EXTENDED ABSTRACTS
of Research Presentations

03rd Annual Research Session

INSTITUTE OF FOOD SCIENCE & TECHNOLOGY SRI LANKA (IFSTSL)

05th August 2017, *Mihilaka Medura,* BMICH, Colombo

Extended Abstracts of the Research Presentations

FoodTechno 2017

Third Annual Research Session of the IFSTSL

05th August 2017

Mihilaka Medura, BMICH Colombo, Sri Lanka

Organized by the Institute of Food Science & Technology Sri Lanka (IFSTSL)

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Third Annual Research Session of the IFSTSL - 2017

(05th August 2017 at Mihilaka Medura, BMICH Colombo, from 8.30 a.m. to 4.30 p.m.)

PROGRAM

8.30 – 9.00 a.m.	Registration of participants
9.00 – 9.05 a.m.	Traditional lighting of the oil lamp
9.05 – 9.15 a.m.	Welcome address by Prof. K.K.D.S. Ranaweera *President/ IFSTSL*
9.15 – 9.40 a.m.	Keynote speech by Mr. Mario De Alwis *Managing Director/ Ma's Tropical Food Processing (Pvt) Ltd*
9.40 – 10.10 a.m.	***TEA***

TECHNICAL SESSION I (*Session Chair: Emeritus Prof. Upali Samarajeewa*)

10.10 – 10.35 a.m.	SAFETY ASSESSMENT AND ANTIBIOTIC SUSCEPTIBILITY OF POTENTIALLY PROBIOTIC YEASTS ISOLATED FROM DAIRY SOURCES OF SRI LANKA
10.35 – 11.00 a.m.	DETERMINING THE FUNCTIONAL PROPERTIES OF HYDROLYZED OVOMUCIN INCORPORATED FISH BURGER PRODUCED FROM CATLA (*Catla catla*)
11.00 – 11.25 a.m.	PRESERVATION OF WHITE COCONUT KERNEL FOR THE PRODUCTION OF DESICCATED COCONUT (DC) IN SRI LANKA
11.25 – 11.50 a.m.	EFFICACY OF ULTRAVIOLET RADIATION AS A NON-THERMAL TECHNIQUE FOR REDUCTION OF MICROORGANISMS IN WATER MELON (*Citrullus lanatus*) JUICE
11.50 – 1.00 p.m.	***LUNCH***

TECHNICAL SESSION II (*Session Chair: Prof. K.K.D.S. Ranaweera*)

1.00 – 1.25 p.m.	BIOWAX COATING IMPROVES QUALITY AND EXTENDS STORAGE LIFE OF LIME FRUITS (*Citrus aurantifolia* L.)

1.25 – 1.50 p.m.	TRANS-FAT FORMATION AND CHANGES IN FATTY ACID PROFILE OF SOY AND COCONUT OIL BLENDS WITH REPEATED FRYING
1.50 – 2.15 p.m.	DEVELOPMENT AND QUALITY EVALUATION OF RICE (*Oryza sativa*) FLAKES INCORPORATED GLUTEN FREE COOKIES FROM MAIZE (*Zea mays* L.) AND SOY BEAN (*Glycine max*) FLOUR BLENDS
2.15 – 2.30 p.m.	Presentation by the gold sponsor
2.30 – 3.00 p.m.	***TEA***

TECHNICAL SESSION III (*Session Chair: Dr. B.E.P. Mendis)*

3.00 – 3.25 p.m.	STUDYING FACTORS AFFECTING WHITE PRECIPITATE FORMED IN ICED TEA WHEN RECONSTITUTED WITH HARD WATER AND FINDING SOLUTIONS
3.25 – 3.50 p.m.	COMPARISON OF FUNCTIONAL PROPERTIES OF STARCHES AVAILABLE IN SRI LANKA
3.50 – 4.15 p.m.	DETERMINATION OF PHYSICAL, CHEMICAL & SENSORY CHARACTERISTICS OF NUWARA ELIYA REGIONAL TEA TO EVALUATE THE QUALITY OF BLACK TEA
4.15 – 4.25 p.m.	Award of certificates
4.25 – 4.35 p.m.	Vote of thanks by Dr. Niranjan Rajapakse/ Coordinator FoodTechno 2017

Message from the President of the IFSTSL

I am privileged and honoured to send this message on behalf of the IFSTSL, which has emerged as a premier center for promoting and advancing the Food Science and Technology knowledge and its applications across the country, where it is needed. The theme of the 3rd Annual Research Session (FoodTechno 2017) is Innovation to Application. Nowadays, the term "innovation" is increasingly used in all science fields. Breakthrough innovation comes slowly and cautiously to the food and beverage industry.

As we know, learning and innovation go hand in hand. The arrogance of success is to think and see that what we did yesterday will be sufficient for tomorrow. This year, the IFSTSL has initiated several very important events such as Sri Lanka Food BIZ Excellence 2017 which will be jointly organized with Sri Lanka Food Processors Association (SLFPA) and Inter-University Food Science Quiz programme 2017 intended for University Undergraduates. The IFSTSL with the help of its membership will take lead in forums where food safety, consumer welfare and the like will be in the focus.

We are extremely proud of our members' devotion and dedication, which enabled the Institute to achieve its prime objectives and most importantly to be a Nationally important body to make the local food sector unique in the region, and because we can and must do better. I wish the 3rd Annual Research Session great success and all the very best for continued progress. The Sri Lankan Food Industry is a complex and diverse trading culture providing goods and services to consumers of our country and beyond. The radical technological changes, innovations and societal upheavals have left their marks also on our food and eating habits. I can visualize that the IFSTSL growing in pursuit of higher standards of research, training, dissemination of knowledge etc. I would like to take this opportunity to thank all those who initiated the programmes of this nature and those who are presently involved in achieving IFSTSL objectives.

Albert Einstein once said "Creativity is contagious. Pass it on". There is no doubt that innovation is the most important human resource of all. Without innovations, there would be no progress, and we would be forever repeating the same patterns. "Millions of rupees worth of research knowledge lies dormant at different Institutions waiting for the right disruptor to come along and create a business." Therefore, 3rd Annual Research Session can certainly be a good forum for prospective food manufacturers to take this knowledge and materialize into business. My blessings and good wishes will always be with the IFSTSL and its crew in making this Research Session a success.

Prof. K.K.D.S. Ranaweera
President of the IFSTSL

Message from the Coordinator of FoodTechno 2017

On behalf of the organizing committee, it is my pleasure to welcome you all to the "FoodTechno 2017: Innovation to application" organized by the Institute of Food Science and Technology, Sri Lanka (IFSTSL). The mandate of IFSTSL clearly identifies its leading role in promoting the linkages among different stakeholder groups of the Sri Lankan food sector for scientific and technical support. Being an independent body, IFSTSL has succeeded in this role over the years through several activities organized such as seminars, workshops, research sessions and training programs and among other stakeholder groups, the working force of the food industry has become one of the main beneficiaries of these activities. This research session is one such activity commenced in 2015 as an annual activity of the institute.

Research studies related to food science and technology are being conducted by several groups including universities, research institutes and other line agencies. The findings are disseminated among scientific communities mainly during research fora. Parallel to this, a line of publications are appearing in scientific research journals. Neither these research fora, nor the scientific publications effectively disseminate the knowledge ascertained by these researches to the food industry, where the highest need exists for food science and technology related scientific knowledge. The main objective of the research session is to provide a common platform for the researchers from Universities and other research institutes in Sri Lanka, to develop a dialogue with the food industry aiming to transfer the research knowledge they gathered from scientific research. It is expected that this scientific research findings could be applied into industry for its improvements. . Research papers presented in the session were chosen from research carried out in National Universities and other research institutes in Sri Lanka.

I wish to extend my gratitude to those contributed in making this research session a success. To our generous sponsors, for their financial support, and the executive committee of the IFSTSL, for their encouragement throughout. A special word of appreciation goes to executive committee members of the Sri Lankan Food Processors Association for always having been there with every possible support. Last but not least, the authors and attendees, for continuing to contribute to, and believing in the success of FoodTechno over the years.

I wish this annual research session all success.

Dr. Niranjan Rajapakse
Coordinator/ FoodTechno 2017

Extended Abstracts of the Research Presentations

FoodTechno 2017- *Third Annual Research Session of the IFSTSL*

(05th August 2017 at Mihilaka Medura, BMICH Colombo, from 8.30 a.m. to 4.30 p.m.)

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SAFETY ASSESSMENT AND ANTIBIOTIC SUSCEPTIBILITY OF POTENTIALLY PROBIOTIC YEASTS ISOLATED FROM DAIRY SOURCES OF SRI LANKA

Rajawardana D.U. [*], Hewajulige I.G.N., Nanayakkara C.M. [1] and Nilukshi1 D.A.V.
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Summary

One of the most recent and significant non-process uses of yeasts in food industry is as probiotics, despite the fact that probiotic potential of yeasts are not well explored yet. This study was carried out to evaluate safety and antibiotic susceptibility of yeasts previously confirmed to possess strong probiotic potentials, for future food applications. Yeasts (28 isolates) belonging to 12 species isolated from dairy sources of Sri Lanka were studied for their DNase and gelatinase production, haemolytic activities and antibiotic susceptibility patterns. None of the studied isolates produced undesirable gelatinase or DNase enzymes. *Candida rugosa* DBMY32, *Candida pararugosa* M172B, *Candida orthopsilosis* Co 90-125, *Kluyveromyces thermotolerans* NRRL Y-8284 and *Pichia kudriavzevii* strain NRRL Y-5396 produced green or clear zones around colonies on blood agar plates (α- or β haemolytic) making them undesirable for food applications. The yeasts were resistant to all tested antibiotics even though most probiotic bacteria reported n literature are susceptible to those antibiotics. Most yeast possess a natural resistance against antibiotics and can be used for patients undergoing antibiotic treatments. The above results suggest that the isolates can be used for the development of yeast based probiotic food and/or starter cultures, though need further in vivo evaluations.

Keywords: Yeast, Probiotic, Antibiotic susceptibility, Safety attributes, Haemolysis

Introduction

The yeasts constitute a large and diverse group of microorganisms that are attracting increased attention from the science and industry. In addition to their role in the food processing industries, yeasts play various roles in livestock feeding, veterinary practices, in medicine, biomedical and pharmaceutical industries. Among yeasts *Saccharomyces boulardi* is successfully used as a commercial probiotic by the food and pharmaceutical industries for human consumption. It reduces the symptoms of acute diarrheal infection, prevents re-infection of *Clostridium difficile*, and lowers the inflammatory response upon infection. Therefore, if new strains of probiotic yeasts were isolated and identified, that may be very beneficial and also attractive exploration for food, feed, as well as pharmaceutical industries. Yeasts are more tolerant to gastro-intestinal (GI) conditions compared to probiotic bacteria and also resistant to most

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antibiotics; hence carry the potential to normalize GI conditions of patients undergoing antibiotic treatments. The main objective of this study was to evaluate the safety of probiotic yeasts isolated from Sri Lankan dairy sources, and to test their antibiotic susceptibilities for future food applications.

Methodology

Maximum possible numbers of yeasts from raw milk and curd were isolated by pour and spread plate techniques using yeast-peptone-dextrose-agar (YPDA) supplemented with 0.1 gl-1 chloramphenicol. Morphological and biochemical characterization of the isolates were carried out based on the methods explained by Nahvi I and Moeini H. (2004). Selected isolates were subjected to standard GI tolerance tests (pH, bile, temperature, gastric enzymes, NaCl and phenol following the methods explained by Aswathy *et al.* (2008). Genomic DNA was extracted from the best 28 isolates, selected region of 18S rRNA gene were PCR amplified with universal primers [primers used to amplify the ribosomal DNA internal transcribed spacer region were ITS1 (GTAGGTGAACCTGCGG) and ITS4 (TCCGCTTATTGATATGC)] and amplified products were subjected to DNA sequencing at Macrogen-South Korea. Antibiotic susceptibility pattern of the isolates were determined for a total of 12 different antibiotics (Oxoid, UK) namely Amoxycillin, Ampicillin, Erythromycin, Amikacin, Norfloxacin, Sulphamethoxazole, Chloramphenicol, Cephalothin, Cefotaxime, Gentamicin,Vancomycin and Tetracycline. Gelatinase production by selected yeast isolates was studied by using tryptone-neopeptone-dextrose (TND) agar containing 0.4% gelatine (Gupta and Malik, 2007). The selected yeast isolates were streaked on DNase agar medium to check for DNase production (Gupta and Malik, 2007). Haemolytic activity was investigated as described by Gerhardt et al. (1981) by streaking into sterile blood agar plates and were examined for signs of α, β and γ haemolysis.

