"Studies on the preparation and evaluation of functionally enriched value added product from malta fruit (*Citrus sinensis* L Osbeck)"

Thesis

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Place: Bharsar, Pauri Garhwal.

Date: July 2016

(Beena Pathak)

CERTIFICATE

This is to certify that the thesis entitled **"Studies on the preparation and evaluation of functionally enriched value added product from malta fruit (*Citrus sinensis* L Osbeck)"** submitted in partial fulfilment of the requirements for the degree of **Master of Science (Food Technology)** with major in **Food Technology** of the College of Horticulture**,** VCSG Uttarakhand University of Horticulture and Forestry, Bharsar is a record of *bonafide* research carried out by **Beena Pathak Id. No. 13181** under my supervision and no part of the thesis has been submitted for any other degree or diploma.

The assistance and help received during the course of this investigation have been acknowledged.

Prof. B. P. Nautiyal
Chairman
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CERTIFICATE

We, the undersigned, members of the Advisory Committee of **Ms. Beena Pathak Id. No. 13181,** a candidate for the degree of **Master of Science (Food Technology)** with major in **Food Technology** agree that the thesis entitled **"Studies on the preparation and evaluation of functionally enriched value added product from malta fruit (*Citrus sinensis* L Osbeck)"** may be submitted in partial fulfilment of the requirements for the degree.

Prof. B. P. Nautiyal
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ABBREVIATIONS

@	:	At the rate
ß	:	Beta
cm	:	Centimeter
CRD	:	Completely randomized design
CD	:	Critical difference
et al	:	Co-workers
°B	:	Degree Brix
^{0} C	:	Degree Celsius
etc	:	Etcetera
g	:	Gram
hrs	:	Hours
Kg	:	Kilogram
Mg	:	Milligram
ml	:	milliliter
mm	:	Millimeter
no.	:	Number
viz	:	Namely
OAA	:	over all acceptability
ppm	:	Parts per Million
%	:	Per cent
$\pm$	:	Positive negative
pH	:	Potential of hydrogen
PDA	:	Potato dextrose agar medium
RBD	:	Randomized block design
RH	:	Relative humidity
Rs	:	Rupees
RTS	:	Ready to serve
RTD	:	ready to drink
SV	:	Source of variation
i.e.	:	That is
T	:	Treatment
TSS	:	Total soluble solid

INTRODUCTION

CHAPTER-1
INTRODUCTION

Malta fruit, a tasty and juicy fruit belonging to the family Rutaceae is botanically known as *Citrus sinensis* (L. Osbeck). Citrus is widely grown tropical, subtropical and Mediterranean regions. It is one of the widely grown fruit in almost 80 countries of the world. Sweet orange tree are widely cultivated for its appetising juice and medicinal value. It is one of the excellent source of ascorbic acid, a powerful natural antioxidant which builds the body immune systems. Important phytochemicals like liminoids, synephrine, hesperdin flavonoid, polyphenol, pectin and sufficient amount of folacin, calcium, potassium, thiamine, niacin and magnesium are also present (Angew, 2007). These biologically active compounds prevent arteriosclerosis, cancer, kidney stones, stomach ulcers and reduction in cholesterol level and high blood sugar level which promote human health.

Honey is a sweet, viscous liquid prepared by bees from nectar collected from plant and stored by them for food (White, 1975). It has been used since ancient times by humans and has gained appreciation as the only concentrated form of sweetener available worldwide (FAO, 1996). Traditionally, its use in food has been as a sweetening agent and due to the variation of botanical origin, honey differs in appearance, sensory perception and composition. Honey is a natural biological product that comprises of simple sugars mainly glucose and fructose (70–80%), water (10–20%), and other minor constituents such as organic acids, mineral salts, vitamins, proteins, phenolic compounds and free amino acids (Ouchemoukh *et al*., 2007). It is highly liked by consumers for its neutraceutical value, characteristic flavour, sweetness, and texture (Subramanian *et al*., 2007).

Table sugar (primarily sucrose) on the other hand has been a part of the daily diet for hundreds of years, but research is now suggesting that more sugar intake can be detrimental to our health. In particular, excessive consumption of table sugars with high glycemic index (GI) has been shown to cause overeating and weight gain. While, honey is known for both its nutritional value and medicinal properties (Juszczak and Fortuna, 2006). It is a complex mixture of carbohydrates and contains organic acids and some amino acids, as well as certain micro- and macro-elements, and it is a rich source of many biologically active compounds (Juszczak, 2006; Ahmed, 2007). It has been recognised as having a number of beneficial health properties, including slower uptake into the

bloodstream, a pharmacological action of reducing blood glucose levels and a high level of bio-available antioxidants, all of which mean that honey could be more beneficial to health than sucrose in the diet (Sethi, 2004).

Honey can also prevent deteriorative oxidation reactions in foods such as enzymatic browning of fruit and vegetables (Chen *et al*., 2000), lipid oxidation in meat (Gheldof and Engeseth, 2002; Nagai *et al*., 2006) and inhibit the growth of foodborne pathogens and food spoilage organisms (Mundo *et al*., 2004; Taormina *et al*., 2001). This property of honey is due to the quantity of antioxidants which vary widely according to the flora and geographical locations. However, processing, handling and storage of honey may influence its quality and composition (Gheldof and Engeseth, 2002).

Food preservation is the process of treating and handling food to stop or slow spoilage and thus allow for longer storage. Preservation of juices involves preventing the growth of bacteria, yeast, fungi and other microorganisms. Food is an essential thing for human survival. Except our own garden plants, all the food used today has some preservatives. Apart from thermal pasteurization, some chemical preservatives are also widely used for the extension of the shelf-life of fruit juices and beverages. The most commonly used preservative are potassium sorbate and sodium benzoate.

An experiment was conducted to study the shelf life of value added product of malta fruit by using different sweetening agents, preservative methods, at different storage conditions ambient and refrigerated temperature. The different value added product prepared in the study was malta fruit RTS, malta fruit iced tea, while, honey and sugar were used as sweetening agents. Little work has been done on the preparation of value added product of malta fruit using honey; henceforth the present study has been carried out with the following objectives:

i) To standardize the optimum quantity of sweetening agents for the preparation of malta RTS and malta iced tea.
ii) To maintain the quality of standardized beverages by the application of different preservative techniques.
iii) To evaluate the effect of storage on quality of standardized Malta beverages.

REVIEW OF LITERATURE

CHAPTER-2
REVIEW OF LITRATURE

Malta is popularly grown at medium to higher hills of Uttarakhand and other parts of India and is popularly known as sweet orange. The fruit is highly nutritive and is the good source of vitamin 'C'. The fruit is also rich in minerals mainly calcium, phosphorus and iron. Many processed products are prepared from sweet oranges to utilize its nutritional and medicinal qualities.

Fruits contain several health-promoting factors including fibers and high concentrations of phenolic acids, flavonoids, vitamins and minerals. Phenolic acids and flavonoids although, not being essential for the survival but may protect us against a number of chronic diseases over the long term. The human diet contains important micronutrients namely vitamins C and E, carotenoids and flavonoids, essential for maintenance of human health. A brief review of the relevant literature on different aspects of present studies has been discussed herewith under the following headings and sub-headings:

2.1 Malta fruit (*Citrus sinensis* L. Osbeck)

Malta fruit is commonly known as sweet orange. It belongs to family Rutaceae and growing in Nigeria and many other tropical and subtropical regions (Piccinelli *et al.*, 2008). The fruit is available in November, December, January and February months.

Clemens and Dubost (2008) reported that malta fruits are valuable sources of vitamins and minerals which contribute to overall dietary quality and thus may reduce the risk of chronic diseases. In addition, they may be a major source of numerous polyphenols, particularly flavonoids in the diet. The fruit is also rich in vitamin A and other flavonoid antioxidants such as alpha and beta carotenes, beta-cryptoxanthin, zeaxanthin and lutein, compounds that have antioxidant properties. Researchers have found that fruit juice consumption can contribute significantly to adequate intakes of essential nutrients including vitamin C, folate, potassium and magnesium. Consumption of 100 per cent pure fruit juices is an effective approach to meet the current dietary recommendations for fruit intake as well as provide essential nutrients and functional components including

antioxidants. Substantial evidences indicate that 100 per cent pure juice intake can be a part of healthy active life style.

2.1.1 Physico-chemical composition of malta fruit

2.1.1.1 Physical Characteristics

The malta fruits are round, ribbed and pale orange in colour and consist of average fruit weight, diameter, number of segments per fruit, 199.0g, 84.06 mm, and 10 respectively (Syed *et al.* 2012). They also found that malta juice contains a good amount of TSS and pH as 10^{o}B and 3.7, respectively.

2.1.1.2 Chemical Characteristics

The chemical composition of malta fruits influenced by environmental factors as like other seasonable fruits and vegetables. The fruit is rich source of vitamin C, sufficient amount of folacin, calcium, potassium, thiamine, niacin, magnesium etc. A complete list of these are illustrated in Table 2.1.

Table 2.1 The nutritional composition of malta fruit (USDA, 2014)

Composition	**Amount**
Energy	197 kJ (47 kcal)
Sugars	9.35 g
Potassium	181 mg (4%)
Dietary fibre	2.4 g
Protein	0.94 g
Water	86.75 g
Vitamin A	11 μg (1%)
Vitamin C	53.2 mg (64%)
Niacin (vit. B3)	0.282 mg (2%)
Thiamine (vit. B1)	0.087 mg (8%)
Calcium	40 mg (4%)
Magnesium	10 mg (3%)
Folate	30 μg (8%)

2.1.2 Medicinal properties of malta fruits

2.1.2.1 Antioxidant and anti-inflammation properties

Ejaz *et al*. (2006) reported that citrus as a good source of ascorbic acid which known as powerful natural antioxidant, bioactive components, like carotenoids and flavonoids that prevent cancer and degenerative diseases. Consumption of such fruits improves body immunity against infectious agents and scavenging harmful, pro-inflammatory free radicals from the blood. Sweet orange contains a variety of phytochemicals like hesperetin and narigenin. Naringenin has a bioactive effect on human health as antioxidant, free radical scavenger, anti-inflammatory, and immune system modulator.

