for plating, and thereafter 15 ml of molten PDA agar was poured aseptically to plates. The samples were spread on petri plates and plates were cooled. The plates kept for incubation in an incubator to maintain temperature $27\pm 0.5^{\circ}\text{C}$ for 72h. Number of colony forming units (CFU/ml) was recorded after every 24h. The total plate count in terms of log colony-forming unit per ml (CFU/ml) was calculated using the following formula (AOAC 2000b). The colonies of 10^{-3} and 10^{-7} were considered for calculations. All the procedure were carried out in aseptic condition and with proper precaution.

$$\text{YMC} \left(\frac{\text{CFU}}{\text{ml}} \right) = \log \left(\frac{\text{Mean Number of colony forming units}}{\text{Volume of samples} \times \text{Dilution factor}} \right) \times 100$$

3.6.9.3 Total plate count (CFU/ml)

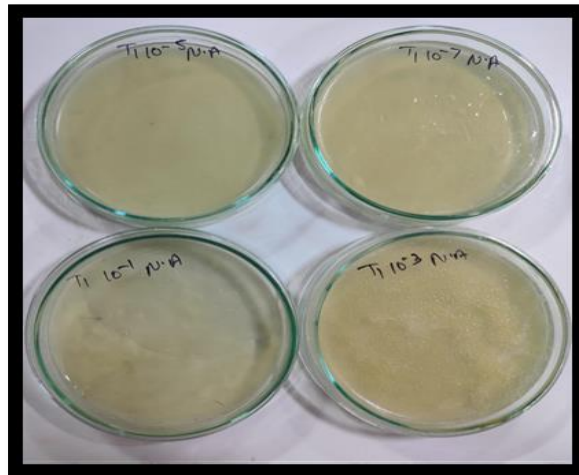
One ml of suitable dilution from each sample spreading to prepared media petri plates. After that, the plates were kept in a incubator for incubation and maintained the temperature $37\pm 0.5^{\circ}\text{C}$ for 48h. Number of colony forming units (CFU/ml) was recorded after every 24h. The total plate count in terms of log colony forming unit per ml

(CFU/ml) was calculated using the following formula (AOAC 2002a). The colonies of 10^{-3} and 10^{-7} were considered for calculations.

$$\text{TPC} \left(\frac{\text{CFU}}{\text{ml}} \right) = \log \left(\frac{\text{Number of colony forming units}}{\text{Volume of samples} \times \text{Dilution factor}} \right) \times 100$$



(A) Preparation of dilutions



(B) Colony count

Plate 3.3 Microbial analysis of treated tender coconut water

3.7 Sensory analysis

Sensory evaluation was conducted for bread. A panel of 15 semi trained panel lists of faculty members and post graduate students of College of Agricultural Engineering and Technology, Junagadh Agricultural University, Junagadh were asked to assess the samples and mark them on a hedonic rating test (1 = dislike extremely, 2 = dislike very much, 3 = dislike moderately, 4 = dislike slightly, 5 = neither like nor dislike, 6 = like slightly, 7 = like moderately, 8 = like very much and 9 = like extremely) in accordance with their opinion for colour, flavour, taste, and overall acceptability (Amerine *et al.* 1965). Sample were kept in petri dish. All tests were performed under uniform lighting conditions, and the subjects were not informed about the background of the study. The samples scoring an overall quality of 5 or above were considered acceptable and those scoring below 5 were considered unacceptable.

3.8 Storage analysis

In the last phase, storage stability was aimed to investigate the quality of processed tender coconut water stored in glass and PET bottles. The samples were kept for 4weeks storage at refrigerated condition at $4\pm 2^{\circ}\text{C}$. At an interval of 1week, the biochemical and microbial analysis were carried out.



(A) Sensory Analysis



(B) Storage Analysis

Plate 3.4 Sensory and Storage Analysis of treated tender coconut water

3.9 Statistical analysis

All the experiments in this study were conducted two times and the mean values were reported. Statistical analysis was done to study the effect of two different factors like treatment and packaging material on dependent parameters, i. e. pH, TSS, titratable acidity, EC, TDS, total sugar, total phenol content, microbial analysis and enzyme activities by Factorial Completely Randomized Design (FCRD by using Microsoft Office Excel 2013) (Pansee and Sukhatme, 1985).

All the treatments were compared at 0.05% level of significance using the Critical Difference test (Microsoft Office Excel 2013). The Analysis of Variance (ANOVA), Standard Error of difference (SEd), Standard Error of mean (SEm) and Critical Difference (CD) for dependent parameter were tabulated and the level of significance was reported.

Fig. 3.1 Process Flow Chart of experiment

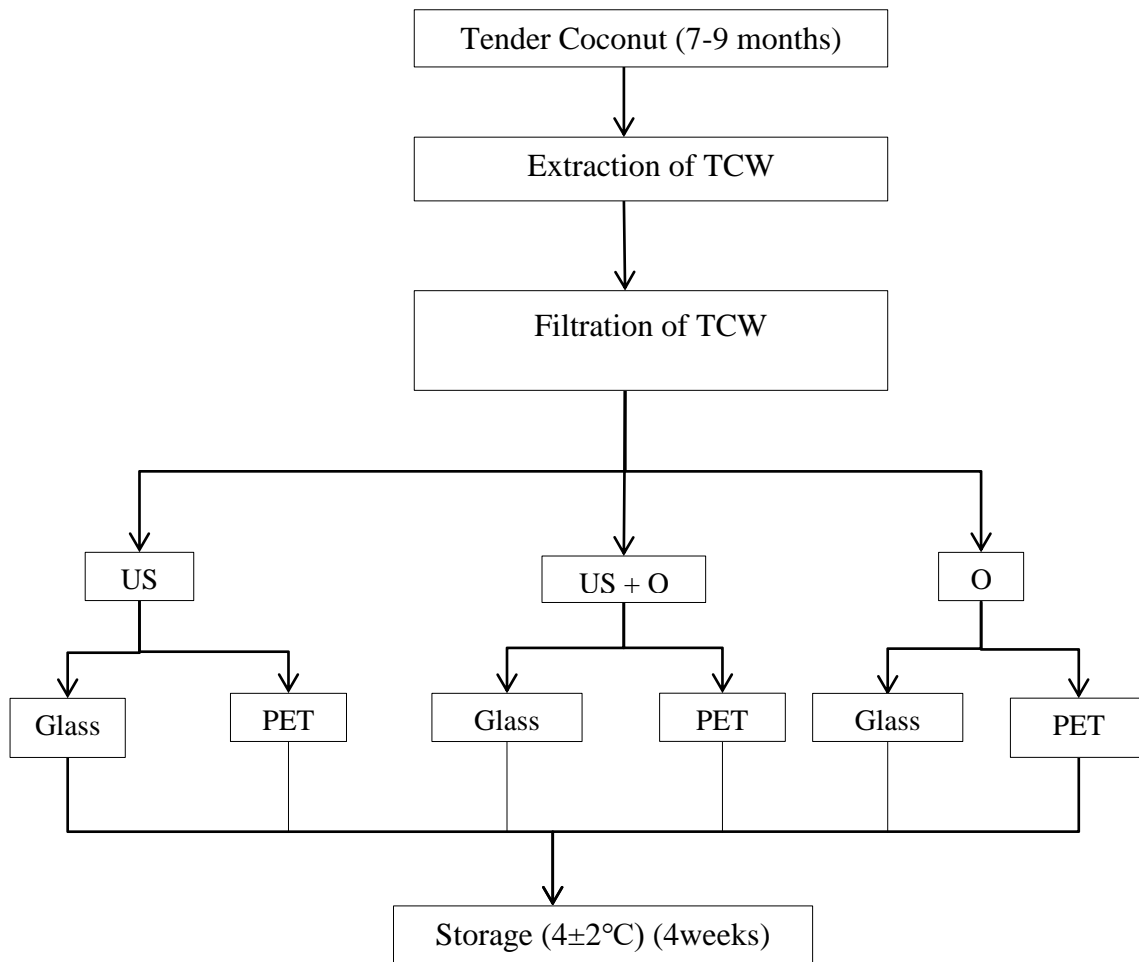


Fig. 3.1 Process flow chart of experiment for tender coconut water

CHAPTER – IV

RESULTS AND DISCUSSION

This chapter deals with the results and discussion part of the experiment, which was carried out for the storage and packaging of tender coconut water using non thermal (ozonation and ultrasonication) treatment with different packaging material as glass and PET bottles stored at $4\pm 2^{\circ}\text{C}$ temperature. Effect of different treatments of ozonation, ultrasonication, packaging material and storage conditions for shelf life extension of tender coconut water, in terms of dependent variables like TSS, pH, total sugar, electrical conductivity, total phenol content, total viable count, yeast and mould count were studied. The quality evaluation was done in terms of sensory evaluation for customer acceptance and microbial count to determine the shelf life of the lime. The data was analyzed using Factorial Completely Randomized Design (FCRD) with three factors and the variations in the parameters were presented in tables and depicted graphically. The results were reported and discussed in the following subsequent paragraphs.

4.1 Physical properties of fresh coconut

The results of physical properties of fresh tender coconut like weight, size, volume, and sphericity of fresh tender coconut fruits are presented in Table 4.1 and described here under. Weight of 10 fruits was measured and average value was recorded. The size, sphericity and volume of coconut was calculated by using the formula as explained in Chapter III. The dimensions (length, width and thickness) of coconut were measured and express in mm. Table 4.1 shows variation in physical dimensions and properties of coconut. The maximum weight, length, width and thickness of the coconut fruit was recorded to be 2.21kg, 296.37, 154.45 and 184.68 mm whereas, minimum was 1.39kg, 274.59, 125.25, and 161.61mm with their standard deviation of 0.227, 7.561, 9.895 and 6.930 respectively. The average value of weight, length, width, thickness of the coconut fruit were measured 1.883kg, 285.19, 140.43 and 180.8 mm respectively. The average size, volume and water recovery of the coconut were found to be 192.96 mm 271.41m^3 and 419.85ml and values ranged from 183 to 203 mm, 190 to 329 m^3 and 363 to 520 ml. The average value of sphericity for coconut was found to be 0.677.

Table 4.1 Variation in weight, length, width, thickness, size, volume, sphericity and water quantity of tender coconut

S. No	weight (kg)	length (l) mm	width (b) mm	thickness (h) mm	Size (mm) $D=(lbh)^{1/3}$	Sphericity $\varepsilon= [(lbh)^{1/3}]/l$	Volume (m ³) $V=4/3 \pi lbh$	Water quantity/per (ml)
1	1.67	287.64	145	161.61	188.7	0.656	282.13	412
2	2.21	276.54	125.25	179.54	183.79	0.665	260.14	405
3	1.86	296.37	153.55	183.71	202.84	0.684	239.41	387.5
4	2.02	283.18	130.67	184.38	189.56	0.669	190.95	363
5	2.06	289.47	138.96	178.8	192.92	0.666	200.56	376.5
6	1.39	291.84	146.03	184.38	198.7	0.681	328.71	457
7	2	293.18	142.76	178.8	195.49	0.667	313.08	432
8	1.91	278.46	154.45	184.68	199.41	0.716	332.2	520.5
9	1.86	280.65	138.23	180.84	191.33	0.682	293.52	427
10	1.85	274.59	129.38	184.03	186.89	0.681	273.41	417.5
AVERAGE	1.883	285.19	140.43	180.08	192.96	0.677	271.41	419.85
RANGE	1.85 to 2.21	274 to 297	125 to 155	161 to 185	183 to 203	0.656 to 0.684	190 to 329	363 to 520
SD	0.227	7.561	9.895	6.930	6.072	0.017	49.405	44.928

4.2 Effect of different process parameters on treated tender coconut water

Processed tender coconut water were prepared as per the combination of different process variables i.e., ozone gas flow rate and time, ultrasonication treatment time, amplitude, frequency and pulse presented in table. Properties such as biochemical, enzyme activities, microbial and sensory were determined.

4.2.1 Effect of different treatment time and different packaging materials on total soluble solids (TSS) of treated tender coconut water

Total soluble solids (TSS) content is the measure of total dissolved solids in terms of mineral in tender coconut water which is the quality parameter of it. An appraisal of data on the effect of different treatment time and different packaging materials on total soluble solids (TSS) of processed tender coconut water is summarized in Table 4.2, 4.3 and 4.4 and graphically depicted in Fig. 4.1, 4.2 and 4.3.

4.2.1.1 Effect of Ozonation treatment on TSS content for tender coconut water

From the Table 4.2 as well as from Fig. 4.1, it is clear that the ozonation treatment processed tender coconut water total soluble solids ranged from 4.85 to 5.19 °Brix increasing with increase in storage period when stored at 4±2°C in different packaging. Total soluble solids (TSS) content was increasing with ozonation treatment time and values were found more in glass bottle as compared to PET bottle.

Ozonation treatment processed tender coconut water the maximum total soluble solids (TSS) of 4th week of storage period was obtained in ozonation treatment 30min stored in glass bottle (O₃S₁), 30min stored in PET bottle (O₂S₂) and 20min stored in glass bottle (O₂S₁) were 5.19, 5.03 and 5.01°Brix respectively. Tender coconut water the minimum total soluble solids (TSS) 4th week of storage period was obtained in ozonation treatment 20min stored in glass bottle (O₂S₁), 10min stored in glass and PET bottle (O₁S₁) and (O₁S₂) were 4.89 and 4.98°Brix respectively (Table 4.2).

The ozonation treated coconut water at different storage period was influenced significantly by different processing times at 1st, 2nd, 3rd and 4th week of storage period. The tender coconut water treatment O₁S₁ and O₃S₁ were 3rd and 4th week showed significantly higher total soluble solids (TSS) of processed tender coconut water at different storage period. Though, it was statistically at par with 1st, 2nd, 3rd and 4th week of

storage period. From the results is also clear that as the storage period increases from 1st week to 4th week, the slight variations observed in TSS of the processed tender coconut also did not characterize relevant change. Similar behaviour has been reported for ozone processing of prebiotic orange juice (Almeida *et al.*, 2015), acai juice (Oliveira *et al.*, 2018), and peach juice (Jaramillo-Sanchez *et al.*, 2017).

Table 4.2 Effect of different ozonation treatment time and different packaging materials on the total soluble solids (TSS) for tender coconut water

(Initial Total Soluble Solids 4.94°Brix)

Mean Total Soluble Solids (TSS) , °Brix				
Treatments	Storage period in weeks			
	1 week	2 week	3 week	4 week
O ₁ S ₁	4.88	4.94	5.19	4.98
O ₁ S ₂	4.92	4.96	4.89	4.98
O ₂ S ₁	4.98	4.97	4.87	4.89
O ₂ S ₂	5.02	5.00	4.93	5.01
O ₃ S ₁	5.10	5.03	5.24	5.19
O ₃ S ₂	4.90	4.85	4.98	5.03
S. Em	0.0054	0.0047	0.0056	0.0053
CD at 5%	0.0168	0.0145	0.0173	0.0162
Test	Sig.	Sig.	Sig.	Sig.
CV %	0.1898	0.1647	0.1937	0.1821

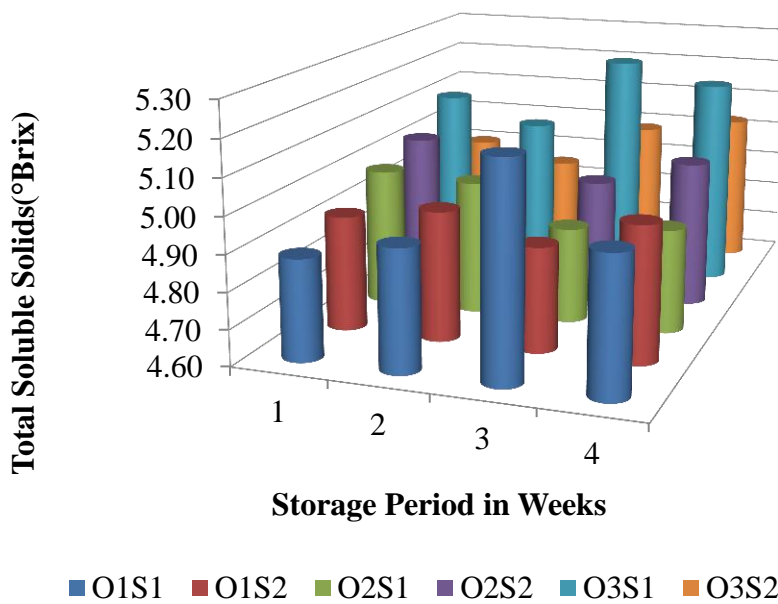


Fig. 4.1 Effect of different ozonation treatment time and different packaging materials on the total soluble solids (TSS) of treated tender coconut water

The interaction of different treatment time and different packaging materials on the total soluble solids (TSS) of processed tender coconut water was found significantly at 1st, 2nd, 3rd and 4th week of storage period at 5% level of significance.

So it is concluded that ozonation treatment should be given for 20 min with glass bottle packaging to get more TSS content in tender coconut water for the storage period of 4 week.

4.2.1.2 Effect of Ultrasonication treatment time and packaging material on TSS for tender coconut water

From the Table 4.3 as well as from Fig. 4.2, it is clear that the ultrasonication treatment processed tender coconut water total soluble solids ranged from 4.83 to 5.23 °Brix increasing with increase in storage period when stored at 4±2°C in different packaging. Total soluble solids (TSS) content was increasing with ultrasonication treatment time and values were found more in PET bottle as compared to glass bottle.

In ultrasonication processed tender coconut water, the maximum total soluble solids (TSS) 4th week of storage period was obtained in treatment 30min stored in PET bottle (U₃S₂), 20min stored in glass bottle (U₂S₁) and 10min treatment stored in PET bottle (U₁S₂) were 5.23, 5.17 and 5.03 °Brix respectively. The minimum total soluble solids (TSS) 4th week of storage period was obtained in treatment 20min stored in PET bottle (U₂S₂), 30min treatment stored in glass bottle (U₃S₁) and 10min stored in glass bottle (U₁S₁) were 4.93, 4.95 and 4.98 °Brix respectively (Table 4.3). Similar behaviour has been reported for ultrasound processing of grape juice for various time intervals also showed no remarkable change in total soluble solids (TSS) during storage period. (Adil *et al.*, 2015)

The ultrasonication processed coconut water at different storage period was influenced significantly by different processing times at 1st, 2nd, 3rd and 4th week of storage period. The processed tender coconut water stored in glass bottle showed significantly higher total soluble solids (TSS) of processed tender coconut water at 1st, 2nd week of storage period and PET bottle showed significantly higher TSS of processed tender coconut water at 3rd, 4th week of storage period. Though, it was statistically at par with 1st, 2nd, 3rd and 4th week storage period.

From the results is also clear that as the storage period increases from 1st week to 4th week, the slight variations observed in TSS of the processed tender coconut also did not characterize relevant change.

Table 4.3 Effect of different treatment time and different packaging materials on the total soluble solids (TSS) of ultrasonication treated tender coconut water

Initial Total Soluble Soids 4.94°Brix

Mean Total Soluble Solids (TSS) , °Brix				
Treatments	Storage period in weeks			
	1 week	2 week	3 week	4 week
U ₁ S ₁	4.91	4.83	4.94	4.98
U ₁ S ₂	4.97	4.99	4.86	5.03
U ₂ S ₁	4.99	5.01	4.86	5.17
U ₂ S ₂	4.92	4.87	4.97	4.93
U ₃ S ₁	4.88	4.94	4.99	4.95
U ₃ S ₂	4.89	4.90	5.08	5.23
S. Em	0.0047	0.0059	0.0051	0.0043
CD at 5%	0.0145	0.0183	0.0157	0.0133
Test	Sig.	Sig.	Sig.	Sig.
CV %	0.1657	0.2086	0.1782	0.1476

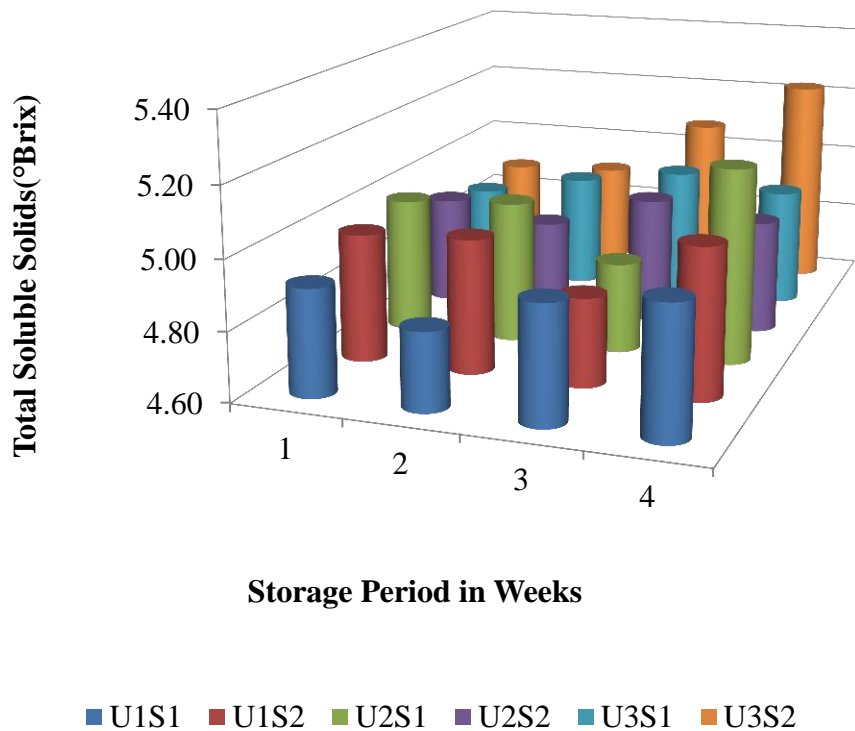


Fig. 4.2 Effect of different treatment time and different packaging materials on the total soluble solids (TSS) of ultrasonication treated tender coconut water

The interaction of different treatment time and different packaging materials on the total soluble solids (TSS) of processed tender coconut water was found significantly at 1st, 2nd, 3rd and 4th week of storage period at 5% level of significance.

So it is concluded that ultrasonication treatment should be given for 30 min with PET bottle packaging to get more TSS content in tender coconut water for the storage period of 4 week.

4.2.1.3 Combined effect of Ozonation and Ultrasonication treatment time and packaging material for TSS on tender coconut water

From the Table 4.4 as well as from Fig. 4.3, it is clear that the combined treatment processed tender coconut water total soluble solids ranged from 4.87 to 5.30 °Brix increasing with increase in storage period when stored at 4±2°C in different packaging. Total soluble solids (TSS) content was increasing with ultrasonication treatment time and values were found more in glass bottle as compared to PET bottle.

Combined treatment processed tender coconut water the maximum total soluble solids (TSS) 4th week of storage period was obtained in treatment 20min ozonation and 20min ultrasonication stored in glass bottle (O₂U₂S₁), 10min ozonation and 30min ultrasonication stored in glass bottle (O₁U₃S₁) and 10min ozonation and 20min ultrasonication stored in PET bottle (O₁U₂S₂) were 5.29, 5.28 and 5.27 °Brix respectively. The minimum total soluble solids (TSS) 4th week of storage period was obtained in treatment 20min ozonation and 30min ultrasonication stored in glass bottle (O₂U₃S₁) and 10min ozonation and 10min ultrasonication stored in PET bottle (O₁U₁S₂) were 4.93 and 4.97 °Brix respectively (Table 4.4).

The ozonation and ultrasonication treatment processed coconut water at different storage period was influenced significantly by different processing times at 1st, 2nd, 3rd and 4th week of storage period. The processed tender coconut water stored in glass bottle showed significantly higher total soluble solids (TSS) of processed tender coconut water at different storage period. Though, it was statistically at par with treatment 1st, 2nd, 3rd and 4th week storage period. From the results is also clear that as the storage period

increases from 1st week to 4th week, the slight variations observed in total soluble solids (TSS) of the processed tender coconut also did not characterize relevant change.

The interaction of different treatment time and different packaging materials on the total soluble solids (TSS) of processed tender coconut water was found significantly at 1st, 2nd, 3rd, and 4th week of storage period at 5% level of significance.

Table 4.4 Effect of different treatment time and different packaging materials on the total soluble solids (TSS) of combined treated tender coconut water

Initial Total Soluble Solids 4.94°Brix

Mean Total Soluble Solids (TSS) , °Brix				
Treatments	Storage period in weeks			
	1 week	2 week	3 week	4 week
O ₁ U ₁ S ₁	4.89	4.91	5.07	5.23
O ₁ U ₁ S ₂	4.93	4.91	4.87	4.97
O ₁ U ₂ S ₁	5.13	4.95	5.02	5.19
O ₁ U ₂ S ₂	5.07	5.13	5.18	5.27
O ₁ U ₃ S ₁	4.95	4.98	5.05	5.28
O ₁ U ₃ S ₂	5.07	5.17	4.98	5.02
O ₂ U ₁ S ₁	5.12	5.04	4.98	5.17
O ₂ U ₁ S ₂	5.02	5.00	4.97	5.01
O ₂ U ₂ S ₁	5.18	4.97	5.26	5.29
O ₂ U ₂ S ₂	5.23	5.27	4.97	4.98
O ₂ U ₃ S ₁	5.14	4.97	4.87	4.93
O ₂ U ₃ S ₂	5.24	5.19	4.97	4.98
O ₃ U ₁ S ₁	5.24	5.02	5.23	5.21
O ₃ U ₁ S ₂	4.95	4.88	4.96	5.02
O ₃ U ₂ S ₁	4.93	5.23	5.01	5.20
O ₃ U ₂ S ₂	4.87	4.94	4.97	5.03
O ₃ U ₃ S ₁	5.30	5.14	4.96	5.20
O ₃ U ₃ S ₂	4.97	5.23	5.26	5.17
S. Em	0.0055	0.005	0.0048	0.0057
CD at 5%	0.0158	0.0144	0.0137	0.0164
Test	Sig.	Sig.	Sig.	Sig.
CV %	0.1879	0.1725	0.1645	0.1935

So it is concluded that combined treatment should be given for 20 min ozonation and 20min ultrasonication with glass bottle packaging to get more TSS content in tender coconut water for the storage period of 4 week.

4.2.2 Effect of different treatment time and different packaging materials on titratable acidity (TA) of treated tender coconut water

The titratable acidity is the factor that affected the acidity of the fruits and as necessary to maintain during the storage of fruits. Results pertaining to titratable acidity in tender coconut water as influenced by different treatments at 7 days interval during storage are presented Table 4.5, 4.6 and 4.7 and graphically depicted in Fig. 4.4, 4.5 and 4.6 for different treatment time and different packaging materials.