Results and Discussion

According to FAO/ WHO, (2002) every probiotic strain needs to be assessed for safety to be used as a food or feed supplement. Yeast isolates (80) which were morphologically and phenotypically identified as belonging to the generas of Kluyveromyces, Pichia and Candida, were subjected to in-vitro GI tolerance tests. Twenty eight isolates that survived under simulated GI conditions were selected for further investigations. Genotypic identifications confirmed that the isolates were different strains of Kluyveromyces, Pichia and Candida confirming the results obtained for phenotypic characterizations. The identified potentially probiotic yeast types tested for their safety attributes are listed in Table 1.

Development of clear zones around the spots against the opaque background indicated a positive reaction for Gelatinase and DNase tests (Gupta and Malik, 2007) as shown in Figure 3. DNA Hydrolysis test or Deoxyribonuclease (DNase) test is used to determine the ability of an organism to produce deoxyribonuclease or DNase enzyme and hydrolyze DNA and utilize it as a source of carbon and energy for growth. A microorganism should not produce gelatinase or DNase enzymes so as to be used as a probiotic in food and feed supplements'. None of the strains studied in this study

produced those two enzymes (Gelatinase and DNase are mostly produced by pathogenic microorganisms) therefore could consider as potentially safe yeast types.

Table 1: Potentially probiotic yeast species isolated from dairy sources of Sri Lanka.

Yeast type	**Presence (%)**
Candida tropicalis voucher	3.70
Candida metapsilosis	3.70
Candida rugosa	3.70
Candida tropicalis	3.70
Candida pararugosa	3.70
Candida orthopsilosis	14.81
Candida versatilis	3.70
Kluyveromyces marxianus	11.11
Pichia kudriavzevii	40.74
Pichia sp. AQGWD 7	3.70
Pichia sp.	3.70
Kluyveromyces thermotolerans	3.70

However, Candida rugosa strain DBMY32 and Candida pararugosa strain M172B as well as Candida orthopsilosis Co 90-125 produced clear zones around the colonies (Figure 1) on blood agar plates (β-haemolytic). Kluyveromyces thermotolerans NRRL Y-8284 and Pichia kudriavzevii strain NRRL Y-5396 produced green zones (Figure 2) around colonies (α-haemolytic). Rest of the isolates did not produce clear zones around colonies (γ-haemolytic) on human blood agar plates (Hargrove and Alford, 1978). Only the γ-haemolytic isolates could be considered as safe for human consumption.

The yeast strains screened for antibiotic tolerance test (ABST) survived in the presence of all of the antibiotics tested as no zone of inhibition was observed around the discs (Amoxycillin, Ampicillin, Erythromycin, Amikacin, Norfloxacin, Sulphamethoxazole, Chloramphenicol, Cephalothin, Cefotaxime, Gentamicin,Vancomycin, Tetracycline). The suggested reason for this resistance is due to physiological or structural peculiarities of the strains such as cell wall characteristics, lack of antibiotic binding sites or antibiotic target function, *etc* in bacteria. According to Kim *et al.* (2004) most of the probiotic microorganisms under discussion are bacteria and many of them are generally susceptible to all of these antibiotics. Since yeasts have a natural resistance against these antibiotics, if probiotic yeasts which are safe for human consumption are found, they could be used for patients undergoing antibiotic treatments for prolonged periods to re-establish the colonic health (Nayak, 2011).

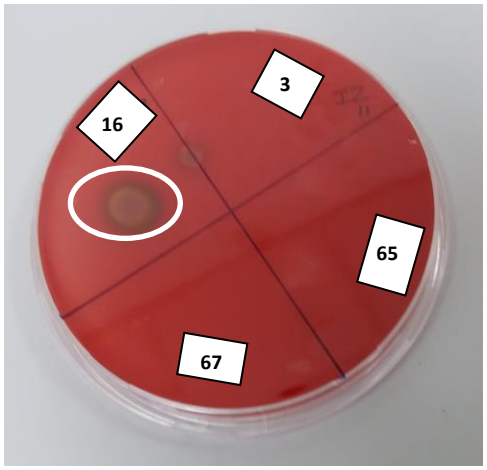

Figure 1: Signs of β-haemolysis

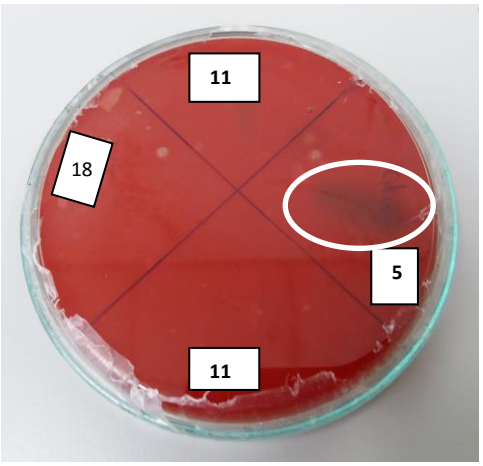

Figure 2: Signs of α-haemolysis

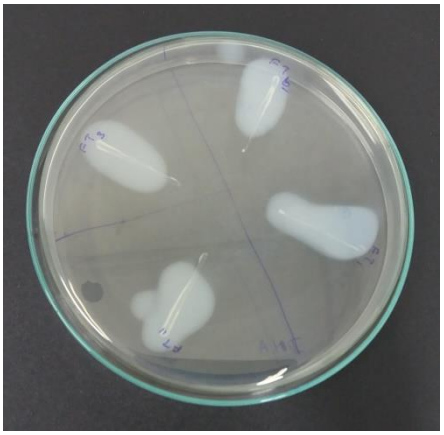

Figure 3: Signs of negative DNase tests

Conclusion

The raw cows' milk and curd are rich sources for the isolation of beneficial yeasts with potential probiotic activities. Majority of the tested isolates proved to be safe for food applications in-vitro, except for few yeast strains. Further in-vivo evaluations are needed for use of them directly or human consumption.

Acknowledgement

Financial support from the NSF Grant (RG/2016/AG/02) is gratefully acknowledged.

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DETERMINING THE FUNCTIONAL PROPERTIES OF HYDROLYZED OVOMUCIN INCORPORATED FISH BURGER PRODUCED FROM CATLA (*Catla catla*)

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Summary

Fish burger is a popular fast food, which contains higher amount of essential proteins, lipids, vitamins, and minerals. Ovomucin is one of proteins the in egg white and can be used to produce bioactive peptides with several functional properties, which can be used in food processing industry. Hydrolysates of ovomucin showed many functional properties once incorporated in to fish burger. The results conclude that hydrolyzed ovomucin can be a good ingredient for fish burger production as it has many functional activities with ability to increase the keeping quality of food through inhibiting microbial activity by chelating the metal ions, and lipid oxidation.

Keywords: Fish burger, Hydrolyzed ovomucin, Antimicrobial, Antioxidant, Metal chelating

Introduction

Fish burgers are popular fast food products, and one of the secondary minced fish based products distributed as frozen form. In addition, it is considered as comminuted raw meat product made from fish flesh. Fish and fisheries products contain high quality protein and other nutrients vitamins and minerals. Fish burger is more susceptible to microbial spoilage and reduces keeping quality mainly due to lipid oxidation. Hydrolysates of ovomucin have strong antioxidant effects by radical scavenging activities which reduce the oxidation by 85% and more than 90% of the antioxidant activity was retained even after 24 hours (Chang *et al*., 2013). In addition, Peptides derived from ovomucin with high iron-chelating activity can inhibit oxidation and microbial growth (Hiidenhovi, 2007). Therefore; hydrolysates have the capability of controlling bacteria (Omana *et al*., 2010). Also with high iron-chelating activity, microbial growth can be reduced (Ko *et al*., 2009). Thus, hydrolysates of ovomucin can be used as antimicrobial agent, antioxidant agent and metal chelating agent in food processing industry (Kobayashi *et al*., 2004). The objective of this study was to determine the functional properties of hydrolysates of ovomucin once it is incorporated in to fish burger.

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Methodology

Ovomucin (20 mg/ml) was hydrolyzed with 1% pepsin (1:100) for 0 hours (0 h incubation refers to immediately after the addition of enzyme) and followed with heat inactivation at 100 ^{o}C for 15 min. Fish burgers were produced using 125 ppm of hydrolysates (Tr1), 125 ppm of ovomucin (Tr2) with incorporating all the other ingredients as fish (64%), bread crumble (17%), ice (12%), vegetable oil (4%), salt (2%), chili (0.5%), black pepper (600 ppm), MSG (10 ppm), cardamom (7 ppm), sugar (200 ppm), milk powder (10 ppm). Samples were cooked at 80 ^{o}C to a core temperature of 72 ^{o}C and cooled in cold water to 20– 25 ^{o}C. All samples were sealed in polyethylene and stored in a freezer (-18 ^{o}C) for 21 days for analysis.

Determining the functional properties

Fish burger samples of each treatment were ground with distilled water and were incubated at 37 ^{o}C for 16 hours. One milliliter of incubated sample was transferred and added with 2 ml of thiobarbituric acid/trichloroacetic acid solution (20 mM TBA/15% TCA) and 50 µl of 10% butylated hydroxyanisole in 90% ethanol, and then vortex-mixed. Properly vortex-mixed samples were incubated in a 90 ^{o}C water bath for 15 min to develop color. The samples were cooled in an ice bath for 10 min and centrifuged at 3000 rpm for 15 minutes at 5 ^{o}C. The absorbance of the solution was measured at 532 nm compared to a blank prepared with 01 ml of distilled water and 02 ml TBA/TCA solution. The amounts of TBARS were expressed as milligram of Malondialdehyde (MDA) per kilogram.

Ferrozine method (Carter, 1971) with some modifications was used to measure the Fe-chelating activity. Absorbance of the solution was measured at 532 nm and activity was measured using the following equation. (Fe^{2+} chelating activity % = {1- (sample absorbance/ blank absorbance)} * 100).

Method of (Kong and Xiong, 2006) with some modifications was used to measure the Cu^{2+} chelating activity. The absorbance of the samples was measured at 632 nm and activity was measured using the following equation. (Cu^{2+} chelating activity % = {1-(sample absorbance/ blank absorbance)} * 100).

Inhibition of locally isolated Escherichia coli was tested after culturing in Macconky agar and inhibition of locally isolated Salmonella was checked using Eosin methylene blue agar in burger samples on 0, 3, 6 and 9 day of storage to detect antimicrobial activity.

Data were analyzed with MINITAB 16.0 statistical software. Differences between means were determined by the least significant difference test ($P<0.05$).

Results and Discussion

Oxidation assessment

Conferring the results of antioxidant activity in fish burger, the oxidation values in two treatments are significantly different from each other ($p<0.005$). Considering the mean comparison, lowest oxidation rate was consisted with treatment 1 (ovomucin 125 ppm). Ovomucin hydrolysates have two peptides, with strong antioxidant activities (Chiang *et al.*, 2006). Hydrolysates produced by heating at 100 ^{O}C for 15 min under pH 12.00 had

a strong antioxidant activity (Abeyrathne *et al*., 2016). However, the oxidation value increased dramatically on day 05 and decreased afterward. Malonaldehyde, is capable of cross-linking with amino acids to form amidine linkages and may also interact with other components of fish such as nucleosides, nucleic acids, amino acids of phospholipids and other aldehydes, which are end products of lipid oxidation. This could help to explain the decrease in levels of TBARS with time (Odote and Obiero, 2009). Furthermore, decrease in the levels of TBARS after an increase is probably due to the carbonyls, which are unstable and react easily with other compounds. It is also known that oxygen accessibility, degree of tissue disruption, and storage temperatures can affect shelf life and are important in rancidity (Odote and Obiero, 2009).

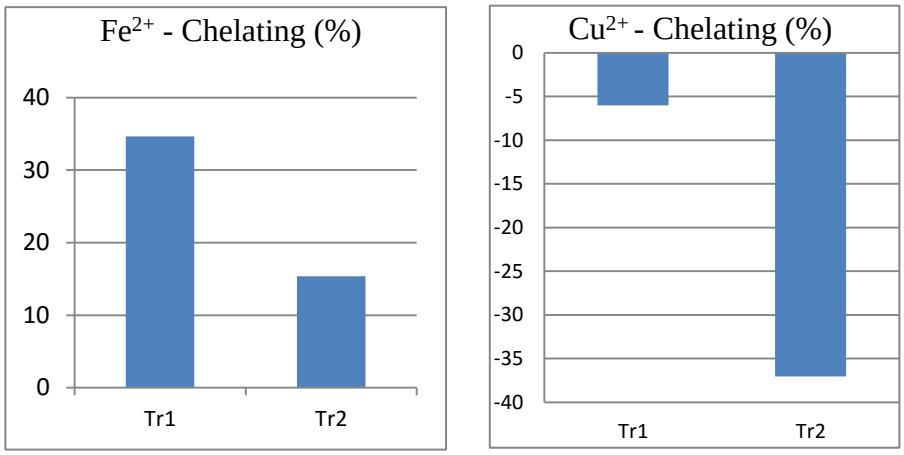

Figure 1. Graphical expression of $Fe^{2+}$and Cu^{2+}chelating activity of treatments.