Malta fruit also known to contain anti-inflammatory properties. Tripoli *et al.* (2007) found that citrus flavonoids contain compounds with anti-inflammatory activity due to the presence of regulatory enzymes (protein kinase C, phosphodiesterase, phospholipase, lipoxygenase, and cyclooxygenase) that control the formation of the biological mediators, responsible for the activation of endothelial cells and specialized cells involved in inflammation. Flavonoid inhibition of the immune and inflammation responses can be associated with their inhibition of these enzymes.

In a study by Cha *et al*. (2001) observed that oranges contians about 170 phytonutrients and over 60 flavonoids with anti-tumor, anti-inflammatory, blood clot inhibiting and antioxidant properties. All these properties help to promote overall health.

2.1.2.2 Anti-obesity

Walton *et al*. (1945) reported that sweet oranges contain low calories and no saturated fats or cholesterol, but is rich in dietary fibre, pectin which is very effective in persons with obesity. Pectin as bulk laxative protects the mucous membrane from exposure to toxic substances, as well as by binding to cancer causing chemicals in the colon. Pectin has also been shown to reduce blood cholesterol levels by decreasing its re-absorption in the colon by binding to bile acids in the colon.

2.2 Sweetening agents

A sugar substitute is a food additive that provides a sweet taste to products. Some sugar substitutes are natural and some are artificial also. A brief review of the relevant

literature on used sweetening agents in present studies has been discussed here with under the following headings and sub-headings:

2.2.1 Table sugar

Cardello and Damasio (1997) reported that Sucrose is mainly used for the sweetening of drinks. Alternative sweeteners can avoid problems with dental decay and other health risks associated with the excessive consumption of caloric sweeteners, such as sucrose In India use of artificial and natural sweeteners such as honey is limited but being economical, these are helpful to control obesity and other potential diseases. A variety of artificial sweeteners are available in the market like, aspartate, cyclamate, sucralose and saccharin *etc*. These are the non-nutritive sweeteners which are not metabolized by the body and do not contribute any energy or calories to the diet but their use is restricted because of health hazards (American Dietary Association, 2004). However, nutritive natural sweeteners like honey can replace artificial sweeteners in the energy diet drinks with health safety.

2.2.2 Honey

The Codex Alimentarius Commission (1981) defines honey as the natural sweet substance produced by honeybees from nectar of blossoms or from secretions of living parts of plants or excretions of plant sucking insects on the living part of plants, which honey bees collect, transform and combine with specific substances of their own, store and leave in the honey comb to ripen and mature. Honey is highly nutritious and is a good source of minerals and vitamins; further it contains antioxidants which destroy free radicals and delay ageing. It is considered as a safe and wholesome food for old, children and adults. Honey is primarily made of water and carbohydrates.

Bogdanov *et al.* (2008) reported that honey is an energizer, helping workers and athletes overcome fatigue and regain energy. Children, young and old can alike take honey, without worrying any side effects. It is the oldest and only available unique natural sweetener to mankind and is the last of natural unprocessed food to be consumed.

Ouchemoukh *et al.* (2007) concluded that when honey derived from the nectar of flowers it known as nectar or blossom honey and can be further categorized as mono/unifloral honey and multifloral honey whereas honey produced from honeydew is known as honeydew honey.

Gheldof *et al.* (2002) reported that the amount and type of these antioxidant compounds depend largely upon the floral source or variety of the honey. In general, darker honeys have been shown to be higher in antioxidant content than lighter honey.

Bath and Singh (1998) reported that the appearance of honey plays an important role on its commercial acceptance. The consumers demand a free flowing, non-crystallized and quality product. Freshly extracted honey is liquid, however, it may crystallize during storage depending on several factors such as botanical flora and geographical location, temperature, moisture and sugar content. In order to delay the natural crystallization process and ensure stability during its shelf-life, fresh honey is usually processed before being packed and stored, with the purpose of dissolving sugar and destroying yeasts. An attempt has been made in this chapter to review the current status of knowledge pertaining to quality of honey, its processing and storage under the following heads.

2.2.2.1 Honey composition

Honey is comprised primarily of fructose (38.2%), glucose (31%) and water (17.1%). However, the remaining 13.7% of honey provides manufacturers with some remarkable benefits. Among those components are a variety of other sugars, enzymes, amino acids, antioxidants, vitamins and minerals. It is this unique blend that gives honey its functional advantages. Honey, on average, has a pH of 3.9.

Mundo *et al.* (2004) reported that acidity can work to enhance flavors, inhibit mold and bacteria growth and extend the shelf-life of a variety of products. The amino-acids that give honey its low pH also serve as precursors to honey antimicrobial capabilities. Within beverages, honey's composition plays a critical role. Beverage manufacturers need to accommodate honey's sweetness levels, as well as the enzymes and potentially active bacteria that the ingredient can impart to an end- product.

Doner, (1977) studied that floral honey is a high sugar food product made by honey bees from the nectar of flowers. The major constituents of honey are fructose (37.5g/100g), glucose (28.1g/100g) and water (18.4g/100g), with the remainder made up of a complex mixture of mono-, di- and tri- saccharides. The main sugars found in honey are fructose and glucose, usually in a ratio of 2.1:1.0, these two sugars together account for major percentage of the total carbohydrate content.

Doner (1977) reported that water content is the main factor that influences quality and storage life of honey. It is also influenced by the time of extraction and ripening process from the comb (Chung *et al*., 1984). Water activity of honey varies between 0.5 (16% moisture) and 0.6 (18.3% moisture) in the 40-100 °F (4-37 °C) temperature range.

Mato *et al*. (2003) reported that honey contains about 30 organic acids. Although the major contributor is gluconic acid in equilibrium with its lactone. This acid is produced by the activity of the enzyme glucose oxidase, with its resultant concentrations ranging from 0.23–0.98 per cent (White, 1975).

HMF (5-hydroxymethylfurfuraldehyde) measurement is used to evaluate the quality of honey; generally it is not present in fresh honey, its content increases during processing and storage. Honey processing, requires heating both to reduce viscosity, and to prevent crystallisation or fermentation (Bath and Singh, 1998).

The macro and trace elements in honey depend on its botanical and geological origin (Bengsch, 1992). Different trace elements (Al, Ba, Sr, Bi, Cd, Hg, Pb, Sn, Te, W, Sb, Cr, Ni, Ti, Co, Mo) and mineral (P, S, Ca, Mg, K, Na, Zn, Fe, Cu, Mn) have been systematically investigated in botanically and geologically defined honey by various authors (Stocker *et al*., 2005). The vitamin content in honey is low. Vitamins such as phyllochinon (K), thiamin (B1), riboflavin (B2), pyridoxin (B6) and niacin are reported in honey but in general the amount of vitamins and minerals is small and the contribution of honey to the RDI(recommended daily intake) of the different trace substances is negligible (Bogdanov *et al*., 2008).

Azeredo *et al.* (2003) reported that honey contains roughly 0.5 per cent proteins, mainly enzymes and free amino acids. Variation in protein content has been reported in honey due to different floral sources. The high protein contents of honey were more than 1000 μg/g.

Iglesias *et al.* (2004) the amount of total free amino acids in honey corresponds between 10 and 200 mg/100g, with proline as their major contributor, corresponding to around 50% of the total free amino acids.

Gheldof *et al*., (2002) reported that honey contains a variety of phytochemicals (as well as other substances such as organic acids, vitamins, and enzymes) that may serve as sources of dietary antioxidants. The amount and type of these antioxidant compounds depend largely upon the floral source or variety of the honey and also reported that generally darker honeys have been shown to be higher in antioxidant content than lighter

honey. They also reported that the antioxidant capacity of honey has been attributed to several factors including α-tocopherol, polyphenolics, organic acids, ascorbic acid, β-carotene and enzymes. They studied a chromatographic analysis of the phenolic fractions of 8 honeys of different floral sources suggested that most honeys have quantitatively different phenolic profiles, and that a linear correlation exists between phenolic content and oxygen radical absorbance. Honey can be used as a healthy alternative to sugar and consumed in substantially smaller portions than other dietary antioxidant sources, thereby serving as a supplementary source of antioxidants. However, if even some of the sugar in the typical diet were to be replaced with honey, this could result in a significant increase in the intake of antioxidants.

2.2.2.2 Consumer's acceptability of honey:

The aroma and flavour of honey are its most important characteristics from the bee keepers and consumer's point of view.

Kaushik *et al.* (1993) concluded that while studying the "Effect of storage conditions on the quality of honey" stored at 40±10C, at room temperature and at 5±10C and reported that maximum deterioration in colour occured in honey stored at 40±10°C, followed by honey stored at room temperature and at 5±10C.

Gupta *et al.* (1995) reported that storage of honey at 40°C resulted in deterioration of colour and found that addition of potassium metabisulphite reduced the darkening effect in honey stored at room temperature. Unheated honey stored at 50°C was found to be the best.

2.3 Value added products from malta and honey

Nagai *et al.* (2006) suggested that honey is a sweet and viscous substance produced by the honeybees from the nectar of floral plants. It is produced in almost every country of the world and is a very important energy food. It is used as an ingredient in hundreds of recipes of food, for sweetness, flavour, colour, caramelisation and viscosity. Due to various favourable properties it is used as an additive in a variety of foods and beverages. The antibacterial effect of honey counteracts microbial spoilage of food, e.g. of meat.