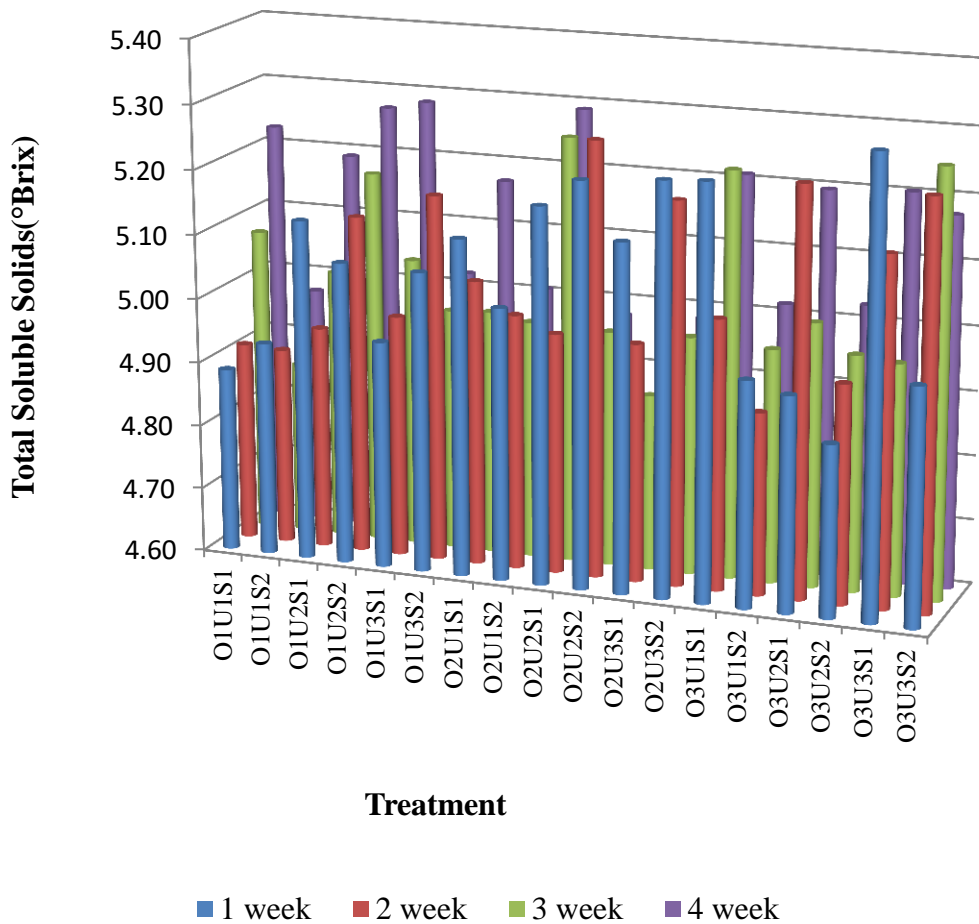


Fig. 4.3 Effect of different treatment time and different packaging materials on the total soluble solids (TSS) of combined treated tender coconut water

4.2.2.1 Effect of Ozonation treatment time and packaging material on TA for tender coconut water

From the Table 4.5 as well as from Fig. 4.4, it is clear that the ozonation treatment processed tender coconut water total soluble solids ranged from 0.07 to 0.09 % increasing and decreasing with increase in storage period when stored at 4±2°C in different packaging.

Titrateable acidity (TA) content was increasing with ozonation treatment time and values were found more in PET bottle as compared to glass bottle.

The results presented in Table 4.5 shows the effects of different time treatment and different packaging in Fig. 4.4. The titrateable acidity (TA) of ozonation treatment processed tender coconut water increases and decreases with the increase in storage period. The maximum titrateable acidity (TA) value of processed tender coconut water was found to be treatment 10min stored in PET bottle (O₁S₂), 20min stored in PET bottle (O₂S₂) and 30min stored in PET bottle O₃S₂ were 0.09 % respectively. The minimum titrateable acidity (TA) value of processed tender coconut water was found to be treatment 10min stored in glass bottle O₁S₁, and 30min stored in glass bottle (O₃S₁) were 0.07 % respectively (Table 4.5).

Table 4.5 Effect of different treatment time and different packaging materials on the titrateable acidity (TA) of ozonation treated tender coconut water
Initial titrateable acidity (TA) = 0.08%

Mean Titrateable Acidity (TA) , %				
Treatments	Storage period in weeks			
	1 week	2 week	3 week	4 week
O ₁ S ₁	0.07	0.07	0.07	0.07
O ₁ S ₂	0.08	0.09	0.08	0.09
O ₂ S ₁	0.09	0.09	0.09	0.08
O ₂ S ₂	0.09	0.08	0.07	0.09
O ₃ S ₁	0.08	0.09	0.09	0.07
O ₃ S ₂	0.08	0.09	0.07	0.09
S. Em	0.0045	0.0047	0.0049	0.0044
CD at 5%	NS	0.0144	NS	0.0137
Test	NS	Sig.	NS	Sig.
CV %	9.8975	9.7461	10.7544	9.6081

The statistical analysis of data revealed that the rate of decrease and increase in titratable acidity (TA) due to different treatment time and different packaging materials were found to be significant with the storage period 2nd and 4th week of storage period respectively, where observed significantly higher due to treatment and different packaging materials. Different treatment in PET bottle though 1st, 2nd, 3rd and 4th week storage period was remained statistically at par with different treatment time. These findings were as par with results reported on ozone treatment of orange (Tiwari *et al.*, 2008) and tomato (Tiwari *et al.*, 2009) juices where a slightly decrease in acidity.

The interaction of different treatment time and different packaging materials on the total soluble solids (TA) of processed tender coconut water was found significantly at 2nd and 4th week of storage period at 5% level of significance.

So it is concluded that combined treatment should be given for 10 min, 20min and 30min ozonation with PET bottle packaging to get more TA content in tender coconut water for the storage period of 4 week.

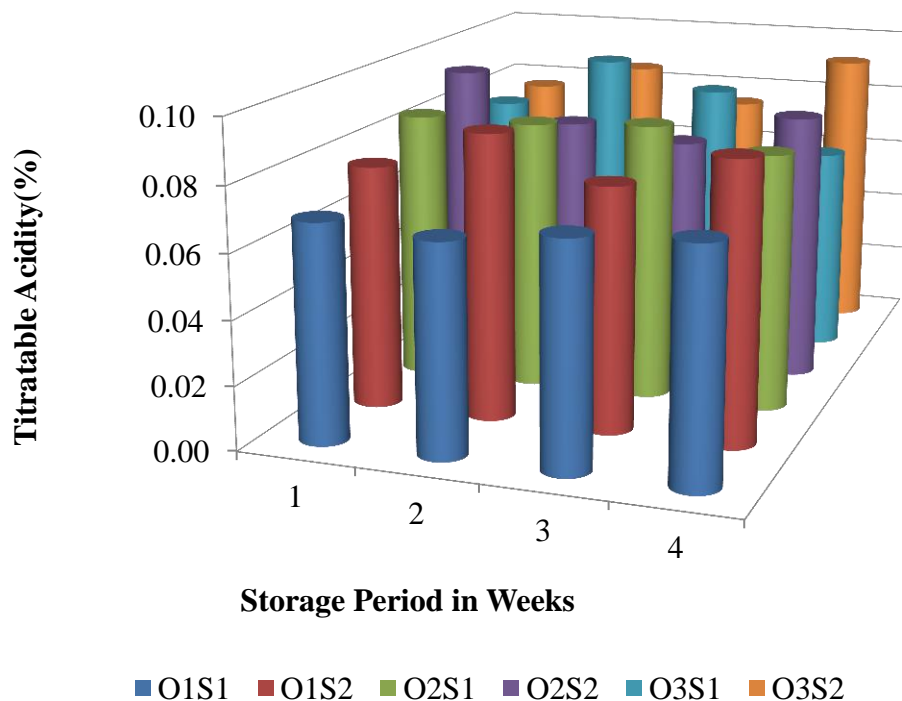


Fig. 4.4 Effect of different treatment time and different packaging materials on the titratable acidity (TA) of ozonation treated tender coconut water

4.2.2.2 Effect of Ultrasonication treatment time and packaging material on TA for tender coconut water

From the Table 4.6 as well as from Fig. 4.5, it is clear that the ultrasonication treatment processed tender coconut water total soluble solids ranged from 0.07 to 0.11 % increasing and decreasing with increase in storage period when stored at 4±2°C in different packaging. Titratable acidity (TA) content was increasing with ultrasonication treatment time and values were found more in PET bottle as compared to glass bottle.

The results presented in Table 4.6 shows the data on the effects of different time treatment and different packaging in Fig. 4.5. The titratable acidity (TA) of ultrasonication treatment processed tender coconut water increases and decreases with the increase in storage period. The maximum titratable acidity (TA) value of processed tender coconut water was found to be treatment 20min stored in PET bottle (U₂S₂) and 30min stored in PET bottle (U₃S₂) 0.10 and 0.09 % after 4th week of storage period respectively. The minimum titratable acidity (TA) value of processed tender coconut water was found to be treatment 10min stored in glass bottle(U₁S₁), 10min stored in PET bottle (U₁S₂) and 30min stored in glass bottle (U₁S₁) 0.08 % after 4th week of storage period respectively. .

Table 4.6 Effect of different treatment time and different packaging materials on the titratable acidity (TA) of ultrasonication treated tender coconut water

Initial Titratable Acidity (TA) = 0.08%

Mean Titratable Acidity (TA) , %				
Treatments	Storage period in weeks			
	1 week	2 week	3 week	4 week
U ₁ S ₁	0.07	0.07	0.09	0.07
U ₁ S ₂	0.09	0.07	0.11	0.07
U ₂ S ₁	0.07	0.08	0.11	0.08
U ₂ S ₂	0.07	0.09	0.11	0.10
U ₃ S ₁	0.07	0.07	0.08	0.07
U ₃ S ₂	0.08	0.07	0.08	0.09
S. Em	0.0045	0.0047	0.005	0.0051
CD at 5%	NS	NS	NS	NS
Test	NS	NS	NS	NS
CV %	10.52	10.74	8.96	10.58

The statistical analysis of data revealed that the rate of increase and decrease in titratable acidity (TA) due to different treatment time and different packaging materials were found to be not significant with the storage period 1st, 2nd, 3rd and 4th week of storage period respectively, where observed significantly higher due to 1st, 2nd, 3rd and 4th week storage period was remained statistically at par with different treatment time. Similar behaviour has been reported for ultrasound processing of grape juice for various time intervals also showed no remarkable change in acidity during storage period. (Adil *et al.*, 2015)

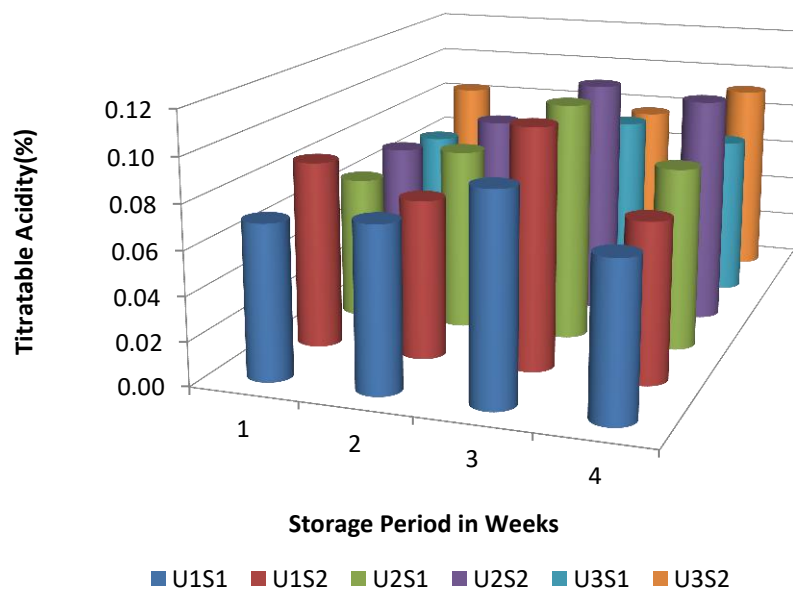


Fig. 4.5 Effect of different treatment time and different packaging materials on the titratable acidity (TA) of ultrasonication treated tender coconut water

The interaction of different treatment time and different packaging materials on the total soluble solids (TA) of processed tender coconut water was found non-significant at 1st, 2nd, 3rd and 4th week of storage period at 5% level of significance.

So it is concluded that ultrasonication treatment should be given for 20min with PET bottle packaging to get more TA content in tender coconut water for the storage period of 4 week.

4.2.2.3 Combined effect of Ozonation and Ultrasonication treatment time and packaging material on TA for tender coconut water

The data presented in Table 4.7 shows the effects of different time treatment and different packaging materials at stored 4±2°C on the titratable acidity (TA) of processed tender coconut water and also shown graphically in Fig. 4.6.

From the Table 4.7 as well as from Fig. 4.6, it is clear that the combined treatment processed tender coconut water total soluble solids ranged from 0.06 to 0.11 % increasing and decreasing with increase in storage period when stored at 4±2°C in different packaging. Titratable acidity (TA) content was increasing with combined treatment time and values were found more in PET bottle as compared to glass bottle.

Table 4.7 Effect of different treatment time and different packaging materials on the titratable acidity (TA) of combined treated tender coconut water

Initial Titratable Acidity (TA) = 0.08%

Mean Titratable Acidity (TA) , %				
Treatments	Storage period in weeks			
	1 week	2 week	3 week	4 week
O ₁ U ₁ S ₁	0.07	0.10	0.07	0.06
O ₁ U ₁ S ₂	0.08	0.10	0.07	0.10
O ₁ U ₂ S ₁	0.09	0.07	0.07	0.08
O ₁ U ₂ S ₂	0.06	0.09	0.08	0.09
O ₁ U ₃ S ₁	0.07	0.11	0.07	0.07
O ₁ U ₃ S ₂	0.09	0.09	0.08	0.11
O ₂ U ₁ S ₁	0.09	0.09	0.07	0.07
O ₂ U ₁ S ₂	0.07	0.07	0.08	0.09
O ₂ U ₂ S ₁	0.08	0.06	0.10	0.08
O ₂ U ₂ S ₂	0.10	0.09	0.09	0.09
O ₂ U ₃ S ₁	0.08	0.10	0.09	0.07
O ₂ U ₃ S ₂	0.09	0.09	0.09	0.09
O ₃ U ₁ S ₁	0.08	0.09	0.07	0.11
O ₃ U ₁ S ₂	0.06	0.07	0.10	0.08
O ₃ U ₂ S ₁	0.07	0.07	0.10	0.07
O ₃ U ₂ S ₂	0.06	0.10	0.07	0.08
O ₃ U ₃ S ₁	0.11	0.10	0.09	0.09
O ₃ U ₃ S ₂	0.09	0.09	0.09	0.10
S. Em	0.0046	0.005	0.005	0.0055
CD at 5%	0.0133	0.0144	0.0145	0.0158
Test	Sig.	Sig.	Sig.	Sig.
CV %	10.078	9.9594	10.7524	11.2653

The titratable acidity (TA) of ozonation and ultrasonication treatment processed tender coconut water increases and decreases with the increase in storage period. The maximum titratable acidity (TA) value of processed tender coconut water was found to be treatment 10min ozonation and 30min ultrasonication stored in PET bottle (O₁U₃S₂) were 0.11 % after 4weeks of storage period respectively. Whereas the minimum titratable acidity (TA) was found to be treatment 10min ozonation and 10min ultrasonication stored in glass bottle (O₁U₁S₁) were 0.06 % after 4th week of storage period respectively.

The statistical analysis of data revealed that the rate of decrease and increase in titratable acidity (TA) due to different treatment time and different packaging materials were found to be significant with the storage period 1st, 2nd, 3rd, and 4th week of storage period respectively, where observed significantly higher due to 1st, 2nd, 3rd and 4th week storage period was remained statistically at par with different treatment respectively.

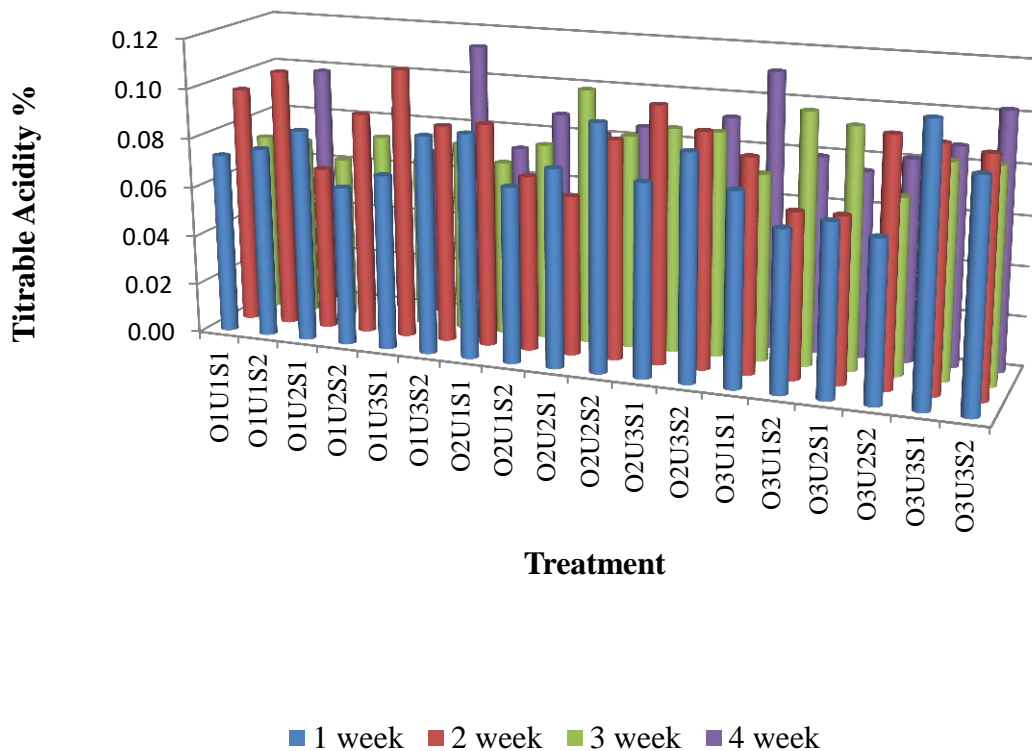


Fig. 4.6 Effect of different treatment time and different packaging materials on the titratable acidity (TA) of combined treated tender coconut water

The interaction of different treatment time and different packaging materials on the titratable (TA) of processed tender coconut water was found significantly at 1st, 2nd, 3rd, and 4th week of storage period at 5% level of significance.

So it is concluded that combined treatment should be given for 10 min ozonation and 30min ultrasonication with PET bottle packaging to get more TA content in tender coconut water for the storage period of 4 week.

4.2.3 Effect of different treatment time and different packaging materials on pH of treated tender coconut water

The letters pH stand for potential of hydrogen, since pH is effectively a measure of the concentration of hydrogen ions in a substance. The pH is the factor that affected the acidity of the fruits and necessary to be maintained during the storage of fruits.

An appraisal of data on the effect of different treatment time and different packaging materials on pH of total processed tender coconut water is summarized in Table 4.8, 4.9 and 4.10 and graphically depicted in Fig. 4.7, 4.8 and 4.9.

4.2.3.1 Effect of Ozonation treatment time and packaging on pH for tender coconut water

The results presented in Table 4.8 shows the effects of different time treatment and different packaging in Fig. 4.7 it is clear that the ozonation treatment processed tender coconut water pH ranged from 4.70 to 5.01 decreasing with increase in storage period when stored at $4\pm 2^{\circ}\text{C}$ in different packaging. The pH was decreasing with ozonation treatment time and values were found more in glass bottle as compared to PET bottle.

The pH of ozonation treatment processed tender coconut water decreases with the increase in storage period. The maximum pH value of processed tender coconut water 4th week of storage period was obtained in treatment 20min stored in glass bottle (O_2S_1), 10min stored in glass bottle (O_1S_1) and 20min stored in PET bottle (O_2S_2) were 4.94, 4.88 and 4.79 respectively. The minimum pH value of processed tender coconut water 4th week of storage period was obtained in treatment 30min stored in PET bottle (O_3S_2), 10min stored in PET bottle (O_1S_2) and 30min stored in glass bottle (O_3S_1) were 4.70 and

4.73 respectively (Table 4.8), which were in confirmation of results found for ozone treatment of orange (Tiwari *et al.*, 2008).

Table 4.8 Effect of different treatment time and different packaging materials on the pH ozonation treated tender coconut water

Initial pH =4.96

Mean pH				
Treatments	Storage period in weeks			
	1 week	2 week	3 week	4 week
O ₁ S ₁	4.95	4.87	4.82	4.88
O ₁ S ₂	4.76	4.73	4.71	4.73
O ₂ S ₁	4.93	4.87	5.02	4.94
O ₂ S ₂	4.80	4.79	4.76	4.79
O ₃ S ₁	4.74	4.76	4.70	4.73
O ₃ S ₂	4.71	4.66	4.73	4.70
S. Em	0.0085	0.0096	0.0104	0.0113
CD at 5%	0.0262	0.0297	0.0319	NS
Test	Sig.	Sig.	Sig.	NS
CV %	0.3057	0.3487	0.3487	0.416

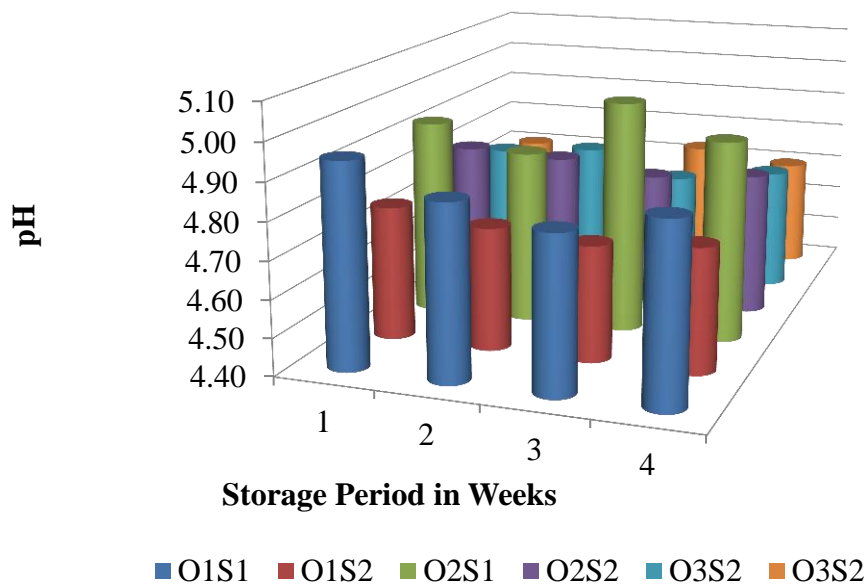


Fig. 4.7 Effect of different treatment time and different packaging materials on the pH ozonation treated tender coconut water

The statistical analysis of data revealed that the rate of decrease in pH due to different treatment time and different packaging materials were found to be significant with the storage period 1st, 2nd and 3rd week of storage period respectively, where

observed significantly higher due to 1st, 2nd, and 3rd week storage period was remained statistically at par with different treatment time respectively where observed significantly higher due to different treatment time O₂S₁ i.e. 20min ozonation treatment and different packaging materials S₁ i.e. glass bottle though 1st, 2nd, and 3rd week storage period was remained statistically at par with different treatment time respectively.

The interaction of different treatment time and different packaging materials on the pH of processed tender coconut water was found significantly at 1st, 2nd and 3rd week of storage period at 5% level of significance.

So it is concluded that ozonation treatment should be given for 20 min ozonation with glass bottle packaging to get more pH in tender coconut water to extend shelf life upto 4 week.

4.2.3.2 Effect of Ultrasonication treatment time and packaging on pH for tender coconut water

The results presented in Table 4.5 shows the effects of different time treatment and different packaging in Fig. 4.8 it is clear that the ultrasonication treatment processed tender coconut water pH ranged from 4.66 to 4.91 decreasing with increase in storage period when stored at 4±2°C in different packaging. pH was decreasing with ozonation treatment time and values were found more in glass bottle as compared to PET bottle.

The pH of ultrasonication treatment processed tender coconut water decreases with the increase in storage period. Ultrasonication treatment processed tender coconut water the maximum pH 4th week of storage period was obtained in treatment 20min stored in glass bottle (U₂S₁), 30min stored in glass bottle (U₃S₁) and 30 min stored in PET bottle (U₃S₂) were 4.87, 4.86 and 4.85 respectively. Ultrasonication treatment processed tender coconut water the minimum pH 4th week of storage period was obtained in treatment 20min stored in PET bottle (U₂S₂), 10min stored in PET bottle (U₁S₂) and 10min stored in glass bottle (U₁S₁) were 4.68, 4.79 and 4.80 respectively (Table 4.9). Similar behaviour has been reported for ultrasound processing of grape juice for various time intervals also showed no remarkable change in pH during storage period. (Adil *et al.*, 2015)

The statistical analysis of data revealed that the rate of decrease in pH due to different treatment time and different packaging materials were found to be significant with the storage period 1st, 2nd and 3rd week of storage period respectively, where observed significantly higher due to different treatment time U₂S₁ i.e. 20min ultrasonication treatment and different packaging materials S₁ i.e. glass bottle though 1st, 2nd, and 3rd week storage period was remained statistically at par with different treatment time respectively.

Table 4.9 Effect of different treatment time and different packaging materials on the pH of ultrasonication treated tender coconut water

Initial pH =4.96

Mean pH				
Treatments	Storage period in weeks			
	1 week	2 week	3 week	4 week
U ₁ S ₁	4.79	4.81	4.80	4.80
U ₁ S ₂	4.80	4.78	4.79	4.79
U ₂ S ₁	4.91	4.82	4.87	4.87
U ₂ S ₂	4.66	4.71	4.68	4.68
U ₃ S ₁	4.86	4.85	4.86	4.86
U ₃ S ₂	4.84	4.86	4.85	4.85
S. Em	0.0085	0.0116	0.0088	0.0119
CD at 5%	0.0262	0.0358	0.0272	NS
Test	Sig.	Sig.	Sig.	NS
CV %	0.3061	0.4198	0.3185	0.4324

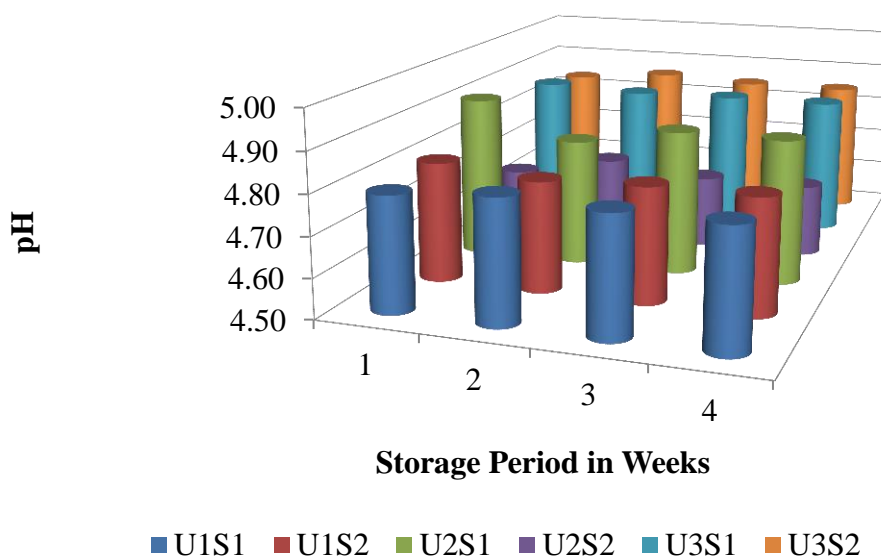


Fig. 4.8 Effect of different treatment time and different packaging materials on the pH of ultrasonication treated tender coconut water

The interaction of different treatment time and different packaging materials on the pH of processed tender coconut water was found significantly at 1st, 2nd and 3rd week of storage period at 5% level of significance. So it is concluded that ultrasonication treatment should be given for 20 min ultrasonication with glass bottle packaging to get more pH in tender coconut water to extend shelf life upto 4 week.

4.2.3.3 Combined effect of Ozonation and Ultrasonication treatment time on pH for tender coconut water

From the Table 4.10 presents the data on the effects of different time treatment and different packaging materials at stored 4±2°C on the pH of processed tender coconut water and also shown graphically in Fig 4.9.