High iron chelating activity was observed in treatment one and significant difference was seen among treatments ($p<0.05$). Ovomucin hydrolyzed by heating at 100 ^{O}C for 15 min under pH 12 showed a high iron-chelating activity (Abeyrathne *et al*., 2016). However Cu^{2+} chelating activity of the treatments was totally different from that of the iron-chelating activity. It was observed that the levels of Cu^{2+} ions in samples were higher in both treatments, indicating that cupper ions were not chelated. Peptides produced from ovomucin have no chelating but releasing activities (Abeyrathne *et al*., 2016). Therefore, hydrolysates from ovomucin are not suitable as copper chelating agents.

All samples were negative for both microbes throughout the study. (Hiidenhovi, 2007) showed that peptides with high iron-chelating activity can prevent oxidation and microbial growth. It was observed that ovomucin has the capability of controlling bacteria such as *E. coli*, *Bacillus dysentericus*, and *Vibrio bacterium* (Omana *et al*., 2010; Ko *et al*., 2009)

Conclusions

Research revealed that hydrolysates of ovomucin can be used in food industry to prevent oxidative changes and microbial spoilage in food, by improving the functional properties in food while enhancing the nutritional content.

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PRESERVATION OF WHITE COCONUT KERNEL FOR THE PRODUCTION OF DESICCATED COCONUT (DC) IN SRI LANKA

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Summary

Study of storage stability in fresh coconut kernel for the production of desiccated coconut is vitally important. This research was conducted to preserve the fresh coconut kernel by application of hurdle technology. Two treatments were applied; treatment 1: a mixture of 0.3% citric acid and 0.02% ascorbic acid and the treatment 2: a mixture of 0.3% citric acid, 0.02% ascorbic acid, 3% sodium chloride, 0.1% tri sodium citrate and 0.05% sodium benzoate. Control samples were evaluated for physico-chemical parameters at every 2 h intervals until 18 h under ambient temperature conditions. Treated kernels were evaluated at 6 h intervals until 48 h. The Color (L), moisture (% db), total soluble solids (^{o}Brix), pH, total sugar (%), fat (%), free fatty acids (%), peroxide value (meq/kg), total plate count (CFU/g) were analyzed as physico-chemical parameters. It was found that there was a significant difference ($P<0.001$) between two treatments and storage time for all tested physico-chemical properties. Sensory evaluation was conducted to assure the quality of final desiccated product further. It was found that there is no significant difference ($P<0.05$) in all sensory properties. It was concluded that the kernels with treatment 2 can be kept for 24 h of storage under ambient temperature conditions, without noticeable changes in quality and sensory attributes.

Keywords: Coconut kernel, Desiccated coconut, Hurdle technology, Physico-chemical properties.

Introduction

The desiccated coconut (DC) is a dried white shredded product manufactured from grated coconut kernel under hygienic processing conditions. Desiccated coconut industry is very popular and having a higher demand across the world. In industries, fresh coconut kernels are cut in tons per day and feed the surge tank to be desiccated. In any case of technical issue or any other delay in production process, fresh kernels should be held in the surge tank without having any quality defects. This has being a real challenge for coconut processing industries. Identification of an appropriate preservation technique for fresh kernels has become an essential need in coconut industry.

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Several research studies have been conducted based on the storage stability of fresh kernels and numerous physico-chemical properties have been checked to determine the quality and shelf life of the kernels (Jangchud, 2007, Appaiah, 2015). According to Gunathilake, 2010 and Obibuzor, 2015 shelf life of fresh kernels is about 8-10 hr of period under room temperature conditions. Though there are ample amount of techniques available to preserve fresh kernels, most of them are not applicable in the production lines due to driers operating at high temperatures. Therefore, chemical preservation is identified as the most reliable technique to preserve kernels.

This research study is an approach to extend the shelf life of fresh coconut kernel using chemical preservation technique, without affecting its organoleptic properties. In this study two different combinations of hurdle technology were used and checked whether they are significantly different or not. Findings of this study will be useful for coconut processing industries in reduction of wastage and secure the possible monetary losses.

Methodology

Fully matured, de-shelled de-paired and chlorinated (1 ppm) fresh coconut kernels were randomly collected from the surge tank without considering varietal variations. Behavior of physiochemical parameters of fresh coconut kernels were checked under ambient temperature (30 ^{o}C±2) conditions with and without treatments (food grade chemicals). Each analysis was performed in triplicates. Treatment 01: 0.3 % citric acid + 0.002 % ascorbic acid and Treatment 02: 0.3 % citric acid+ 0.002 % ascorbic acid+ 3 % NaCl + 0.1 % tri sodium citrate + 0.05 % sodium benzoate.

Fresh kernels were cut into small cubes and kept in ambient temperature (30 ^{o}C±2), sealed with 2 h interval until 18 h of period for the control samples and with 6 h interval until 48 h of period for treated samples. All sealed samples were stored in freezer for the analysis of physico-chemical properties.

The moisture content was measured on dry basis (AOCS method no. Ac 2–41 1997). Total sugar was measured using method AOCS, 1998. Fat was extracted from fresh kernel according to AOCS Official Butt-tube method Ac3–44 (AOCS 1998). The indicator method specified by ISO 965.33 in 1997 as outlined by AOCS (1984-1997) was used in determination of peroxide value (PV).

Color of the finely ground fresh coconut kernels was measured using colorimeter (CM 350d, Tokyo, Japan) with L, a, b values. pH of the coconut milk which is extracted from the fresh coconut kernels was measured using pH meter (Orion, EA 920, Boston) at 30±2 ^{o}C. TSS was measured using the brix meter (Atago, N1-E, Japan) and Total Plate Count was checked according to Sri Lanka Standard 516: part 1:1991.

Sensory properties of desiccated coconut samples were subjected to paired comparison to check the compatibility of its sensory properties with control sample. Thirty trained panelists were used to assess the color, aroma, texture (crispiness) and flavor of preserved desiccated coconut.

Results and Discussion

Variations of Physiochemical properties and sensory properties were analyzed in fresh coconut kernels over the time with different treatments. In here two treatments were used including control sample and in treatment one; 0.3 % citric acid as an acidulant, 0.02 % ascorbic acid as an anti-oxidant were used. In treatment two; 0.3 % citric acid as an acidulant, 0.002 % ascorbic acid as an anti-oxidant, 3 % sodium chloride as a humectant, 0.1 % tri sodium citrate as a buffer and 0.05 % sodium benzoate as an anti-microbial agent were used.

Shelf life of the fresh coconut kernel was determined by using specification (USDA national nutrient database, 2004) for each physiochemical parameter. In addition to basic physico-chemical parameters, sensory properties play a vital role in consumer perspectives. Therefore, being acceptable in sensory attributes also an essential requirement for a particular product.Variations of each physico-chemical parameters of fresh coconut kernels over the time was observed by this research study and following figure 1.1 shows the variation pattern of each physico-chemical parameters with time.

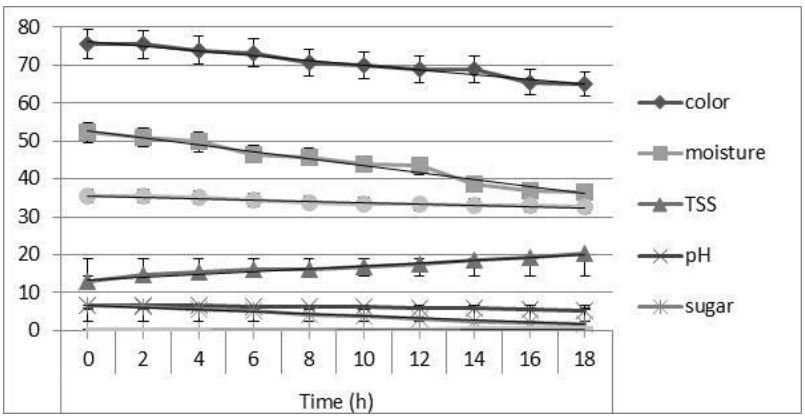

Figure 1.1: Variation pattern of each physico-chemical parameters with time in fresh coconut kernels.

Effect of treatments in preservation of quality attributes over 48 h period of time were checked and following tables 1.1, 1.2 show the mean observations of each physico-chemical parameters with regard of treatments.

Table 1.1: Mean comparisons of physico-chemical parameters based on treatments in coconut kernels.

	Color ("L")	Moisture%	Fat%	Total sugars%	pH
Control	70.36±0.56^{A}	48.91±0.85^{A}	33.94±0.65^{A}	4.12±0.23^{A}	6.3±0.19±^{A}
Treatment 1	71.05±0.40^{A}	46.95±1.25^{B}	34.72±0.18^{B}	4.59±0.36^{B}	4.8±0.15±^{B}
Treatment 2	74.52±0.82^{C}	45.08±0.66^{C}	35.30±0.13^{C}	5.68±0.15^{C}	4.4±0.23±^{C}

Table 1.2: Mean comparisons of FFA, PV, TSS and TPC based on treatments in coconut kernels.

Treatments	FFA%	PV (meq/1000g)	TSS%	TPC ($\times 10^4$)
Control	0.08 ± 0.001^{A}	0.50 ± 0.02^{A}	16.90 ± 0.1^{A}	102.33 ± 8.6^{A}
Treatment 1	0.08 ± 0.003^{A}	0.28 ± 0.05^{B}	13.25 ± 1.5^{B}	83.16 ± 6.4^{B}
Treatment 2	0.04 ± 0.001^{B}	0.26 ± 0.06^{C}	10.62 ± 1.8^{C}	27.41 ± 7.5^{C}

According to above table 1.1 all tested physico-chemical parameters showed significant difference ($P<0.05$) in all treatments. The "Treatment 2" showed a better performance in respect of preservation of kernels. It was able to maintain the standard levels (USDA specification, 2004) of each physico-chemical properties of fresh coconut kernels over time.

As well as the treatment effect, effect of storage time also plays a vital role in preservation of physico-chemical properties in coconut kernels. Following table 1.3 and 1.4 show the mean comparisons of all tested physico-chemical properties in coconut kernels based on storage time.

Table 1.3: Mean comparison of color, moisture, fat, total sugars and pH in coconut kernels based on storage period.

*Time (h)	Color (L)	Moisture%	Fat%	Total sugars%	pH
0	75.50 ± 0.26^{A}	52.46 ± 1.31^{A}	35.56 ± 1.20^{A}	6.61 ± 0.90^{A}	5.5 ± 0.06^{A}
6	72.36 ± 0.40^{A}	48.17 ± 1.20^{B}	35.13 ± 1.24^{B}	5.27 ± 0.56^{B}	5.3 ± 0.03^{B}
12	71.18 ± 0.21^{B}	45.15 ± 0.80^{C}	34.41 ± 0.56^{C}	4.35 ± 0.84^{C}	5.01 ± 0.01^{C}
18	67.86 ± 2.10^{B}	42.14 ± 1.46^{D}	33.52 ± 0.80^{D}	2.95 ± 1.3^{D}	4.94 ± 0.02^{D}

* At ambient temperature conditions during 18 h of tested period

Table 1.4: Effect of time in preservation of coconut kernel based on FFA, PV, TSS, and TPC.

*Time (h)	FFA%	PV (meq/1000g)	TSS%	TPC (10^4)
0	0.01 ± 0.002^{A}	0.20 ± 0.03^{A}	12.13 ± 0.02^{A}	3.3 ± 1.0^{A}
6	0.04 ± 0.004^{B}	0.28 ± 0.01^{B}	13.10 ± 0.56^{B}	60.0 ± 4.3^{B}
12	0.09 ± 0.001^{C}	0.38 ± 0.06^{C}	13.94 ± 0.85^{C}	106.6 ± 8.1^{C}
18	0.12 ± 0.005^{D}	0.53 ± 0.07^{D}	15.26 ± 0.95^{D}	137.8 ± 9.2^{D}

*At ambient temperature conditions during 18 h of tested period.