Singh *et al.* (1988) suggested that honey is the nectar and saccharin exudation of plants, gathered, modified and stored in the combs by honey bees. In India, the consumption of honey is mainly restricted for medicinal purposes though it has been used

in a number of preparations elsewhere. Apart from the food value, it is also known for its medicinal value in burns, infections, wounds and its antimicrobial properties. The use of honey in beverage and baked food industry can be given impetus by determining suitability for use in products already being produced and developing newer honey products. According to FDA standards, of identity honey is the optimum sweetening agent for fruit butters, jellies, jams and preserves providing it either to be the sole ingredient or represent 20 per cent of solid mixtures with certain other optional sweetness. It is unlikely that any appreciable amount of honey is presently in such use.

Bogdanov (2010) found that the nectar utilizing honey as a substitute of sugar was a viable alternative for opening up new vistas for surplus honey utilization in food industry is in baking, cereal and the confectionary industries

2.3.1 Ready-to-Serve

Fladae *et al.* (2003) reported that citrus fruits have attractive bright colour, appealing taste and flavor. The composition of citrus fruit juice is beneficial with respect to its mineral and ascorbic acid contents. There is a great potential to use this fruit in value added products such as diet drinks.

Nchez-moreno *et al.* (2003) studied that citrus drinks are probably the most recognized and globally accepted fruit drinks Sucrose is mainly used for the sweetening of these drinks. Fruit beverages are highly nutritive, refreshing, thirst quenching, appetizing and easily digestible. Squash, nectar and other forms of ready to serve (RTS) beverages are prepared from different fruits like mango, apple, papaya, pineapple, guava, citrus, grapes and other minor fruits (Deka *et al*., 2004).

Sweetening agent used in fruit nectars is sugar, but honey can also be used as a substitute for sugar for making different vitaminised drinks and other nutritious beverages. The beverages made with honey have a significant flavour and nutrition than that of sugar products (Singh *et al.*, 1998).

2.3.2 Iced Tea

Tea, a product made up from leaf and bud of the plant *Camellia sinensis*, is the second most consumed beverage in the world, well ahead of coffee, beer, wine and carbonated soft drinks. Rao *et al.* (2011) Tea is the most consumed drink in the world after water. Green tea is a ‘non-fermented’ tea, and contains more catechins, than black tea or

oolong tea. Catechins are *in vitro* and *in vivo* strong antioxidants. In addition, its content of certain minerals and vitamins increases the antioxidant potential of this type of tea. Since ancient times, green tea has been considered by the traditional Chinese medicine as a healthful beverage. Recent human studies suggest that green tea may contribute to a reduction in the risk of cardiovascular disease and some forms of cancer, as well as to the promotion of oral health and other physiological functions such as anti-hypertensive effect, body weight control, antibacterial and antivirasic activity, solar ultraviolet protection, bone mineral density increase, anti-fibrotic properties, and neuroprotective power. Increasing interest in its health benefits has led to the inclusion of green tea in the group of beverages with functional properties. However, although all the evidence from research on green tea is very promising, future studies are necessary to fully understand its contributions to human health, and advise its regular consumption in Western diets, in which green tea consumption is nowadays limited and sporadic.

Tea contain caffeine (1 to 5%) of its dry weight depending on type, brand (Bennett and Bonnie, 2001) and brewing method (Hicks *et al*., 1996). Caffeine belongs to a family of naturally occurring components known as xanthenes which are known to be stimulants. In addition to its popularity as a refreshing beverage, tea is now receiving scientific attention because the flavanols it contains appear to have beneficial effects on human health. (Arts *et al*., 2002; Dora *et al*., 2003; Geleijnse *et al*., 2002; Hakim *et al*., 2003; Higdon and Frei, 2003; Lambert and Yang, 2003; McKay and Blumberg, 2002; Sun *et al*., 2002; Vita, 2003; Wu *et al*., 2003; Zhang *et al*., 2002). Black teas have a characteristic profile of flavonoids. The dimeric theaflavins and polymeric thearubigins are present in large quantities and virtually exclusively in black tea.

Due to a connection with the health promoting properties of brewed tea, the consumption of ready-to-serve tea beverages has also increased significantly (Starling, 2007).

Dinsmore and Nagy, (1971; 1972) observed higher storage temperature yields more furfural while fresh non-pasteurized juice contains no furfural. A good correlation between furfural accumulation and the appearance of off-flavours was found in citrus juices.

Due to its sweetness, color and flavor, honey is often used as a replacement for sugar, as a component, or as a natural protective agent, in a lot of industrially produced foodstuffs (Pyrzynska and Biesaga, 2009). Its composition depends on the kind of flowers (plants) from which honeybees collect the nectars, and other factors (such as

environmental conditions and climate, for example). Besides sweeteners, honey contains numerous other components, of which many, including the polyphenols which exhibits antioxidative properties (Pyrzynska and Biesaga, 2009; Schramm *et al.*, 2003; Scalbert *et al.*, 2005). The average intake of sweeteners per person is estimated to be more than 70 kg per year, replacements of the traditional sweeteners in some foodstuffs with honey can result in improved antioxidative defense for healthy people (Bogdanov *et al.*, 2008; Schramm *et al.*, 2003).

2.4 Method of preservation

Clifford (2000) reported that food processing such as pasteurization plays an important role in the destruction of bioactive compounds. Processing can alter and often damage fruit antioxidants. Maceration, heating and various separation steps can result in oxidation, thermal degradation, leaching and other events that lead to lower level of antioxidants in processed foods compared with fresh. This is particularly true in case of vitamin C and phenolic antioxidants. It is well known that phenolic contents present in fruits and vegetables easily undergo enzymatic or chemical oxidation during processing and storage.

Murakami *et al*. (2002) reported that during the processing of foods, various transformations of phenolics occur to produce yellowish to brownish pigments. These chemical changes has been attributed to post-harvest treatments which could lead to the formation of various compounds having antioxidant and pro-oxidant properties and could exert complex effects on the antioxidant properties of phenolic compounds.

Vikram *et al.* (2005) Reported that thermal pasteurization can produce undesirable quality changes like loss of colour and flavour in addition to reducing the nutritional quality of juice.

2.5 Storage study of products

Among factors involved in quality loss are Maillard reaction and loss of ascorbic acid. Ascorbic acid degradation depends on processing parameters, type of packaging material used, handling and storage conditions. Formation of furfural and 5-hydroxymethyl-2-furfuraldehyde usually accompanies degradation of ascorbic acid.

Furfural accumulation parallels with non-enzymic browning in the preparation of orange powder (Tatum *et al.,* 1967; 1969).

Lanjhiyana *et al*. (2010) have recorded slight increase in TSS in lime-ginger RTS/blended RTS, during storage and he concluded that it might be due to the conversion of polysaccharides like pectin, cellulose, starch etc into simple sugars.

Saravanan *et al*. (2004) also observed slight increase in TSS during storage in his study on Standardization of recipe for papaya nectar, and concluded that might be attributed to breakdown of the complex carbohydrates into simple soluble carbohydrates.

Mehta *et al.* (1983) reported decline in acidity during storage of citrus juice and concluded that titratable acidity decrease in juice could be attributed to chemical interaction between organic constituent of juice induced by temperature and action of enzymes.

Hussain *et al*. (2011) observed decrease in ascorbic acid of the processed products during the storage and concluded that ascorbic acid content in strawberry pulp was affected by treatments *viz*., freezing, heating and accelerated storage. These losses of ascorbic acid were attributed to the effect of processing, storage time and exposure to light.

Das (2009) observed a significant reduction in ascorbic acid content of jamun beverages during storage and he suggested that this decrease may be due to the oxidation of ascorbic acid into dehyroascorbic acid by trapped oxygen. Ahire *et al.* (2010) recorded higher decrease in ascorbic acid content in juices at ambient storage conditions.

Raj *et al*. (2011) reported a significant decrease in the phenolic content of sand pear and apple juice blends during six months storage.

MATERIALS AND METHODS

CHAPTER -3
MATERIALS AND METHODS

The present investigation entitled, "Studies on preparation and evaluation of value added product from malta fruit (*citrus sinensis* L. Osbeck)" was conducted under different experiments in the department of Food Science and Technology, College of Horticulture, VCSGUUHF, Bharsar Pauri Garhwal (Uttarakhand) during the years 2014-2015 and 2015-2016. The material used, experimental details and techniques employed in the investigation are furnished in this chapter.

3.1 Malta fruits

Malta fruits were collected from the surrounding villages of College of Horticulture, VCSGUUHF, Bharsar Pauri Garhwal.

3.2 Ingredients

Table sugar (sucrose), tea leaves and honey were purchased from the local market of Pauri Garhwal, Uttarakhand.

3.3 Detail of experiments

3.3.1 Development of malta honey based products

3.3.1.1 Ready-to-serve malta fruit drink

The malta RTS beverage was prepared according to the minimum specifications prescribed for RTS under FSSAI specifications (FSSAI, 2006) using different sweetening agents *i.e.* honey and sugar and the complete operation followed was shown in Figure 3.1. Further to standardize the best combination of pulp concentration and TSS for malta RTS different combinations treatments were tried as shown in Table 3.1. The prepared malta RTS were packed in previously sterilized glass bottles then subjected to pasteurization or chemical treatment using 120ppm sodium benzoate or a combined treatment of both (pasteurization with 60 ppm sodium benzoate) to maintain the quality during storage of six months. The prepared RTS then stored at ambient (17-25^{o}C) or refrigerated (4-7^{o}C) temperature for six months to assess the feasibility of different preservation methods.

Table 3.1 Different treatment combination of sweetening agent, pulp concentration and TSS for the standardization of malta RTS

Sweetening agents	Pulp Concentrations (%)	TSS (%)
Sugar	10, 12, 14	10, 12, 14
Honey	10, 12, 14	10, 12, 14

Pre-trials were conducted to adjudge the best concentration of juice and TSS of prepared malta beverages through sensory evaluation on hedonic scale (Appendix-I). The best treatment then evaluated for various physico-chemical and antioxidant parameters at three months intervals for a total period of six months.