Table 4.10 Effect of different treatment time and different packaging materials on the pH of combined treated tender coconut water

Initial pH =4.96

Mean pH				
Treatments	Storage period in weeks			
	1 week	2 week	3 week	4 week
O ₁ U ₁ S ₁	5.03	4.95	4.86	4.95
O ₁ U ₁ S ₂	4.96	4.91	4.84	4.90
O ₁ U ₂ S ₁	4.92	4.93	4.88	4.91
O ₁ U ₂ S ₂	4.66	4.73	4.72	4.70
O ₁ U ₃ S ₁	4.84	4.77	4.82	4.81
O ₁ U ₃ S ₂	4.91	4.86	4.81	4.86
O ₂ U ₁ S ₁	4.82	4.86	4.87	4.85
O ₂ U ₁ S ₂	4.91	4.90	4.80	4.87
O ₂ U ₂ S ₁	4.71	4.75	4.79	4.75
O ₂ U ₂ S ₂	4.49	4.83	4.86	4.72
O ₂ U ₃ S ₁	4.83	4.98	4.90	4.90
O ₂ U ₃ S ₂	4.77	4.74	4.72	4.74
O ₃ U ₁ S ₁	4.76	4.81	4.77	4.78
O ₃ U ₁ S ₂	4.82	4.75	4.74	4.77
O ₃ U ₂ S ₁	4.71	4.71	4.72	4.72
O ₃ U ₂ S ₂	4.76	4.67	4.77	4.74
O ₃ U ₃ S ₁	4.80	4.87	4.84	4.84
O ₃ U ₃ S ₂	4.89	4.77	4.78	4.81
S. Em	0.0092	0.0131	0.0102	0.0073
CD at 5%	0.0263	0.0375	0.0292	0.0209
Test	Sig.	Sig.	Sig.	Sig.
CV %	0.3299	0.4697	0.367	0.2657

The results presented in Table 4.10 shows the effects of different time treatment and different packaging in Fig. 4.9 it is clear that the combined treatment processed tender coconut water pH ranged from 4.49 to 5.03 decreasing with increase in storage period when stored at $4\pm 2^{\circ}\text{C}$ in different packaging. pH was decreasing with combined treatment time and values were found more in glass bottle as compared to PET bottle.

The pH of combined treatment processed tender coconut water decreases with the increase in storage period. Combined treatment processed tender coconut water the maximum pH 4th week of storage period was obtained in treatment 10min ozonation and 20min ultrasonication stored in glass bottle ($O_1U_2S_1$), 20min ozonation and 20 min ultrasonication stored in glass bottle ($O_2U_2S_1$) and 30min ozonation and 20min ultrasonication stored in glass bottle ($O_3U_2S_1$) were 4.95 and 4.91 respectively.

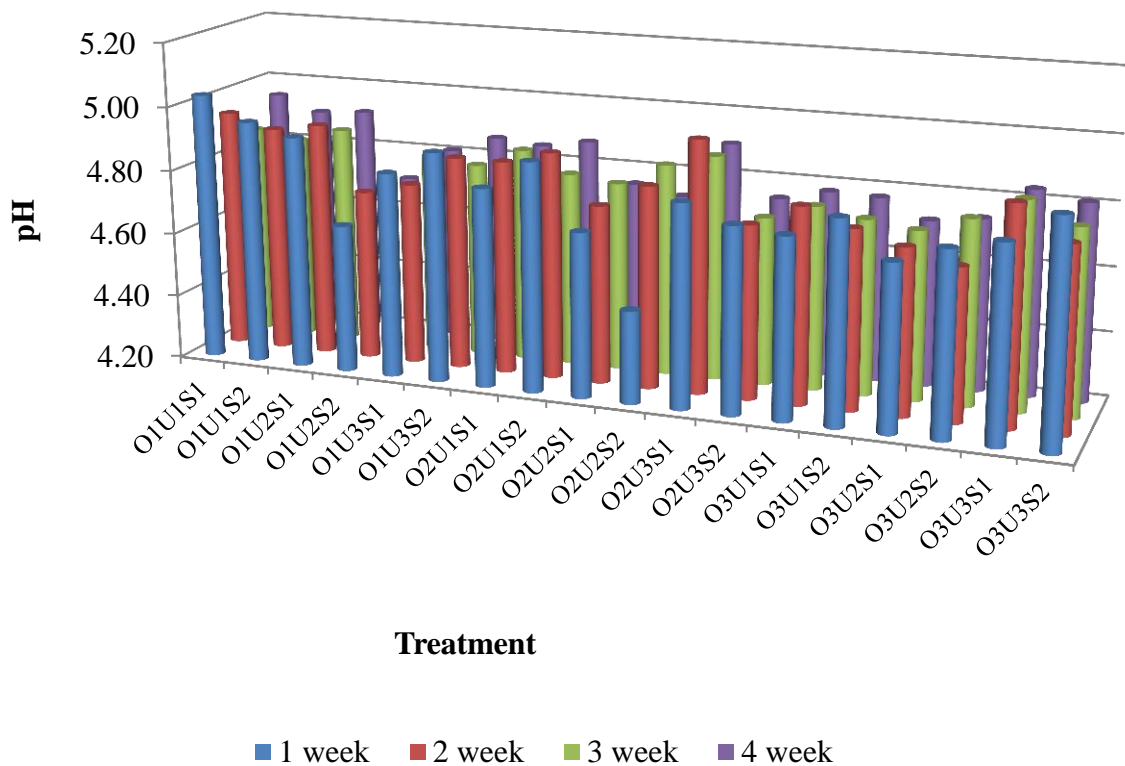


Fig. 4.9 Effect of different treatment time and different packaging materials on the pH of combined treated tender coconut water

Combined treatment processed tender coconut water the minimum pH 4th week of storage period was obtained in treatment 10min ozonation and 20min ultrasonication stored in PET bottle ($O_1U_2S_2$), 20min ozonation and 20min ultrasonication stored in PET

bottle (O₂U₂S₂) and 30min ozonation and 20min ultrasonication stored in glass bottle (O₃U₂S₁) were 4.70 and 4.72 respectively (Table 4.10).

The statistical analysis of data revealed that the rate of decrease in pH due to different treatment time and different packaging materials were found to be significant with the storage period 1st, 2nd, 3rd, and 4th week of storage period respectively, where observed significantly higher due to different treatment time O₁U₁S₁ i.e. 10min ozonation treatment 10min ultrasonication treatment and different packaging materials S₁ i.e. glass bottle though 1st, 2nd, and 3rd week storage period was remained statistically at par with different treatment time respectively.

So it is concluded that combined treatment should be given for 10 min ozonation and 10min ultrasonication with glass bottle packaging to get more pH in tender coconut water. The interaction of different treatment time and different packaging materials on the pH of processed tender coconut water was found significantly at 1st, 2nd, 3rd and 4th week of storage period at 5% level of significance.

4.2.4 Effect of different treatment time and different packaging materials on Electrical Conductivity (EC) of treated tender coconut water

Electrical Conductivity (EC) is a measure of the ability of water to pass an electrical current. Because dissolved salts and other inorganic chemicals conduct electric current, conductivity increases as salinity increases. An appraisal of data on the effect of different treatment time and different packaging materials on Electrical Conductivity (EC) of total processed tender coconut water is summarized in Table 4.11, 4.12 and 4.13 and graphically depicted in Fig 4.10, 4.11 and 4.12.

4.2.4.1 Effect of Ozonation treatment on EC content for tender coconut water

From the Table 4.11 presents the data on the effects of different time treatment and different packaging in Fig. 4.10 it is clear that the ozonation treatment processed tender coconut water EC ranged from 5.10 to 6.02 mS/cm decreasing with increase in storage period when stored at 4±2°C in different packaging. EC was decreasing with ozonation treatment time and values were found more in PET bottle as compared to glass bottle.

Results and discussion

The Electrical Conductivity (EC) of ozonation treatment processed tender coconut water decreases with the increase in storage period. The maximum Electrical Conductivity (EC) value of processed tender coconut water 4th week of storage period was obtained in treatment 20min stored in PET bottle (O₂S₂), 30min stored in glass bottle (O₃S₁) and 10min stored in PET bottle (O₁S₂) were 5.84, 5.82 and 5.64 mS/cm respectively. The minimum Electrical Conductivity (EC) value of processed tender coconut water 4th week of storage period was obtained in treatment 20min stored in glass bottle (O₂S₁), 10min stored in glass bottle (O₁S₁) and 30min stored in PET bottle (O₃S₂) were 5.10, 5.32 and 5.41 mS/cm respectively (Table 4.11). These similar findings were reported by Kannangara *et al.*, (2018) in comparative analysis of coconut water in four different maturity stages.

The statistical analysis of data revealed that the rate of decrease in Electrical Conductivity (EC) due to different due to different treatment time and different packaging materials were found to be significant with the storage period 1st, 2nd, 3rd, and 4th week of storage period respectively, where observed significantly higher due to different treatment time O₂S₂ i.e. 20min ozonation treatment and different packaging materials S₂ i.e. PET bottle though 1st, 2nd, and 3rd week storage period was remained statistically at par with different treatment time respectively.

Table 4.11 Effect of different treatment time and different packaging materials on the Electrical Conductivity (EC) of ozonation treated tender coconut water

Initial Electrical Conductivity (EC) = 5.84 mS/cm

Mean Electrical Conductivity (EC) , mS/cm				
Treatments	Storage period in weeks			
	1 week	2 week	3 week	4 week
O ₁ S ₁	5.52	5.38	5.38	5.32
O ₁ S ₂	5.74	5.15	5.87	5.64
O ₂ S ₁	5.68	5.88	5.23	5.10
O ₂ S ₂	5.95	5.08	5.89	5.84
O ₃ S ₁	5.89	6.02	5.76	5.82
O ₃ S ₂	5.65	5.65	5.04	5.41
S. Em	0.0021	0.0035	0.0024	0.0114
CD at 5%	0.0065	0.0108	0.0073	0.035
Test	Sig.	Sig.	Sig.	Sig.
CV %	0.0636	0.1099	0.0743	0.3561

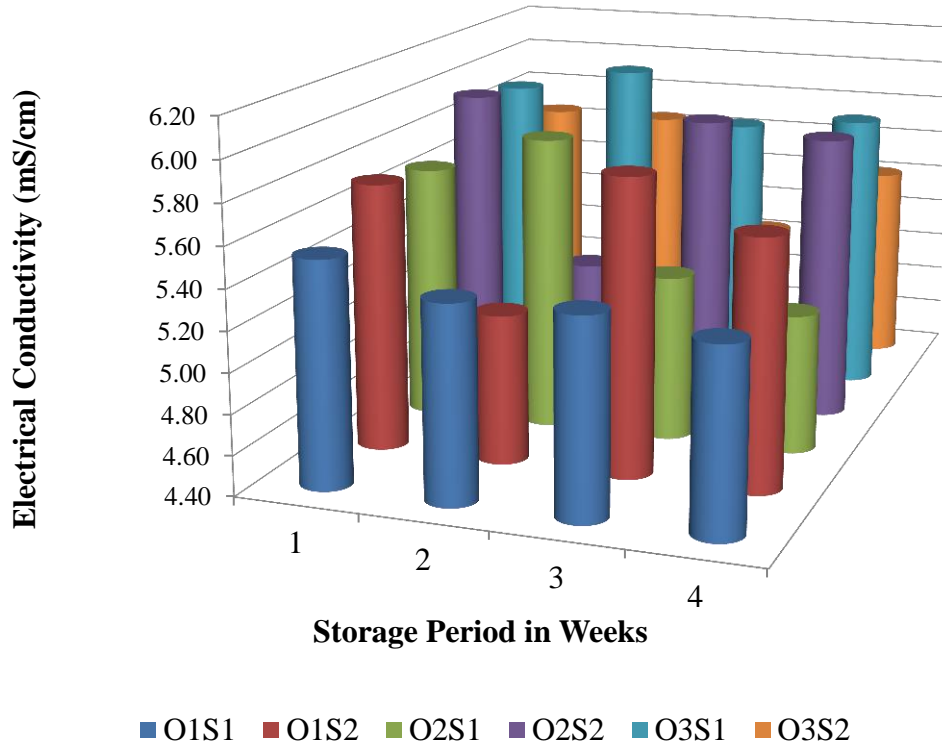


Fig. 4.10 Effect of different treatment time and different packaging materials on the Electrical Conductivity (EC) of ozonation treated tender coconut water

The interaction of different treatment time and different packaging materials on the EC of processed tender coconut water was found significantly at 1st, 2nd, 3rd, and 4th week of storage period at 5% level of significance.

So it is concluded that ozonation treatment should be given for 20 min with PET bottle packaging to get more EC in tender coconut water for extending shelf life upto 4 week.

4.2.4.2 Effect of Ultrasonication treatment time on EC for tender coconut water

The results presented in Table 4.12 shows the effects of different time treatment and different packaging materials at stored 4±2°C on the Electrical Conductivity (EC) of processed tender coconut water and also shown graphically in Fig. 4.11. The Electrical Conductivity (EC) of ultrasonication treatment processed tender coconut water decreases with the increase in storage period.

Results and discussion

Ultrasonication treatment processed tender coconut water EC ranged from 5.10 to 6.03 mS/cm decreasing with increase in storage period when stored at $4\pm 2^{\circ}\text{C}$ in different packaging. EC was decreasing with ozonation treatment time and values were found more in PET bottle as compared to glass bottle.

Ultrasonication treatment processed tender coconut water the maximum Electrical Conductivity (EC) 4th week of storage period was obtained in treatment 30min stored in PET bottle (U₃S₂), 10min stored in PET bottle (U₁S₂) and 30min stored in glass bottle (U₃S₁) were 5.83, 5.38 and 5.29 mS/cm respectively. Ultrasonication treatment processed tender coconut water the minimum Electrical Conductivity (EC) 4th week of storage period was obtained in treatment 20min stored in PET bottle (U₂S₂), 10min stored in glass bottle (U₁S₁) and 20min stored in glass bottle (U₂S₁) were 5.15, 5.18 and 5.24 mS/cm respectively (Table 4.12).

The statistical analysis of data revealed that the rate of decrease in Electrical Conductivity (EC) due to different due to different treatment time and different packaging materials were found to be significant with the storage period 1st, 2nd, 3rd, and 4th week of storage period respectively, where observed significantly higher due to different treatment time U₃S₂ i.e. 30min ultrasonication treatment and different packaging materials S₂ i.e. PET bottle though 1st, 2nd, 3rd and 4th week storage period was remained statistically at par with different treatment time respectively.

Table 4.12 Effect of different treatment time and packaging materials on the Electrical Conductivity (EC) of ultrasonication treated tender coconut water

Initial Electrical Conductivity (EC) = 5.84 mS/cm

Mean Electrical Conductivity (EC) , mS/cm				
Treatments	Storage period in weeks			
	1 week	2 week	3 week	4 week
U ₁ S ₁	5.63	5.22	5.13	5.18
U ₁ S ₂	5.12	5.50	5.57	5.38
U ₂ S ₁	5.03	6.03	6.33	5.24
U ₂ S ₂	5.15	5.33	5.97	5.15
U ₃ S ₁	5.86	5.15	5.28	5.29
U ₃ S ₂	5.90	5.25	5.16	5.83
S. Em	0.0025	0.0019	0.0027	0.0073
CD at 5%	0.0077	0.006	0.0084	0.0225
Test	Sig.	Sig.	Sig.	Sig.
CV %	0.0791	0.0619	0.0852	0.237

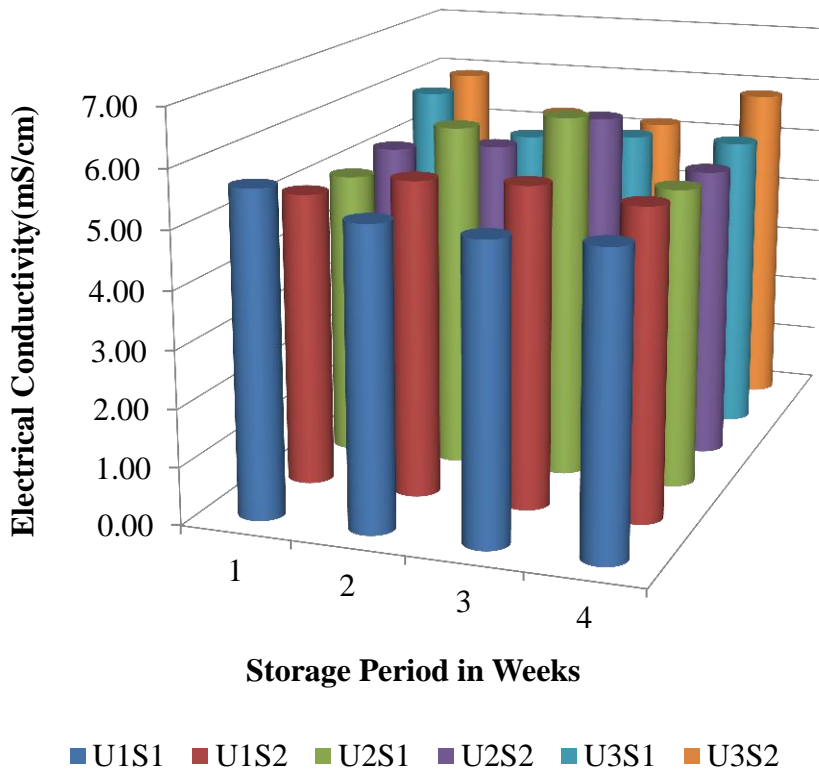


Fig. 4.11 Effect of different treatment time and different packaging materials on the Electrical Conductivity (EC) of ultrasonication treated tender coconut water

The interaction of different treatment time and different packaging materials on the EC of processed tender coconut water was found significantly at 1st, 2nd, 3rd, and 4th week of storage period at 5% level of significance.

So it is concluded that ultrasonication treatment should be given for 30 min ultrasonication and with PET bottle packaging to get more EC in tender coconut water for extending shelf life upto 4 week.

4.2.4.3 Combined effect of Ozonation and Ultrasonication treatment time on EC for tender coconut water

From the Table 4.13 presents the data on the effects of different time treatment and different packaging materials at stored 4±2°C on the Electrical Conductivity (EC) of processed tender coconut water and also shown graphically in Fig. 4.12.

Results and discussion

Combined treatment processed tender coconut water EC ranged from 5.12 to 6.49 mS/cm decreasing with increase in storage period when stored at 4±2°C in different packaging. EC was decreasing with combined treatment time and values were found more in glass bottle as compared to PET bottle. The Electrical Conductivity (EC) of combination treatment processed tender coconut water increases with the increase in storage period. Combination treatment processed tender coconut water the maximum Electrical Conductivity (EC) 4th week of storage period was obtained in treatment 30min ozonation and 10min ultrasonication stored in glass bottle (O₃U₁S₁), 30min ozonation and 10 min ultrasonication stored in PET bottle (O₃U₁S₂) and 20min ozonation and 30min ultrasonication stored in PET bottle (O₂U₃S₂) were 6.49, 6.32 and 6.19 mS/cm respectively.

Table 4.13 Effect of different treatment time and different packaging materials on the Electrical Conductivity (EC) of combined treated tender coconut water
Initial Electrical Conductivity (EC) = 5.84 mS/cm

Mean Electrical Conductivity (EC) , mS/cm				
Treatments	Storage period in weeks			
	1 week	2 week	3 week	4 week
O ₁ U ₁ S ₁	5.59	5.34	5.53	5.44
O ₁ U ₁ S ₂	5.68	5.63	5.46	5.12
O ₁ U ₂ S ₁	5.38	5.96	6.28	5.71
O ₁ U ₂ S ₂	5.19	5.84	6.05	5.59
O ₁ U ₃ S ₁	5.78	5.68	5.64	5.69
O ₁ U ₃ S ₂	5.34	5.73	6.07	5.73
O ₂ U ₁ S ₁	5.59	5.87	5.59	5.74
O ₂ U ₁ S ₂	5.86	5.95	5.19	5.88
O ₂ U ₂ S ₁	5.40	5.91	5.38	5.82
O ₂ U ₂ S ₂	5.12	5.65	5.19	5.93
O ₂ U ₃ S ₁	5.54	5.82	5.84	5.78
O ₂ U ₃ S ₂	5.15	5.76	5.86	6.19
O ₃ U ₁ S ₁	5.73	5.69	5.23	6.49
O ₃ U ₁ S ₂	5.37	5.64	5.59	6.32
O ₃ U ₂ S ₁	5.94	5.72	5.70	5.98
O ₃ U ₂ S ₂	5.64	5.72	5.24	5.45
O ₃ U ₃ S ₁	5.87	5.19	5.31	6.03
O ₃ U ₃ S ₂	5.34	5.38	5.41	5.63
S. Em	0.0024	0.0321	0.0022	0.009
CD at 5%	0.0069	0.0922	0.0064	0.0259
Test	Sig.	Sig.	Sig.	Sig.
CV %	0.0754	0.9772	0.0688	0.2687

Combined treatment processed tender coconut water the minimum EC 4th week of storage period was obtained in treatment 10min ozonation and 10min ultrasonication stored in PET bottle (O₁U₁S₂), 10min ozonation and 10min ultrasonication stored in glass bottle (O₁U₁S₁) and 30min ozonation and 20min ultrasonication stored in PET bottle (O₃U₂S₂) were 5.12, 5.44 and 5.45 mS/cm respectively (Table 4.13).

The ozonation and ultrasonication treatment processed coconut water at different storage period was influenced significantly by different processing times at 1st, 2nd, 3rd and 4th week of storage period. The processed tender coconut water stored in glass bottle O₃U₁S₁ i.e. 30min ozonation treatment and 10min ultrasonication treatment showed significantly higher Electrical Conductivity (EC) of processed tender coconut water at different storage period. From the results is also clear that as the storage period increases from 1st week to 4th week.

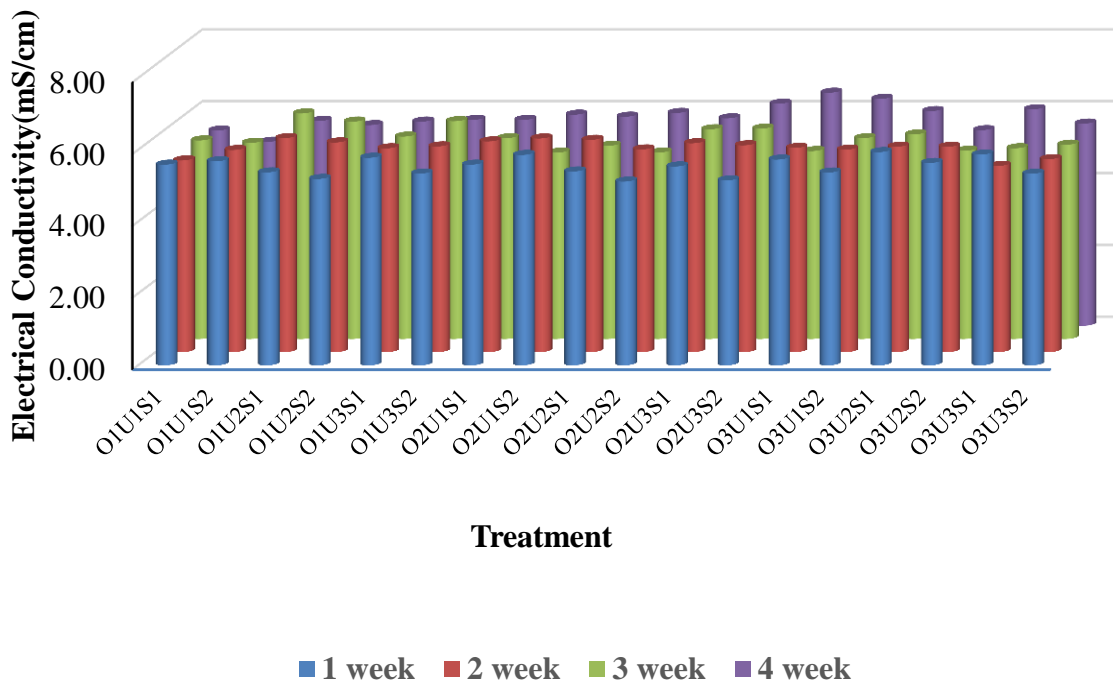


Fig 4.12 Effect of different treatment time and different packaging materials on the Electrical Conductivity (EC) of combined treated tender coconut water

The interaction of different treatment time and different packaging materials on the EC of processed tender coconut water was found significantly at 1st, 2nd, 3rd, and 4th week of storage period at 5% level of significance.

So it is concluded that combined treatment should be given for 30 min ozonation and 10min ultrasonication with glass bottle packaging to get more EC in tender coconut water for extending shelf life upto 4 week.

4.2.5 Effect of different treatment time and different packaging materials on total dissolved solids (TDS) of treated tender coconut water

Total Dissolved Solids (TDS) is a measure of the dissolved combined content of all inorganic and organic substances present in a liquid in molecular, ionized, or micro granular suspended form. An appraisal of data on the effect of different treatment time and different packaging materials on total dissolved solids (TDS) of processed tender coconut water is summarized in Table 4.14, 4.15 and 4.16 and graphically depicted in Fig. 4.13, 4.14 and 4.15.

4.2.5.1 Effect of Ozonation treatment on TDS content for tender coconut water

The results presented in Table 4.14 shows the effects of different time treatment and different packaging materials at stored $4\pm 2^\circ\text{C}$ on the Total Dissolved Solids (TDS) of processed tender coconut water and also shown graphically in Fig. 4.13. The Total Dissolved Solids (TDS) of ozonation treatment processed tender coconut water decreases with the increase in storage period.

Ozonation treatment processed tender coconut water TDS ranged from 4.02 to 4.70 ppm decreasing with increase in storage period when stored at $4\pm 2^\circ\text{C}$ in different packaging. TDS was decreasing with ozonation treatment time and values were found more in PET bottle as compared to glass bottle.

Total Dissolved Solids (TDS) of ozonation treatment processed tender coconut water decreases with the increase in storage period. The maximum Total Dissolved Solids (TDS) value of processed tender coconut water 4th week of storage period was obtained in treatment 30min stored in PET bottle (O_3S_2), 30min stored in glass bottle (O_3S_1) and 10min stored in PET bottle (O_1S_2) were 4.61, 4.25 and 4.18 ppm respectively. The

minimum Total Dissolved Solids (TDS) value of processed tender coconut water 4th week of storage period was obtained in treatment 20min stored in glass bottle (O₂S₁), 10min stored in glass bottle (O₁S₁) and 30min stored in PET bottle (O₃S₂) were 4.06, 4.09 and 4.14 ppm respectively (Table 4.14).

Table 4.14 Effect of different treatment time and different packaging materials on the Total Dissolved Solids (TDS) of ozonation treated tender coconut water

Initial Total Dissolved Solids (TDS) = 4.44ppm

Mean Total Dissolved Solids (TDS), ppm				
Treatments	Storage period in weeks			
	1 week	2 week	3 week	4 week
O ₁ S ₁	4.36	4.25	4.25	4.09
O ₁ S ₂	4.53	4.07	4.64	4.25
O ₂ S ₁	4.49	4.64	4.13	4.14
O ₂ S ₂	4.70	4.02	4.66	4.06
O ₃ S ₁	4.67	4.75	4.55	4.18
O ₃ S ₂	4.46	4.46	3.98	4.61
S. Em	0.0058	0.0028	0.0019	0.0058
CD at 5%	0.0179	0.0085	0.006	0.0179
Test	Sig.	Sig.	Sig.	Sig.
CV %	0.2213	0.1092	0.0769	0.2383

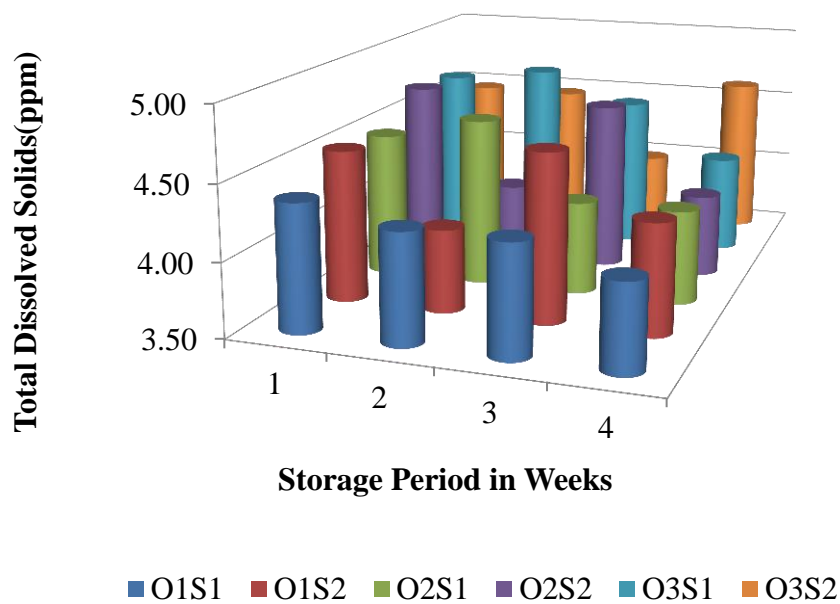


Fig. 4.13 Effect of different treatment time and different packaging materials on the Total Dissolved Solids (TDS) of ozonation treated tender coconut water

The statistical analysis of data revealed that the rate of decrease in Total Dissolved Solids (TDS) due to different due to different treatment time and different packaging materials were found to be significant with the storage period 1st, 2nd, 3rd, and 4th week of storage period respectively, where observed significantly higher due to different treatment time O₃S₂ i.e. 30min ozonation treatment and different packaging materials S₂ i.e. PET bottle though 1st, 2nd, and 3rd week storage period was remained statistically at par with different treatment time respectively

The interaction of different treatment time and different packaging materials on the TDS of processed tender coconut water was found significantly at 1st, 2nd, 3rd and 4th week of storage period at 5% level of significance.