According to above tables 1.3 and 1.4 all tested physico-chemical properties in each storage periods were significantly different ($P<0.05$) and it was found that the 24 h sample treated with "Treatment 02" has shown the maximum shelf life which is in the specific range of each pysico-chemical parameters.
In physico-chemical analysis it is found that the "Treatment 02" is the best method in preserving fresh kernels and the maximum shelf life of treated samples is 24 h of period. DC samples which were preserved for 24 h using "Treatment 02" was checked for sensory properties.
Sensory evaluation indicated that there is no significant difference between ($P<0.05$) color, aroma, texture and flavor of treated samples and those attributes of control samples. Therefore, it was concluded that the 24 h sample of "Treatment 02" is acceptable in all sensory attributes.

Conclusions

Physical, chemical and microbiological parameters were greatly influenced by treatments which were applied in preservation of fresh coconut kernel. The stability of fresh coconut kernel can be maintained up to 24-30 h of period under ambient temperature conditions with application of hurdle technology without disturbing the sensory attributes.

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EFFICACY OF ULTRAVIOLET RADIATION AS A NON-THERMAL TECHNIQUE FOR REDUCTION OF MICROORGANISMS IN WATER MELON (*Citrullus lanatus*) JUICE

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Summary

The efficacy of UV-C treatment as a non-thermal technique on the reduction of microorganisms in watermelon juice was evaluated. A pilot scale UV-C sterilizer (40W) was used to apply UV-C dosages from 1.59 Jcm^{-2} to 57.18 Jcm^{-2} at two flow rates (0.1 m3/h) to well water, potable water and watermelon juice. The maximum microbial reduction of watermelon juice was obtained at the highest UV dose 57.18 Jcm-2. No significant alteration ($p > 0.05$) was observed in titratable acidity, brix, total phenolic compounds and antioxidant capacity. UV technology could be an alternate instead of pasteurization. Studies are in progress using a range of juices to test the effect of particle sizes, suspended and soluble solids indicating encouraging results for commercial application.

Keywords: Fruit juices, Non-thermal technique, Ultraviolet radiation, Microbial reduction

Introduction

Thermal processing causes significant changes in the quality and organoleptic characteristics of juices (Turtoi and Borda, 2013). Non-thermal techniques have received increasing attention for preservation of juices, due to their potential for inactivating spoilage and pathogenic microorganisms, while minimizing the organoleptic characteristics. Among these non-thermal technologies, Ultraviolet radiation has many advantages over thermal pasteurization (Noci *et al.*, 2008; Keyser *et al.*, 2008). Ultraviolet-C (200-280 nm) is considered to be germicidal against microorganisms as it penetrates the cell membrane and results in electron shifting and DNA structure breakdown. The wavelength of 254 nm is used for the disinfection of fruit juice (Koutchma *et al*., 2004; Falguera *et al*., 2011; Shama *et al.*, 1996). Application of UV- C (200-280) is still not reported in the juice industry of Sri Lanka.

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Methodology

A system composed of juice re-circulation with an overhead tank, collecting bottom tank, UV sterilizer (GYC-UUVE-40 W, Guan Yu, China) and pumps was fabricated. UV sterilizer was thoroughly cleaned by using potable water, 200 ppm Opal solution and then with potable water. A sample of well water and portable water (100L) were separately treated with UV light under two flow rates of 0.1 m^3/h and 0.6 m^3/h at 30 + 2^oC and sample was collected into a sterilized glass bottles. Watermelons fruits were cleaned by washing with potable water, 200 ppm Opal solution and then with potable water. Fruits were cut into pieces and blended. Juice was filtered through a clean muslin cloth. The extracted juice was diluted with potable water to obtain 100 L of 33.33 % watermelon juice. Prepared watermelon juice (100 L) was treated with UV-C dosages from 1.59 Jcm^{-2} to 57.18 Jcm^{-2} under two different flow rates (0.1 m^3/h, 0.6 m3/h). Samples for microbiological analysis were collected in to sterilized stomacher bags after each UV dose. A sample of fresh water melon juice was heated at 80 ^oC for 5 minutes. SMS was added in accordance with the SLS 729 standard. The heated juice was then filled in to the glass bottles (200 mL) and capped. The sealed bottles were pasteurized for 30 minutes by immersing in a water bath maintained at boiling water. The bottles were allowed to cool, and stored in the refrigerator (4 ^oC). Samples were tested for microbial safety by subjecting them to Aerobic Plate Count (APC) and Yeast and Mold count (YM). The statistical analysis was carried out using ANOVA to test the significance of each variable (α=0.05) and followed by comparisons performed using the Turkey test by the statistical software MINITAB ®17. One way ANOVA was used to determine the effect of treatments, and UV doses in UV treatments on different juice parameters. The UV dosage for each cycle of treatment was calculated according to the method used by Keyser *et al.* (2008).

$$\text{Dosage} = \frac{\text{UV Intensity (I)} \times \text{Exposure time (t)}}{\text{Exposure surface Area (A)}}$$

Table 1: UV Doses in different flow rates and circulations

Number of Circulations	UV Doses (Jcm^{-2}) (Flow rate 0.1 m^3/h)	UV Doses (Jcm^{-2}) (Flow rate 0.6 m^3/h)
1	9.53	1.59
2	19.06	3.18
3	28.59	-
4	38.12	-
5	47.65	-
6	57.18	-
7	66.71	-
8	76.24	-
9	85.77	-

Results and Discussion

Results of the well water UV –C treatment (Table 2) showed that all the samples treated under UV dosage of 9.53 Jcm^{-2} and, 19.06 Jcm^{-2} , under low flow rate (: 0.1 m^3/h) and UV dose of 1.59 Jcm^{-2} and: 3.18 Jcm^{-2} under high flow rate (0.6 m^3/h) rates has an effect to reduce both APC and yeast and mold counts. Moreover, the effect increased with the increase of UV dose both by re-circulation.

Table 2: Microbiological test results of UV treated well water and potable water.

Treatment			APC (CFU/mL)	Yeast and Mold (CFU/mL)
SLS Standards			< 50	Nil
Well water	Control		2.4×10^2	2×10^1
	UV treatment	dose: 9.53 Jcm^{-2}	5.4×10^1	<1
		dose: 19.06 Jcm^{-2}	2.6×10^1	Nil
		dose: 1.59 Jcm^{-2}	1.5×10^2	2.5
		dose: 3.18 Jcm^{-2}	2.5×10^1	Nil
	Pasteurization		Nil	Nil
Potablewater				
	Control		<1	Nil
	UV treatment	dose 9.53 Jcm^{-2}	<1	Nil
		dose 19.06 Jcm^{-2}	<1	Nil
	Pasteurization		Nil	Nil

Results (Table 3) of UV treated watermelon juice showed that single UV-C treatment (no re-circulation) in both flow rates have very large number of APC and Yeast and Mold counts compared to SLS 729-2010 standards for ready to serve beverage. Results of APC and yeast and mould counts of UV-C treated watermelon juices with the increased number of re-circulation (Table 3) to increase the dose clearly showed that reduction of both APC and yeast and mould count, and gave encouraging results. However, even after the increased dose level of 57.18 Jcm^{-2} (after 5^{th} circulation) microbial load of the watermelon juice is not complying with the SLS 729-2010 standard.

Results indicated that the exposure time (UV-C dosage) and the low transmittance of UV through the watermelon juice containing high amount of suspended solids, soluble solids have a critical effect on inactivation of microbes. The greater the amount of soluble solids and suspended solids, the lower the intensity of penetration of the UV-C light through the liquid being treated and need to increase UV dosage via re-circulation. Moreover, the high concentration of initial microorganisms in the juice may cause the aggregation of cells and this aggregation forms a shield which can prevent the adjacent cells to be irradiated (shadowing effect). Moreover, effect of UV-C radiation depends on the type of liquid, its UV-C absorptive.

Table 3: Microbial test results of watermelon juice.

Treatment	Flow rate of UV treatment	APC (CFU/mL)	Yeast and mold count (CFU/mL)
	SLS Standards	< 50	Nil
UV treatment	Control	Uncountable	7.0×10^3
	dose : 1.59 Jcm^{-2}	Uncountable	$5.1x\ 10^3$
	dose : 9.53 Jcm^{-2}	Uncountable	6.9×10^3
	Pasteurized	Nil	Nil

Table 4: Microbial results for UV treated watermelon juice with recirculation (0.1 m3/h flow rate)

Treatment		APC (CFU/ml)	Yeast and mold (CFU/ml)
SLS standards		<50	Nil
Control		1.8×10^5	1.5×10^2
		Uncountable	$7.0\ x10^3$
UV treatment	UV Dose: 28.59 Jcm^{-2}	$6.25x\ 10^4$	$6.9\ \times 10^1$
	UV Dose: 38.12Jcm^{-2}	4.1×10^4	$7.4\ \times 10^1$
	UV Dose : 47.65 Jcm^2	$3.0\ x\ 10^4$	$6.6\ x\ 10^1$
	UV Dose: 57.18 Jcm^{-2}	9.7×10^3	5.4×10^1
Pasteurized		Nil	Nil

The penetration depth of UV-C light through the liquids is very low and reported that typically about 1 mm (Falguera *et al.*, 2011). Thus, the organisms being treated must be directly exposed to the rays (Caron *et al*., 2007). For the treatment to be effective, the juice has to be exposed as a thin film. Since, the inner diameter of the used UV sterilizer model could not be adjusted, it is not possible to expose as a thin film and recirculation of juice and processing at a very low flow rate can be done to increase the exposure time to UV-C. Results showed that there is a possibility to reduce microbial load with further recirculation at low flow rates. The reduction rate of yeast and mold due to the UV-C light is lesser than the reduction rate of APC. This could be attributed to smaller size of bacteria than yeast and molds which causes easier UV passage, increases the chance of cross linkage of neighboring thiamin and cytosine due to different cell wall constructions of bacteria with existence of higher levels of pyrimidine in bacterial DNA (Torkamani and Niakousari, 2011; Montgometry, 1985; Miller *et al.*, 1999). Studies are in progress using curry leaf and aloe vera juice to affect

the particle sizes, suspended solids and total soluble solids without adding sugar and with sugar showed encouraging results.

Conclusion

In UV-C microbial inactivation of juices the microbial reduction was dependent on the suspended solids and colour pigments of juices. Increased dosage of UV-C is necessary for juices. yeast and moulds show higher resistance to UV-C light than bacteria. UV-C light found as a combined use of the technique with other preservation methods for the disinfection of juice.

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BIOWAX COATING IMPROVES QUALITY AND EXTENDS STORAGE LIFE OF LIME FRUITS (*Citrus aurantifolia* L.)

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Summary

A study was conducted to evaluate the effect of carnauba and cinnamon oil based bio wax coating on lime fruits (*Citrus aurantifolia* Swingle) for extending the postharvest life. Lime fruits cv. Local at their optimum maturity were manually harvested from a commercial orchard, and brought to the laboratory immediately. The bulk sample was divided into four lots each containing 120 fruits. Two lots of fruits were dipped in the wax formulation for 45 seconds and the other two lots were kept as the control (no wax). Treated and control samples were stored in cold room (13°C ± 2°C ; RH 90%) and ambient conditions (32°C±1°C ; RH 70%) in plastic crates. Evaluation of physicochemical attributes were conducted before treatment (day 0) and thereafter at 7 and 15 day intervals for fruits stored in cold room and ambient conditions respectively, until the fruits exhibit limit of marketability. Bio wax treatment extended the storage life of lime fruits up to 15 days under ambient conditions in contrast to untreated fruits where marketable life ended within a week. The marketability of treated fruits stored at 13°±2°C was extended up to 60 days in contrast to untreated counterparts where marketable life was 45 days. Further experiments are in progress.

Keywords: Cold storage, Quality, Shelf life, Tropical fruits

Introduction

Lime (*Citrus aurantifolia* Swingle) is one of the high priority crops that is used in daily culinary and in food industry. The production is seasonal thus during off season, price per kg of lime rises to a level that is unaffordable by the consumers while during peak time, it drops to a level that is insufficient to recover the cost of production. The shelf life of mature limes is limited to a maximum of 5 to 6 days under ambient conditions. Therefore, there is an urgent need to develop postharvest treatments and storage strategies to prevent the market glut and to meet the year round demand. Generally, lime fruits show high rate of water loss during storage. Our observations proved that lime fruit peel become thinner and thinner as the fruit matures (Samaradiwakara *et al*., 2017) and increased surface area to volume ratio contributing to higher rate of water loss. It is reported that rate of water loss can be reduced by 30-50% when fruits are coated with wax (Wills *et al.,* 2007). Therefore, this study was conducted with the

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objective of evaluating the effect of the new Hexanal incorporated ITI bio wax formulation for extending the postharvest life of lime.