3.3.1.2 Malta iced tea

Iced tea is a form of cold tea, usually served in a glass with ice. It may or may not be sweetened. The malta iced tea was prepared by standard method as given in Figure 3.2. The malta iced tea was prepared by using Tata premium tea leaves and two different sweetening agents honey and sugar were used. The different Tea concentration used for the preparation of malta iced tea was 0.5, 0.75 and 1%. As per the concentration prescribed the measured quantity of tea was taken in muslin cloth and kept inside the boiling juice for three minutes. After three minutes the muslin cloth containing tea is squeezed and removed. The developed malta beverages were packed in 500 ml capacity glass bottles and stored at two different temperatures viz., ambient (17-25^{o}C) or refrigerated (4-7^{o}C) and then subjected to pasteurization or chemical treatment using 120ppm sodium benzoate or a combined treatment of both (pasteurization with 60 ppm sodium benzoate) to preserve the quality during storage.

Organoleptic evaluation was carried out in similar manner as done in case of malta RTS to adjudge the best concentration of tea for the preparation of malta iced tea. The best concentration was used for further storage study of six months at an interval of three months.

3.4 Physico-chemical characteristics

3.4.1 Physical parameters

3.4.1.1 Total soluble solids (TSS)

TSS was determined with the help of hand refractometer of range 0-32^{o}B (Model ERMA). The TSS was recorded by placing 1-2 drops of extract on the prism of a hand refractrometer. The results were expressed as ^{o}Brix (Ranganna, 2007) using reference table for temperature correction.

3.4.1.2 pH

pH was measured with the help of digital pH meter standardized with buffer solution.

3.4.2 Bio-chemical parameters

3.4.2 .1 Titratable acidity

Titratable acidity was estimated by titrating a known volume of the sample against standard 0.1 N NaOH solution using phenolphthalein as an indicator up to the end point (pink colour). The titratable acidity was expressed as per cent citric acid (AOAC, 2004).

$$\text{Titratable acidity (\%)} = \frac{\text{Titre} \times \text{normality of alkali} \times \text{volume made up} \times \text{equivalent weight of acid}}{\text{Volume of sample taken} \times \text{volume of aliquot taken} \times 1000} \times 100$$

3.4.2 .2 Sugars

A known weight of sample (25 g/ml) was taken in a 250 ml volumetric flask and 100 ml water was added to it. Solution was neutralized with 1 N NaOH and 2 ml of 45% lead acetate was added to it and kept for 10 min. Excess of lead acetate was removed from the sample by using 2 ml of 22% potassium oxalate in 250 ml volumetric flask. After diluting it up to the mark, the solution was filtered and clear filtrate was taken to estimate reducing sugars by titrating against a known quantity of Fehling's A and Fehling's B solution using methylene blue as an indicator (Lane and Eynon, 1923). Reducing sugars were estimated as per cent and calculated as given below:

a) % Reducing sugars = $\frac{\text{mg of invert sugar x dilution}}{\text{Titre x weight or volume of sample}}$ x 100

Total sugars were estimated by adding 5 g of citric acid to 50 ml filtrate from the reducing sugar estimation and heating it for 10 min., then neutralizing the sample with 1N NaOH using phenolphthalein as indicator and making volume 250 ml in volumetric flask with distilled water. The total sugars were estimated as per cent and calculated as given as under:

b) % Total sugars as invert sugars = Calculated as in (a) making use of titre value as obtained in the determination of total sugars after inversion

c) % Reducing sugars = (% total invert sugars - % reducing sugars) x 0.95

d) % Total sugars = (% reducing sugars + % sucrose)

3.4.3 Antioxidant parameters

3.4.3.1 Ascorbic acid

Ascorbic acid content was determined as per AOAC (2004) method using2, 6-dichlorophenol indophenol dye. A known volume of the sample extracted in 3% meta phosphoric acid was titrated with dye to pink colour as end point. Results were expressed as mg per 100 g of sample and calculated by using the following formula:

$$\text{Ascorbic acid (mg/100 g)} = \frac{\text{Titre} \times \text{dye factor} \times \text{volume made up}}{\text{Aliquot of extract taken} \times \text{weight of sample taken}} \times 100$$

3.4.3.2 Total phenols

The amount of total phenols in the sample were determined with the Folin-Ciocalteu reagent according to the method of Bray and Thorpe (1954) using catechol as a standard. One gram of sample was taken and ground with 10 ml of 80 per cent ethanol in pestle and mortar, and centrifuged for 20 min at 1000 rpm and filtered. Filtrate was evaporated in oven up to dryness and residue was dissolved in 5 ml distilled water. 0.2-2.0 ml aliquot was taken in separate test tubes and volume was made up to 3 ml with water. Then 0.5 ml Folin-Ciocalteu reagent was added. After 3 min 2 ml of Na_2CO_3 (20%) was added and mixed. Test tubes were placed in boiling water bath for one min and then cooled. Optical density of the sample was recorded at 650 nm with the help of UV-VIS spectrophotometer (Model Shimadzu, Japan). The concentration was determined as per the standard procedure from the standard curve. The standard curve was prepared using different concentrations (8-32 μg/mL) of catechol and results were expressed as mg per 100 g on fresh weight basis.

3.4.3.3 β-carotenoids

According to the method given by Kemmerer and Fraps (1943), 25 gm of sample was weighed and ground in a pestle and mortar with acetone and filtered through cotton into a conical flask. Extraction was continued till the sample became colourless. Filtrate was then transferred to a separating funnel and 10-15 ml of petroleum ether was added. The coloured liquid phase was transferred into the petroleum ether phase which was filtered through anhydrous Na_2SO_4 and final volume was made (25ml). Colour was measured at 452 nm. Acetone 3 % in petroleum ether was used as a blank.

Standard curve was prepared using 25mg of β-carotene weighed and dissolved in 2.5ml of chloroform and volume made up to 250 ml with petroleum ether. 10 ml of this solution was taken and diluted to 100 ml with petroleum ether. Then 5, 10, 15, 20, 25 and 30 ml of this solution were pipette out to separate 100ml volumetric flasks, each containing 3ml of acetone and these were diluted to mark with petroleum ether. The concentrations

were 0.5, 1.0, 1.5, 2.0, 2.5, and 3 µg/ ml. Now the colour was measured at 452 nm. Acetone 3% in petroleum ether as blank. Absorbance was then plotted against concentration and expressed as µg of carotene/100 g.

3.4.4 Microbial parameters

Total plate count was carried out by using nutrient agar and potato dextrose agar. Total plate count was carried out by aseptically inoculating 0.1 ml of serially diluted samples in standard plate count agar medium prepared according to Ranganna (1997). An aliquot (0.1 mL) of the sample after serial dilution (10^{-2}, 10^{-4}, 10^{-6} and 10^{-8}) was aseptically inoculated in pre-sterilized plates followed by pouring total plate count agar (10-15 mL) under sterilized environment of laminar air flow. The plates were then incubated at 37^{o}C for 72 h prior to counting of microbes (Bacteria, yeasts and moulds). The results of the total plate count (TPC) were expressed as CFU/ml of sample

3.5 Sensory evaluation

Sensory evaluation of honey and sugar based malta beverages were conducted on the basis of colour, flavour, taste, body, taste, consistency and overall acceptability on a 9 point hedonic scale (Appendix-I) as per the method prescribed by Amerine *et al.* (1965).

Taste panel (5 members at a time) comprised of faculty members and PG students of department of Food Science and Technology, College of Horticulture, VCSGUUHF, Bharsar (Pauri Garhwal). Efforts were made to keep the same panel for sensory analysis throughout the entire period of study. Plain water was provided to the panelists for mouth rinsing in between the sensory evaluation.

3.6 Statistical analysis

The data pertaining to the sensory evaluation of the sugar and honey based food products were analyzed according to Randomized Complete Block Design as described by Mahony (1985), while the data on chemical characteristics of different products before and during storage were analyzed statistically by following Completely Randomized Design (CRD) at 95 per cent level of significance (Cochran and Cox, 1967).

APPENDIX-I

HEDONIC RATING TEST

Evaluation for sensory quality of malta products

Name: _____

Product: _____ Date: _____

Please evaluate the following samples as per standard scale:

Attributes	Samples							
	1	2	3	4	5	6	7	8
Colour								
Flavour								
Taste								
Consistency								
Overall acceptability								

9 Point Hedonic Scale

9 Like extremely
8 Like very much
7 Like moderately
6 Like slightly
5 Neither like nor dislike
4 Dislike slightly
3 Dislike moderately
2 Dislike very much
1 Dislike extremely

Signature of evaluator

APPENDIX-II

Temperature corrections for the standard Model of Sugar Refractometer calibration for 20^0 C

Percentage of dry substances														
Temperatures (^{o}C)	5	10	15	20	25	30	35	40	45	50	55	60	65	70
Subtract from dry substances percentage														
15	0.29	0.31	0.33	0.34	0.34	0.35	0.36	037	0.37	0.38	0.39	0.39	0.40	0.40
16	0.24	0.25	0.26	0.27	0.28	0.28	0.29	0.30	0.30	0.30	0.31	0.31	0.32	0.32
17	0.18	0.19	0.20	0.21	0.21	0.21	0.22	0.22	0.23	0.23	0.23	0.23	0.24	0.24
18	0.13	0.13	0.14	0.14	0.14	0.14	0.15	0.15	0.15	0.15	0.16	0.16	0.16	0.16
19	0.06	0.06	0.07	0.07	0.07	0.07	0.08	0.08	0.08	0.08	0.08	0.08	0.08	0.08
Add to dry substances percentage														
21	0.07	0.07	0.07	0.07	0.08	0.08	0.08	0.08	0.08	0.08	0.08	0.08	0.08	0.08
22	0.13	0.14	0.14	0.15	0.15	0.15	0.15	0.15	0.16	0.16	0.16	0.16	0.16	0.16
23	0.20	0.21	0.22	0.22	0.23	0.23	0.23	0.23	0.24	0.24	0.24	0.24	0.24	0.24
24	0.27	0.28	0.29	0.30	0.30	0.31	0.31	0.31	0.31	0.31	0.32	0.32	0.32	0.32
25	0.35	0.36	0.37	0.38	0.38	0.39	0.40	0.40	0.40	0.40	0.40	0.40	0.40	0.40
26	0.42	0.43	0.44	0.45	0.46	0.47	0.48	0.48	0.48	048	0.48	0.48	0.48	0.48
27	0.50	0.52	0.53	0.54	0.55	0.55	0.56	0.56	0.56	0.56	0.56	0.56	0.56	0.56
28	0.57	0.60	0.61	0.62	0.63	0.63	0.64	0.64	0.64	0.64	0.64	0.64	0.64	0.64
29	0.66	0.68	0.69	0.71	0.72	0.72	0.73	0.73	0.73	0.73	0.73	0.73	0.73	0.73
30	0.74	0.77	0.78	0.79	0.80	0.80	0.81	0.81	0.81	0.81	0.81	0.81	0.81	0.81