So it is concluded that ozonation treatment should be given for 30 min with PET bottle packaging to get more TDS in tender coconut water for prolonging shelf life upto 4 week.

4.2.5.2 Effect of Ultrasonication treatment time on TDS for tender coconut water

The results presented in Table 4.15 shows the effects of different time treatment and different packaging materials at stored 4±2°C on the Total Dissolved Solids (TDS) of processed tender coconut water and also shown graphically in Fig. 4.14. The Total Dissolved Solids (TDS) of ultrasonication treatment processed tender coconut water decreases with the increase in storage period.

Ultrasonication treatment processed tender coconut water TDS ranged from 3.97 to 5.01 ppm decreasing with increase in storage period when stored at 4±2°C in different packaging. TDS was decreasing with ozonation treatment time and values were found more in PET bottle as compared to glass bottle.

Total Dissolved Solids (TDS) of ultrasonication treatment processed tender coconut water decrease with the increase in storage period. Ultrasonication treatment processed tender coconut water the maximum Total Dissolved Solids (TDS) 4th week of storage period was obtained in treatment 20min stored in PET bottle (U₂S₂), 30min stored in PET bottle (U₃S₂) and 10min stored in PET bottle (U₁S₂) were 4.62, 4.61 and 4.46 ppm respectively. Ultrasonication treatment processed tender coconut water the minimum Total Dissolved Solids (TDS) 4th week of storage period was obtained in treatment 20min

stored in glass bottle (U₂S₁), 10min stored in glass bottle (U₁S₁) and 30min stored in PET bottle (U₃S₂) were 4.03, 4.20 and 4.27 ppm respectively (Table 4.15).

Table 4.15 Effect of different treatment time and packaging materials on the total dissolved solids (TDS) of ultrasonication treated tender coconut water

Initial Total Dissolved Solids (TDS) = 4.44ppm

Mean Total Dissolved Solids (TDS), ppm				
Treatments	Storage period in weeks			
	1 week	2 week	3 week	4 week
U ₁ S ₁	4.45	4.12	4.05	4.20
U ₁ S ₂	4.05	4.35	4.40	4.46
U ₂ S ₁	3.97	4.76	5.01	4.03
U ₂ S ₂	4.07	4.21	4.71	4.62
U ₃ S ₁	4.63	4.07	4.17	4.61
U ₃ S ₂	4.66	4.15	4.08	4.27
S. Em	0.0021	0.0015	0.0023	0.01
CD at 5%	0.0065	0.0047	0.0072	0.0307
Test	Sig.	Sig.	Sig.	Sig.
CV %	0.0847	0.0624	0.0923	0.3953

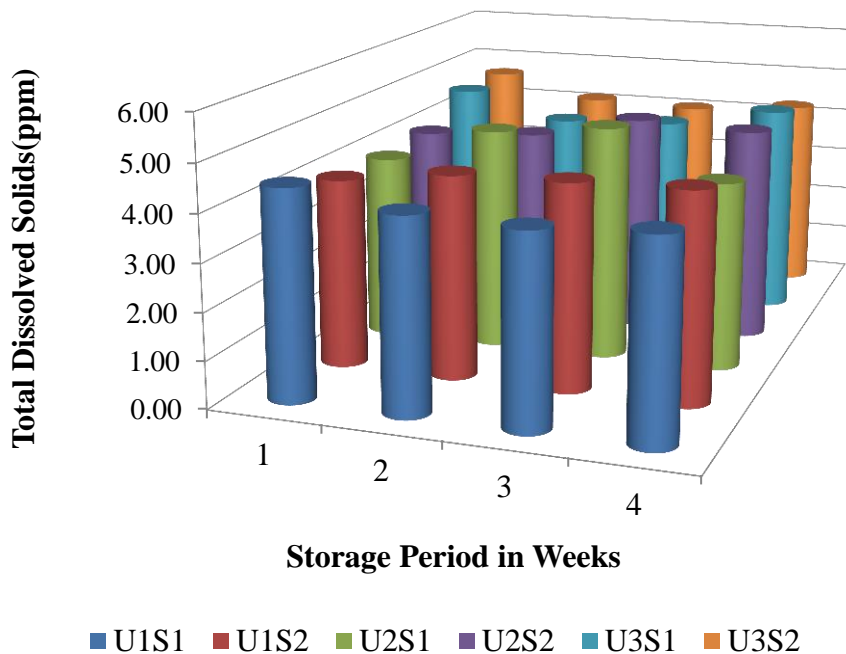


Fig. 4.14 Effect of different treatment time and different packaging materials on the total dissolved solids (TDS) of ultrasonication treated tender coconut water

The statistical analysis of data revealed that the rate of decrease in Total Dissolved Solids (TDS) due to different due to different treatment time and different packaging materials were found to be significant with the storage period 1st, 2nd, 3rd, and 4th week of storage period respectively, where observed significantly higher due to different treatment time U₂S₂ i.e. 20min ultrasonication treatment and different packaging materials S₂ i.e. PET bottle though 1st, 2nd, 3rd and 4th week storage period was remained statistically at par with different treatment time respectively.

The interaction of different treatment time and different packaging materials on the TDS of processed tender coconut water was found significantly at 1st, 2nd, 3rd, and 4th week of storage period at 5% level of significance.

As a result, it is inferred that ultrasonication treatment should be given for 20 min with PET bottle packaging to get more TDS in tender coconut water for prolonging shelf life upto 4 week.

4.2.5.3 Combined effect of Ozonation and Ultrasonication treatment time on TDS for tender coconut water

From the Table 4.16 presents the data on the effects of different time treatment and different packaging materials at stored 4±2°C on the Total Dissolved Solids (TDS) of processed tender coconut water and also shown graphically in Fig. 4.15.

The Total Dissolved Solids (TDS) of ozonation and ultrasonication treatment processed tender coconut water increases with the increase in storage period. Combined treatment processed tender coconut water TDS ranged from 4.04 to 5.13 ppm increasing with increase in storage period when stored at 4±2°C in different packaging. TDS was decreasing with combined treatment time and values were found more in glass bottle as compared to PET bottle.

Combination treatment processed tender coconut water the maximum Total Dissolved Solids (TDS) 4th week of storage period was obtained in treatment 30min ozonation and 10min ultrasonication stored in glass bottle (O₃U₁S₁), 30min ozonation and 10 min ultrasonication stored in PET bottle (O₃U₁S₂) and 20min ozonation and 30min ultrasonication stored in PET bottle (O₂U₃S₂) were 5.13, 4.99 and 4.89 ppm respectively.

Combined treatment processed tender coconut water the minimum Total Dissolved Solids (TDS) 4th week of storage period was obtained in treatment 10min ozonation and 10min ultrasonication stored in PET bottle (O₁U₁S₂), 10min ozonation and 10min ultrasonication stored in glass bottle (O₁U₁S₁) and 30min ozonation and 20min ultrasonication stored in PET bottle (O₃U₂S₂) were 4.05 and 4.30 ppm respectively (Table 4.16).

Table 4.16 Effect of different treatment time and different packaging materials on the total dissolved solids (TDS) of combined treated tender coconut water

Initial Total Dissolved Solids (TDS) = 4.44ppm

Mean Total Dissolved Solids (TDS), ppm				
Treatments	Storage period in weeks			
	1 week	2 week	3 week	4 week
O ₁ U ₁ S ₁	4.42	4.22	4.37	4.30
O ₁ U ₁ S ₂	4.49	4.45	4.31	4.05
O ₁ U ₂ S ₁	4.25	4.71	4.96	4.51
O ₁ U ₂ S ₂	4.10	4.61	4.78	4.42
O ₁ U ₃ S ₁	4.57	4.48	4.45	4.49
O ₁ U ₃ S ₂	4.22	4.53	4.79	4.53
O ₂ U ₁ S ₁	4.42	4.64	4.42	4.53
O ₂ U ₁ S ₂	4.63	4.70	4.10	4.65
O ₂ U ₂ S ₁	4.27	4.67	4.25	4.60
O ₂ U ₂ S ₂	4.04	4.46	4.10	4.68
O ₂ U ₃ S ₁	4.38	4.60	4.61	4.57
O ₂ U ₃ S ₂	4.07	4.55	4.63	4.89
O ₃ U ₁ S ₁	4.53	4.50	4.13	5.13
O ₃ U ₁ S ₂	4.24	4.46	4.42	4.99
O ₃ U ₂ S ₁	4.69	4.52	4.50	4.72
O ₃ U ₂ S ₂	4.45	4.52	4.14	4.30
O ₃ U ₃ S ₁	4.64	4.10	4.20	4.76
O ₃ U ₃ S ₂	4.22	4.25	4.27	4.45
S. Em	0.0019	0.0254	0.0018	0.0071
CD at 5%	0.0054	0.073	0.0052	0.0203
Test	Sig.	Sig.	Sig.	Sig.
CV %	0.075	0.9788	0.0712	0.2674

The ozonation and ultrasonication treatment processed coconut water at different storage period was influenced significantly by different processing times at 1st, 2nd, 3rd and 4th week of storage period. The processed tender coconut water stored in glass bottle O₃U₁S₁ i.e. 30min ozonation treatment and 10min ultrasonication treatment showed significantly higher Total Dissolved Solids (TDS) of processed tender coconut water at

different storage period. From the results is also clear that as the storage period increases from 1st week to 4th week.

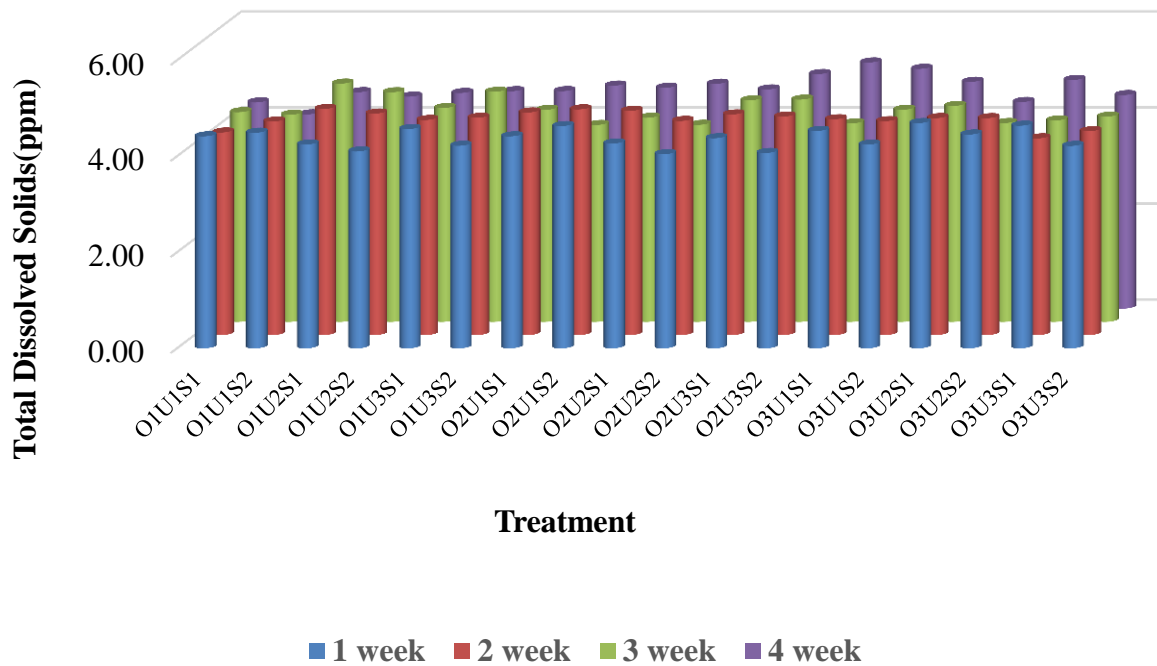


Fig. 4.15 Effect of different treatment time and different packaging materials on the total dissolved solids (TDS) of combined processed tender coconut water

The interaction of different treatment time and different packaging materials on the TDS of processed tender coconut water was found significantly at 1st, 2nd, 3rd, and 4th week of storage period at 5% level of significance.

Given a result, it is determined that combined treatment should be given for 30 min ozonation and 10min ultrasonication with glass bottle packaging to get more TDS in tender coconut water storage period of 4 week.

4.2.6 Effect of different treatment time and different packaging materials on total sugar (TS) of treated tender coconut water

Total Sugar include sugars naturally present in many nutritious foods and beverages. Such as sugar in milk and fruits as well as any added sugars that may be present in the product. An appraisal of data on the effect of different treatment time and

different packaging materials on total sugar (TS) of processed tender coconut water is summarized in Table 4.17, 4.18 and 4.19 and graphically depicted in Fig. 4.16, 4.17 and 4.18.

4.2.6.1 Effect of Ozonation treatment on TS content for tender coconut water

An appraisal of data on Total Sugar of stored tender coconut water are given Table 4.17 and graphically depicted in Fig. 4.16. From the table it is clear that total sugar (TS) in processed tender coconut water decreases with the increase in storage period. The decrease in total sugar in processed tender coconut water with the increase in storage period results in the deteriorating of the quality of coconut water. However, a decrease in the total sugar (TS) of strawberries stored in an ozonated atmosphere was reported earlier (Perez, Sanz, Rios, Olias & Olias,1999).

Ozonation treatment processed tender coconut water TS ranged from 3.33 to 4.97 % decreasing with increase in storage period when stored at $4\pm 2^{\circ}\text{C}$ in different packaging. TS was decreasing with ozonation treatment time and values were found more in glass bottle as compared to PET bottle.

Total Sugar (TS) of ozonation treatment processed tender coconut water decreases with the increase in storage period. The maximum Total Sugar (TS) value of processed tender coconut water 4th week of storage period was obtained in treatment 10min stored in glass bottle (O_1S_1), 30min stored in PET bottle (O_3S_2) and 10min stored in PET bottle (O_1S_2) were 4.57, 4.29 and 4.35 % respectively. The minimum Total Sugar (TS) value of processed tender coconut water 4th week of storage period was obtained in treatment 20min stored in PET bottle (O_2S_2), 30min stored in glass bottle (O_3S_1) and 20min stored in glass bottle (O_2S_1) were 3.62, 4.08 and 4.13 % respectively (Table 4.17).

The statistical analysis of data revealed that the rate of decrease in total sugar (TS) due to different due to different treatment time and different packaging materials were found to be significant with the storage period 1st, 2nd, 3rd, and 4th week of storage period respectively, where observed significantly higher due to different treatment time O_1S_1 i.e. 10min ozonation treatment and different packaging materials S_1 i.e. glass bottle though 1st, 2nd, and 3rd week storage period was remained statistically at par with different treatment time respectively.

The interaction of different treatment time and different packaging materials on the TS of processed tender coconut water was found significantly at 1st, 2nd, 3rd, and 4th week of storage period at 5% level of significance. So it is concluded that ozonation treatment should be given for 10 min with glass bottle packaging to get more TS in tender coconut water to extend shelf life up to 4 week.

Table 4.17 Effect of different treatment time and different packaging materials on the total sugar (TS) of ozonation treated tender coconut water

Initial Total Sugar (TS) =4.71%

Treatments	Mean Total Sugar (TS), %			
	Storage period in weeks			
	1 week	2 week	3 week	4 week
O ₁ S ₁	4.66	4.45	4.56	4.57
O ₁ S ₂	4.60	4.62	4.53	4.25
O ₂ S ₁	4.45	4.38	4.25	4.13
O ₂ S ₂	4.97	4.23	4.28	3.62
O ₃ S ₁	3.33	4.29	3.48	4.08
O ₃ S ₂	4.44	4.06	5.16	4.29
S. Em	0.0116	0.0131	0.011	0.0133
CD at 5%	0.0358	0.0402	0.0338	0.0409
Test	Sig.	Sig.	Sig.	Sig.
CV %	0.4568	0.5226	0.434	0.5528

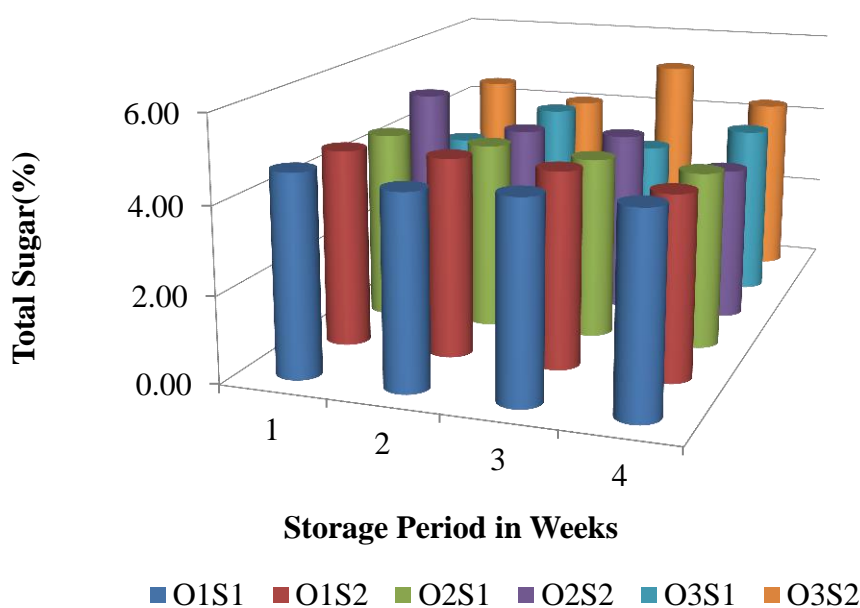


Fig. 4.16 Effect of different treatment time and different packaging materials on the total sugar (TS) of ozonation treated tender coconut water

4.2.6.2 Effect of Ultrasonication treatment time on TS for tender coconut water

An appraisal of data on Total Sugar (TS) of stored tender coconut water are given Table 4.18 and graphically depicted in Fig. 4.17. From the table it is clear that total sugar in processed tender coconut water decreases with the increase in storage period in both case i.e. different treatment time as well as materials. The decrease in total sugar (TS) in processed tender coconut water with the increase in storage period results in the deteriorating of the quality of coconut water. Similar reductions were reported for ultrasound treated carrot juice also (Zou and Jiang, 2016). A slight increase in total sugar (TS) content of TCW in the present study may be due to the higher release of sugars from tissues by ultrasound treatment (Gomez-Lopez *et al.*, 2018).

Ultrasonication treatment processed tender coconut water TS ranged from 3.36 to 5.49 % decreasing with increase in storage period when stored at 4±2°C in different packaging. TS was decreasing with ultrasonication treatment time and values were found more in PET bottle as compared to glass bottle.

Table 4.18 Effect of different treatment time and different packaging materials on the total sugar (TS) of ultrasonication treated tender coconut water

Initial Total Sugar (TS) = 4.71%

Mean Total Sugar (TS), %				
Treatments	Storage period in weeks			
	1 week	2 week	3 week	4 week
U ₁ S ₁	4.74	4.90	3.46	4.95
U ₁ S ₂	4.84	4.36	4.55	4.44
U ₂ S ₁	3.74	4.40	4.67	4.37
U ₂ S ₂	4.57	4.34	3.82	4.63
U ₃ S ₁	4.23	4.13	3.36	3.72
U ₃ S ₂	5.49	5.13	4.22	5.16
S. Em	0.0156	0.0158	0.0087	0.0126
CD at 5%	0.048	0.0487	0.0269	0.0389
Test	Sig.	Sig.	Sig.	Sig.
CV %	0.5862	0.6027	0.3762	0.4809

Total Sugar (TS) of ultrasonication treatment processed tender coconut water decrease with the increase in storage period. Ultrasonication treatment processed tender coconut water the maximum Total Sugar (TS) 4th week of storage period was obtained in treatment 30min stored in PET bottle (U₃S₂), 10min stored in glass bottle (U₁S₁) and 20min stored in PET bottle (U₂S₂) were 5.16, 4.95 and 4.63 % respectively.

Ultrasonication treatment processed tender coconut water the minimum Total Sugar (TS) 4th week of storage period was obtained in treatment 30min stored in glass bottle (U₃S₁), 20min stored in glass bottle (U₂S₁) and 10min stored in PET bottle (U₁S₂) were 3.72, 4.37 and 4.44 % respectively (Table 4.18).

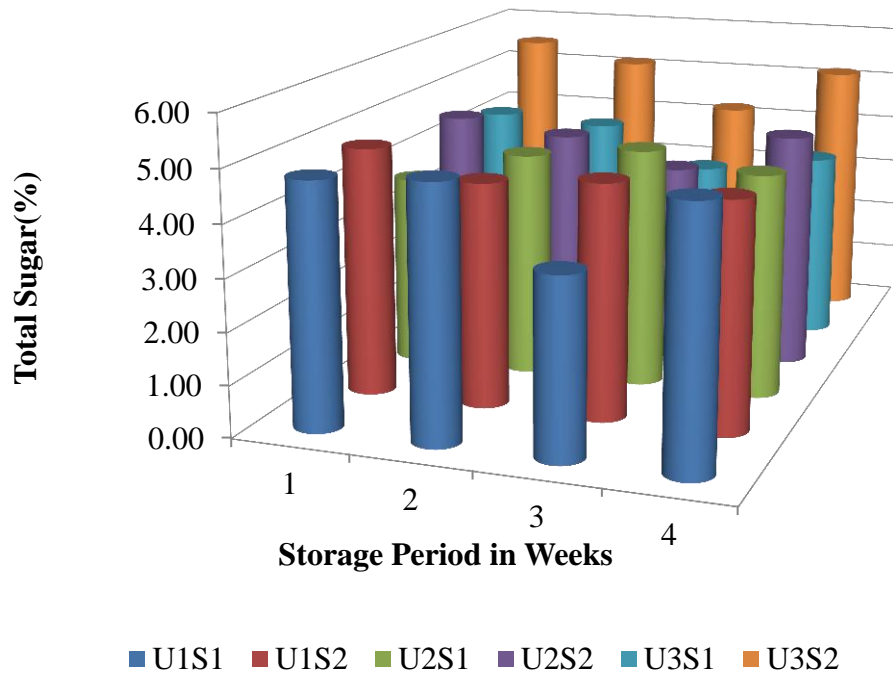


Fig. 4.17 Effect of different treatment time and different packaging materials on the total sugar (TS) of ultrasonication treated tender coconut water

The statistical analysis of data revealed that the rate of decrease in total sugar (TS) due to different due to different treatment time and different packaging materials were found to be significant with the storage period 1st, 2nd, 3rd, and 4th week of storage period respectively, where observed significantly higher due to different treatment time U₃S₂ i.e. 30min ultrasonication treatment and different packaging materials S₂ i.e. PET bottle though 1st, 2nd, 3rd and 4th week storage period was remained statistically at par with different treatment time respectively.

The interaction of different treatment time and different packaging materials on the TS of processed tender coconut water was found significantly at 1st, 2nd, 3rd, and 4th week of storage period at 5% level of significance.

As a result, it is inferred that Ultrasonication treatment should be given for 30 min with PET bottle packaging to get more TS in tender coconut water to extend shelf life up to 4 week.

4.2.6.3 Combined effect of Ozonation and Ultrasonication treatment time on TS for tender coconut water

From the Table 4.19 presents the data on the effects of different time treatment and different packaging materials at stored $4\pm 2^{\circ}\text{C}$ on the Total sugar (TS) of processed tender coconut water and also shown graphically in Fig. 4.18. The total sugar (TS) of ozonation and ultrasonication treatment processed tender coconut water decreases with the increase in storage period.

Table 4.19 Effect of different treatment time and different packaging materials on the total sugar (TS) of combined treated tender coconut water

Initial Total Sugar (TS) = 4.71%

Mean Total Sugar (TS), %				
Treatments	Storage period in weeks			
	1 week	2 week	3 week	4 week
O ₁ U ₁ S ₁	3.91	3.59	4.49	3.55
O ₁ U ₁ S ₂	4.22	5.53	4.64	3.75
O ₁ U ₂ S ₁	3.79	4.14	4.07	3.66
O ₁ U ₂ S ₂	4.26	4.40	4.25	4.54
O ₁ U ₃ S ₁	3.58	5.04	4.38	4.22
O ₁ U ₃ S ₂	3.68	5.06	4.23	3.70
O ₂ U ₁ S ₁	4.33	4.46	4.82	3.79
O ₂ U ₁ S ₂	3.43	4.51	5.39	3.94
O ₂ U ₂ S ₁	3.60	4.24	4.06	3.55
O ₂ U ₂ S ₂	3.98	3.72	3.78	3.60
O ₂ U ₃ S ₁	4.08	4.37	4.30	4.36
O ₂ U ₃ S ₂	4.15	4.36	4.22	5.16
O ₃ U ₁ S ₁	3.50	4.16	3.46	5.27
O ₃ U ₁ S ₂	4.56	4.04	3.25	3.53
O ₃ U ₂ S ₁	3.50	4.01	4.17	5.17
O ₃ U ₂ S ₂	4.56	3.75	4.07	3.48
O ₃ U ₃ S ₁	4.39	4.06	3.67	3.54
O ₃ U ₃ S ₂	4.47	3.59	5.52	4.06
S. Em	0.0148	0.027	0.0093	0.0155
CD at 5%	0.0424	0.0776	0.0267	0.0444
Test	Sig.	Sig.	Sig.	Sig.
CV %	0.6393	1.0938	0.3774	0.662

Combined treatment processed tender coconut water TS ranged from 3.25 to 5.27 % increasing with increase in storage period when stored at $4\pm 2^{\circ}\text{C}$ in different packaging. TDS was decreasing with combined treatment time and values were found more in glass bottle as compared to PET bottle.

Combination treatment processed tender coconut water the maximum Total Sugar (TS) 4th week of storage period was obtained in treatment 30min ozonation and 10min ultrasonication stored in glass bottle ($\text{O}_3\text{U}_1\text{S}_1$), 30min ozonation and 20 min ultrasonication stored in glass bottle ($\text{O}_3\text{U}_2\text{S}_1$) and 20min ozonation and 30min ultrasonication stored in PET bottle ($\text{O}_2\text{U}_3\text{S}_2$) were 5.27, 5.17 and 5.16 % respectively. Combined treatment processed tender coconut water the minimum Total Sugar (TS) 4th week of storage period was obtained in treatment 30min ozonation and 30min ultrasonication stored in glass bottle ($\text{O}_3\text{U}_3\text{S}_1$), 10min ozonation and 10min ultrasonication stored in glass bottle ($\text{O}_1\text{U}_1\text{S}_1$) and 20min ozonation and 20min ultrasonication stored in glass bottle ($\text{O}_2\text{U}_2\text{S}_1$) were 3.54 and 3.55 % respectively (Table 4.19).