Methodology

Lime fruits (*C. aurantifolia*) cv. Local was obtained from a commercial orchard in Anuradhapura. Fruits were harvested manually at mature green stage (weight 41.6±6.4 g and TSS ≈8.4 ±0.1% and TA ≈7.6±0.01%), packed in plastic crates of 18-20 kg capacity and transported to the laboratory. Fruits free from any visible defects were selected and further sorted based on weight, diameter and colour to have uniformity. The selected fruits were divided into 4 lots each containing 120 fruits (per experimental unit). Two lots of fruits were dipped in the bio-wax formulation for about 45 seconds. The other two lots were kept as the control (no wax). After that, treated and control samples were stored in cold room (13°C±2°C, RH 90%) and under ambient conditions (32°C±2°C, RH 70%) respectively in medium size (600x300x320 mm) plastic crates. Evaluation of physicochemical attributes were made before treatment (day 0) and thereafter at 7 & 15 day intervals for fruits stored under ambient and cold room conditions respectively.

Data collection: Peel colour was measured using Hunter lab colour difference meter (CR 400, Konica Minolta) and the values of L*, a*, b* were recorded. Fruit firmness (with peel) was measured using bench top type digital fruit firmness tester (53205, Turoni). Total soluble solids (TSS) of the extracted juice was measured by a temperature compensated digital refractometer (3810, Atago PAL-1) and expressed as percentage. Titratable acidity (TA) was determined as per AOAC (2005) and expressed as grams of citric acid equivalents per 100 ml of juice. Juice pH was measured by a pH meter (230A+, Thermoorion). Weight loss was recorded by subtracting final weights from the initial weights of stored fruits and expressed as percent weight loss with reference to the initial weight. Decay incidence was evaluated and expressed as the proportion (by number) of rotten fruits relative to total number of fruits stored in each treatment. Visual quality of fruits were rated by using 1-5 scale i.e. 5=excellent (green & fresh), 4=good (yellow with slight green & fresh), 3=fair (all yellow & fresh), 2=poor (yellow with small brown patches, unmarketable, but usable), 1=unusable (all brown).

Results and Discussion

At optimum harvest maturity lime fruits are characterized by light green peel colour, at which CIE lab parameters were 54.2±2.9 (L*), 20.3±1.4 (a*), 36.9±2.0 (b*). Untreated fruits stored under ambient conditions demonstrated rapid degradation in peel colour as indicated by significant increase in L* and b* while reduction in a* on the contrary to wax coated counterparts (Table 1). More than 50% of the uncoated ambient stored fruits exhibited dark brown peel colour after 15 days whereas it was 4% in the treated counterparts. No signs of peel browning were observed in treated and control fruits stored in the cold room until 45 days. By the end of 60 days in cold storage, more than 80% of untreated fruits exhibited peel browning in contrast to its treated counterparts

where only 4% of fruits showed browning of peel. No incidence of disease was observed in both treated and control fruits stored in the cold room for 60 days. However, at ambient temperature storage, both wax coated and control fruits showed disease development of 6% and 11% respectively by the end of 15 days (data not shown).

Fruit firmness increased significantly during storage except the fruits coated with wax and stored at low temperature (Table 1). This might be due to toughening of the fruit peel exhibiting leathery texture. Fruit peel showed withered appearance indicating signs of water loss which contributed to high rate of physiological loss in weight (PWL, Table 1). The highest PWL was observed in uncoated ambient temperature stored fruits whereas the rate of weight loss was reduced by 50% in wax coated fruits. By the end of 60 days of cold storage period, the PWL of treated fruits was reduced by 14% in contrast to uncoated fruits which showed 28.5% PWL (data not shown).

Table 1. Physicochemical parameters of bio wax treated lime under different storage conditions at 15 days after storage

Treatment and storage condition	Peel colour			Fruit Firmness (N)	PWL (%)	Chemical properties of juice		
	*L**	*a**	*b**			TSS (%)	TA (% citric acid)	pH
Wax + ambient	63.0±2.8^{b}	15.9±4.4^{a}	47.1±3.3^{b}	168.3±1.8 ^{a}	16.9±2.0^{b}	8.3±0.3	7.5±0.4	2.24±0.04
No wax + ambient	75.4±1.2^{a}	6.7±0.7^{b}	53.2±3.2^{a}	163.3±4.2 ^{ab}	33.7±5.3^{a}	8.5±0.5	7.3±0.4	2.26±0.08
Wax + cold room	59.8±2.5^{b}	20.3±1.2^{a}	44.2±2.3^{b}	135.7±3.7 ^{c}	8.9±0.7^{c}	7.7±0.3	7.0±0.4	2.25±0.03
No wax + cold room	62.7±0.7^{b}	19.8±1.3^{a}	46.4±1.4^{b}	160.0±1.5 ^{b}	11.6±0.7^{bc}	8.0±0.3	7.5±0.3	2.16±0.03
LSD (P < 0.05)	3.75	4.53	5.00	5.73	5.44	NS	NS	NS

Initial (day 0): L* = 54.2±2.9, a* = 20.3±1.4, b*=36.9±2.0, Firmness = 135.8±2.7N, TSS = 8.3±0.09, TA = 7.6±0.05, pH = 2.14±0.03. L* = 0: black, 100: white; a* = (-): greenness, (+): redness; b* = (-) blueness, (+): yellowness. Means in a column with the same letter are not significantly different (at P < 0.05) according to LSD; Each value represent mean ± S.D. of three replicates (n=40). Ambient storage condition: 32±1 ^{o}C, RH 70%, cold storage condition: 13°C±2^{o}C, RH 90, NS: not significantly different

Internal fruit quality parameters measured as TSS, TA and pH were not affected by the wax treatment significantly (P > 0.05) and these three parameters remained almost unchanged during 15 days of ambient and 60 days of cold storage conditions (Tables 1 and 2). Similar results have been reported by Gunasekara *et al*. (2016) for mango cv. Karuthacolomban.

Table 2. Juice chemical parameters of bio wax treated lime at 30, 45 and 60 days in cold room conditions (13±2°C, RH 90%.).

Parameters and treatments		Time (days after storage)		
		30	**45**	**60**
TSS (%)	Wax	7.9±0.6	8.3±0.1	8.6±0.3
	No wax	7.7±0.2	8.6±0.2	8.7±0.6
	P(α =0.05)	0.7^{NS}	0.1^{NS}	0.8^{NS}
TA (% citric acid)	Wax	7.0±0.3	7.9±0.5	7.6±0.2
	No wax	7.2±0.3	8.3±0.2	7.8±0.3
	P(α =0.05)	0.6^{NS}	0.3^{NS}	0.5^{NS}
pH	Wax	2.3±0.09	2.27±0.02	2.30±0.05
	No wax	2.4±0.07	2.32±0.02	2.34±0.05
	P(α =0.05)	0.2^{NS}	0.05^{NS}	0.4^{NS}

Data was analyzed by two sample t test at P = 0.05 using MINITAB; Each value represent mean ± S.D. of three replicates (n=40). Cold storage condition: 13±2 °C, RH 90%.

This is a very good positive point, because if we can take measures to minimize the rate of degradation in peel colour and rate of water loss, the storage life of lime fruits could be extended beyond 60 days under low temperature. Therefore, further experiments are in progress with modifications.

Conclusion

The bio wax treatment extended storage life of lime fruits up to 15 days under ambient conditions (32°C±2oC, RH 70%) in contrast to its untreated counterpart of which marketable life was only 7 days. However, if the treated fruits are stored at low temperature (13°C±2°C ; RH 85-90%), the storage life can be extended for up to 02 months on the contrary to untreated ambient temperature stored fruits and by 15 days when compared to its untreated low temperature stored counterpart.

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TRANS FAT FORMATION AND CHANGES IN FATTY ACID PROFILE OF SOY AND COCONUT OIL BLENDS WITH REPEATED FRYING

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Summary

Fried foods are very popular worldwide. Cooking oil with high unsaturated fatty acid forms trans-fat during frying which increases with repeated usage. Soy oil possess high unsaturation, and is prone to isomerization during frying. Blending soy oil with another cooking oil is a better option to minimize trans-fat formation.

Keywords: Frying, Soy oil, Blending, Coconut oil, Trans-fat

Introduction

Frying is a very common food preparation method. Oxidation, isomerization, polymerization and hydrolysis of fats occur during frying and form free fatty acids, trans-fat and polar molecules (Paul and Mittal, 1997). Absorption of them into fried food is more problematic. Trans-fat is recognized as a critical compound formed during frying. The major ingredient of fried food is cooking oil. Therefore, cost of oil becomes the most important factor in terms of economy. To reduce cost, people use same oil repeatedly for frying (Rani *et al.*, 2010). Chemical reactions during repeated frying make fried food unsuitable in terms of nutritional facts (Ghidurus, 2010). Moreno *et al*. (1999) revealed, trans fat formation during frying is closely related to process temperature and oil use time. Palm, soy bean, canola and sunflower oil are the major cooking oils consumed by people worldwide. Among the oils containing unsaturated fatty acids soybean oil is widely available. Being oil rich in unsaturated fat, soy oil is highly susceptible for trans fat formation during frying. Blending soy oil with coconut oil increases the stability of oil blends against isomerization. It can also improve the keeping quality of soy oil facilitating the market demand. The present study was aimed to study the behaviour of fatty acids in soy and coconut oil blend over repeated frying.

Methodology

The study was carried out at the Food and Nutrition Research Centre, CIC Agribusiness (Pvt) Ltd, Pelwehera, Sri Lanka. Branded, refined soy bean oil and coconut oil were used. Three blends having 75%, 50% and 25% of soy oil were prepared and labelled as

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S75, S50, and S25 respectively. Pure soy oil and pure coconut oil samples were labelled as S100 and S0 respectively. Each of the above oil samples were of 200ml in volume. All samples were separately mixed well using a mechanical stirrer at 180 rpm for 15 minutes. All were heated to 180 ^{O}C. Potato slices (200 g) were continuously deep fried for 30 minutes in a stainless steel pan. Heated oil was obtained after completion of 30 minutes continuous deep frying. The process was repeated two times to obtain three times heated oil with five hours cooling interval to reach room temperature. No fresh oil was added between the frying processes. After being heated and cooled, small amount of oil was taken for the fatty acid profile analysis. Fatty acid composition was analysed by Gas Chromatography (Agilent 7890A). Fatty acid methyl esters (FAME) of oil samples were prepared according to AOAC 969.33 method (Chu and Kung, 1998). CRM 47885 SUPELCO 37FAME was initially analyzed as the reference standard FAME mix to determine the peak identity.

Results and Discussion

Table 01: Fatty acid profile changes in oil samples with frying cycles.