EXPERIMENTAL RESULTS

CHAPTER-4
EXPERIMENTAL RESULTS

The present investigations entitled "Studies on the preparation and evaluation of functionally enriched value added product from malta fruit (*Citrus sinensis* L Osbeck)" were carried out in the department of Food Science and Technology, College of Horticulture, VCSG UUHF, Bharsar (Pauri Garhwal). The results obtained are discussed in this chapter under the following heads:

4.1 Physico-chemical characteristics of fresh malta fruits

4.2 Physico-chemical characteristics of honey

4.3 Standardization of prepared beverages

4.4 Physico-chemical characteristics of fresh products

4.5 Storage study of developed products of malta fruit

4.6 Effect of different treatments, storage duration and conditions on microbial quality characteristics of malta products

4.7 Cost of production of malta fruit beverages.

4.1 Physico-chemical characteristics of fresh malta fruit

4.1.1 Physical parameters

The data pertaining to various physico-chemical characteristics of malta fruits has been shown in Table 4.1. The malta fruit had an average fruit weight, length and width as 149.62 ± 10.46 g, 6.56 ± 0.70 cm and 6.80 ± 0.78 cm, respectively. The average number of ten fruit segments and number of seeds/fruit was observed to be 10.00 ± 0.00 and 31.33 ± 1.53, respectively. The length, width and weight peeled fruits was 5.41 ± 0.54 cm, 5.26 ± 0.35 cm and 130.99 ± 17.30 g, respectively. The average weight of peel was found 40.89 ± 3.01 g and thickness was 0.54 ± 0.11 cm. The total soluble solids and pH of malta fruit juice was10.33 ± 1.15°B and 2.97 ± 0.02.

4.1.2 Chemical parameters

The data on chemical analysis of malta fruit contained total titratable acidity (as citric acid) 2.18 ± 0.13 %. The reducing sugars, non-reducing sugars and total sugars were found to be 4.83 ± 0.22 % 3.99 ± 0.75 % and 8.82 ± 0.65 % respectively.

Table 4.1 Physico-chemical characteristics of fresh malta fruit

Parameters	**Mean ± S.D.**
Physical parameters	
Weight of fruit(g)	149.62 ± 10.46
Length of fruit (cm)	6.56 ± 0.70
Width of fruit (g)	6.80 ± 0.78
Number of segment /fruit	10.00 ± 0.00
Number of seed/ fruit	31.33 ± 1.53
Length of peeled fruit (cm)	5.41 ± 0.54
Width of peeled fruit (cm)	5.26 ± 0.35
Weight of peeled fruit (g)	130.99 ± 17.30
Thickness of peel (cm)	0.54 ± 0.11
Weight of peel (g)	40.89 ± 3.01
T.S.S of fruit (°B)	10.33 ± 1.15
Chemical parameters	
Titratable acidity (as % Citric acid)	2.18 ± 0.13
pH	2.97 ± 0.02
Ascorbic acid (mg/100mL)	41.35 ± 0.13
Reducing sugars (%)	4.83 ± 0.22
Non-reducing sugars (%)	3.99 ± 0.75
Total sugars (%)	8.82 ± 0.65
Antioxidant parameters	
Total phenols (mg/100mL)	7.98 ± 0.74
β-carotenoids of fruit (mg/100mL)	8.45 ± 7.38
β-carotenoids of peel (mg/100mL)	9.75 ± 2.46

4.1.3 Antioxidants parameters

Ascorbic acid was observed to be 41.35 ± 0.13 mg/100mL. β-carotenoids content of fruit and peel was recorded to be 8.45 ± 7.38 mg/100mL and 9.75 ± 2.46 mg/100mL respectively. The total phenols content of fruit was 7.98 ± 0.74 mg/100mL.

4.2 Physico-chemical characteristics of honey

4.1.1 Physical parameters

Physico-chemical characteristics of honey shown in Table 4.2. The total soluble solids of honey was found to be 81.1 ± 1.15 °B.

Table 4.2: Physico- chemical characteristics of honey

Parameters	**Mean ± S.D.**
Physical parameters	
T.S.S (°B)	81.1 ± 1.15
Chemical parameters	
Titratable Acidity (as % Citric acid)	4.28 ± 0.13
Reducing sugars (%)	65.54 ± 0.22
Non-reducing sugars (%)	8.82 ± 0.75
Total sugars (%)	74.36 ± 0.65
Antioxidant parameters	
Total phenols (mg/100mL)	18.30 ± 2.46
β-carotenoids (mg/100mL)	89.47 ± 7.38

4.1.2 Chemical parameters

The data on chemical analysis of honey contained titratable acidity (as citric acid) 4.28 ± 0.13 %. The reducing sugars, non-reducing sugars and total sugars were found to be 65.54 ± 0.22 %, 8.82 ± 0.75 % and 74.77 ± 0.65 % respectively.

4.1.3 Antioxidant parameters

Ascorbic acid was observed to be 38.74 ± 0.13 mg/100mL. β-carotenoids and total phenols content of honey was recorded to be 89.47 ± 7.38 mg/100mL and 18.30 ± 2.46 mg/100mL.

4.3 Standardization of prepared beverages

4.3.1 Sensory characteristics of freshly prepared sugar malta RTS

It is evident from the (fig 4.1) that the maximum sensory score were 8.31, 8.10, 8.20, 8.60 and 8.70 for colour, flavour, taste, consistency and overall acceptability were recorded in T5 (12^{o} B TSS and 12 % juice concentration) and minimum score 6.38, 5.83,

5.10, 6.30 and 4.90 for colour, flavour, taste, consistency, and overall acceptability was obtained in T1 (10 °B TSS and 10 % juice).

4.3.2 Sensory characteristics of freshly prepared honey malta RTS

It is clear from the figure (Fig 4.2) that the maximum sensory score were 8.31, 8.12, 8.32, 8.39,and 8.36 for colour, flavour, taste, consistency and overall acceptability were recorded in T9 (14°B TSS and 14 % juice concentration) and minimum score were 5.36, 5.93, 5.23, 5.23 and 6.21 for colour flavour, taste, consistency, and overall acceptability was obtained in T1 (10°B TSS and 10 % juice).

4.3.3 Sensory characteristics of freshly prepared malta iced tea

The best combination from both the sweetening agents were further treated with different concentration of tea T1 (0.50 %), T2 (0.75 %) and T3 (1.00 %) maximum sensory score 8.13, 8.34, 8.67, 8.76 and 8.15 for colour, flavour, taste, consistency and overall acceptability were recorded in T3 (1.00 %) for sugar malta iced tea. Sensory evaluation graphically represented in fig 4.3. As well as T3 (1.00 %) also reached highest sensory scores 8.31, 8.36, 8.34, 8.36 and 8.13 were recorded for colour, flavour, taste, consistency and overall acceptability for honey malta iced tea. The graphical representation of sensory parameters represented in fig 4.4.

4.4 Physico-chemical characteristics of fresh products

4.4.1 Sugar malta RTS

4.4.1.1 Physical parameters

The effect of various treatments viz. pasteurization (65 °C for 30 minutes), preservative (sodium benzoate 120 ppm) and pasteurization with half preservative (65°C for 30 minutes with sodium benzoate 60 ppm) on sugar malta RTS is presented in Table 4.3. A slight change in the TSS of sugar malta RTS is there. The highest TSS was observed in treatment T3 (65 °C for 30 minutes pasteurization with sodium benzoate 60 ppm) 12.03 ± 0.09 °B followed by treatment T2 (Sodium benzoate 120 ppm) and least in treatment T1 (pasteurization 65 °C for 30 minutes) 12.03 ± 0.09 °B. pH of prepared sugar malta RTS was also influenced by the various treatments and was found in range of 3.57 ± 0.26 to 3.76 ± 0.09.