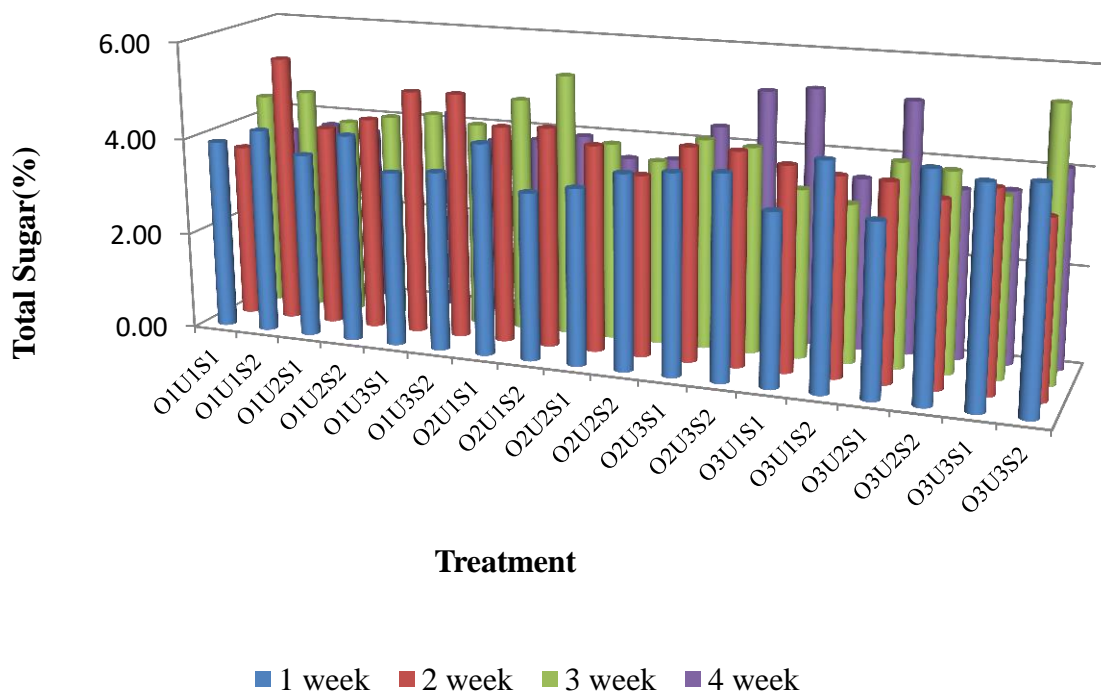


Fig. 4.18 Effect of different treatment time and different packaging materials on the total sugar (TS) of combined treated tender coconut water

The ozonation and ultrasonication treatment processed coconut water at different storage period was influenced significantly by different processing times at 1st, 2nd, 3rd

and 4th week of storage period. The processed tender coconut water stored in glass bottle O₃U₁S₁ i.e. 30min ozonation treatment and 10min ultrasonication treatment showed significantly higher total sugar (TS) of processed tender coconut water at different storage period. From the results is also clear that as the storage period increases from 1st week to 4th week.

The interaction of different treatment time and different packaging materials on the TDS of processed tender coconut water was found significantly at 1st, 2nd, 3rd, and 4th week of storage period at 5% level of significance.

So it is concluded that combined treatment should be given for 30 min ozonation and 10min ultrasonication with glass bottle packaging to get more TDS in tender coconut water to extend shelf life up to 4 week.

4.2.7 Effect of different treatment time and different packaging materials on total phenol content (TPC) of treated tender coconut water

Total phenol content activity is the process to figure out the amount of phenolic content in the samples. Phenolic compounds that contained in the plants have redox properties, and the properties allow them acting as antioxidants. An appraisal of data on the effect of different treatment time and different packaging materials on total phenol content (TPC) of processed tender coconut water is summarized in Table 4.20, 4.21 and 4.22 and graphically depicted in Fig. 4.19, 4.20 and 4.21.

4.2.7.1 Effect of Ozonation treatment time and different packaging materials on TPC for tender coconut water

The results presented in Table 4.20 shows the effects of different time treatment and different packaging materials at stored 4±2°C on the total phenol content of processed tender coconut water and also shown graphically in Fig. 4.19. The total phenol content of ozonation treatment processed tender coconut water decreases with the increase in storage period.

Ozonation treatment processed tender coconut water TPC ranged from 1.97 to 3.47 (mg GAE/ml) decreasing with increase in storage period when stored at 4±2°C in

different packaging. TPC was decreasing with ozonation treatment time and values were found more in glass bottle as compared to PET bottle.

Total Phenol Content (TPC) of ozonation treatment processed tender coconut water decreases with the increase in storage period. The maximum Total Phenol Content (TPC) value of processed tender coconut water 4th week of storage period was obtained in treatment 10min stored in glass bottle (O₁S₁), 20min stored in PET bottle (O₂S₂) and 20min stored in glass bottle (O₂S₁) were 2.90, 2.67 and 2.37 (mg GAE/ml) respectively. The minimum Total Phenol Content (TPC) value of processed tender coconut water 4th week of storage period was obtained in treatment 10min stored in PET bottle (O₁S₂), 30min stored in glass bottle (O₃S₁) and 30min stored in PET bottle (O₃S₂) were 1.97, 2.00 and 2.23 (mg GAE/ml) respectively (Table 4.20). Ozone treatment causes oxidation of the phenolics (Allende *et al.*, 2007), resulting in a reduction in their contents, and a decrease in polyphenol content, flavonoid content, and antioxidant capacity was observed in ozone treated guava fruit after 10min of treatment (Alothman *et al.*2010).

Table 4.20 Effect of different treatment time and different packaging materials on the total phenol content (TPC) of ozonation treated tender coconut water

Initial Total Phenol Content (TPC) = 3.75(mg GAE/ml)

Mean Total Phenol Content (TPC), mgGAE/ml				
Treatments	Storage period in weeks			
	1 week	2 week	3 week	4 week
O ₁ S ₁	3.23	3.20	3.33	2.90
O ₁ S ₂	3.30	2.77	2.13	1.97
O ₂ S ₁	3.47	2.90	2.97	2.37
O ₂ S ₂	3.37	2.73	2.93	2.67
O ₃ S ₁	3.47	2.97	2.97	2.00
O ₃ S ₂	3.40	2.60	3.03	2.23
S. Em	0.1834	0.1	0.1269	0.0793
CD at 5%	NS	NS	0.3911	0.2445
Test	NS	NS	Sig.	Sig.
CV %	9.4034	6.0538	7.5955	5.8346

The statistical analysis of data revealed that the rate of decrease in total phenol content (TPC) due to different due to different treatment time and different packaging materials were found to be significant with the storage period 3rd, and 4th week of storage period respectively, where observed significantly higher due to different treatment time O₁S₁ i.e. 10min ozonation treatment and different packaging materials S₁

i.e. glass bottle though 1st, 2nd, and 3rd week storage period was remained statistically at par with different treatment time respectively.

The interaction of different treatment time and different packaging materials on the TPC of processed tender coconut water was found significantly at 3rd and 4th week of storage period at 5% level of significance. Given a result, it is determined that ozonation treatment should be given for 10 min with glass bottle packaging to get more TPC in tender coconut water for prolonging shelf life upto 4 week.

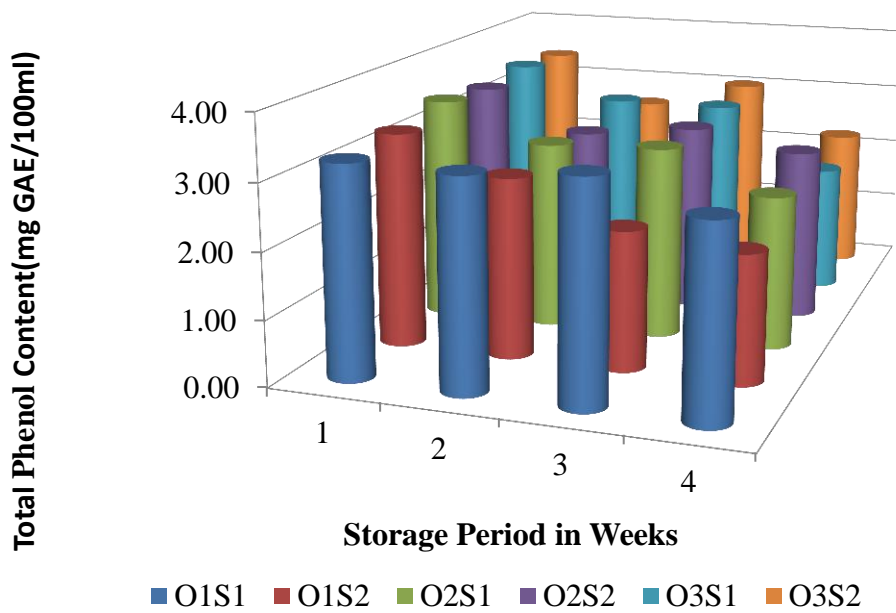


Fig. 4.19 Effect of different treatment time and different packaging materials on the total phenol content (TPC) of ozonation treated tender coconut water

4.2.7.2 Effect of Ultrasonication treatment time and packaging material on TPC for tender coconut water

From the Table 4.21 presents the data on the effects of different time treatment and different packaging materials at stored 4±2°C on the total phenol content of processed tender coconut water and also shown graphically in Fig. 4.20. The total phenol content of ultrasonication treatment processed tender coconut water decreases with the increase in storage period.

Ultrasonication treatment processed tender coconut water TPC ranged from 1.77 to 3.60 (mg GAE/ml) decreasing with increase in storage period when stored at $4\pm 2^\circ\text{C}$ in different packaging. TPC was decreasing with ultrasonication treatment time and values were found more in PET bottle as compared to glass bottle.

Total Phenol Content (TPC) of ultrasonication treatment processed tender coconut water decrease with the increase in storage period. Ultrasonication treatment processed tender coconut water the maximum Total Phenol Content (TPC) 4th week of storage period was obtained in treatment 30min stored in PET bottle (U_3S_2), 10min stored in glass bottle (U_1S_1) and 20min stored in glass bottle (U_2S_1) were 3.17, 2.83 and 2.43 (mg GAE/ml) respectively. Ultrasonication treatment processed tender coconut water the minimum Total Phenol Content (TPC) 4th week of storage period was obtained in treatment 20min stored in PET bottle (U_2S_2), 10min stored in PET bottle (U_1S_2) and 30min stored in glass bottle (U_3S_1) were 1.77, 1.83 and 1.90 (mg GAE/ml) respectively (Table 4.21). Adil *et al.* (2015) also observed a significant decrease in total antioxidant capacity total phenols (approx. 2%) during 15 days storage of ultrasound treated grape juice.

The statistical analysis of data revealed that the rate of decrease in total phenol content (TPC) due to different due to different treatment time and different packaging materials were found to be significant with the storage period 2nd, 3rd, and 4th week of storage period respectively, where observed significantly higher due to different treatment time U_3S_2 i.e. 30min ultrasonication treatment and different packaging materials S_2 i.e. PET bottle though 2nd, 3rd and 4th week storage period was remained statistically at par with different treatment time respectively.

The interaction of different treatment time and different packaging materials on the TPC of processed tender coconut water was found significantly at 2nd, 3rd, and 4th week of storage period at 5% level of significance. As a result, it is inferred that ultrasonication treatment should be given for 30 min with PET bottle packaging to get more TPC in tender coconut water for prolonging shelf life upto 4 week.

Table 4.21 Effect of different treatment time and different packaging materials on the total phenol content (TPC) of ultrasonication treated tender coconut water

Initial Total Phenol Content (TPC) = 3.75(mg GAE/ml)

Mean Total Phenol Content (TPC), mg GAE/ml				
Treatments	Storage period in weeks			
	1 week	2 week	3 week	4 week
U ₁ S ₁	3.13	3.47	3.33	2.83
U ₁ S ₂	3.47	3.57	2.03	1.83
U ₂ S ₁	3.30	2.83	2.37	2.43
U ₂ S ₂	3.10	3.60	2.23	1.77
U ₃ S ₁	3.50	2.50	2.90	1.90
U ₃ S ₂	3.50	2.37	3.23	3.17
S. Em	0.1503	0.0864	0.1052	0.0843
CD at 5%	NS	0.2649	0.3221	0.2585
Test	NS	Sig.	Sig.	Sig.
CV %	7.8102	4.8787	6.7471	6.2568

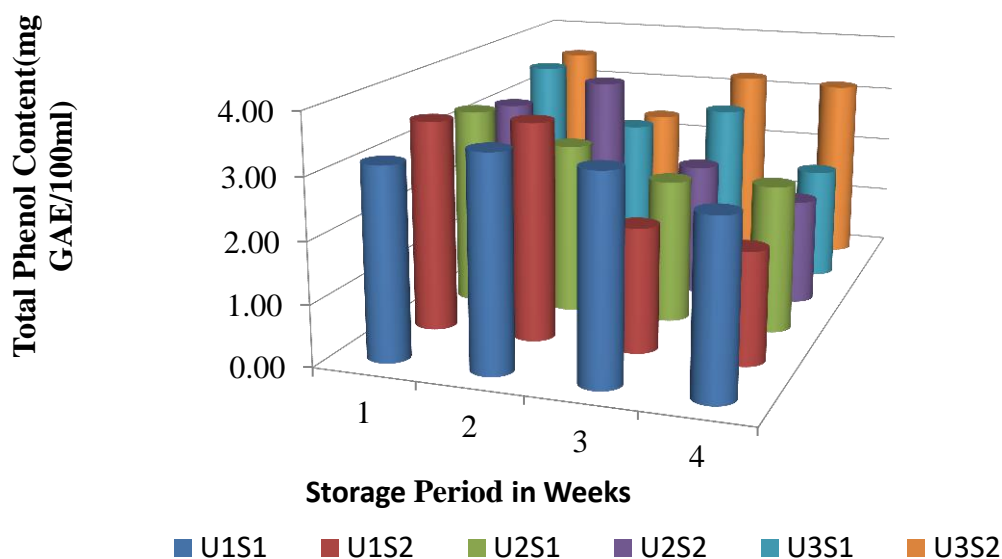


Fig. 4.20 Effect of different treatment time and different packaging materials on the total phenol content (TPC) of ultrasonication treated tender coconut water

4.2.7.3 Combined effect of Ozonation and Ultrasonication treatment time and packaging material on TPC for tender coconut water

The results presented in Table 4.22 shows the effects of different time treatment and different packaging materials at stored 4±2°C on the total phenol content of processed

tender coconut water and also shown graphically in Fig. 4.21. The total phenol content of ozonation and ultrasonication treatment processed tender coconut water decreases with the increase in storage period. Combined treatment processed tender coconut water TPC ranged from 1.73 to 1.80 (mg GAE/ml) decreasing with increase in storage period when stored at 4±2°C in different packaging. TPC was decreasing with combined treatment time and values were found more in PET bottle as compared to glass bottle.

Table 4.22 Effect of different treatment time and different packaging materials on the total phenol content (TPC) of combined treated tender coconut water

Initial Total Phenol Content (TPC) = 3.75(mg GAE/ml)

Mean Total Phenol Content (TPC), mg GAE/ml				
Treatments	Storage period in weeks			
	1 week	2 week	3 week	4 week
O ₁ U ₁ S ₁	3.17	2.93	2.47	2.00
O ₁ U ₁ S ₂	3.47	2.90	2.30	3.00
O ₁ U ₂ S ₁	3.10	2.93	3.13	1.87
O ₁ U ₂ S ₂	3.67	2.90	2.50	2.00
O ₁ U ₃ S ₁	3.50	2.50	1.93	2.97
O ₁ U ₃ S ₂	3.37	2.80	2.50	1.73
O ₂ U ₁ S ₁	3.57	2.87	2.40	2.67
O ₂ U ₁ S ₂	3.27	2.57	2.30	1.87
O ₂ U ₂ S ₁	3.33	3.50	2.30	1.80
O ₂ U ₂ S ₂	2.97	3.33	2.10	2.07
O ₂ U ₃ S ₁	3.47	2.47	2.10	2.17
O ₂ U ₃ S ₂	3.27	2.93	2.30	1.90
O ₃ U ₁ S ₁	3.17	2.87	2.13	2.53
O ₃ U ₁ S ₂	3.73	2.57	3.20	1.83
O ₃ U ₂ S ₁	3.30	3.00	2.90	2.27
O ₃ U ₂ S ₂	3.30	3.13	2.13	1.83
O ₃ U ₃ S ₁	3.37	2.37	2.10	2.20
O ₃ U ₃ S ₂	3.67	2.80	2.37	2.50
S. Em	0.073	0.1252	0.0754	0.0594
CD at 5%	0.2021	NS	0.2143	0.1692
Test	Sig.	NS	Sig.	Sig.
CV %	3.6114	7.6	5.3833	4.6761

Combination treatment processed tender coconut water the maximum Total Phenol Content (TPC) 4th week of storage period was obtained in treatment 10min ozonation and 10min ultrasonication stored in PET bottle (O₁U₁S₂), 10min ozonation and 30min ultrasonication stored in glass bottle (O₁U₃S₁) and 20min ozonation and 10min

ultrasonication stored in glass bottle (O₂U₁S₁) were 3.00, 2.97 and 2.67 (mg GAE/ml) respectively. Combined treatment processed tender coconut water the minimum Total Phenol Content (TPC) 4th week of storage period was obtained in treatment 10min ozonation and 30min ultrasonication stored in PET bottle (O₁U₃S₂), and 20min ozonation and 20min ultrasonication stored in glass bottle (O₂U₂S₁) were 1.73 and 1.80 (mg GAE/ml) respectively (Table 4.22).

The ozonation and ultrasonication treatment processed coconut water at different storage period was influenced significantly by different processing times at 1st, 3rd and 4th week of storage period. The processed tender coconut water stored in glass bottle O₁U₁S₂ i.e. 10min ozonation treatment and 10min ultrasonication treatment Stored in PET bottle showed significantly higher total phenol content (TPC) of processed tender coconut water at different storage period. From the results is also clear that as the storage period decreases from 1st week to 4th week.

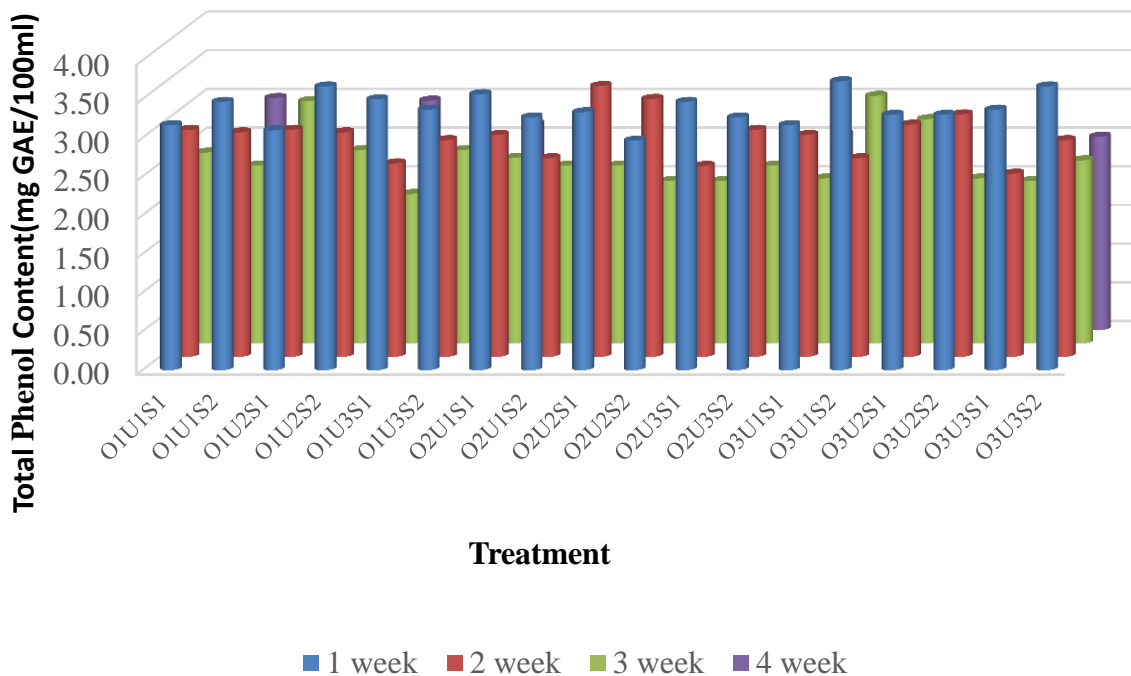


Fig. 4.21 Effect of different treatment time and different packaging materials on the total phenol content (TPC) of combined treated tender coconut water

The interaction of different treatment time and different packaging materials on the TPC of processed tender coconut water was found significantly at 1st, 3rd, and 4th week of storage period at 5% level of significance.

So it is concluded that combined treatment should be given for 10 min ozonation and 10min ultrasonication with PET bottle packaging to get more TPC in tender coconut water for prolonging shelf life upto 4 week.

4.2.8 Effect of different treatment time and different packaging materials on Peroxidase activity (POD) of treated tender coconut water

Peroxidase (POD) is a heme-containing enzyme. POD associated with the development of off-flavors and browning pigments. An appraisal of data on the effect of different treatment time and different packaging materials on Peroxidase activity (POD) of processed tender coconut water is summarized in Table 4.23, 4.24 and 4.25 and graphically depicted in Fig. 4.22, 4.23 and 4.24.

4.2.8.1 Effect of Ozonation treatment and packaging material on POD for tender coconut water

From the Table 4.23 presents the data on the effects of different time treatment and different packaging materials at stored $4\pm 2^{\circ}\text{C}$ on the peroxidase activity (POD) of processed tender coconut water and also shown graphically in Fig. 4.22. The peroxidase activity of ozonation treatment processed tender coconut water decreases with the increase in storage period.

Ozonation treatment processed tender coconut water POD ranged from 0.050 to 0.014 (Δ O.D./min/ml) decreasing with increase in storage period when stored at $4\pm 2^{\circ}\text{C}$ in different packaging. POD was decreasing with ozonation treatment time and values were found more in glass bottle as compared to PET bottle.

The peroxidase activity (POD) of ozonation treatment processed tender coconut water decreases with the increase in storage period. The maximum peroxidase activity (POD) value of processed tender coconut water 4th week of storage period was obtained in treatment 20min stored in glass bottle (O_2S_1), 20min stored in PET bottle (O_2S_2) and 10min stored in PET bottle (O_1S_2) were 0.038, 0.034 and 0.032 (Δ O.D./min/ml) respectively.

Table 4.23 Effect of different treatment time and different packaging materials on the peroxidase activity (POD) of ozonation treated tender coconut water

Initial Peroxidase activity(POD) = 0.12 (Δ O.D./min/ml)

Mean Peroxidase activity(POD), Δ O.D./min/ml				
Treatments	Storage period in weeks			
	1 week	2 week	3 week	4 week
O ₁ S ₁	0.039	0.032	0.024	0.022
O ₁ S ₂	0.046	0.038	0.034	0.032
O ₂ S ₁	0.050	0.044	0.039	0.038
O ₂ S ₂	0.045	0.040	0.034	0.034
O ₃ S ₁	0.028	0.022	0.015	0.014
O ₃ S ₂	0.033	0.027	0.022	0.020
S. Em	0.0013	0.0009	0.0009	0.001
CD at 5%	0.004	0.0028	0.0027	0.0031
Test	Sig.	Sig.	Sig.	Sig.
CV %	3.3374	2.7902	3.2135	3.8609

The minimum peroxidase activity (POD) value of processed tender coconut water 4th week of storage period was obtained in treatment 30min stored in glass bottle (O₃S₁), 30min stored in PET bottle (O₃S₂) and 10min stored in glass bottle (O₁S₁) were 0.014, 0.020 and 0.022 (Δ O.D./min/ml) respectively (Table 4.23). These findings are similar to Rico, Martin- Diana, Frias, Gary and Barry-Ryan(2006) also reported the efficacy of ozone against PPO and POD enzymes in fresh cut lettuce.

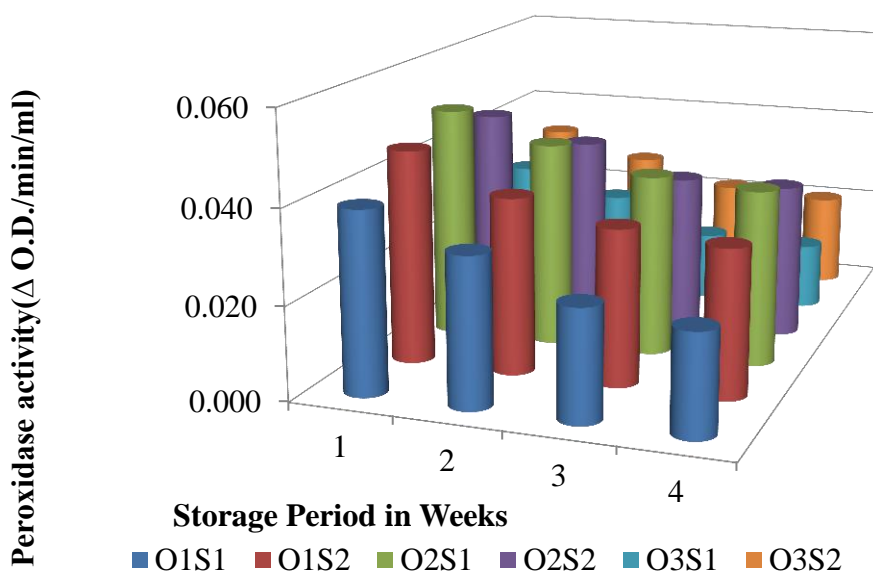


Fig. 4.22 Effect of different treatment time and different packaging materials on the peroxidase activity (POD) of ozonation treated tender coconut water

The statistical analysis of data revealed that the rate of decrease in peroxidase activity(POD) due to different due to different treatment time and different packaging materials were found to be significant with the storage period 1st, 2nd, 3rd, and 4th week of storage period respectively, where observed significantly higher due to different treatment time O₂S₁ i.e. 20min ozonation treatment and different packaging materials S₁ i.e. glass bottle though 1st, 2nd, and 3rd week storage period was remained statistically at par with different treatment time respectively.

The interaction of different treatment time and different packaging materials on the POD of processed tender coconut water was found significantly at 1st, 2nd, 3rd, and 4th week of storage period at 5% level of significance.

As a result, it is inferred that ozonation treatment should be given for 20 min with glass bottle packaging to get more POD in tender coconut water for extending the storage period of 4 week.

4.2.8.2 Effect of Ultrasonication treatment time and packaging material on POD for tender coconut water

The results presented in Table 4.24 shows the effects of different time treatment and different packaging materials at stored 4±2°C on the peroxidase activity (POD) of processed tender coconut water and also shown graphically in Fig. 4.23. The peroxidase activity of ultrasonication treatment processed tender coconut water decreases with the increase in storage period.

Ultrasonication treatment processed tender coconut water POD ranged from 0.031 to 0.012 (Δ O.D./min/ml) decreasing with increase in storage period when stored at 4±2°C in different packaging. POD was decreasing with ultrasonication treatment time and values were found more in PET bottle as compared to glass bottle.

Ultrasonication treatment processed tender coconut water the maximum peroxidase activity (POD) 4th week of storage period was obtained in treatment 20min stored in PET bottle (U₂S₂), 10min stored in PET bottle (U₁S₂) and 30min stored in PET bottle (U₃S₂) were 0.021, 0.020 and 0.018 (Δ O.D./min/ml) respectively. Ultrasonication treatment processed tender coconut water the minimum peroxidase activity (POD) 4th week of storage period was obtained in treatment 30min stored in glass bottle (U₃S₁),

20min stored in glass bottle (U₂S₁) and 10min stored in glass bottle (U₁S₁) were 0.012, 0.014 and 0.016 (Δ O.D./min/ml) respectively (Table 4.24).