Oil sample	Frying	Caprylic	Capric	Lauric	Myristic	Palmitic	Stearic	Oleic	Linoleic	Linolenic
S100	F0	ND	ND	ND	0.07	9.74	4.41	24.35	52.98	6.20
	F1	ND	ND	ND	0.09	9.74	4.41	24.35	52.98	6.20
	F2	ND	ND	ND	0.10	11.28	5.12	26.70	49.16	4.76
	F3	ND	ND	ND	0.12	13.10	5.90	23.96	44.48	3.52
S75	F0	1.26	1.08	9.12	4.31	10.07	4.16	20.80	41.88	4.88
	F1	1.33	1.11	1.11	4.15	10.42	4.36	21.48	41.34	4.60
	F2	1.45	1.12	9.65	4.32	11.08	4.70	22.33	39.45	4.01
	F3	1.61	1.18	10.41	4.80	12.55	5.66	24.38	34.20	2.77
S50	F0	2.68	2.35	20.08	8.67	10.11	4.08	17.06	30.17	3.41
	F1	2.86	2.45	20.77	9.01	10.66	4.29	17.51	28.07	2.83
	F2	2.81	2.57	23.26	10.37	12.19	4.91	17.57	22.79	1.78
	F3	3.34	2.80	24.48	10.91	13.00	5.34	18.19	18.65	1.17
S25	F0	4.36	3.75	31.60	13.60	10.22	3.95	12.88	16.92	1.79
	F1	5.07	4.13	33.67	14.25	10.43	3.90	12.30	13.95	1.28
	F2	4.72	4.01	34.33	15.00	11.32	4.33	12.66	11.39	0.84
	F3	4.97	4.27	36.92	16.21	12.08	4.63	11.66	7.24	0.35
S0	F0	6.31	5.33	44.84	19.16	10.16	3.58	7.94	2.03	ND
	F1	5.46	5.12	45.04	19.16	10.30	3.58	7.17	1.38	ND
	F2	5.62	5.30	47.02	20.30	10.55	3.66	6.03	0.63	ND
	F3	7.47	5.83	46.90	19.77	10.25	3.58	4.57	0.27	ND

*S100- soy 100%, S75- soy 75%, S50- soy 50%, S25- soy 25%, S0- coconut 100%, F0- frying 0, F1- frying 1, F2- frying 2, F3-frying 3, ND- Not detected

Table 1 clearly illustrates the soy oil comprises unsaturated fatty acids such as oleic, linoleic and linolenic while coconut oil is comprised with saturated fatty acids such as lauric, myristic and palmitic acid. Due to high unsaturation (83.54%), soy oil has high susceptible to isomerize during frying and makes trans-fat. Since coconut oil comprises low level of unsaturated fatty acids, it gives less chance to trans-fat formation during frying. The obtained percent yield for essential fatty acids; both linoleic and linolenic in pure soy oil (59.2%) was higher than coconut oil (2.0%). Furthermore results indicated

that the soy oil was deficient with medium chain fatty acid (~0%) while coconut oil had higher proportion of medium chain fatty acid (56.48%). Medium chain fatty acids are health beneficial fatty acids because they can readily burn and produce energy to body, never have they stored at adipose tissue as fat. The results of analysis showed that the contribution of coconut oil proportion to soy oil, affect to reduce unsaturation of oil blends. This may mean that the blending of high unsaturated oil with high saturated oil leads to reduce the unsaturation and modify the fatty acid profiles of oil blends which comprise properties of both oils. Li *et al.*, (2014) found that blending of sunflower kernel oil with Sclerocaryabirrea oil could modify the fatty acid profile of the blends. The obtained results in this analysis were similar to cited finding.

Table 02: Fatty acid profile changes with repeated frying.

Sample	Frying	Unsaturated fat	Saturated fat	Trans fat
S100	F0	83.54	14.22	ND
	F1	81.91	15.48	0.05
	F2	80.62	16.5	0.08
	F3	76.95	19.12	0.21
S75	F0	67.56	30.01	ND
	F1	67.42	31.46	0.04
	F2	65.79	32.95	0.08
	F3	61.35	36.21	0.2
S50	F0	50.64	47.98	ND
	F1	48.41	50.03	0.04
	F2	42.14	56.11	0.13.
	F3	38.01	59.87	0.19
S25	F0	31.59	67.47	ND
	F1	27.53	71.45	0.04
	F2	24.89	73.73	0.05
	F 3	19.24	79.08	0.06
S0	F0	9.97	89.37	ND
	F1	8.54	90.27	ND
	F2	6.65	93.72	ND
	F 3	4.84	93.81	ND

*S100- soy 100%, S75- soy 75%, S50- soy 50%, S25- soy 25%, S0- coconut 100%, F0- frying 0, F1- frying 1, F2- frying 2, F3-frying 3, ND- Not detected

As shown in table 2, unsaturation was reduced with the increasing frequency of frying in all oil samples. In addition saturation of the oil increased with the increasing number of frying. According to the compiled study linolelaidic acid (C18:2 trans) was identified as the trans-fat. It is trans-isomer of linoleic acid. It was shown in the present study that the trans-fat was formed and it was increased with the increasing frequency of frying in soy oil and its three blends.

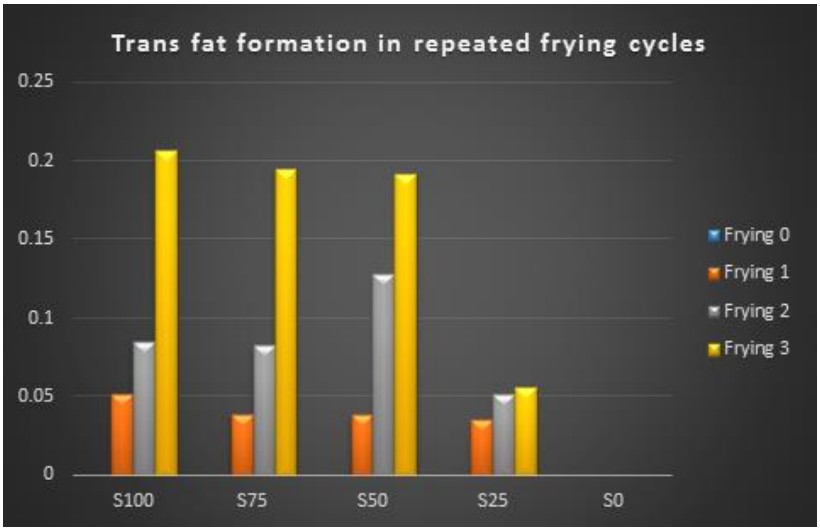

Figure 01: Trans fat formation with repeated frying.

The result of the analysis indicated that there was no trans-fat in S0 oil sample in any stages of frying. This may mean that there has a negligible chance to form trans-fat in coconut oil due to the lack of unsaturated fatty acids. As presented in the graph, the percentages of trans-fat in soy oil sample was obtained the highest values in every frying cycles compared to other three blends. There was usually a good positive relationship between frying frequency and trans-fat formation. With the increase of number of frying, trans-fat formation increased. Moreno *et al.* (1999) cited trans-fat formation during food frying is closely related to the process temperature and oil use time. Further it was mentioned that trans unsaturation started to increase at 150 ^{0}C, and became much more significant from 250 ^{O}C onwards. This may mean that it will be a good chance to form trans-fat in unsaturated oils at higher temperature.
In addition, this analysis indicated that the contribution of coconut oil may affect to reduce the trans-fat formation. Trans fat formation in S25 sample was lower than S75 and S50 oil blends. It was clearly depicted that blending leads to minimize trans-fat formation in oils.

Conclusions

Trans-fat formation increased with increase of frying frequency. However pure coconut oil did not form trans-fat during frying. With the contribution of coconut oil, trans-fat formation in blends led to decrease. Therefore, blending of soy oil with coconut oil is a better option to reduce trans-fat formation during frying.

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DEVELOPMENT AND QUALITY EVALUATION OF RICE (*Oryza sativa*) FLAKES INCORPORATED GLUTEN FREE COOKIES FROM MAIZE (*Zea mays* L.) AND SOY BEAN (*Glycine max*) FLOUR BLENDS

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Summary

Cookies are one of the most convenient and mostly consumed food. It has been identified that gluten free food products and high protein products, have a high market demand. This research has focused on the development and quality evaluation of rice (*Oryza sativa*) flakes incorporated gluten free cookies from maize (*Zeamays* .L) and soy bean (Glycine max) flour. The nutritional value of the developed non-gluten baked cookies were found to be; moisture 2.83 % ±0.01, crude protein 10.07 % ±1.54, crude fat 27.19 % , total ash 1.97 % ±0.19, crude fibre 22.22 %, total carbohydrate content 22.22 %, and total energy value 516.42 Kcal. Gluten (wet, dry) content of raw materials and final sample were recorded as zero. The physical properties determined were; average diameter 5.36 cm±0.09, average thickness 0.55 cm±0.05, lowest spread ratio 9.01 cm±0.06, average raw weight 6.52 g±0.23, average dry weight 7.54 g±0.21, average stack length 113.66 mm±2.08 and average stack weight were 160.49 g±0.89. *E-coli*, *Salmonella* and *Staphylococcus aureus* were not recorded in the final sample, the final test results for total plate count was 2.2×10^2 CFU/g. The cut length, reel width and price of the packaging material were calculated as 19.36 cm, 19.83 cm and Rupees 2.83 respectively. The cost of (160 g for 21 cookies) was approximately Rupees 70.00.

Keywords: Cookies, Gluten free, Maize, Soybean

Introduction

Cookies are consumed by a wide range of people. Cookies are known to have desirable organoleptic properties, safety and nutrition. Usually cookies have a longer shelf life and low in cost. Manufacturers change the nutritional content and organoleptic properties of biscuits according to different trends and demands of the consumers. It has been identified that gluten free food products have a high market demand in the present society (Mona and Hinar, 2015). Gluten allergy is a chronic disease which is a genetic disorder found in human beings (Lorena *et al.*, 2010). Gluten is a protein found in wheat, barley and rye like grains. It is the major component of the bakery products responsible for better structural developments and the other organoleptic properties. (Mishra *et al.*, 2012). Rice flakes, maize flour and soybean flour do not contain gluten (Prakiriti *et al*., 2016). Gluten free is the voluntary term, for food items which contains

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less than 20 ppm of gluten (Simona *et al*., 2014). There is a recorded protein malnutrition problem among some communities in the country, mainly the estate sector which leads to malnutrition conditions. Since cookies are low in costs it can be identified as a good source to be introduced among the children of such communities. Cookies can also be defined as the excellent source for the fortification and value addition of nutrients such as proteins and fibre. So this study is designed to incorporate the maize and soy bean flour blends incorporated with rice flakes to make gluten free high protein cookies.

Methodology

Several sensory evaluation tests were carried out to find out the suitable formulation of the cookie. The first sensory evaluation was carried out to determine the suitable soy bean and maize flour blends. Nine scale hedonic test for appearance, aroma, sweetness, sourness, crispiness, after taste and overall acceptability were tested among 11 samples using 23 untrained panelists from the plant. The 11 cookies samples (Coded: 218, 408, 561, 739, 426, 8 49, 501, 782, 721, 306, 519) were prepared using only soybean flour and maize flour without the addition of rice flakes. The ratios of maize flour (MF) to soy flour (SF) were (MF:SF;100:0, 90:10, 80:20, 70:30, 60:40, 50:50, 40:60, 30:70, 20:80, 10:90, 0:100). The second sensory evaluation was carried out to find the suitable rice flake proportion. This was selected among 7 samples by using 25 untrained panelists from the plant. 9 scale hedonic test for appearance, aroma, sweetness, color, crispiness, after taste and overall acceptability were tested. 7 cookies samples (Coded as 297, 471, 746, 827, 948, 518, 516) were prepared using the selected soybean flour and maize flour blend with the addition of rice flakes in different ratios. (FM:RF; 100:0, 90:10, 80:20, 70:30, 60:40, 50:50, 40:60, where FM: Flour Mixture, RF: Rice Flakes). Thus, the most accepted proportion of the rice flakes and the flour blend were selected. The results of all sensory evaluation tests were statistically analyzed according to the kruskal wallis test using SAS 9.3 software. For the cookie preparation the flour mixtures and rice flakes were mixed with 2 % leavening agent, 40 % shortening, 40 % powdered sugar, 2 % vanilla and 2 % salt. Mixed flour mixture was subjected to molding and baking under 160 ^{O}C for 20 min using electric oven. The proximate analysis of the final cookie sample was carried out according to the AOAC methods for moisture, crude fat, crude fibre, crude protein, total ash content, and carbohydrate content and total energy content were calculated by FAO (2003) methods. Microbiological analysis was carried out according to the SLS standard methods for, *E-Coli, Salmonella and Staphylococcus aureus*. Gluten test (Wet, dry) was carried out according to AACC methods. The physical properties of the cookies were determined according to the AACC (2000) method; diameter, thickness, spread ratio, raw weight, dry weight, length, stack length and stack weight were the physical properties measured.

Results and Discussion

Sample no. 721 (soy: maize = 4:1) was selected as best sample through the first sensory evaluation. Among the sensory attributes tested it was found that there were no significant difference between appearance ($p=0.24$) and aroma ($p=0.51$), and there was

a significant difference among the attributes such as sweetness (p=0.01), sourness (p=0.02), crispiness (p=0.01), after taste (p=0.01) and overall acceptability (p=0.01). Sample.no 746 (flour blend: rice flakes = 4:1) was selected as best sample and final sample through second sensory evaluation. Among the sensory attributes tested it was found that there were significant difference in all the sensory attributes tested appearance, aroma, sweetness, color, crispiness, after taste and overall acceptability (for all sensory attributes p<0.001). The results of the proximate analysis were as follows, moisture (analysed through moisture meter) 2.83 % ± 0.01, crude protein 10.07 % ±1.54, crude fat 27.19 %, total ash 1.97 % ± 0.19, Crude fiber 22.22%, total carbohydrate content 22.22 %, and total energy value 516.42 Kcal. But in comparison of commercially available whole wheat based cookies with final selected sample of the study, 6.30 % of moisture, 7.30 % of crude protein, 3.90 % of total ash and 27.18 % were obtained. The final sample was noted to have a high shelf life, great potential to overcome the protein malnutrition problem among the people. Gluten (wet, dry) content of raw materials and final sample were recorded as zero. Therefore the developed cookie can be recommended for gluten intolerance people. The average diameter, average thickness, lowest spread ratio, average raw weight, average dry weight, average stack length and average stack weight were recorded as 5.36 cm±0.09, 0.55 cm ± 0.05, 9.01 cm ± 0.06, 6.52 g ± 0.23, 7.54 g ± 0.21, 113.66 mm ± 2.08 and 160.49 g ± 0.89 respectively. *E-coli, Salmonella* and *Staphylococcus aureus* were not recorded in the final sample. The final test results for total plate count was 2.2×10^2 CFU/g. The shelf life of the cookies can be extended for more than 6 months. The cut length, reel width and price of the packaging material was calculated as 19.36 cm, 19.83 cm and Rs. 2.83 respectively. The final cost of cookies was evaluated as approximately Rs. 70.00 (160 g for 21 cookies).