Table 4.3 Physico-chemical characteristics of fresh products

Parameters		Sugar malta RTS	Honey malta RTS	Sugar malta iced tea	Honey malta iced tea
Physical parameters					
T.S.S of fruit (°B)	T1	12.03 ± 0.09	14.10 ± 0.01	12.03 ±0.04	14.10 ± 0.12
	T2	12.10 ± 0.04	14.20 ± 0.01	12.03±0.12	14.20 ± 0.20
	T3	12.20 ± 0.09	14.12± 0.10	12.06±0.09	14.03 ± 0.03
pH	T1	3.57 ± 0.26	3.68 ± 0.05	3.39±0.01	3.17 ± 0.30
	T2	3.76 ± 0.09	3.75 ± 0.05	3.56±0.01	3.56 ± 0.20
	T3	3.63 ± 0.10	3.59 ± 0.13	3.63±0.03	3.21 ± 0.40
Bio-chemical parameters					
Titratable Acidity (%)	T1	0.40 ± 0.01	0.45 ± 0.04	0.40 ± 0.01	0.50 ± 0.02
	T2	0.43 ± 0.01	0.44 ± 0.9	0.43 ± 0.16	0.53 ± 0.17
	T3	0.43 ± 0.01	0.44 ± 0.7	0.36 ± 0.01	0.63 ± 0.08
Reducing sugars (%)	T1	4.72 ± 0.06	5.54 ± 0.58	4.42 ± 0.77	5.08 ± 0.04
	T2	4.77 ± 0.14	5.92 ± 0.17	4.30 ± 0.79	5.05 ± 0.11
	T3	4.85 ± 0.07	5.67 ± 0.08	4.13 ± 0.83	5.15 ± 0.15
Non-reducing sugars (%)	T1	5.16 ± 0.49	5.64 ± 0.59	5.54 ± 0.58	5.80 ± 0.49
	T2	5.18 ± 0.17	5.05 ± 0.22	5.92 ± 0.17	5.93 ± 0.17
	T3	5.26 ± 0.08	5.72 ± 0.66	5.67 ± 0.08	5.67 ± 0.08
Total sugars (%)	T1	9.88 ± 0.75	10.72 ± 0.59	9.96 ± 0.58	10.88 ± 0.45
	T2	9.95 ± 0.15	10.97 ± 0.22	10.22 ± 0.15	10.98 ± 0.15
	T3	10.11 ± 0.16	11.39 ± 0.78	9.80 ± 0.06	10.82 ± 0.06
Antioxidant parameters					
Ascorbic acid (mg/100mL)	T1	19.44 ± 0.01	22.79 ±0.74	19.73 ± 0.75	22.93 ± 0.75
	T2	19.49 ± 0.01	23.26 ± 0.75	20.26 ± 0.75	23.26 ± 0.75
	T3	19.33 ± 0.01	22.73 ± 0.75	18.30 ±2.46	22.73 ± 0.75
Total Phenols (mg/100mL)	T1	7.69 ± 0.25	8.30 ± 0.30	9.32 ± 0.58	11.69 ± 0.25
	T2	7.41 ± 0.30	8.29 ± 0.27	9.55 ± 0.15	11.41 ± 0.30
	T3	7.34 ± 0.09	8.61 ± 0.60	9.27 ± 0.06	12.34 ± 0.01
β-Carotenoids (mg/100mL)	T1	8.31 ± 0.92	9.94 ± 0.01	9.30 ± 0.30	11.31 ± 0.92
	T2	8.87 ± 1.21	9.61 ± 0.27	9.37 ± 0.01	11.87 ± 0.32
	T3	8.51 ± 0.20	9.97 ± 0.02	9.40 ± 0.60	11.25 ± 0.01

4.4.1.2 Bio-chemical parameters

Various bio-chemical characteristics viz. titratable acidity, reducing sugars, non-reducing sugars and total sugars were also studied and presented in the Table.4.3. The titratable acidity was found to be in the range of 0.40 ± 0.01 to 0.43 ± 0.01 % as citric acid. The highest reducing sugars 4.85 ± 0.07 % was observed in treatment T3 (65°C for 30 minutes pasteurization with sodium benzoate 60 ppm) 4.85 ± 0.07 % followed by treatment T2 (Sodium benzoate 120 ppm) and least was in treatment T1 (65°C for 30 minutes pasteurization) 4.72 ± 0.06 %.The highest non reducing sugars was observed in treatment T3 (pasteurization with half preservative) followed by treatment T2 (Sodium benzoate 120 ppm) and least in treatment T1 (pasteurization) 5.16 ± 0.49 %. The highest total sugars was also observed in treatment T3 (pasteurization (65°Cfor 30 minutes with half preservative) 5.72 ± 0.66 % and least was in treatment T2 (sodium benzoate) 5.05 ± 0.22 %.

4.4.1.3 Antioxidant parameters

Ascorbic acid, total phenols and β-carotenoids contents as an antioxidant parameter study was carried out to evaluate the prepared sugar malta RTS and data obtained was presented in the Table.4.3. The highest ascorbic acid was observed in treatment T2 (Sodium benzoate 120 ppm) 23.26 ± 0.75 mg/100mL, followed by T1 (pasteurization) 22.79 ± 0.74 mg/100mL and least in treatment T3 (pasteurization with half preservative) 22.73 ± 0.75 mg/100mL. The β-carotenoids was found in the range of 8.31 ± 0.92 mg/100mL to 9.20 ± 0.2 mg/100mL and total phenol of sugar malta RTS was recorded in range of 7.34 ± 0.09 mg/100mL to 7.69 ± 0.25 mg/100mL.

4.4.2 Honey malta RTS

4.4.2.1 Physical parameters

The effect of various treatments viz. pasteurization (65°C for 30 minutes), preservative (sodium benzoate 120 ppm) and pasteurization with half preservative (65°C for 30 minutes with sodium benzoate 60 ppm) on honey malta RTS. The research finding showed a slight change in the TSS of honey malta RTS is presented in Table 4.3. The maximum TSS 14.20 ± 0.01 °B was observed in treatment T2 (sodium benzoate 120 ppm) followed by treatment T3 (65°C for 30 minutes pasteurization with sodium benzoate 60 ppm) and minimum TSS 14.10 ± 0.01 °B was observed in treatment T1 (pasteurization

65°C for 30 minutes). pH of prepared honey malta RTS was also influenced by the various treatments and ranged between 3.59 ± 0.13 to 3.75 ± 0.05.

4.4.2.2 Bio-chemical parameters

Various biochemical characteristics viz. titratable acidity, reducing sugars, non-reducing sugars and total sugars of honey malta RTS were also studied and presented in the Table.4.3. The titratable acidity was found in the range of 0.44 ± 0.7 % to 0.45 ± 0.04 %. The highest reducing sugar 5.92 ± 0.17 % was observed in treatment T2 (Sodium benzoate 120 ppm) followed by treatment T3 (65 °C for 30 minutes pasteurization with sodium benzoate 60 ppm) 4.85 ± 0.07 %, and least was in treatment T1 (65 °C for 30 minutes pasteurization) 5.54± 0.58 %. The highest non reducing sugars was observed in treatment T3 (pasteurization with half preservative 5.26 ± 0.08 %, followed by treatment T2 (Sodium benzoate 120 ppm) and least in treatment T1 (pasteurization) 5.16 ± 0.49 %. The highest total sugars was also observed in treatment T3 (pasteurization 65 °C for30 minutes with half preservative) 10.11 ± 0.16 % and least was in treatment T1 (pasteurization) 9.86 ± 0.08 %.

4.4.2.3 Antioxidant parameters

Antioxidant parameters, Ascorbic acid, total phenols and β-carotenoids contents are the studied under current study to evaluate the prepared honey malta RTS and data was presented in the Table.4.3. The highest ascorbic acid was observed in treatment T2 (Sodium benzoate 120 ppm) 20.26 ± 0.75 mg/100mL, followed by T1 (pasteurization) 19.73 ± 0.75 mg/100mL and least in treatment T3 (pasteurization with half preservative) 18.30 ± 2.46 mg/100mL. The β carotenoids was found in the range of 8.31 ± 0.92 mg/100mL to 9.20 ± 0.21 mg/100mL and total phenol of honey malta RTS was recorded in range of 7.34 ± 0.09 mg/100mL to 7.69 ± 0.25 mg/100mL.

4.4.3 Sugar malta iced tea

4.4.3.1 Physical parameters

The data pertaining effect of various preservative techniques on sugar malta iced tea is presented in Table 4.3. TSS of prepared sugar malta iced tea was also influenced by the various treatments and was found in range of 12.03 ± 0.04 to 12.06 ± 0.09°B.

4.4.3.2 Bio-chemical parameters

Titratable acidity, reducing sugars, non-reducing sugars and total sugars of prepared sugar malta iced tea has been presented in the Table 4.3. The titratable acidity was found in the range of 0.36 ± 0.01 % to 0.43 ± 0.16 %The highest reducing sugars 4.42 ± 0.77 % was observed in treatment T1 (pasteurization 65 ^{o}C for 30 minutes) followed by treatment T2 (Sodium benzoate 120 ppm) 4.30±0.79 %, and least was in treatment T3 (pasteurization with half preservative) 4.13 ± 0.83 %. The highest non reducing sugars was found to be ranged between 5.54 ± 0.58 to 5.92 ± 0.17 %. The highest total sugars was also observed in treatment T2 (9.55 ± 0.15 %) and least was found in treatment T3 (9.27 ± 0.06 %).

4.4.3.3 Antioxidant parameters

Ascorbic acid, total phenols and β-carotenoids contents are the antioxidant parameter was carried out to evaluate the prepared sugar malta iced tea and data was presented in the Table.4.3. The highest ascorbic acid was observed in treatment T2 (Sodium benzoate 120 ppm) 19.49 ± 0.01 mg/100mL, followed by T1 (pasteurization) 19.44 ± 0.01 mg/100mL and least in treatment T3 (pasteurization with half preservative) 19.33 ± 0.01 mg/100mL. The Carotene was found in the range of 9.18 ± 0.60 mg/100mL to 9.61 ± 0.27 mg/100mL and total phenol of sugar malta iced tea was recorded in range of 9.27 ± 0.06 mg/100mL to 9.55 ± 0.15 mg/100mL.

4.4.4 Honey malta iced tea

4.4.4.1 Physical parameters

The effect of various treatments viz. pasteurization (65 ^{o}C for 30 minutes), preservative (sodium benzoate 120 ppm) and pasteurization with half preservative (65 ^{o}C for 30 minutes with sodium benzoate 60 ppm) on honey malta iced tea presented in Table 4.3. A slight difference in the TSS of honey malta iced was observed. The highest TSS was recorded in treatment T2 (sodium benzoate 120 ppm) 14.20 ± 0.01 ^{o}B followed by treatment T1 (pasteurization 65 ^{o}C for 30 minutes) and least in treatment T3 (Sodium benzoate 60 ppm with pasteurization 65 ^{o}C for 30 minutes) 14.03 ± 0.03 ^{o}B. pH of prepared honey malta iced was also influenced by the various treatments and was found in range of 3.17 ± 0.3 to3.56 ± 0.2.