Table 4.24 Effect of different treatment time and different packaging materials on the peroxidase activity (POD) of ultrasonication treated tender coconut water

Initial Peroxidase activity(POD) = 0.12 (Δ O.D./min/ml)

Mean Peroxidase Activity (POD) , Δ O.D./min/ml				
Treatments	Storage period in weeks			
	1 week	2 week	3 week	4 week
U ₁ S ₁	0.029	0.024	0.019	0.016
U ₁ S ₂	0.030	0.028	0.024	0.020
U ₂ S ₁	0.025	0.021	0.018	0.014
U ₂ S ₂	0.031	0.026	0.022	0.021
U ₃ S ₁	0.029	0.021	0.016	0.012
U ₃ S ₂	0.030	0.024	0.020	0.018
S. Em	0.0016	0.001	0.001	0.0011
CD at 5%	0.005	NS	NS	NS
Test	Sig.	NS	NS	NS
CV %	5.8447	4.4674	4.958	6.7849

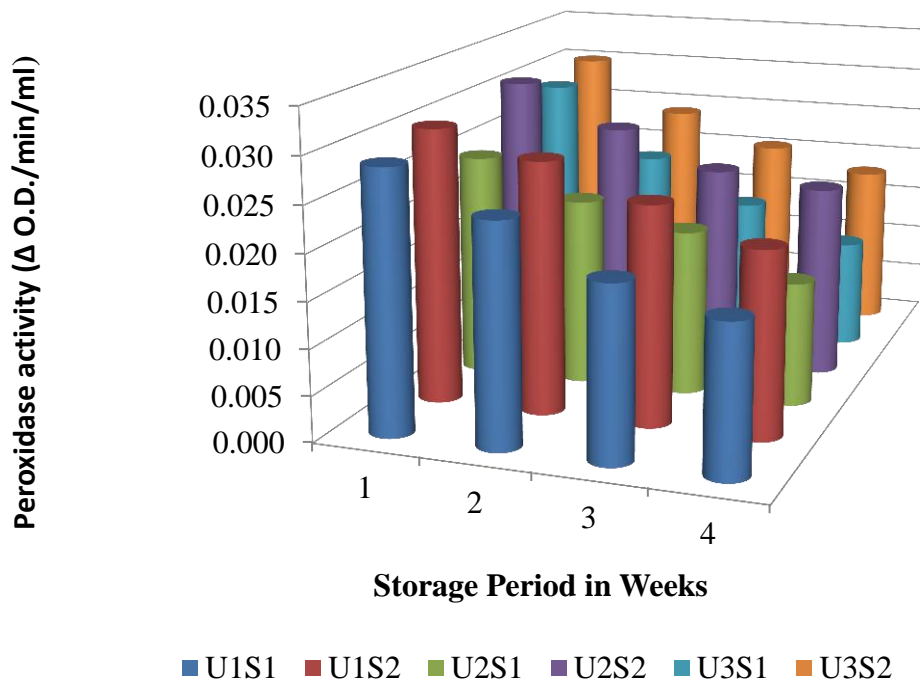


Fig. 4.23 Effect of different treatment time and different packaging materials on the peroxidase activity (POD) of ultrasonication treated tender coconut water

The statistical analysis of data revealed that the rate of decrease in peroxidase activity (POD) due to different treatment time and different packaging materials were found to be significant with the storage period 1st week of storage period respectively, where observed significantly higher due to different treatment time U₂S₂ i.e. 20min ultrasonication treatment and different packaging materials S₂ i.e. PET bottle though 1st, 2nd, 3rd and 4th week storage period was remained statistically at par with different treatment time respectively.

The interaction of different treatment time and different packaging materials on the POD of processed tender coconut water was found non significantly at 2nd, 3rd, and 4th week of storage period at 5% level of significance.

So it is concluded that ultrasonication treatment should be given for 20 min ultrasonication and with PET bottle packaging to get more POD in tender coconut water for extending the storage period of 4 week.

4.2.8.3 Combined effect of Ozonation and Ultrasonication treatment time and packaging material on POD for tender coconut water

The results presented in Table 4.25 shows the effects of different time treatment and different packaging materials at stored 4±2°C on the peroxidase activity (POD) of processed tender coconut water and also shown graphically in Fig. 4.24. The peroxidase activity of ozonation and ultrasonication treatment processed tender coconut water decreases with the increase in storage period.

Combined treatment processed tender coconut water peroxidase activity (POD) ranged from 0.051 to 0.014 (Δ O.D./min/ml) decreasing with increase in storage period when stored at 4±2°C in different packaging. peroxidase activity (POD) was decreasing with combined treatment time and values were found more in PET bottle as compared to glass bottle.

The peroxidase activity (POD) of combination treatment processed tender coconut water increases with the increase in storage period. Combination treatment processed tender coconut water the maximum peroxidase activity (POD) 4th week of storage period was obtained in treatment 30min ozonation and 20min ultrasonication stored in PET bottle (O₃U₂S₂), 30min ozonation and 10 min ultrasonication stored in glass bottle (O₃U₂S₁) and 20min ozonation and 10min ultrasonication stored in PET bottle (O₂U₁S₂)

were 0.039, 0.037 and 0.036 (Δ O.D./min/ml) respectively. Combined treatment processed tender coconut water the minimum peroxidase activity (POD) 4th week of storage period was obtained in treatment 20min ozonation and 20min ultrasonication stored in glass bottle ($O_2U_2S_1$) and 20min ozonation and 20min ultrasonication stored in PET bottle ($O_2U_2S_2$) were 0.014 and 0.016 (Δ O.D./min/ml) respectively (Table 4.25).

Table 4.25 Effect of different treatment time and different packaging materials on the peroxidase activity (POD) of combined treated tender coconut water

Initial Peroxidase activity(POD) = 0.12 (Δ O.D./min/ml)

Mean Peroxidase Activity (POD), Δ O.D./min/ml				
Treatments	Storage period in weeks			
	1 week	2 week	3 week	4 week
$O_1U_1S_1$	0.037	0.031	0.025	0.023
$O_1U_1S_2$	0.042	0.040	0.034	0.028
$O_1U_2S_1$	0.030	0.028	0.018	0.017
$O_1U_2S_2$	0.031	0.030	0.024	0.019
$O_1U_3S_1$	0.032	0.029	0.023	0.018
$O_1U_3S_2$	0.037	0.031	0.025	0.022
$O_2U_1S_1$	0.049	0.042	0.040	0.032
$O_2U_1S_2$	0.051	0.048	0.045	0.036
$O_2U_2S_1$	0.025	0.020	0.015	0.014
$O_2U_2S_2$	0.028	0.023	0.018	0.016
$O_2U_3S_1$	0.038	0.035	0.029	0.023
$O_2U_3S_2$	0.043	0.037	0.031	0.024
$O_3U_1S_1$	0.046	0.040	0.033	0.030
$O_3U_1S_2$	0.050	0.046	0.035	0.030
$O_3U_2S_1$	0.049	0.047	0.038	0.037
$O_3U_2S_2$	0.051	0.049	0.043	0.039
$O_3U_3S_1$	0.024	0.022	0.018	0.015
$O_3U_3S_2$	0.028	0.024	0.018	0.017
S. Em	0.0012	0.0012	0.0016	0.001
CD at 5%	0.0033	NS	NS	0.003
Test	Sig.	NS	NS	Sig.
CV %	3.1457	3.6646	5.7301	4.4454

The ozonation and ultrasonication treatment processed coconut water at different storage period was influenced significantly by different processing times at 1st and 4th week of storage period. The processed tender coconut water stored in glass bottle $O_3U_2S_2$ i.e. 30min ozonation treatment and 20min ultrasonication treatment Stored in PET bottle showed significantly higher peroxidase activity(POD) of processed tender coconut water

at different storage period. From the results is also clear that as the storage period decreases from 1st week to 4th week.

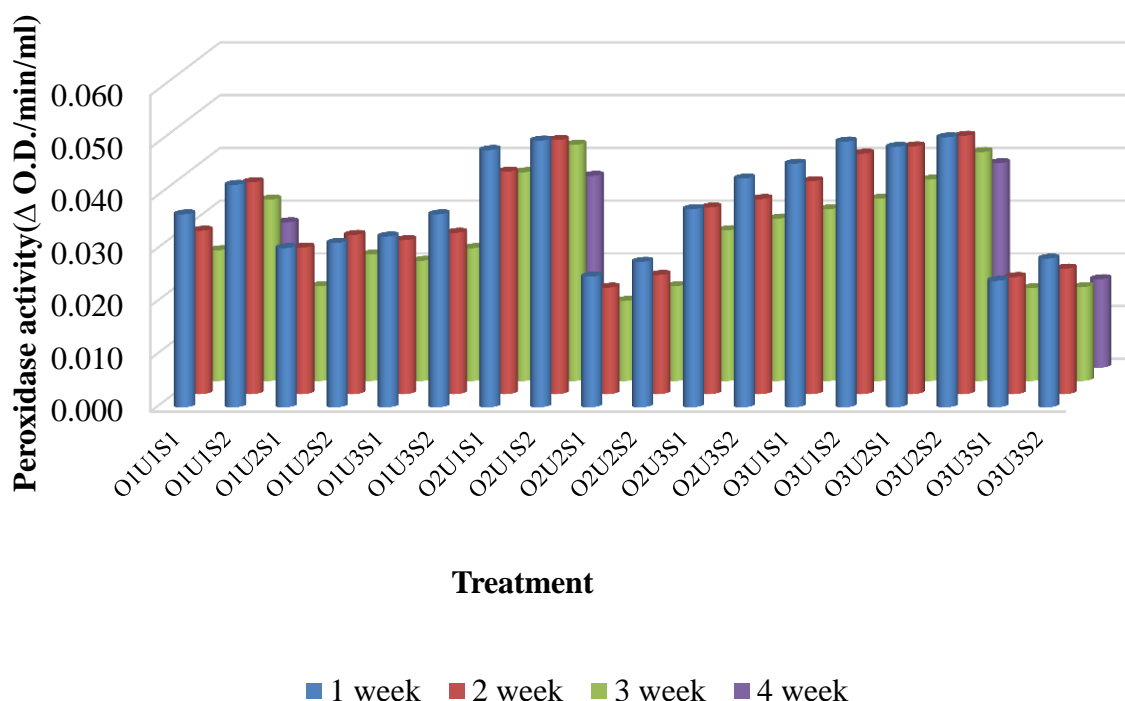


Fig. 4.24 Effect of different treatment time and different packaging materials on the peroxidase activity (POD) of combined treated tender coconut water

The interaction of different treatment time and different packaging materials on the POD of processed tender coconut water was found significantly at 1st and 4th week of storage period at 5% level of significance.

Given a result, it is determined that combined treatment should be given for 30 min ozonation and 20min ultrasonication with PET bottle packaging to get more POD in tender coconut water to prolong shelf life upto 4 week.

4.2.9 Effect of different treatment time and different packaging materials on Poly Phenol Oxidase (PPO) of treated tender coconut water

Poly Phenol Oxidase (PPO) is a copper-containing enzyme that causes enzymatic browning in fresh fruits and vegetables products such as juices. An appraisal of data on the effect of different treatment time and different packaging materials on Poly Phenol

Oxidase (PPO) of processed tender coconut water is summarized in Table 4.26, 4.27 and 4.28 and graphically depicted in Fig 4.25. 4.26 and 4.27.

4.2.9.1 Effect of Ozonation treatment and packaging materials on PPO for tender coconut water

The results presented in Table 4.26 shows the effects of different time treatment and different packaging materials at stored $4\pm 2^{\circ}\text{C}$ on the Poly Phenol Oxidase (PPO) activity of processed tender coconut water and also shown graphically in Fig. 4.25. The Poly Phenol Oxidase (PPO) activity of ozonation treatment processed tender coconut water decreases with the increase in storage period.

Ozonation treatment processed tender coconut water PPO ranged from 0.039 to 0.010(Δ O.D./min/ml) decreasing with increase in storage period when stored at $4\pm 2^{\circ}\text{C}$ in different packaging. PPO was decreasing with ozonation treatment time and values were found more in PET bottle as compared to glass bottle.

The Poly Phenol Oxidase (PPO) of ozonation treatment processed tender coconut water decreases with the increase in storage period. The maximum Poly Phenol Oxidase (PPO) value of processed tender coconut water 4th week of storage period was obtained in treatment 10min stored in PET bottle (O_1S_2), 20min stored in PET bottle (O_2S_2) and 30min stored in PET bottle (O_3S_2) were 0.033 and 0.032 (Δ O.D./min/ml) respectively.

Table 4.26 Effect of different treatment time and different packaging materials on the poly phenol oxidase (PPO) of ozonation treated tender coconut water

Initial Poly Phenol Oxidase (PPO) = 0.151(Δ O.D./min/ml)

Mean Poly Phenol Oxidase (PPO), Δ O.D./min/ml				
Treatments	Storage period in weeks			
	1 week	2 week	3 week	4 week
O_1S_1	0.035	0.037	0.026	0.021
O_1S_2	0.037	0.044	0.036	0.033
O_2S_1	0.010	0.032	0.028	0.020
O_2S_2	0.014	0.040	0.039	0.033
O_3S_1	0.010	0.036	0.031	0.025
O_3S_2	0.020	0.039	0.038	0.032
S. Em	0.0006	0.0006	0.0011	0.0013
CD at 5%	0.0018	0.0019	NS	0.004
Test	Sig.	Sig.	NS	Sig.
CV %	2.9326	1.7213	3.6298	4.9892

The minimum Poly Phenol Oxidase (PPO) value of processed tender coconut water 4th week of storage period was obtained in treatment 20min stored in glass bottle (O₂S₁), 10min stored in glass bottle (O₁S₁) and 30min stored in glass bottle (O₃S₁) were 0.020, 0.021 and 0.025 (Δ O.D./min/ml) respectively (Table 4.26).

The statistical analysis of data revealed that the rate of decrease in Poly Phenol Oxidase (PPO) activity due to different due to different treatment time and different packaging materials were found to be significant with the storage period 1st, 2nd, and 4th week of storage period respectively, where observed significantly higher due to different treatment time O₁S₂ i.e. 10min ozonation treatment and different packaging materials S₂ i.e. PET bottle though 1st, 2nd, and 3rd week storage period was remained statistically at par with different treatment time respectively.

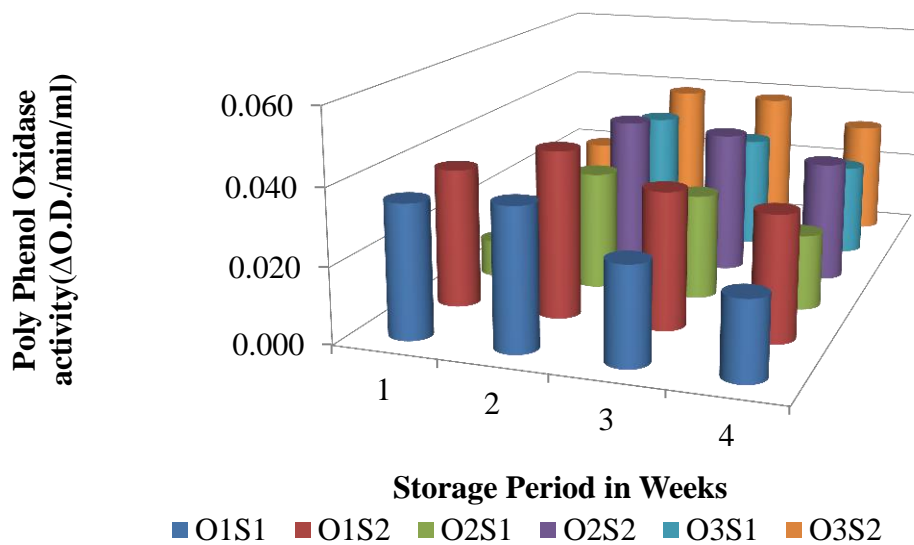


Fig. 4.25 Effect of different treatment time and different packaging materials on the polphenol oxidase (PPO) of ozonation treated tender coconut water

The interaction of different treatment time and different packaging materials on the POD of processed tender coconut water was found significantly at 1st, 2nd, 3rd, and 4th week of storage period at 5% level of significance.

So it is concluded that ozonation treatment should be given for 20 min with glass bottle packaging to get more POD in tender coconut water.

4.2.9.2 Effect of Ultrasonication treatment time packaging materials on PPO for tender coconut water

The results data presented in Table 4.27 shows the effects of different time treatment and different packaging materials at stored $4\pm 2^\circ\text{C}$ on the Poly Phenol Oxidase (PPO) activity of processed tender coconut water and also shown graphically in Fig. 4.26. The Poly Phenol Oxidase (PPO) activity of ultrasonication treatment processed tender coconut water decreases with the increase in storage period.

Ultrasonication treatment processed tender coconut water PPO ranged from 0.040 to 0.010 (Δ O.D./min/ml) decreasing with increase in storage period when stored at $4\pm 2^\circ\text{C}$ in different packaging. PPD was decreasing with ultrasonication treatment time and values were found more in PET bottle as compared to glass bottle.

Table 4.27 Effect of different treatment time and different packaging materials on the poly phenol oxidase (PPO) of ultrasonication treated tender coconut water

Initial Poly Phenol Oxidase (PPO) = 0.151(Δ O.D./min/ml)

Mean Poly Phenol Oxidase (PPO) activity, Δ O.D./min/ml				
Treatments	Storage period in weeks			
	1 week	2 week	3 week	4 week
U ₁ S ₁	0.035	0.019	0.013	0.017
U ₁ S ₂	0.037	0.021	0.024	0.032
U ₂ S ₁	0.010	0.018	0.014	0.017
U ₂ S ₂	0.014	0.027	0.019	0.021
U ₃ S ₁	0.010	0.031	0.025	0.036
U ₃ S ₂	0.020	0.032	0.026	0.040
S. Em	0.0006	0.0006	0.0004	0.005
CD at 5%	0.0018	0.002	0.0013	0.0014
Test	Sig.	Sig.	Sig.	Sig.
CV %	2.9326	2.7655	2.2242	2.9842

Ultrasonication treatment processed tender coconut water the maximum Poly Phenol Oxidase (PPO) 4th week of storage period was obtained in treatment 30min stored in PET bottle (U₃S₂), 30min stored in glass bottle (U₃S₁) and 10min stored in PET bottle (U₁S₂) were 0.040, 0.036 and 0.032 (Δ O.D./min/ml) respectively. Ultrasonication

treatment processed tender coconut water the minimum Poly Phenol Oxidase (PPO) 4th week of storage period was obtained in treatment 10min stored in glass bottle (U₁S₁), 20min stored in glass bottle (U₂S₁) and 20min stored in PET bottle (U₂S₂) were 0.017 and 0.021 (Δ O.D./min/ml) respectively (Table 4.27).

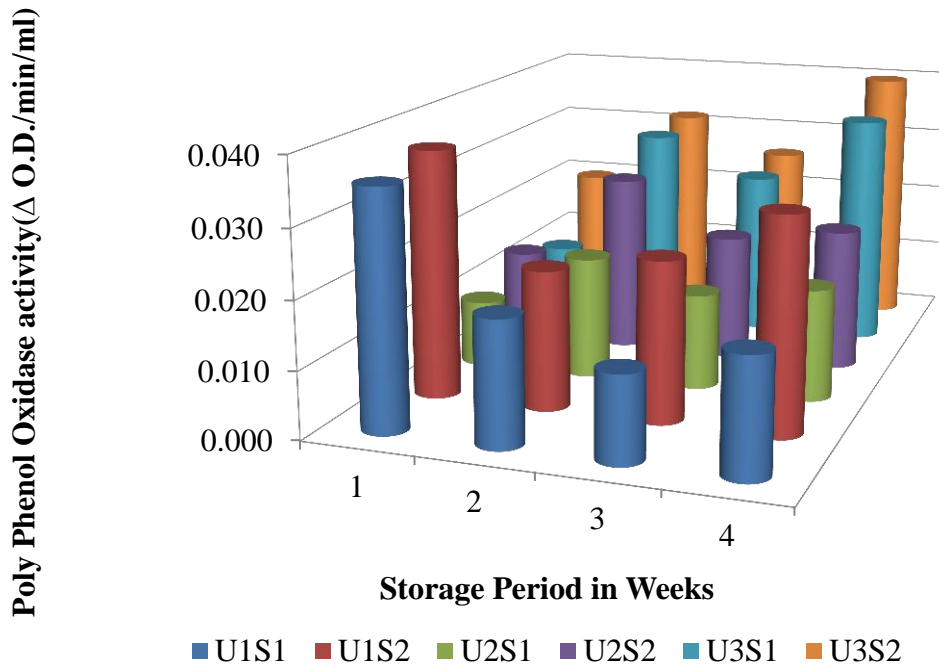


Fig. 4.26 Effect of different treatment time and different packaging materials on the poly phenol oxidase (PPO) of ultrasonication treated tender coconut water

The statistical analysis of data revealed that the rate of decrease in Poly Phenol Oxidase (PPO) activity due to different due to different treatment time and different packaging materials were found to be significant with the storage period 1st, 2nd, 3rd and 4th week of storage period respectively, where observed significantly higher due to different treatment time U₃S₂ i.e. 30min ultrasonication treatment and different packaging materials S₂ i.e. PET bottle though 1st, 2nd, and 3rd week storage period was remained statistically at par with different treatment time respectively.

The interaction of different treatment time and different packaging materials on the PPO of processed tender coconut water was found non significantly at 1st, 2nd, 3rd, and 4th week of storage period at 5% level of significance.

Given a result, it is determined that ultrasonication treatment should be given for 30min ultrasonication and with PET bottle packaging to get more PPO in tender coconut water.

4.2.9.3 Combined effect of Ozonation and Ultrasonication treatment time and packaging materials for PPO on tender coconut water

The results data presented in Table 4.28 shows the effects of different time treatment and different packaging materials at stored $4\pm 2^\circ\text{C}$ on the Poly Phenol Oxidase (PPO) activity of processed tender coconut water and also shown graphically in Fig. 4.27. The Poly Phenol Oxidase (PPO) activity of ozonation and ultrasonication treatment processed tender coconut water decreases with the increase in storage period.

Combined treatment processed tender coconut water Poly Phenol Oxidase (PPO) ranged from 0.063 to 0.007 (Δ O.D./min/ml) decreasing with increase in storage period when stored at $4\pm 2^\circ\text{C}$ in different packaging. Poly Phenol Oxidase (PPO) was decreasing with combined treatment time and values were found more in PET bottle as compared to glass bottle.

The Poly Phenol Oxidase (PPO) of combination treatment processed tender coconut water increases with the increase in storage period. Combination treatment processed tender coconut water the maximum Poly Phenol Oxidase (PPO) 4th week of storage period was obtained in treatment 30min ozonation and 10min ultrasonication stored in PET bottle ($\text{O}_3\text{U}_1\text{S}_2$), and 30min ozonation and 20min ultrasonication stored in PET bottle ($\text{O}_3\text{U}_2\text{S}_2$) were 0.039 and 0.038 (Δ O.D./min/ml) respectively. Combined treatment processed tender coconut water the minimum Poly Phenol Oxidase (PPO) 4th week of storage period was obtained in treatment 20min ozonation and 20min ultrasonication stored in glass bottle ($\text{O}_2\text{U}_2\text{S}_1$) and 20min ozonation and 30min ultrasonication stored in glass bottle ($\text{O}_2\text{U}_3\text{S}_1$) were 0.010 (Δ O.D./min/ml) respectively (Table 4.28).

The ozonation and ultrasonication treatment processed coconut water at different storage period was influenced significantly by different processing times at 1st, 2nd, 3rd and 4th week of storage period. The processed tender coconut water stored in glass bottle $\text{O}_3\text{U}_1\text{S}_2$ i.e. 30min ozonation treatment and 10min ultrasonication treatment Stored

in PET bottle showed significantly higher Poly Phenol Oxidase (PPO) activity of processed tender coconut water at different storage period. From the results is also clear that as the storage period decreases from 1st week to 4th week.

Table 4.28 Effect of different treatment time and packaging materials on the Poly Phenol Oxidase (PPO) activity of combined treated tender coconut water

Initial Poly Phenol Oxidase (PPO) = 0.151(Δ O.D./min/ml)

Mean Poly Phenol Oxidase (PPO), Δ O.D./min/ml				
Treatments	Storage period in weeks			
	1 week	2 week	3 week	4 week
O ₁ U ₁ S ₁	0.043	0.039	0.031	0.027
O ₁ U ₁ S ₂	0.049	0.038	0.036	0.029
O ₁ U ₂ S ₁	0.040	0.038	0.036	0.026
O ₁ U ₂ S ₂	0.041	0.039	0.032	0.029
O ₁ U ₃ S ₁	0.045	0.042	0.039	0.032
O ₁ U ₃ S ₂	0.044	0.041	0.040	0.033
O ₂ U ₁ S ₁	0.049	0.042	0.040	0.035
O ₂ U ₁ S ₂	0.050	0.042	0.041	0.037
O ₂ U ₂ S ₁	0.017	0.015	0.007	0.010
O ₂ U ₂ S ₂	0.026	0.014	0.013	0.014
O ₂ U ₃ S ₁	0.027	0.015	0.008	0.010
O ₂ U ₃ S ₂	0.038	0.033	0.026	0.020
O ₃ U ₁ S ₁	0.049	0.044	0.041	0.037
O ₃ U ₁ S ₂	0.062	0.050	0.045	0.039
O ₃ U ₂ S ₁	0.050	0.044	0.039	0.036
O ₃ U ₂ S ₂	0.063	0.051	0.044	0.038
O ₃ U ₃ S ₁	0.027	0.016	0.008	0.011
O ₃ U ₃ S ₂	0.029	0.017	0.010	0.011
S. Em	0.0009	0.0009	0.0009	0.0007
CD at 5%	0.0026	0.0025	0.0025	0.0019
Test	Sig.	Sig.	Sig.	Sig.
CV %	2.2325	2.6184	2.6184	2.6838

The interaction of different treatment time and different packaging materials on the PPO of processed tender coconut water was found significantly at 1st, 2nd, 3rd and 4th week of storage period at 5% level of significance.

As a result, it is inferred that combined treatment should be given for 30 min ozonation and 10min ultrasonication with PET bottle packaging to get more PPO in tender coconut water.

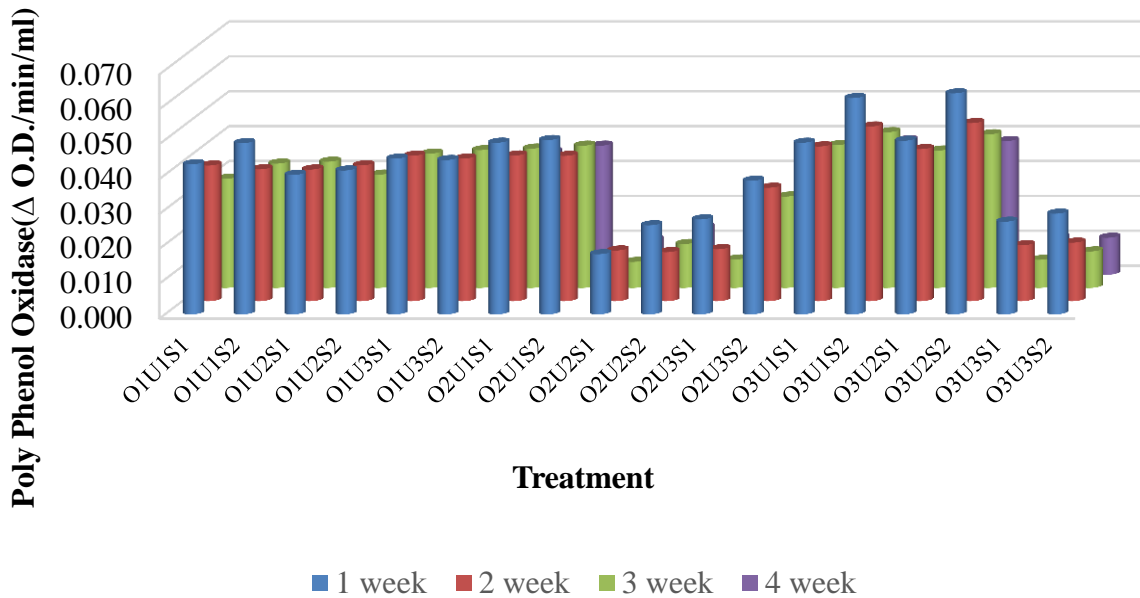


Fig. 4.27 Effect of different treatment time and different packaging materials on the Poly Phenol Oxidase PPO activity of combined treated tender coconut water

4.2.10 Effect of different treatment and packaging materials on microbiological parameters for stored tender coconut water

The results data for microbial parameters viz. total plate count, yeast and mould count of processed tender coconut water for different treatment like ozonation and ultrasonication with packaging material are presented in the table 4.29.