Conclusions

According to the results of this finding, the developed rice flaked incorporated, soy bean and maize flour gluten free cookie can be recommended for gluten intolerance people and it can also be used as a nutritional source for protein malnutrition. And the final product was observed as microbiologically safe comparing with the commercially available whole wheat flour cookies.

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STUDYING FACTORS AFFECTING WHITE PRECIPITATE FORMED IN ICED TEA WHEN RECONSTITUTED WITH HARD WATER AND FINDING SOLUTIONS

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Summary

The study was conducted in an organization who engages in value added tea manufacturing and exporting. One of company's newly established product category is iced teas. The product is being manufactured in two phases. In the 1st phase a concentrated liquid tea extract is manufactured in a factory that has been established within a tea garden. In the 2nd phase the concentrated liquor is being diluted to make "Ready to Drink Tea" (RTD) and packed in the bottling plant. Hardness of water used for diluting is critical at this stage as hard water causes create a white insoluble matter in the end product leading to an unacceptable hazy product. The reason for above was assumed to be a reaction between Ca^{2+} ions present in hard water and some constituent containing in the product. As per literature, tea is a source of oxalic acid which can combine with Ca^{2+} ions in hard water to form calcium oxalate and create sediment in the beverage (Shimada, 2012). The objective of the study was to investigate on components in the product interacting with hard water and to investigate a treatment to make the product tolerant to hard water to a practically feasible limit. Favoring the forward reaction ($Ca^{2+} + C_2O_4^{--} \rightleftharpoons CaC_2O_4 \downarrow$) by increased Ca^{2+} concentration in the extracted liquor in order to exhaust oxalate ions in the media before coming to the end product and centrifugal removal of sediment is the theory behind the study. Ca^{2+} was introduced through calcium chloride.

Keywords: Iced tea, Turbidity, Sediment, Haze, Hardness

Introduction

Tea is a natural herb which is made from Camellia plant. It is the mostly consumed beverage on earth after water. Sri Lanka has the best climatic conditions for tea cultivation where no other country can compete with Ceylon tea, due to its distinct taste and aroma. A range of iced tea is manufactured on site at the tea garden without using any chemical processing aids. This tea is rich in antioxidants and full of natural goodness, while giving a taste of real garden fresh Ceylon Tea. However, the iced tea is found to have comparatively lower shelf life and higher storage necessity. The problem associated with the studied iced tea, was the formation of a white color haze / sediment, when tea concentrate was diluted with hard water. Therefore, the study was designed

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with the overall objective of finding a solution to the formation of haze in the finished iced tea upon reconstitution with hard water due to white sedimentation. The specific objectives of the study were to identify the components of iced tea affecting white color sedimentation and to identify the most suitable options to overcome this problem.

Materials and Methods

Hard water at different hardness levels were prepared by method described by Environmental Protection Agency of USA (US EPA). Samples of black tea were collected from company owned Rilhena Estate. Samples of citric acid, ascorbic acid and cane sugar were collected from the factory. Degree of formation of sediment was measured through turbidity measured by a digital turbidity meter, where the degree of turbidity is expressed as Nephalometric Turbidity Units (NTU). Total Hardness was measured in ppm by the method described by US EPA.
Contributing ingredients of tea concentrate to form sediment with hard water was determined through a series of combinations of major ingredients as given in Table 1.

Table 1: Different combinations of ingredients to identify contributing source of factor of sedimentation in the drink.

Treatment	Ingredients
Treatment 1:	Citric acid and hard water
Treatment 2:	Sugar and hard water
Treatment 3:	Tea extract and hard water
Treatment 4:	Tea extract, sugar and hard water
Treatment 5:	Tea extract, citric acid and hard water
Treatment 6:	Tea extract, citric acid, ascorbic acid and hard water

Calcium ions were introduced to the tea extract at different levels as indicated in the table 2. Tea concentrate was produced following the process shown in figure 1. The end product was made by diluting with water at different hardness as shown in Table 2. Degree of sedimentation was measured in terms of turbidity in NTU immediately after making the product, after 1 week of storage, and after 2 weeks of storage. A sensory evaluation was carried out to validate sensorial acceptance of the treatments. Experimental data were statistically analyzed with SPSS.

Figure 1: Process Adapted to Make Tea Concentrate and then to Make RTD Iced Tea

Table 2: Different Treatment and Hardness of Water Used for Making RTD

Treatment	**Hardness of Diluting water**					
	25 ppm	**50 ppm**	**100 ppm**	**200 ppm**	**300 ppm**	**400 ppm**
Control (used soft water)	NTU	NTU	NTU	NTU	NTU	NTU
Hard Water Hardness: 200 ppm	NTU	NTU	NTU	NTU	NTU	NTU
$CaCl_2$ 20 ppm	NTU	NTU	NTU	NTU	NTU	NTU
$CaCl_2$ 40 ppm	NTU	NTU	NTU	NTU	NTU	NTU
$CaCl_2$ 80 ppm	NTU	NTU	NTU	NTU	NTU	NTU
$CaCl_2$ 160 ppm	NTU	NTU	NTU	NTU	NTU	NTU
$CaCl_2$ 20 ppm	NTU	NTU	NTU	NTU	NTU	NTU

Results and Discussion

Figure 2 shows turbidity of mixtures of different combinations that are being used to make iced tea (BMF means tea extract). Turbidity of mixtures containing tea extract are significantly high compared to mixtures without tea extract. The results indicate that the contributing compound to sedimentation with hard water is linked with tea.

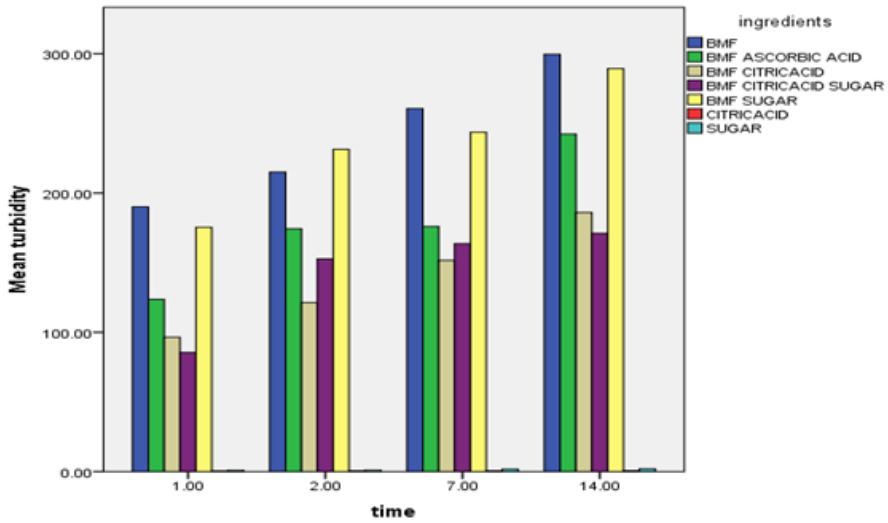

Figure 2: Turbidity in NTU of Mixtures of Ingredients of Iced Tea at different time of storage in days.

Turbidity of RTD in storage for products made with added calcium ions are shown graphically in figure 3 below. Calcium ion concentration of water used to extract tea in manufacturing tea concentrate has a significant effect on turbidity of the resulting RTD iced tea. The best treatment was use of treated water (soften through an ion ethane column) with added $CaCl_2$ at the dosage of 160 mg/L.

Conclusion

Oxalate present in tea leaves reacts with Ca^{2+} ions present in water to cause haze in iced tea if reconstituted with water with a high degree of hardness. Pre-treatment of extraction water of teas with calcium ions can be used as a remedy of turbidity development of Iced Teas.

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COMPARISON OF FUNCTIONAL PROPERTIES OF STARCHES AVAILABLE IN SRI LANKA

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Summary

Starch contributes 50 to 70% of energy in the human diet, providing a direct source of glucose, and glycemic response to excessive consumption of starch may be a factor in some diet-related illnesses. Rice is the principle staple food in Sri Lankan diet and other carbohydrates rich flour types are also consumed in a considerable quantity. Higher consumption of white rice may increase the prevalence of diabetes in Sri Lanka. There are an increasing number of evidences that physical and morphological characteristics of starch plays major role in relation to the occurrence of diet related diseases. The objective of this study is to compare the functional properties of 16 available starches in Sri Lanka by observing the granular structure, determining Water Solubility Index (WSI), Water Absorption Index (WAI), and Water Swelling Capacity (WSC). Particle size 100-150 μm dry powder obtained for analysis. The hydrolyzing rate was determined by GOD method. The granule morphology was determined using Scanning Electron Microscopy (SEM) and WSI, WAI and WSI was determined according to method described in Noor et al, 2014. Hydrolyzing rates of the starches for amyloglucosidase and α-amylase ranged from 4.78±3.04 to 85.69±8.18 μM glucose/min and 2.10±1.25 to 174.37±9.96 μM maltose/min respectively. The highest glucose and maltose releasing rates were observed respectively in Oats and Palmyra starches while the least rate was observed in Soy starch on both occasions. The average granule size of the starches ranged from 12.22 to 1457.20 μm2. Largest granule sizes were found in Mandu, Kithul, Chickpea and Oats while White Basmati, White Raw Rice and Red Basmati had markedly smaller granule sizes. The highest Average WAI showed Undu and while Wheat showed least average WAI and ranged from 0.8±0.02 – 2.73±0.11. Highest WSI was recorded in Soy while Olu recorded least WSI. Kithul demonstrated highest WSC while Soy Demonstrated least WSC ranged from 11.31±0.1 – 2.4±0.03. Further studies and analysis are required to determine relationship between these attributes to conclude by confirming the obtained observations.

Keywords: Starch, Granule morphology, Water solubility index, Water swelling capacity

Introduction

Starch contributes 50 to 70% of the energy in the human diet, providing a direct source of glucose, which is an essential substrate for the brain and red blood cells for generating metabolic energy (Copeland *et al*., 2006). The glycemic response to excessive consumption of rapidly digesting starch is a factor in some diet-related

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illnesses such as Diabetes, Obesity, Weight gain, etc. (Jayawardana *et al*, 2013). Rice is the principal staple of Sri Lankan diet and other carbohydrates are consumed in lesser quantities. The high consumption of white rice is thought to increase the incidence and prevalence of diabetes (Medagama *et al*, 2015). More than a fifth of Sri Lankan adults are dysglycemic and the prevalence of diabetes is alarmingly high (Jayawardana *et al*, 2013). Therefore, identifying and analyzing the characteristics of commonly available starches in Sri Lanka will be able to create a descriptive profile and a reliable source to prepare dietary guidelines for patients with diabetes mellitus and other dietary illnesses, and further it will be useful in product development, and value addition of certain foods.

Methodology

The Selected samples were Atta, Chickpea, Corn, Kithul, Kurakkan, Mandu, Oats, Olu, Palmira, Raw Red Rice, Red Basmati, Soy, Undu, Wheat, White Basmati, White Raw Rice and particle size 100-150 μm dry powder obtained for analysis. The hydrolyzing rate was determined by GOD method described in Visvanathan *et al*, 2016. The granule morphology was determined using Scanning Electron Microscopy (SEM). Other tests such as WSC, WAI and WSI was determined according to a method described in Noor *et al*, 2014.