4.4.4.2 Bio-chemical parameters

Various biochemical characteristics viz. titratable acidity, reducing sugars, non-reducing sugars and total sugars of honey malta iced tea were studied and presented in the Table.4.3. The titratable acidity was ranged between 0.50 ± 0.02 % to 0.63 ± 0.08 %. The highest reducing sugar 5.15 ± 0.15 % was observed in treatment T3 (65 ^{o}C for 30 minutes pasteurization with sodium benzoate 60 ppm) followed by treatment T1 (pasteurization 65 ^{o}C for 30 minutes) 5.08 ± 0.04 %, and least was in treatment T2 (sodium benzoate 120 ppm) 5.54 ± 0.58 %. The highest non reducing sugars was observed in treatment T2 (Sodium benzoate 120 ppm) 5.26 ± 0.08 %, followed by treatment T1 (pasteurization 65 ^{o}C for 30 minutes) and least in treatment T3 (pasteurization with half preservative) 5.67 ± 0.08 %. The highest total sugars was also observed in treatment T1 (pasteurization 65 ^{o}C for 30 minutes) 10.58 ± 0.45 % followed by (T2 (Sodium benzoate 120 ppm) 10.55 ± 0.15 % followed by (pasteurization 10.11 ± 0.16 % and least was in treatment T3 (65 ^{o}C for 30 minutes with half preservative) 10.27 ± 0.06 %.

4.4.4.3 Antioxidant parameters

Ascorbic acid, total phenols and β-carotenoids contents are the antioxidant parameters were studied under present study to evaluate the prepared honey malta iced tea and data was presented in the Table.4.3. The highest ascorbic acid was observed in treatment T2 (Sodium benzoate 120 ppm) 23.26 ± 0.75 mg/100 mL, followed by T1 (pasteurization 65 ^{o}C for 30 minutes) 22.93 ± 0.75 mg/100mL and least in treatment T3 (pasteurization with half preservative) 22.73 ± 0.75 mg/100mL. The β-carotenoids was found in the range of 8.31 ± 0.92 mg/100mL to 9.25 ± 0.01 mg/100mL and total phenols of honey malta iced tea was recorded in range of 7.34 ± 0.01 mg/100mL to 7.69 ± 0.25 mg/100mL.

4.5 Storage study of developed products of malta fruit

4.5.1 Effect of different treatments, storage duration and conditions on quality characteristics of sugar malta RTS

4.5.1.1Total Soluble Solids (^{0}B)

The effect of storage duration and conditions on TSS content of sugar malta RTS is depicted by the Table 4.4 there was a progressive and continuous significant increase was observed in TSS content of all treatments with an increase in storage period upto six months. Maximum TSS 12.490 ^{o}B was recorded in T2 (sodium benzoate 120 ppm) during

six month storage while minimum TSS 12.210 ^{o}B was recorded in T1 (Pasteurization 65 ^{o}C for 30 minutes) during three month storage. Among different preservative methods the highest changes was observed under ambient storage conditions and least was under refrigerated conditions. The interaction of various factors revealed the significant results for change in TSS during storage at both ambient and refrigerated temperature.

Table: 4.4 Effect of different treatments, storage duration and conditions on TSS (^{0}B) of sugar malta RTS

Treatments	3 Months			6Months		
	Ambient	Refrigerated	Mean	Ambient	Refrigerated	Mean
T1	12.420	12.210	**12.385**	12.460	12.350	**12.405**
T2	12.460	12.220	**12.340**	12.490	12.340	**12.415**
T3	12.380	12.230	**12.305**	12.380	12.340	**12.360**
Mean	**12.420**	**12.343**		**12.443**	**12.343**	
CD $_{0.05}$						
Time (A)		0.006	**A x B**		0.009	
Conditions (B)		0.006	**A x T**		0.011	
Treatments (T)		0.008	**B x T**		0.011	
			AxBxT		0.015	

4.5.1.2 Titratable Acidity (%)

The data obtained for change in titratable acidity is presented in Table 4.5 .There was a gradual declining trend in titratable acidity content was observed with an advancement in storage periods under all treatments. The titratable acidity was maintained at range of 0.340 to 0.490 %. Maximum acidity 0.490 % was recorded in T2 (Sodium benzoate 120 ppm) at ambient temperature during three month storage duration whereas minimum 0.340 % was recorded in T3 (Pasteurization with half preservative) during six month storage. Among different preservative methods the highest changes was observed under ambient storage conditions and least was under refrigerated conditions.

Table: 4. 5 Effect of different treatments, storage duration and conditions on titratable acidity (%) of sugar malta RTS

Treatments	3 Months			6 Months		
	Ambient	Refrigerated	Mean	Ambient	Refrigerated	Mean
T1	0.460	0.427	**0.440**	0.413	0.357	**0.409**
T2	0.490	0.407	**0.462**	0.457	0.343	**0.417**
T3	0.387	0.380	**0.384**	0.383	0.340	**0.362**
Mean	**0.419**	**0.425**		**0.444**	**0.347**	
$CD_{0.05}$						
Time (A)		0.006	A x B		0.009	
Conditions (B)		0.006	A x T		0.011	
Treatments (T)		0.008	B x T		0.011	
			AxBxT		0.015	

4.5.1.3 Ascorbic acid (mg/100mL)

Table: 4. 6 Effect of different treatments, storage duration and conditions on ascorbic acid (mg/100mL) of sugar malta RTS

Treatments	3 Months			6 Months		
	Ambient	Refrigerated	Mean	Ambient	Refrigerated	Mean
T1	16.800	17.700	**17.250**	9.580	10.610	**10.095**
T2	19.050	19.350	**19.200**	9.560	10.630	**10.095**
T3	17.650	17.800	**17.725**	9.580	10.510	**10.045**
Mean	**17.983**	**18.133**		**9.573**	**10.583**	
$CD_{0.05}$						
Time (A)		0.006	A x B		0.009	
Conditions (B)		0.006	A x T		0.011	
Treatments (T)		0.008	B x T		0.011	
			AxBxT		0.015	

Data representing the effect of various treatments on ascorbic acid contents at different storage intervals are presented in Table 4.6. A perusal of the data reveals that there was a continuous significant decrease in the mean ascorbic acid content with an

increase in storage periods upto six months at both storage conditions. However, significantly higher ascorbic acid content was observed in T2 (sodium benzoate 120 ppm) 19.05 mg/100 mL followed by T1 (pasteurization 65 ^{o}C for 30 minutes) during three month storage and least 10.510 mg/100mL in T3 (pasteurization with half preservative) during three months storage in sugar malta RTS. However, there was a significantly higher decrease at ambient conditions than under refrigerated conditions.

4.5.1.4 Total sugars (%)

Table: 4. 7 Effect of different treatments, storage duration and conditions on total sugars (%) of sugar malta RTS

Treatments	3 Months			6 Months		
	Ambient	Refrigerated	Mean	Ambient	Refrigerated	Mean
T1	9.530	9.430	**9.480**	10.570	10.130	**9.930**
T2	9.630	9.123	**9.376**	10.660	10.240	**10.110**
T3	9.800	9.369	**9.585**	10.610	10.300	**10.095**
Mean	**9.457**	**9.284**		**10.583**	**9.573**	
CD $_{0.05}$						
Time (A)		0.006	**A x B**		0.009	
Conditions (B)		0.006	**A x T**		0.011	
Treatments (T)		0.008	**B x T**		0.011	
			AxBxT		0.015	

It is evident from Table 4.7 that sugar malta RTS showed a progressive and continuous significant increase in the total sugar content was observed upto six months storage under all the treatments, with less change under refrigerated temperature than under ambient temperature. Which ranges between 9.240 % in T1 (pasteurization 65 ^{o}C for 30 minutes) to 10.840 % in T3 (pasteurization with half preservative) and 9.123 % in T1 (pasteurization 65 ^{o}C for 30 minutes) to 9.580 % in T3 (pasteurization with half preservative) for ambient and refrigerated temperature respectively. Besides, interaction of various factors revealed the significant results for change in total sugars.

4.5.1.5 Reducing sugars (%)

Table: 4. 8 Effect of different treatments, storage duration and conditions on reducing sugars (%) of sugar malta RTS

Treatments	3 Months			6 Months		
	Ambient	Refrigerated	Mean	Ambient	Refrigerated	Mean
T1	4.410	4.120	**4.265**	5.850	5.720	**5.785**
T2	4.470	4.160	**4.315**	5.900	5.770	**5.835**
T3	4.550	4.210	**4.425**	5.760	5.850	**5.805**
Mean	**4.443**	**4.227**		**5.837**	**5.78**	
CD $_{0.05}$						
Time (A)		0.019	**A x B**		0.026	
Conditions (B)		0.019	**A x T**		0.032	
Treatments (T)		0.023	**B x T**		0.032	
			AxBxT		0.046	

Perusal data presented in Table 4.8 reveals that there was a progressive and continuous significant increasing trend in the mean reducing sugars content in all three treatments of sugar malta RTS with an increase in storage periods upto six month at both conditions. Maximum reducing sugars 5.900 % was recorded in T3 (pasteurization with half preservative) during six month storage and minimum reducing sugars 4.120 % was recorded in T1 (pasteurization 65 ^{o}C for 30 minutes) during three month storage. However, higher increase at ambient conditions than under refrigerated conditions.

4.5.1.6 Non- reducing sugars (%)

A perusal of data in Table 4.9 shows that there was a significant decline of non-reducing sugars for different treatments in sugar malta RTS. The maximum non-reducing sugars 5.400 % was recorded in T3 (pasteurization with half preservative) and minimum 4.410 % was recorded in T1 (pasteurization 65 ^{o}C for 30 minutes). However, the decrease in non-reducing sugars was higher at ambient temperature than refrigerated temperature.