4.2.10.1 Total plate count

The data of total plate count of processed tender count prepared through ozonation and ultrasonication technology is presented in Table 4.29. According to table no microbial growth within first two weeks was observed in samples. From the table, it can be observed that the total plate count of processed tender coconut water ranged from 0 log (CFU/ml) to 3.681log (CFU/ml). The maximum total plate count (3.681 log (CFU/ml))

was observed for the treatment No. 20 having a combination of 20min ozonation and 10 min ultrasonication treatment stored in PET bottles and stored 4weeks of processed tender coconut water. However, their counts were recorded below the satisfactory level as recommended for the ozonation treated fruit juice 5 log reduction by Patil *et al.*, (2010) and 3.4 log reduction in ultrasound orange juice was reported by Valero *et al.* (2007). Hence, the prepared processed tender coconut water is found safe for the consumptions.

Table 4.29 Effect of different treatment time and different packaging materials on the microbial parameters treated tender coconut water

T. No	Total plate count, Log (CFU/ml)		Yeast and Mould count, Log (CFU/ml)		T. No	Total plate count, Log (CFU/ml)		Yeast and Mould count, Log (CFU/ml)	
	Storage period		Storage period			Storage period		Storage period	
	3rd week	4th week	3rd week	4th week		3rd week	4th week	3rd week	4th week
1	1.342	2.1	1.602	2.431	16	1.301	2.505	1.342	2.447
2	1.602	2.079	1.447	2.147	17	ND	1.397	2.376	3.256
3	1.384	2.485	1.681	2.253	18	1.342	2.613	ND	1.778
4	1.726	2.762	2.398	3.181	19	2.164	3.654	2.485	3.153
5	2.143	3.095	2.613	2.672	20	2.477	3.681	2.762	3.231
6	2.23	3.183	2.653	3.156	21	ND	2.472	1.301	2.019
7	ND	1.573	1.384	2.690	22	ND	1.681	ND	1.726
8	1.301	2.376	ND	1.397	23	ND	1.726	2.230	3.185
9	ND	1.756	1.778	2.579	24	2.431	3.447	ND	1.602
10	ND	2.418	ND	1.384	25	2.472	3.623	2.431	3.105
11	ND	2.164	1.397	2.472	26	2.623	3.643	2.613	3.343
12	1.477	2.23	ND	1.301	27	2.491	3.503	2.477	3.147
13	1.778	2.477	1.726	2.491	28	2.613	3.554	2.762	3.219
14	1.699	2.019	ND	2.519	29	2.447	3.447	2.398	2.505
15	ND	1.374	1.301	2.623	30	2.580	3.531	2.519	3.243

4.2.10.2 Yeast and mould count

The data of yeast and mould count of processed tender coconut water as prepared through ozonation and ultrasonication technology is presented in Table 4.23. According to table no microbial growth within first two weeks was observed in samples. From the table, it can be observed that the yeast and mould count of processed tender coconut water ranged from 0 log (CFU/ml) to 3.343log (CFU/ml). The maximum yeast and mould count

(3.343 log (CFU/ml)) was observed for the treatment No. 26 having a combination of 30min ozonation and 10 min ultrasonication treatment stored in PET bottles and stored 4weeks of processed tender coconut water.

Looking to the microbial analysis, it was suggested to give ultrasonication treatment for 20 min with glass bottle packaging to extend shelf life of 4 week to get good quality tender coconut water without microbial infection.

4.3 Sensory analysis of treated tender coconut water

Sensory evaluation for different treatment and packaging materials of processed tender coconut water was carried by 9-point hedonic scale by panel of experts and results obtained were presented in Table 4.30, 4.31 and 4.32 also shown graphically in Fig. 4.29, 4.30 and 4.31 for overall acceptability including color, flavor and taste.

Based on 9 Point Hedonic scale

9 - Like extremely

8 - Like very much

7 - Like moderately

6 - Like slightly

5 - Neither like nor dislike

4 - Dislike slightly

3 - Dislike moderately

2 - Dislike very much

1 - Extremely dislike

4.3.1 Effect of different treatment time and different packaging materials on the Overall acceptability of treated tender coconut water

4.3.1.1 Effect of Ozonation treatment on overall acceptability for tender coconut water

The ranks of overall acceptability were given to the treatment by taking average of the all sensory characteristics (color, flavor and taste). The mean sensory score assigned by the panelists to the overall acceptability attribute of stored ozonation treatment processed tender coconut water, packed in different packaging materials are given in Table 4.30 and Fig. 4.28.

From the Table it is clear that the maximum overall acceptability 4th week of storage period was obtained in treatment 20min stored in glass bottle (O₂S₁), 20min stored in PET bottle (O₂S₂) and 10min stored in glass bottle (O₁S₁) were 6.70, 6.67 and 6.50 respectively. The minimum overall acceptability 4th week of storage period was

obtained in treatment 30min stored in PET bottle (O_3S_2), 30min stored in glass bottle (O_3S_1) and 10min stored in PET bottle (O_1S_2) were 5.47, 5.50 and 6.33 respectively (Table 4.30).

As shown in table 4.30, it is clearly indicated that the initial mean score of 8.33 was declined gradually 5.47 during storage indicating the inferior response. This may be due to biochemical and microbiological reaction after 4weeks of the storage. The control was observed higher in overall acceptability as compared to all the treatments.

Table 4.30 Effect of different treatment time and different packaging materials on the overall acceptability of ozonation treated tender coconut water

Initial score of overall acceptability = 8.50

Mean Overall Acceptability				
Treatments	Storage period in weeks			
	1 week	2 week	3 week	4 week
O_1S_1	7.60	7.00	6.77	6.50
O_1S_2	7.37	6.90	6.73	6.33
O_2S_1	7.67	7.10	7.00	6.70
O_2S_2	7.67	6.80	6.67	6.67
O_3S_1	8.20	7.50	7.20	5.50
O_3S_2	8.33	7.20	7.13	5.47
S. Em	0.085	0.0892	0.0828	0.0913
CD at 5%	NS	NS	NS	NS
Test	NS	NS	NS	NS
CV %	1.8912	2.1092	2.0728	2.5525

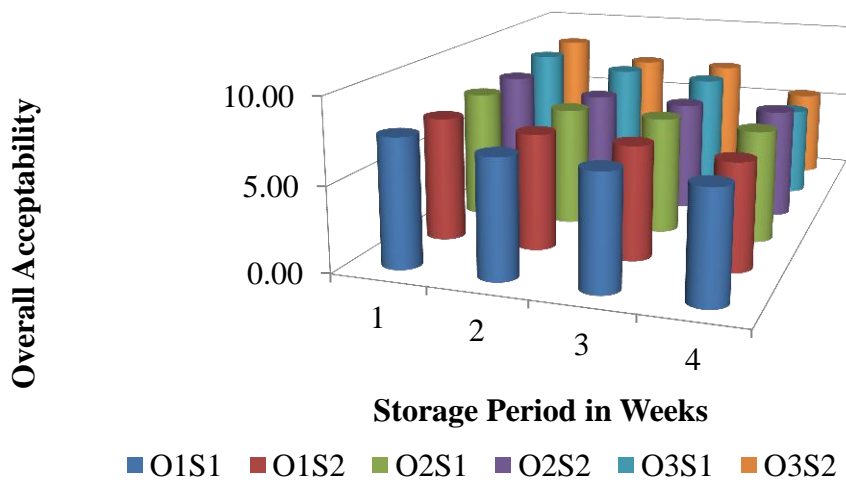


Fig. 4.28 Effect of different treatment time and different packaging materials on the overall acceptability of ozonation treated tender coconut water

The interaction between treatment time and packaging materials gives statistically non- significant results.

Further it may concluded that, looking of acidity, TSS, pH, total sugar, total phenol content, TDS, EC, total plate count, enzyme activity and overall acceptability, it may be concluded that the prepared tender coconut water treated ozonation time ($O_2 S_1$) i.e. 20 min and stored in glass bottle up to 4weeks.

4.3.1.2 Effect of Ultrasonication treatment time on overall acceptability for tender coconut water

The ranks of overall acceptability were given to the treatment by taking average of the all sensory characteristics (color, flavor and taste). The mean sensory score assigned by the panelists to the overall acceptability attribute of stored ultrasonication treatment processed tender coconut water, packed in different packaging materials are given in table 4.31 and Fig. 4.29.

From the table it is clear that the maximum overall acceptability 4th week of storage period was obtained in treatment 20min stored in glass bottle (U_2S_1), 20min stored in PET bottle (U_2S_2), and 30min stored in PET bottle (U_3S_2) were 8.13 and 7.50 respectively. The minimum overall acceptability 4th week of storage period was obtained in treatment 30min stored in glass bottle (U_3S_1), 10min stored in PET bottle (U_1S_2) and 10min stored in glass bottle (U_1S_1) were 7.30, 7.37 and 7.47 respectively (Table 4.31).

As shown in table 4.31, it is clearly indicated that the initial mean score of 8.37 was declined gradually 7.30 during storage indicating the inferior response. This may be due to biochemical and microbiological reaction after 4weeks of the storage. The control was observed higher in overall acceptability as compared to all the treatments.

The interaction between treatment time and packaging materials gives statistically non- significant results.

Further it may concluded that, looking of acidity, TSS, pH, total sugar, total phenol content, TDS, EC, total plate count, enzyme activity and overall acceptability, it may be

concluded that the prepared tender coconut water treated ultrasonication time U₂S₁ and U₂S₂ i.e. 20 min and stored in glass bottle and PET bottles up to 4weeks

Table 4.31 Effect of different treatment time and different packaging materials on the overall acceptability of ultrasonication treated tender coconut water

Initial score of overall acceptability = 8.50

Mean Overall Acceptability				
Treatments	Storage period in weeks			
	1 week	2 week	3 week	4 week
U ₁ S ₁	7.83	7.70	7.60	7.47
U ₁ S ₂	7.77	7.60	7.50	7.37
U ₂ S ₁	8.37	8.37	8.20	8.13
U ₂ S ₂	8.13	8.27	8.30	8.13
U ₃ S ₁	7.73	7.50	7.40	7.30
U ₃ S ₂	7.87	7.60	7.50	7.50
S. Em	0.0745	0.0694	0.0638	0.0882
CD at 5%	NS	NS	NS	NS
Test	NS	NS	NS	NS
CV %	1.6239	1.5332	1.4234	2.0322

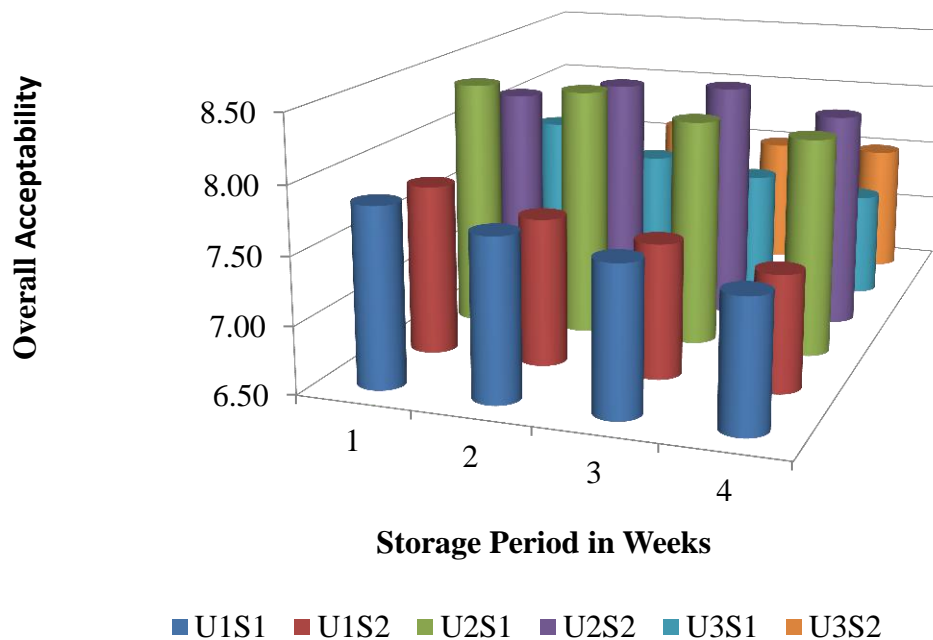


Fig 4.29 Effect of different treatment time and different packaging materials on the overall acceptability of ultrasonication treated tender coconut water

4.3.1.3 Combined effect of Ozonation and Ultrasonication treatment time for overall acceptability tender coconut water

The ranks of overall acceptability were given to the treatment by taking average of the all sensory characteristics (color, flavor and taste). The mean sensory score assigned by the panelists to the overall acceptability attribute of stored ozonation and ultrasonication treatment processed tender coconut water, packed in different packaging materials are given in table 4.32 and Fig. 4.30.

From the table it is clear that the maximum over all acceptability 4th week of storage period was obtained in treatment 20min ozonation and 10min ultrasonication stored in PET bottle (O₂U₁S₂), 20min ozonation and 10min ultrasonication stored in glass bottle (O₂U₁S₁) and 10min ozonation and 10min ultrasonication stored in PET bottle (O₁U₁S₂) were 6.93, 7.03 and 7.20 respectively. The minimum over all acceptability 4th week of storage period was obtained in treatment 10min ozonation and 10min ultrasonication stored in PET bottle (O₁U₁S₂), 20min ozonation and 10min ultrasonication stored in glass bottle (O₂U₁S₁) and 10min ozonation and 10min ultrasonication stored in glass bottle (O₁U₁S₁) were 5.63, 5.97 and 6.07 respectively (Table 4.32).

As shown in table 4.32, it is clearly indicated that the initial mean score of 8.40 was declined gradually 5.63 during storage indicating the inferior response for ozonation and ultrasonication treatment . This may be due to biochemical and microbiological reaction after 4weeks of the storage. The control was observed higher in overall acceptability as compared to all the treatments.

The interaction between treatment time and packaging materials gives statistically non- significant results for 1st, 2nd, 3rd and 4th week.

Further it may concluded that, looking of acidity, TSS, pH, total sugar, total phenol content, TDS, EC, total plate count, enzyme activity and overall acceptability, it may be concluded that the prepared tender coconut water treated O₂U₂S₂ i.e. ozonation and ultrasonication treated 20 min and stored in PET bottles up to 4weeks.

Table 4.32 Effect of different treatment time and different packaging materials on the overall acceptability of combined treated tender coconut water

Initial score of overall acceptability = 8.50

Mean Overall Acceptability				
Treatments	Storage period in weeks			
	1 week	2 week	3 week	4 week
O ₁ U ₁ S ₁	7.27	7.00	6.67	6.07
O ₁ U ₁ S ₂	7.20	7.03	6.57	5.63
O ₁ U ₂ S ₁	7.97	7.80	7.43	6.93
O ₁ U ₂ S ₂	8.07	7.80	7.63	7.33
O ₁ U ₃ S ₁	7.30	7.10	6.83	6.63
O ₁ U ₃ S ₂	7.30	7.23	6.90	6.57
O ₂ U ₁ S ₁	7.03	6.80	6.50	5.97
O ₂ U ₁ S ₂	6.93	6.80	6.57	6.13
O ₂ U ₂ S ₁	8.40	8.10	7.83	7.33
O ₂ U ₂ S ₂	8.20	8.10	7.60	7.50
O ₂ U ₃ S ₁	7.87	7.73	7.40	7.20
O ₂ U ₃ S ₂	7.80	7.73	7.50	7.27
O ₃ U ₁ S ₁	7.33	6.87	6.67	6.40
O ₃ U ₁ S ₂	7.37	6.97	6.83	6.67
O ₃ U ₂ S ₁	7.50	7.20	7.03	6.80
O ₃ U ₂ S ₂	7.53	7.07	6.80	6.40
O ₃ U ₃ S ₁	8.00	7.73	7.60	7.27
O ₃ U ₃ S ₂	7.73	7.60	7.40	7.10
S. Em	0.0724	0.079	0.0809	0.0892
CD at 5%	NS	NS	NS	NS
Test	NS	NS	NS	NS
CV %	1.6509	1.8556	1.9738	2.2955

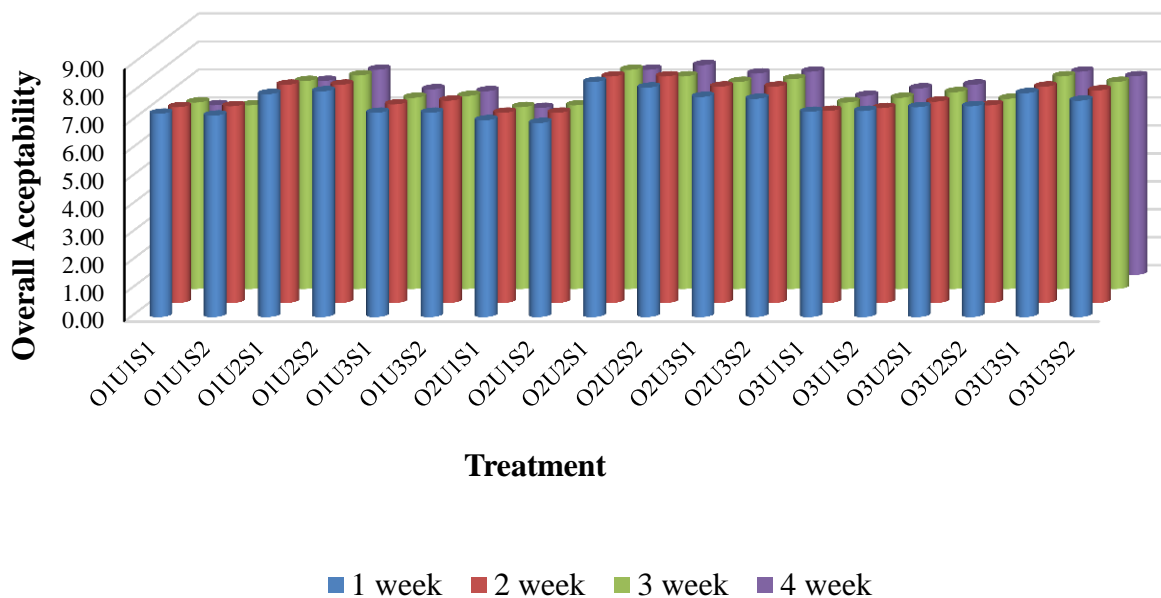


Fig 4.30 Effect of different treatment time and different packaging materials on the overall acceptability of combined treated tender coconut water

4.4 Comparison of different treatment time and different packaging materials for shelf life extension of tender coconut water

The best treatment for ozonation treatment, ultrasonication treatment and combined treatment was compared in terms of shelf life, biochemical parameters like pH, TSS, titratable acidity, EC, TDS, total sugar, total phenol content, microbial count, enzyme activities and sensory analysis. The comparison data pertaining to treated tender coconut water was presented in Table 4.33. The results for ozonation treatment 20min treatment time and glass bottle compared with 10min treatment time and glass bottle, ultrasonication treatment 20min treatment time and glass bottle compared with 20min treatment time and PET bottle, combined treatment 20min ozonation treatment, 20 min ultrasonication treatment and PET bottle compared with glass bottle stored at 4±2°C upto 4weeks.

The storage life of tender coconut water was extended up to 4weeks for ozonation treatment, ultrasonication treatment and combined treatment stored in glass and PET bottle packing and at refrigerated condition. This had shown prolongation of shelf life of

tender coconut water by incorporating treatment, packaging and refrigeration ($4\pm 2^{\circ}\text{C}$) storage.

In case of biochemical parameters, there was a little difference in pH, TSS, and titratable acidity for different treatment treatments, packing and storage conditions but total sugar, total phenol content and enzyme activities were decreased and microbial count increased with storage period. In respect of enzyme activities, ozonation, ultrasonication and combined treatment retained maximum reduced enzyme activities selected for best treatment for prolonging shelf life of tender coconut water up to 4 weeks.

During the sensory analysis ozonation treatment recorded 6.70 score, combined treatment 7.33 score, while it was for 8.13 score ultrasonication treatment would be favourable treatment in respect of other all treatment, packing and storage conditions as it maintained the customer appeal in terms of appearance. Browning percent was observed more in control at ambient condition as compared to coated fruits. There was limited level (satisfactory level) attack of microbes in ozonation, ultrasonication and combined treatments mentioned in Table 4.33.

Table 4.33 Comparison of different treatment time and different packaging materials for shelf life extension of tender coconut water

S. No.	Particulars	Ozonation Treatment		Ultrasonication treatment		Combined treatment	
		O ₂ S ₁	O ₁ S ₁	U ₂ S ₁	U ₂ S ₂	O ₂ U ₂ S ₂	O ₂ U ₂ S ₁
(1)	(2)	(3)	(4)	(5)	(6)	(7)	(8)
1	Storage life	4 th week	1 st week	4 th week	1 st week	4 th week	1 st week
2	Biochemical parameters						
	TSS Initial=4.94°Brix	4.89	4.88	5.17	4.92	4.98	5.18
	Titrateable acidity Initial=0.08%	0.08	0.07	0.08	0.07	0.09	0.08
	pH Initial=4.96	4.94	4.95	4.87	4.66	4.72	4.71
	EC= 5.64 mS/cm	5.10	5.52	5.24	5.15	5.93	5.40
	TDS= 4.17ppm	4.14	4.36	4.03	4.07	4.68	4.27
	TS=4.51%	4.13	4.66	4.37	4.57	3.94	4.33
	Phenol content Initial= 3.75mg GAE/100ml	2.37	3.23	2.43	3.10	2.07	3.33
	Total plate count (log CFU/ml)	2.485	Nil	1.756	Nil	1.681	Nil
	Yeast and mould count (log CFU/ml)	2.253	Nil	2.579	Nil	1.726	Nil
3	Enzyme Activities						
	PPO (Δ O.D./min/ml)	0.020	0.035	0.017	0.014	0.014	0.017
	POD (Δ O.D./min/ml)	0.038	0.039	0.014	0.031	0.016	0.025
4	Sensory analysis						
	Overall acceptability	6.70	7.60	8.13	8.13	7.50	8.40

4.5 Recommendation of pretreatment for prolonging shelf life of tender coconut water

From the analysis of observations taken during experiment, it was observed that amongst all the treatments, the ultrasonication treatment for 20 min with glass bottle packing at 4 ± 2 °C storage temperature yielded better results. The shelf life of tender coconut water could be increased to 4 weeks by packing in glass bottle and storage at 4 ± 2 °C.

It was recommended to give treatment of ultrasonication to tender coconut water for 20 min to store at 4 ± 2 °C temperature in glass bottle for prolonging the shelf life up to 4 weeks for maintaining freshness and quality of tender coconut water with TSS (5.17 %), pH (4.87), titratable acidity (0.08%), total sugar (4.37%), electrical conductivity (5.24mS/cm), total dissolved solids (4.03ppm), total phenol content (2.43 mg GAE/100ml) minimum infection of level (satisfactory level) microbes, poly phenol oxidase activity (0.017 Δ O.D./min/ml), peroxidase activity(0.014 Δ O.D./min/ml).

CHAPTER – V

SUMMARY AND CONCLUSIONS

The present research work entitled “**Study on packaging and storage of tender coconut water**” was carried at Department of processing and Food Engineering, College of Agricultural Engineering and Technology, Junagadh Agricultural University, Junagadh.

Coconut (*Cocos nucifera* L.) has been described as the most important and extensively grown palm tree worldwide. Every part of the plant is useful and, in many cases, human life would be impossible in its absence. The leaves and trunk are used for construction, and the root is employed in medicine. The fruit is the most valuable portion; the envelope (mesocarp), also known as the husk, is used to make rope, carpets, geotextiles, and growing media. The hard brown shell (endocarp) of the plant can be processed into extremely high-quality activated charcoal. The nut's interior (endosperm) is separated into two edible parts: a transparent liquid (coconut water) with a white kernel.

Coconut pulp or kernel has been regarded a cash crop for more than a century due to its high fat content; nevertheless, coconut is now more than just an oil seed. Copra, the dried kernel, was a vital worldwide product in the early twentieth century. The lauric oil derived from copra was processed into margarine and detergent by the food and chemical industries. However, over the last 20 years, the volume of global copra trade has declined by 75% while exports of "fresh coconuts" have soared by 300%. The canned coconut milk, coconut cream, and coconut juice/water business is rapidly expanding. Coconut is becoming more valuable as a fresh fruit rather than only an international oil commodity.

Tender coconut water (TCW) is the sweet, clear, watery portion of the juvenile green coconut. It is used to quench thirst and as a sports drink. TCW contains a good amount of carbohydrates, B vitamins, and a tiny amount of proteins and fats, in addition to electrolytes such as sodium, potassium, calcium, and magnesium, which provides balanced nutrition to boost health. Polyphenols (phenolics and flavonoids) have been linked to a variety of health benefits, and these beneficial chemicals have been found in trace concentrations in TCW. These chemicals also influence food colour and sensory

qualities. The principal browning enzymes responsible for changes in the sensory and nutritional quality of TCW are polyphenol oxidase (PPO) and peroxidase (POD).

PPO uses polyphenol as a substrate and through rapid polymerization of O-quinones produces brown, black, or red pigment, whereas POD catalyses phenolic oxidation reactions to produce browning. Browning occurring in foods due to the reactions between sugar and amino acids, vitamin C decomposition, and pigment destruction is measured in terms of the browning index, which is an indicator of nonenzymatic browning.

TCW is in high demand due to its unique nutritional properties. TCW begins to deteriorate once exposed to air and stored at room temperature due to microbial contamination as well as oxidation reactions, which cause changes in its sensory properties. As a result, proper processing technique is required to produce a stable product without altering its taste, nutritional, and nutraceutical contents. Several preservation strategies have been tried, including filtration, reverse osmosis, spray drying, carbonation, preservative addition, UV treatment, pulsed electric field, and high pressure carbon dioxide, but preserving the TCW with its wholesome natural property remains a challenge.

Keeping above in view the present studies was undertaken to Study on packaging and storage of tender coconut water with the following objectives:

1. To standardize the process parameters for the tender coconut water preservation by ozonation and ultrasonication.
2. To study the shelf life of tender coconut water packed in different packaging material.
3. To study quality parameters during storage of tender coconut water.

The tender coconuts was purchased from the Junagadh Agricultural University, Junagadh, Gujarat, India. Coconut water samples were extracted from tender coconuts (*Cocos nucifera* L.) at the 7-9th maturation month. Coconuts were immersion in water for 20min. The extraction was done manually with the aid of a sterile stainless steel knife. After extraction, the samples were homogenized and then filtered on standard muslin cloth. The filtered tender coconut water was treated with ozonation and ultrasonication methods for different times and variables. Treated samples were packed in glass and PET bottles. Then the packed samples were refrigerated at $4\pm 2^{\circ}\text{C}$ and stored for 4weeks.

The experiment was designed using Factorial Completely Randomized Design (FCRD) with three replications and had fifteen treatments viz., in first factor ozone treatment different time (10,20,30 min) and second factor processed tender coconut water stored in two different packaging materials (glass and PET bottles) for ozone and ultrasound treatment. Combination treatment first factor ultrasound treatment three different time, second factor ozonation treatment three different time and third factor processed tender coconut water stored in two different packaging materials.

The different physical, biochemical, enzyme activities and sensory properties of coconut and coconut water viz weight, size, sphericity, volume, titratable acidity (TA), Total Soluble Solids, pH, Total Sugar, Electrical conductivity, Total viable count, Yeast and mould count, Total phenol content, Total Dissolved Solids, Peroxidase, Poly Phenol Oxidase, Taste, Color, Flavor and Overall acceptability of each treatment of processed tender coconut water were determined as standard techniques and procedures.