Results and Discussion

The average area distribution of starch granules from different starch samples ranged from 12.22 to1457.20μm^2. Typical micrographs show polygonal, lenticular and spherical shaped granules in different samples. Largest granule sizes were found in Mandu, Kithul, Chickpea and Oats while White Basmati, White Raw Rice and Red Basmati had markedly smaller granule sizes. Hydrolyzing rates of the starches for α-amylase ranged from 4.78±3.04 to 85.69±8.18 μM maltose/min. The highest maltose releasing rates were observed in Oats while the least maltose releasing rate was observed in Soy. Hydrolyzing rates of the starches for amyloglucosidase ranged from 2.10±1.25 to 174.37±9.96 μM glucose/min and the highest glucose releasing rates were observed in Palmyra and Oats while the least rate was observed in Soy.

Differences among swelling behaviors of various starches can be attributed to changes conferred to the structure of starch granules. Kithul demonstrated highest Water Swelling Capacity (WSC), while Soy Demonstrated least WSC ranged from 11.31±0.1 to 2.4±0.03. Water Absorption Index (WAI) indicates the extent of starch gelatinization. The highest average WAI was found in Undu and Palmyra, while Wheat (All Purpose) showed least average WAI ranged from 0.8±0.02 to 2.73±0.11.

Solubility of starch varies with temperature and it can be generally concluded that solubility increases with temperature increment for starch but Undu shows slight inverse difference of solubility than other starch varieties.

Conclusion

In this study, according to the average area distribution of starch granules Mandu found as the largest granule while White Basmati found as the smallest granule. It may be because of the differences in the structure of starch granules of various botanical species or due to structural alterations of domestic and commercial processing of foods. Hydrolyzing rates detects the maltose or glucose releasing rates per minute. In both occasions Soy has been recorded as the lowest maltose and glucose releasing rates which are desirable for preventing non-communicable disease conditions. Moreover the behavior of starch with water has been observed and further studies are required to determine and to confirm above observed data. This would facilitate to predict functionality of starch structure and explain how it interacts with other food constituents which is a significant challenge in food science and nutrition. Finding of this study may be useful to develop different functional foods targeting patients with diabetes mellitus.

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DETERMINATION OF PHYSICAL, CHEMICAL & SENSORY CHARACTERISTICS OF BLACK TEA IN NUWARA ELIYA REGION

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Summary

Tea is the most popular non-alcoholic beverage in the global market, second only to water. According to the tea characteristics and origin, Sri Lankan tea growing regions have divided in to 7 different regions. Current study was designed to determine the physical, sensory and chemical parameters of Nuwara Eliya regional black tea, which is popular for its delicatefragrance, to establish a database and with the aim of producing specialty tea. In order to achieve that, black tea samples from three different Nuwara Eliya regional tea factories were analyzed for their physical, sensory and chemical parameters from different grades as BOPF, BOP, PEKOE and Dust 1. The variation of chemical composition among the tea estates and grades were analyzed. According to the results, it was revealed that all the tested chemical parameters were complied with ISO standards and volatile compounds trans-2-hexenal, Pentanal and Linalool showed higher concentrations. When considering the correlation a significant positive correlation showed between TF, TR and color and those are in agreement with the results of previous studies

Keywords: Regional tea, Correlation, Chemical composition

Introduction

Tea is made by using the young tender shoots of *Camellia sinensis (L.) o. Kuntze* is a unique aromatic beverage prepared by pouring hot water over cured leaves. It is the main export crop in Sri Lanka (Hara *et al*, 1995; Wright *et al*, 2002). Tea is known as the most widely consumed non-alcoholic beverage after water (Venkateswaran et al, 2002). The chemical, physical and sensory characteristics of the tea vary according to the different regions present in Sri Lanka. The soil type, climatic conditions and agricultural practices are the factors affect the regional tea quality. There are seven different tea regions as, Dimbula, Nuwara Eliya, Kandy, Uva, Udapussellawa, Ruhuna and Sabaragamuwa in Sri Lanka. Nuwara Eliya tea infusion in the cup is the lightest of all the types of Sri Lankan tea with the delicately fragrant flavor. WU 3 agro ecological region consider as the Nuwara Eliya region. It is located at an elevation of approximately 6000 ft above the sea level where rainfall is modest except during the dry season. This unique climatic condition of the region produces the tea that is unique

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in quality and finest. Combined with low temperature, this produces teas of wonderful bunch.

The objectives of this study are to develop a data base on the chemical, physical and sensory parameters of Nuwara Eliya region and to correlate these parameters to each other if there are any. Also these data can be used to produce specialty tea with unique characters and identify the factors affecting the different qualities of black tea in different regions.

Methodology

The research was conducted at the Biochemistry Division of Tea Research Institute of Sri Lanka, Talawakelle. Samples were collected from the three different factories (Estate 1, Estate 2 and Estate 3) in WU 3 region, which has nearly similar manufacturing dates and 4 grades such as BOP, BOPF, PEKOE and Dust 1from each factory.

Physical quality parameters of the black tea samples were analyzed based on the particle size, color of the particles, appearance, texture, and the brew color of the black tea in fusion.

Highly trained sensory panelists were used to analyze the attributes such as infused leaf, color of brew, strength, and flavor of black tea samples.

Chemical parameters of the Black products were analyzed according to the methods given in the Table 1.

Table 1: Measured chemical parameters and used methods.

Parameter	Tested Method
Moisture content	ISO 18134-1
Water extract %	ISO 9768 (% m/m 32 min)
Total Polyphenol Content	ISO 14502-1 (minimum11)
Individual Catechin content	ISO-14502-2
Caffeine Content	ISO-14502-2
Amino acid content	Ninhydrin Method (Yemm and Cocking, 1955)
Fluoride ion content	Fluoride Ion Selective Method (Giljanovic et al., 2012),
Aluminum ion content	Spectrophotometric Method (Michael et al., 2012),
TF/TR content	Spectrophotometric Method (Roberts and Smith, 1963)
Antioxidant activity	DPPH Assay (Blois ,1958)
Aroma (volatile) compounds	Gas chromatography (Yamanishi et al, 1989)

Two-way ANOVA was done to detect the variation of tea among the estates and different grades with chemical composition by Minitab 17 software. Correlation analysis was conducted by using Non- parametric correlation (Spearman's rank correlation) to identify the correlation of the sensory parameters with the chemical composition.

Results and Discussion

All the samples have the typical light brown color. When comparing with the other samples Pekoe samples were consisted of lighter infusion color. Particle sizes of the samples were varied between estates and were in standard order Pekoe, BOP, BOPF and Dust 1. According to the quality scores given for infused leaf, Color of the infusion, Strength of the infusion and quality, Estate 1 samples were found to be higher in values comparatively.

Table 2: Chemical composition ranges of Nuwara Eliya regional black tea.

Chemical composition	Range
Moisture content	4.07±0.11 - 6.20±0.07 %
Water Extract	38.32±0.03 - 46.65±0.06 %
Total amino acids	0.71±0.09 - 3.35±0.32 %
Total polyphenol	13.85±3.39 - 24.45±1.13 %
Aluminum	0.0001 - 0.0003 %
Fluoride	2.27±0.05 - 4.03±0.05 mg/l
Theaflavin (TF)	0.25±0.01 - 0.51±0.00 %
Thearubigins (TR)	9.35±0.02 - 14.70±1.24 %
Caffeine	1.92±0.05 - 4.25±0.24 %
Antioxidant Activity	7.71±0.04 - 2.66±0.06 %
Volatile compounds	
Linalool	7.66±1.10 - 22.48±1.31 ppm
Pentanal	17.61±1.93 - 40.29±5.66 ppm
trans-2-hexenal	7.12±0.65 - 21.08±1.52 ppm
cis-3-hexenal	5.13±1.30 - 13.591.03± ppm
Methyl Salicylate	4.30±0.61 - 6.51±0.21 ppm
2-Phenyl ethanol	0.97±0.04 - 3.00±0.00 ppm

(Data represented as mean±SE (n=3) in dry weight basis)

Moisture content of the tested black tea samples were varied as 4.07% to 6.20%. Samarasingham (1990) found that an increase of moisture content is related with quality loss of the tea. Othieno and Owuor, (1984); Robinson and Owuor, (1993) recommended that the moisture content of the teas must be controlled to lie under 6.5% which complies with the present study. There is no significant variation among the estates (0.842) and grades (0.505).

Total amino acid content (0.094, 0.145), total polyphenol content (0.248, 0.781), total catechin content (0.133, 0.551), water extract (0.06, 0.34), TF content (0.099, 0.074) didn't show any significant difference among the estate or grades. Whereas the Fluoride ion content (0.004, 0.014) and Aluminum Ion content (0.007, 0.012) showed a significant difference between estate and grades both. Antioxidant activity (0.00, 0.808)

and Caffeine content (0.014, 0.2285) showed a significant difference among estates but not among grades and vice versa for TR (0.103, 0.019). The water extract in black tea must be more than or equal to 32% (ISO 3720, 1986). In this study the water extract of all the samples were obeyed the standard. Compared with the water extract of black teas produced in other countries (Owuor *et al*., 1986), India 36.89 to 41.95%, Kenya 44.12% and China 36.79%, it can be seen that the average water extract in the black tea samples from Nuwara Eliya regional teas is relatively similar.

According to the World Health Organization recommendations, the recommended dietary allowance (RDA) of fluoride (F^-) for children is two milligrams per day, and for adult four milligram per day. When consider the fluoride content of all the tested samples, even consumption five cups of black tea per day will not exceed the RDA, and it can be confirmed that by consuming the tested samples would not cause such health risk. Sinija and Mishra, (2008) stated that the variation of polyphenol content in black tea could be due to the variation of the chemical composition of the tea leaves with the season, climate, horticultural practice, and position of the leaf on the harvested shoot. As the samples are from the same region the difference is less and complies with the ISO requirement.

As it is stated by Kottawa-Arachi *et al.* (2011), free amino acid content of black tea varied from 0.72 to 2.67% in the wet season and 0.86 to 2.00% in the dry season which complies with the results. As recommended by WHO, the recommended dietary allowance (RDA) of Aluminum for children is 2-6 mg/day and for adult 6-14 mg/day and the values of present study don't exceed the requirement.

The major antioxidants in tea are Thearubigins, Theaflavins, Catechins, Flavanols for example Kaempferol, Myricetin, Quercetin; Oxyaromatic acids; Flavones for example Apigens; Gallic acid derivatives such as Tannins, etc. (Yashin *et al*.,2011). The tested samples were in considerable level, according to previous studies related to black tea (Cabrera et al., 2013). The variation between estates may due to the micro climatic factors as these affect on the concentrations of above compounds.

The content of the Theaflavin in black tea was reported to be in the range of 0.3-1.8%, on dry weight basis (Modder, 2003) which complies with the results of present study and TR too. But their ratio is high confirming the semi-fermenting tea processing method in Nuwara Eliya region. Normally Nuwara Eliya tea is popular for its fragrance and volatiles are the responsible chemicals for this. Trans-2-hexanal, Cis-3-hexanal, Methyl Salicylate and 2-Phenyl Ethanol there was no significant difference between the estates and grades. But Pentanal and Linalool has a significant difference between estates and for grades only Pentanal shows a difference. When consider comparatively, Linalool, Pentanal and Trans-2-hexanal has high concentrations.

Caffeine and total Catechins play a significant role in quality of black tea. Earlier studies showed that caffeine content and catechin content of black teas was affected by clone, season and stage of plucking (Hara et al., 1995; Harbowy and Balentine, 1997; Owuor and Chavanji, 1986). According to the ISO 14502-2 standard the minimum

percentage of total catechins should be 7% and the samples showed compliance with this except one sample.

In order to determine the relationship among chemical parameters and the sensory attributes, correlation analysis was done among the results of sensory evaluation and chemical composition. Significant positive correlations have been seen between TPP content and quality, (0.446) and AOA and quality (0.580) and WE and quality (0.304). Also a significant negative correlation have been between quality and TF content, (-0.473), TAA and quality (-0.178) and also among quality and color (-0.321).

The content of individual TF shows a significant correlation with tea taster scores and value (Erol *et al*., 2010). When current results were considered, sensory characters such as infusion color and strength have been showed a significant positive relationship with TF where, lower the TF content, higher quality of the brew of the black tea. The infusion color is positively correlated with the TF content, (0.736). According to the literatures, AOA should have positive correlation with quality. In the current study also, a positive was found between the AOA and quality. AOA is positively correlated with the TC. Also TPP is positively correlated with TC. TF has considerable positive correlation with color of brew. TR has shown positive correlation with color of black tea brew.

Conclusions

When the overall quality is considered, Nuwara Eliya tea complies with the requirements. The results of the study show the relationship between color and the TF content. Nuwara Eliya tea can be promoted as a specialty tea with health benefits using above results.

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