Based on the data collected and its analysis, the following conclusions were drawn from the present investigation.

1. Physical property of tender coconut viz. weight, size, sphericity, volume were measured as 1.883kg, 285.19, 140.43 and 180.8 mm respectively.
2. Titratable acidity (TA) was found ranged from 0.06 to 0.11%. The maximum TA was observed for the treatment maximum O_1S_2 , O_2S_2 , O_3S_2 (0.09%) and minimum O_1S_1 , O_3S_1 (0.07%) for ozonation treatment and ultrasonication treatment maximum U_2S_2 (0.10%) and minimum U_1S_1 , U_1S_2 , U_3S_1 (0.07%) titratable acidity and combination treatment maximum $O_1U_3S_2$, $O_3U_1S_1$ (0.11) and minimum $O_1U_1S_1$ (0.06%) for 4th week storage period.
3. Total Soluble Solids was found ranged from 4.83 to 5.30°Brix. The maximum TSS was observed for the treatment O_3S_1 (5.19°Brix) and minimum O_2S_1 (4.89°Brix) for ozonation treatment and ultrasonication treatment maximum U_3S_2 (5.23°Brix) and minimum U_2S_2 (4.93°Brix) TSS and combination treatment maximum $O_1U_1S_1$ (5.23°Brix) and minimum $O_2U_3S_1$ (4.93°Brix) for 4th week of storage period.
4. pH was found ranged from 5.03 to 4.66. The maximum pH was observed for the treatment O_2S_1 (4.94) and minimum O_1S_1 (4.88) for ozonation treatment and ultrasonication treatment maximum U_2S_1 (4.87) and minimum U_2S_2 (4.68) pH and

- combination treatment maximum $O_1U_1S_1$ (4.95) and minimum $O_1U_2S_2$ (4.70) for 4th week of storage period.
5. Electrical conductivity (EC) was found ranged from 6.49 to 5.08 mS/cm. The maximum EC was observed for the treatment O_2S_2 (5.84mS/cm) and minimum O_2S_1 (5.10mS/cm) for ozonation treatment and ultrasonication treatment maximum U_3S_2 (5.83 mS/cm) and minimum U_2S_2 (5.15 mS/cm) EC and combination treatment maximum $O_3U_1S_1$ (6.49mS/cm) and minimum $O_1U_1S_2$ (5.12 mS/cm) for 4th week of storage period.
 6. Total Dissolved Solids (TDS) was found ranged from 5.13 to 3.97 ppm. The maximum TDS was observed for the treatment O_3S_2 (4.61 ppm) and minimum O_2S_2 (4.06 ppm) for ozonation treatment and ultrasonication treatment maximum U_2S_2 (4.62 ppm) and minimum U_2S_1 (4.03 ppm) TDS and combination treatment maximum $O_3U_1S_1$ (5.13 ppm) and minimum $O_1U_1S_2$ (4.05 ppm) for 4th week of storage period.
 7. Total Sugar (TS) was found ranged from 5.49 to 3.33%. The maximum TS was observed for the treatment O_1S_1 (4.57%) and minimum O_2S_2 (3.62%) for ozonation treatment and ultrasonication treatment maximum U_3S_2 (5.16%) and minimum U_3S_1 (3.72%) TS and combination treatment maximum $O_3U_1S_1$ (5.27%) and minimum for $O_3U_1S_2$ (3.53%) 4th week of storage period.
 8. Total Phenol Content (TPC) was found ranged from 3.67 to 1.73 (mg GAE/100ml). The maximum TPC was observed for the treatment O_2S_2 (2.67 mg GAE/100ml) and minimum O_1S_2 (1.97 mg GAE/100ml) for ozonation treatment and ultrasonication treatment maximum U_3S_2 (3.17 mg GAE/100ml) and minimum U_2S_2 (1.77 mg GAE/100ml) TPC and combination treatment maximum $O_1U_1S_2$ (3.00 mg GAE/100ml) and minimum $O_1U_3S_2$ (1.73 mg GAE/100ml) for 4th week of storage period.
 9. Total plate count of processed tender coconut water ranged from 0 log (CFU/ml) to 3.681log (CFU/ml). The maximum total plate count (3.681 log (CFU/ml)) was observed for the treatment No. 20 having a combination of 20min ozonation (T_2) and 10 min (T_1) ultrasonication treatment stored in PET bottles (S_2) and stored 4weeks of processed tender coconut water.
 10. The yeast and mould count of processed tender coconut water ranged from 0 log (CFU/ml) to 3.343log (CFU/ml). The maximum yeast and mould count (3.343 log (CFU/ml)) was observed for the treatment No. 26 having a combination of 30min

ozonation (T_3) and 10 min (T_1) ultrasonication treatment stored in PET bottles (S_2) and stored 4 weeks of processed tender coconut water.

11. Peroxidase (POD) was found ranged from 0.051 to 0.012 (Δ O.D./min/ml). The maximum POD was observed for the treatment O_2S_1 (0.038 Δ O.D./min/ml) and minimum O_3S_1 (0.014 Δ O.D./min/ml) for ozonation treatment and ultrasonication treatment maximum U_2S_2 (0.021 Δ O.D./min/ml) and minimum U_3S_1 (0.012 Δ O.D./min/ml) POD and combination treatment maximum $O_3U_2S_2$ (0.039 Δ O.D./min/ml) and minimum $O_2U_2S_1$ (0.014 Δ O.D./min/ml) for 4th week of storage period.
12. Poly Phenol Oxidase (PPO) was found ranged from 0.062 to 0.010 (Δ O.D./min/ml). The maximum PPO was observed for the treatment O_1S_2 , O_2S_2 (0.033 Δ O.D./min/ml) and minimum O_1S_1 (0.021 Δ O.D./min/ml) for ozonation treatment and ultrasonication treatment maximum U_3S_2 (0.040 Δ O.D./min/ml) and minimum U_1S_1 , U_2S_1 (0.017 Δ O.D./min/ml) PPO and combination treatment maximum $O_3U_1S_2$ (0.039 Δ O.D./min/ml) and minimum $O_2U_2S_1$ (0.010 Δ O.D./min/ml) for 4th week of storage period.
13. The sensory characteristics of processed tender coconut water *viz.* taste, flavor, color and overall acceptability of three different treatment and packaging materials compared with control tender coconut water. The maximum score of overall acceptability was found in O_2S_1 (6.70) of ozonation treatment and of ultrasonication U_2S_1 , U_2S_2 (8.13) treatment and combination treatment $O_2U_2S_2$ (7.50) tender coconut water stored at $4 \pm 2^\circ\text{C}$ for 4th week of storage period.

Based on above results, it may be concluded that, tender coconut water treated with 20min ultrasonication treatment stored at $4 \pm 2^\circ\text{C}$ temperature in glass bottle for prolonging the shelf life up to 4 weeks for maintaining freshness and quality of tender coconut water with reduce the enzyme activities. The objective of this study is achieved because of reduce the enzyme activity and microbial count of tender coconut water.

BIBLIOGRAPHY

- Adil, R. M., Zeng, X. A., Abbasi, A. M., Khan, M. S., Khalid, S., Jabbar, S. and Abid, M. (2015). Influence of power ultrasound on the quality parameters of grapefruit juice during storage. *Scientific Letters*, **3**: 6–12.
- Ali, A., Muhammad, M. T. M., Sijam, K. and Siddiqui, Y. (2010). Potential of chitosan coating in delaying the postharvest anthracnose (*Colletotrichum gloeosporioides* Penz.) of Eksotika II papaya. *International Journal of Food Science and Technology*, **45(10)**: 2134-2140.
- Allende, A., Marin, A., Buendia, B., Thomas-Barberan, F. and Gil, M. I. (2007). Impact of combined postharvest treatments (UV-C light, gaseous O₃, super atmospheric O₂ and high CO₂) on health promoting compounds and shelf-life of strawberries. *Postharvest Biology and Technology*, **46**: 201–211.
- Alleyne, T., Roache, S., Thomas, C. and Shirley, A. (2005). The control of hypertension by use of coconut water and mauby, two tropical food drinks. *West Indian Medicinal Journal*, **54**: 3-8.
- Almeida, F. D. L., Cavalcante, R. S., Cullen, P. J., Frias, J. M., Bourke, P., Fernandes, F. A. N. and Rodrigues, S. (2015). Effects of atmospheric cold plasma and ozone on prebiotic orange juice. *Innovative Food Science and Emerging Technologies*, **32**: 127–135.
- Alothman, M., Kaur, B., Fazilah, A., Bhat, R., and Karim, A. A. (2010). Ozone induced changes of antioxidant capacity of fresh-cut tropical fruits. *Innovative Food Science and Emerging Technologies*, **11(4)**: 666–671.
- Amerine, M. A., Pangborn, R. M. and Roessler, E. B. (1965). Principles of sensory evaluation of food. *In: Food Science and Technology Monographs*. pp. 338-339.
- Anonymous. Using Electrical Conductivity and Total Dissolved Solids Meters to Field Test Water Quality. Available at <https://www.ag.ndsu.edu>. Accessed on 12th June, 2023.
- AOAC (2002a). AOAC Official Method 990.12, Aerobic Plate Counts in Food, Official Methods of Analysis of AOAC International, 17th edition, AOAC International, Gaithersburg, MD, United States. 2:9.

- AOAC (2000b). AOAC Official Method 997.02, Yeast and Mould Counts in Food, Official Methods of Analysis Of AOAC International, 17th edition, AOAC International, Gaithersburg, MD, United States, 2:7.
- 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(1)**: 1-9.
- Campos, C. F., Souza, P. E. A., Coelho, J. V. and Glória, M. B. A. (1996). Chemical composition, enzyme activity and effect of enzyme inactivation on flavor quality of green coconut water 1. *Journal of Food Processing and Preservation*, **20(6)**: 487-500.
- Cappelletti, M., Ferrentino, G., Endrizzi, I., Aprea, E., Betta, E. and Corollaro, M. (2015). High pressure carbon dioxide pasteurization of coconut water: A sport drink with high nutritional and sensory quality. *Journal of Food Engineering*, **145**: 73–81.
- Chattopadhyay, N., Samanta, M. K., Hore, J. K. and Alam, K. (2013). Evaluation of coconut cultivars for tender nut water. *Acta Horticulture*, **975**: 255-262.
- Chemat, F. and Khan, M. K. (2011). Applications of ultrasound in food technology: processing, preservation and extraction. *Ultrasonics sonochemistry*, **18(4)**: 813-835.
- Chuku, L. C. and Kalagbor, G. I. (2014). Protein and mineral element content of coconut (*Cocos nucifera*) water from different species. *American Journal of Advanced Drug Delivery*, **2(4)**: 451-453.
- De Gennaro, L., Cavella, S., Romano, R. and Masi, P. (1999). The use of ultrasound in food technology I: inactivation of peroxidase by thermosonication. *Journal of food engineering*, **39(4)**: 401-407.
- DebMandal, M. and Mandal, S. (2011). Coconut (*Cocos nucifera* L.): In health promotion and disease prevention. *Asian Pacific Journal of Tropical Medicine*, **4(3)**: 241-247.
- Dubois, M., Gilles, K. A., Hamilton, J. K., Rebers, P. T. and Smith, F. (1956). Colorimetric method for determination of sugars and related substances. *Analytical Chemistry*, **28(3)**: 350-356.

- Dwiloka, B., Rizqiati, H. and Setiani, B. E. (2020). Physicochemical and sensory characteristics of green coconut (*Cocos nucifera* L.) water kefir. *International Journal of Food Studies*, **9(2)**: 346-359.
- El-Sheikha, A. F., Ribeyre, F., Larroque, M., Reynes, M. and Montet, D. (2009). Quality of physalis (*Physalis pubescens* L.) juice packaged in glass bottles and flexible laminated packs during storage at 5°C. *African Journal of Food, Agriculture, Nutrition and Development*, **9(6)**.
- Ekasari, C. P. and Widyarti, S. (2019). The physicochemical properties comparison of the natural coconut water and the packaging coconut water. *IOP Conference Series: Earth and Environmental Science*, **391**.
- Fonteles, T. V., Costa, M. G. M., de Jesus, A. L. T., de Miranda, M. R. A., Fernandes, F. A. N. and Rodrigues, S. (2012). Power ultrasound processing of cantaloupe melon juice: Effects on quality parameters. *Food Research International*, **48(1)**: 41–48.
- Gómez-López, V. M., Buitrago, M. E. and Martínez-Yepez, A. (2018). Effect of ultrasonication on sensory and chemical stability of passion fruit juice during refrigerated storage. *Emirates Journal of Food and Agriculture*, **30**: 85–89.
- Hoe, T. K. (2018). The current scenario and development of the coconut industry. *The Planter*, **94(1108)**: 413- 426.
- IBEF, (2022). Coconut Industry and Exports 2021-2022. Available at <https://www/ibef.org/exports/coconut/industry/india>, India Brand Equity Foundation, Accessed on 11th January, 2023.
- Iqbal, A., Nadeem, M., Ainee, A., Ameer, K., Ather Nadeem, M., Sultan, M. and Siddeeg, A. (2022). The impact of ozonation on the physicochemical properties, antioxidant potential and shelf life of Kinnow (*Citrus Reticulata* Blanco) juice. *International Journal of Food Properties*, **25(1)**: 2551-2560.
- Jackson, J. C., Gordon, A., Wizzard, G., McCook, K. and Rolle, R. (2004). Changes in chemical composition of coconut (*Cocos nucifera* L.) water during maturation of the fruit. *Journal of the Science of Food and Agriculture*, **84**: 1049-1052.
- Jacob, A., Sudagar, I. P., Pandiselvam, R., Rajkumar, P. and Rajavel, M. (2023). Effect of packaging materials and storage temperature on the physicochemical and microbial properties of ultrasonicated mature coconut water during storage. *Food Control*, **149**:109693.

- Jaramillo-Sánchez, G. M., Garcia Loredó, A. B., Gómez, P. L. and Alzamora, S. M. (2017). Ozone processing of peach juice: Impact on physicochemical parameters, color, and viscosity. *Ozone: Science and Engineering*, **40(4)**: 305–312.
- Jaramillo Sanchez, G. M., Garcia Loredó, A. B., Contigiani, E. V., Gómez, P. L. and Alzamora, S. M. (2018). Inactivation kinetics of peroxidase and polyphenol oxidase in peach juice treated with gaseous ozone. *International Journal of Food Science & Technology*, **53(2)**: 347-355.
- Jean, W. H., Yong, L. G., Yan, F. N. and Swee, N. T. (2009). The chemical composition and biological properties of coconut (*Cocos nucifera* L.) water. *Molecules*, **14**: 5144-5164.
- Kailaku, S. I., Syah, A. N. A., Risfaheri, Setiawan, B. and Sulaeman, A. (2015). Carbohydrates electrolytes characteristics of coconut water from different varieties and its potential as natural isotonic drink. *International Journal on Advanced Science Engineering Information Technology*, **5**: 23-26.
- Kannangara, A. C., Chandrajith, V. G. G. and Ranaweera, K. K. D. S. (2018). Comparative analysis of coconut water in four different maturity stages. *Journal of Pharmacognosy and Phytochemistry*, **7(3)**: 1814-1817.
- Khadre, M. A., Yousef, A. E. and Kim, J. G. (2001). Microbiological aspects of ozone applications in food: a review. *Journal of Food Science*, **66(9)**: 1242-1252.
- Kim, J. G., Yousef, A. E. and Dave, S. (1999). Application of ozone for enhancing the microbiological safety and quality of foods: a review. *Journal of Food Protection*, **62(9)**: 1071-1087.
- Kwiatkowski, A., Clemente, E., Scarcelli, A. and Vida, J. B. (2008). Quality of coconut water ‘in nature’ belonging to Green Dwarf fruit variety in different stages of development, in plantation on the northwest area of Parana, Brazil. *Journal of Food, Agriculture and Environment*, **6(1)**: 102-105.
- Lopes, M. A. and Larkins, B. A. (1993). Endosperm origin, development, and function. *The Plant Cell*, **5(10)**: 1383.
- Lowry, O. H., Rosebrough, N. J., Farr, A. L. and Randall, R. J. (1951). Protein measurement with the Folin phenol reagent. *Journal of Biological Chemistry*, **193**: 265-275.

- Ma, Y., Xu, L., Wang, S., Xu, Z., Liao, X. and Cheng, Y. (2019). Comparison of the quality attributes of coconut waters by high-pressure processing and high-temperature short time during the refrigerated storage. *Food Science and Nutrition*, **7(4)**: 1512-1519.
- Maciel, M. I., Oliveira, S. L. and Da Silva, I. P. (1992). Effects of different storage conditions on preservation of coconut (*Cocos nucifera*) water. *Journal of Food Processing and Preservation*, **16(1)**: 13-22.
- Mahayothee, B., Koomyart, I., Khuwijitjaru, P., Siriwongwilaichat, P., Nagle, M. and Mullar, O. (2016). Phenolic compounds, antioxidant activity and medium chain fatty acids profiles of coconut water and meat at different maturity stages. *International Journal of Food Properties*, **19**: 2041-2051.
- Mahnot, N. K., Kalita, D., Mahanta, C. L. and Chaudhuri, M. K. (2014). Effect of additives on the quality of tender coconut water processed by nonthermal two stage microfiltration technique. *LWT-Food Science and Technology*, **59(2)**: 1191-1195.
- Mahnot, N. K., Gupta, K. and Mahanta, C. L. (2019). Shelf life enhancement and associated quality and sensory changes on refrigerated storage of tender coconut water subjected to non-thermal microfiltration and treated with additives. *Journal of Food Science and Technology*, **56(7)**: 3408–3421.
- Malick, C. P. and Singh, M. B. (1980). In *Plant Enzymology and Histoenzymology*. Kalyani Publishers, New Delhi, 286.
- Moyssiadi, T., Badeka, A., Kondyli, E., Vakirtzi, T., Savvaidis, I. and Kontominas, M. G. (2004). Effect of light transmittance and oxygen permeability of various packaging materials on keeping quality of low-fat pasteurized milk: Chemical and sensorial aspects. *International Dairy Journal*, **14(5)**: 429-436.
- Naik, M., Sunil, C. K. and Rawson, A. (2020). Tender coconut water: A review on recent advances in processing and preservation. *Food Reviews International*, **1**: 1–22.
- Nwafo, O. E. and Ikenebomeh, M. J. (2009). Effects of different packaging materials on microbiological, physio-chemical and organoleptic quality of zobo drink storage at room temperature. *African Journal of Biotechnology*, **8(12)**.

- O'Donnell, C. P., Tiwari, B. K., Bourke, P. and Cullen, P. J. (2010). Effect of ultrasonic processing on food enzymes of industrial importance. *Trends in Food Science and Technology*, **21**(7): 358-367.
- Oliveira, A. F. A., Mar, J. M., Santos, S. F., da Silva Júnior, J. L., Kluczkovski, A. M., Bakry, A. M. and Campelo, P. H. (2018). Non-thermal combined treatments in the processing of açai (*Euterpe oleracea*) juice. *Food Chemistry*, **265**: 57–63.
- Pandiselvam, R., Sunoj, S., Manikantan, M. R., Kothakota, A. and Hebbar, K. B. (2017). Application and kinetics of ozone in food preservation. *Ozone: Science and Engineering*, **39**(2): 115-126.
- Panase, V. G. and Sukhatme, P. V. (1985). Statistical methods for agricultural research. ICAR, New Delhi, **8**: 308-318.
- Patil, S., Valdramidis, V. P., Cullen, P. J., Frias, J. and Bourke, P. (2010). Ozone inactivation of acid stressed *Listeria monocytogenes* and *Listeria innocua* in orange juice using a bubble column. *Food Control*, **21**: 1723–1730.
- Patrick, J.W. and Offler, C.E. (2001). Compartmentation of transport and transfer events in developing seed. *Journal Experimental Botany*, **52**: 551-564.
- Perez, A. G., Sanz, C., Rios, J. J., Olias, R. and Olias, J. M. (1999). Effects of ozone treatment on postharvest strawberry quality. *Journal of Agricultural and Food Chemistry*, **47**: 1652–1656.
- Pérez-Vicente, A., Serrano, P., Abellán, P. and García-Viguera, C. (2004). Influence of packaging material on pomegranate juice colour and bioactive compounds, during storage. *Journal of the Science of Food and Agriculture*, **84**(7): 639-644.
- Pimentel, T. C., Madrona, G. S., Garcia, S. and Prudencio, S. H. (2015). Probiotic viability, physicochemical characteristics and acceptability during refrigerated storage of clarified apple juice supplemented with *Lactobacillus paracasei* ssp. *paracasei* and *Oligofructose* in different package type. *LWT-Food Science and Technology*, **63**(1): 415-422.
- Piyasena, P., Mohareb, E. and McKellar, R. C. (2003). Inactivation of microbes using ultrasound: a review. *International Journal of Food Microbiology*, **87**(3): 207-216.

- Porto, E., Alves Filho, E. G., Silva, L. M. A., Fonteles, T. V., do Nascimento, R. B. R., Fernandes, F. A. and Rodrigues, S. (2020). Ozone and plasma processing effect on green coconut water. *Food Research International*, **131**:109000.
- Prades, A., Dornier, M., Diop, N. and Pain J. P. (2012). Coconut water uses, composition and properties: A review. *Fruits*, **67(2)**: 87-107.
- Priya, S. R. and Ramaswamy L. (2014). Tender coconut water – nature's elixir to mankind. *International Journal of Recent Scientific Research*, **5(8)**: 1485-1490.
- Rajashri, K., Roopa, B. S., Negi, P. S. and Rastogi, N. K. (2020_a). Effect of ozone and ultrasound treatments on polyphenol content, browning enzyme activities, and shelf life of tender coconut water. *Journal of Food Processing and Preservation*, **44(3)**: 14363.
- Rajashri, K., Rastogi, N. K. and Negi, P. S. (2020). Non- thermal processing of tender coconut water - a review. *Food Reviews International*, **38**: 1–22.
- Ranganna, S. (2000). Handbook of Analysis and Quality Control for Fruits and Vegetables Products, (6th ed.), Tata McGraw-Hill Publishing Company Limited, New Delhi.
- Reddy, K. V., Das, M. and Das, S. K. (2007). Nonthermal sterilization of green coconut water for packaging. *Journal of Food Quality*, **30(4)**: 466-480.
- Revi, M., Badeka, A., Kontakos, S. and Kontominas, M. G. (2014). Effect of packaging material on enological parameters and volatile compounds of dry white wine. *Food Chemistry*, **152**: 331-339.
- Ribeiro, M. de M., Valdramidis, V. P., Nunes, C. A. and de Souza, V. R. (2017). Synergistic effect of thermosonication to reduce enzymatic activity in coconut water. *Innovative Food Science and Emerging Technologies*, **41**: 404-410.
- Rico, D., Martin-Diana, A. B., Frias, J. M., Gary, T. M. H. and Barry-Ryan, C. (2006). Effect of ozone and calcium lactate treatments on browning and texture properties of fresh-cut lettuce. *Journal of Science of Food and Agriculture*, **86**, 2179–2188.
- Rojas, M. L., Trevilin, J. H., dos Santos Funcia, E., Gut, J. A. W. and Augusto, P. E. D. (2016a). Using ultrasound technology for the inactivation and thermal sensitization of peroxidase in green coconut water. *Ultrasonics Sonochemistry*, **36**: 173-181.

- Rojas, M. L., Hellmeister Trevilin, J. and Duarte Augusto, P. E. (2016b). The ultrasound technology for modifying enzyme activity. *Scientia Agropecuaria*, **7(2)**: 145-150.
- Sandhya, V.G. and Rajamohan, T. (2008). Comparative evaluation of the hypolipidemic effect of coconut water and lovastatin in rats fed fat-cholesterol enriched diet. *Food Chem. Toxicol.* **45**: 3585-3592.
- Sanganamoni, S., Mahant, N. K. and Rao, S. P. (2018). Modeling of polyphenol oxidase and peroxidase inactivation in coconut water during thermal treatment. *International Journal of Chemical Studies*, **6(6)**: 1953-1958.
- Seow, C. C. and Gwee, C. N. (1997). Coconut milk, chemistry and technology. *International Journal of Food Science and Technology*, **32**: 189-201.
- Shenkin, A. (2006) The key role of micronutrients. *Clinical Nutritional*, **25**: 1-13.
- Soares, N. F. F. and Hotchkiss, J. H. (1999). Comparative effects of de-aeration and package permeability on ascorbic acid loss in refrigerated orange juice. *Packaging Technology and Science: An International Journal*, **12(3)**: 111-118.
- Sun, Y., Zhong, L., Cao, L., Lin, W. and Ye, X. (2015). Sonication inhibited browning but decreased polyphenols contents and antioxidant activity of fresh apple (*Malus pumila* Mill, cv. Red Fuji) juice. *Journal of Food Science and Technology*, **52(12)**: 8336–8342.
- Tan, T. C., Cheng, L. C., Bhat, R., Rusul G. and Easa A. M. (2014). Composition, physicochemical properties and thermal inactivation kinetics of polyphenol oxidase and peroxidase from coconut water obtain from immature, mature and overly mature coconut. *Food Chemistry*, **142**: 121-128.
- Tan, T. C. and Easa, A. M. (2021). The evolution of physicochemical and microbiological properties of green and mature coconut water (*Cocos nucifera*) under different storage conditions. *Journal of Food Measurement and Characterization*, **15(4)**: 3523-3530.
- Tazeen, H., Vardharaju, N. and Chandrasekar, V. (2016). Influence of ozonation on the some physicochemical properties of tender coconut water. *Advances in Life Sciences*, **5(10)**: 4153-4159.
- Tetra Pack. (2019). *The Coconut Handbook: Technology, engineering, Agriculture*. pp. 1-183. Tetra Pak International SA.

- Tiwari, B. K., Muthukumarappan, K., O' Donnell, C. P. and Cullen, P. J. (2008). Kinetics of freshly squeezed orange juice quality changes during ozone processing. *Journal of Agricultural and Food Chemistry*, **56(15)**: 6416-6422.
- Tiwari, B. K., O'Donnell, C. P., Brunton, N. P. and Cullen, P. J. (2009). Degradation kinetics of tomato juice quality parameters by ozonation. *International Journal of Food Science and Technology*, **44**: 1199– 1205.
- Torres, B., Tiwari, B. K., Patras, A., Wijngaard, H. H., Brunton, N., Cullen, P. J. and O'donnell, C. P. (2011). Effect of ozone processing on the colour, rheological properties and phenolic content of apple juice. *Food Chemistry*, **124(3)**: 721-726.
- Valero, M., Recrosio, N., Saura, D., Muñoz, N., Martí, N. and Lizama, V. (2007). Effects of ultrasonic treatments in orange juice processing. *Journal of Food Engineering*, **80(2)**: 509-516.
- Vigliar, R., Sdepanian, V. L. and Fagundes N. U. (2006). Biochemical profile of coconut water from coconut palms planted in an inland region. *Journal of Pediatrics (Rio J)*, **82**: 308-312.
- Vila, N. and Ampuero, O. (2007). The role of packaging in positioning an orange juice. *Journal of Food Products Marketing*, **13(3)**: 21-48.
- U.S. Food and Drug Administration (USFDA). 2001. Hazard analysis and critical control point (HACCP). Procedures for the safe and sanitary processing and importing of juice. *Federal Regulations*. **66**: 6137-202.
- Wu, J., Chen, H., Chen, W., Zhong, Q., Zhang, M. and Chen, W. (2021). Effect of ultrasonic treatment on the activity of sugar metabolism relative enzymes and quality of coconut water. *Ultrasonics Sonochemistry*, **79**: 105780.
- Zenker, M., Heinz, V. and Knorr, D. (2003). Application of ultrasound-assisted thermal processing for preservation and quality retention of liquid foods. *Journal of Food Protection*, **66(9)**: 1642-1649.
- Zou, Y. and Jiang, A. (2016). Effect of ultrasound treatment on quality and microbial load of carrot juice. *Food Science and Technology*, **36**: 111-115.