Table: 4. 9 Effect of different treatments, storage duration and conditions on non-reducing sugars (%) of sugar malta RTS

Treatments	3 Months			6 Months		
	Ambient	Refrigerated	Mean	Ambient	Refrigerated	Mean
T1	5.120	5.410	**5.265**	4.720	4.410	**4.565**
T2	5.166	5.470	**5.318**	4.760	4.470	**4.615**
T3	5.400	5.450	**5.425**	4.850	4.450	**4.650**
Mean	**5.443**	**5.229**		**4.777**	**4.443**	
CD $_{0.05}$						
Time (A)		0.006	**A x B**		0.008	
Conditions (B)		0.006	**A x T**		0.010	
Treatments (T)		0.007	**B x T**		0.013	
			AxBxT		0.014	

4.5.1.7 pH

Table: 4. 10 Effect of different treatments, storage duration and conditions on pH of sugar malta RTS

Treatments	3 Month			6 Month		
	Ambient	Refrigerated	Mean	Ambient	Refrigerated	Mean
T1	3.420	3.350	**3.385**	3.470	3.450	**3.460**
T2	3.460	3.340	**3.400**	3.510	3.460	**3.485**
T3	3.340	3.380	**3.360**	3.410	3.350	**3.380**
Mean	**3.343**	**3.420**		**3.443**	**3.440**	
CD $_{0.05}$						
Time (A)		0.006	**A x B**		0.009	
Conditions (B)		0.006	**A x T**		0.011	
Treatments (T)		0.011	**B x T**		0.015	
			AxBxT		0.03	

Data representing the effect of various treatments on pH at different storage intervals are presented in Table 4.10. A perusal of the data reveals that there was a progressive and continuous increase in the mean pH value with an increase in storage

periods upto 6 months at both conditions. The mean pH value was ranging from 3.340 to 3.510. Among different preservative methods the highest changes was observed under ambient storage conditions and least was under refrigerated conditions.

4.5.1.8 Total phenols (mg/100mL)

Table: 4. 11 Effect of different treatments, storage duration and conditions on total phenols of sugar malta RTS

Treatments	3 Month			6 Months		
	Ambient	Refrigerated	Mean	Ambient	Refrigerated	Mean
T1	6.740	6.760	**6.750**	5.520	5.970	**5.740**
T2	6.260	6.460	**6.360**	5.570	5.850	**5.715**
T3	6.660	6.710	**6.685**	5.750	5.960	**5.850**
Mean	**6.553**	**6.637**		**5.613**	**5.930**	
CD $_{0.05}$						
Time (A)		0.013	**A x B**		0.018	
Conditions (B)		0.013	**A x T**		0.022	
Treatments (T)		0.016	**B x T**		0.022	
			AxBxT		0.032	

The data of total phenols of sugar malta RTS during different storage intervals and conditions is given in Table 4.11. There was a significant decrease was observed for different treatments in sugar malta RTS. The maximum total phenol 6.760 mg/100mL was recorded inT1 (pasteurization 65 ^{o}C for 30 minutes) and minimum 5.520 mg/100mL was recorded in T3 (pasteurization with half preservative). However, the decrease in total phenols content was higher for ambient and least for refrigerated storage.

4.5.1.9 β carotenoids (mg/100mL)

The total β-carotenoids content of different treatments are given in Table 4.12 which reveals that carotenoid content decreasing during storage. The maximum β-carotenoids 7.940 mg/100mL were recorded in T1 (pasteurization 65 ^{o}C for 30minutes) during 3three month storage and minimum 6.360 mg/100mL was recorded in T3 (pasteurization with half preservative). However the decrease in the β-carotenoids content was higher under ambient temperature than under refrigerated temperature.

Table: 4.12 Effect of different treatments, storage duration and conditions on β-carotenoids (mg/100mL) of sugar malta RTS

Treatments	3 Month			6 Months		
	Ambient	**Refrigerated**	**Mean**	**Ambient**	**Refrigerated**	**Mean**
T1	7.930	7.940	**7.935**	6.930	6.960	**6.945**
T2	7.140	7.250	**7.195**	6.760	6.750	**6.755**
T3	7.760	7.320	**7.540**	6.310	6.360	**6.335**
Mean	**7.613**	**7.500**		**6.667**	**6.690**	
CD $_{0.05}$						
Time (A)		0.011	**A x B**		0.013	
Conditions (B)		0.011	**A x T**		0.019	
Treatments (T)		0.015	**B x T**		0.019	

4.5.1.10 Effect of different treatments, storage duration and conditions on sensory parameters of sugar malta RTS

The sensory evaluation of sugar malta RTS is graphically represented in (fig 4.5) The sensory evaluation for colour, flavour, taste and overall acceptability of sugar malta RTS reveals that highest mean were after three and six months of storage, among different treatments, the sensory scores were higher T3 (pasteurization with half preservative) stored under refrigerated temperature.

4.5.2 Effect of different treatments, storage duration and conditions on quality characteristics of honey malta RTS

4.5.2.1Total Soluble Solids (^{0}B)

Table: 4.13 Effect of different treatments, storage duration and conditions on TSS (^{0}B) of honey malta RTS

Treatments	3 Months			6 Months		
	Ambient	Refrigerated	Mean	Ambient	Refrigerated	Mean
T1	14.413	14.357	**14.385**	14.460	14.427	**14.444**
T2	14.453	14.343	**14.398**	14.490	14.467	**14.479**
T3	14.387	14.340	**14.364**	14.383	14.380	**14.382**
Mean	**14.418**	**14.347**		**14.444**	**14.425**	
CD $_{0.05}$						
Time (A)		0.006	**AXB**		0.008	
Conditions (B)		0.006	**A x T**		0.01	
Treatments (T)		0.007	**B x T**		0.01	
			AxBxT		0.015	

Data presented in Table 4.13 reveals that the TSS content of honey malta RTS increased with a progressive significant increase in storage periods upto six months at both ambient and refrigerated conditions. Maximum TSS 14.490 ^{o}B was recorded in T2 (sodium benzoate120 ppm) and minimum 14.357 ^{o}B was recorded in T1 (pasteurization 65 ^{o}C for 30 minutes). Among different preservative methods the highest changes was observed under ambient storage conditions and least was under refrigerated conditions.

4.5.2.2Titratable Acidity (%)

The titratable acidity was maintained at range of 0.441 % to 0. 590 %. Maximum titratable acidity 0.590 % was recorded in T2 (sodium benzoate 120 ppm) during 3 month storage and minimum acidity 0.441 % was recorded in T1 (pasteurization 65 ^{o}C for 30 minutes) during six month storage. However, the data in Table 4.14 shows that there was a gradual decline in the acid content during storage. It is evident from the data that the decrease reveals that advancement of storage duration, irrespective of storage conditions. Among different preservative methods the highest changes was observed under ambient storage conditions and least was under refrigerated conditions.

Table: 4.14 Effect of different treatments, storage duration and conditions on titratable acidity (%) of honey malta RTS

Treatments	3 Months			6 Months		
	Ambient	**Refrigerated**	**Mean**	**Ambient**	**Refrigerated**	**Mean**
T1	0.560	0.527	**0.544**	0.557	0.441	**0.480**
T2	0.590	0.567	**0.579**	0.513	0.457	**0.485**
T3	0.483	0.480	**0.482**	0.487	0.443	**0.465**
Mean	**0.544**	**0.525**		**0.519**	**0.448**	
CD $_{0.05}$						
Time (A)		0.006	**AXB**		0.008	
Conditions (B)		0.006	**A x T**		0.019	
Treatments (T)		0.007	**B x T**		0.010	
			AxBxT		0.015	

4.5.2.3 Ascorbic acid (mg/100mL)

Table: 4.15 Effect of different treatments, storage duration and conditions on ascorbic acid (mg/100mL) of honey malta RTS

Treatments	3 Months			6 Months		
	Ambient	**Refrigerated**	**Mean**	**Ambient**	**Refrigerated**	**Mean**
T1	19.700	20.617	**20.159**	12.583	13.617	**13.100**
T2	22.183	22.283	**22.233**	13.583	13.633	**13.563**
T3	20.750	20.800	**20.775**	12.583	13.517	**13.050**
Mean	**21.183**	**20.928**		**12.583**	**13.589**	
CD $_{0.05}$						
Time (A)		0.051	**AXB**		0.072	
Conditions (B)		0.051	**A x T**		0.088	
Treatments (T)		0.062	**B x T**		0.010	
			AxBxT		0.124	

The data of ascorbic acid content of honey mala RTS during different stages of storage are presented in Table 4.15 However, significantly higher ascorbic acid content in T2 (sodium benzoate 120 ppm)19.05 mg/100 mL followed by T1 (pasteurization 65 ^{o}C for 30 minutes and least 13.517 mg/100mL in T3 (pasteurization with half preservative) in

honey malta RTS. However, there was a significantly higher decrease at ambient conditions than under refrigerated conditions.

4.5.2.4 Total sugars (%)

Table: 4.16 Effect of different treatments, storage duration and conditions on total sugars (%) of honey malta RTS

Treatments	3 Months			6 Months		
	Ambient	Refrigerated	Mean	Ambient	Refrigerated	Mean
T1	12.303	11.912	**12.098**	12.012	12.507	**12.610**
T2	12.393	11.990	**12.200**	12.813	12.580	**12.697**
T3	12.423	12.310	**12.358**	12.880	12.860	**12.870**
Mean	**12.519**	**11.852**		**12.802**	**12.620**	
CD $_{0.05}$						
Time (A)		0.012	**AXB**		0.017	
Conditions (B)		0.012	**A x T**		0.021	
Treatments (T)		0.015	**B x T**		0.021	
			AxBxT		0.029	

Table 4.16 shows the effect of storage on the total sugar content of honey malta RTS. There was a gradual significant increase in total sugar with advancement of storage period. Which ranges between 11.657 % in T1 (pasteurization 65 ^{o}C for 30 minutes) to 12.880 % in T3 (pasteurization with half preservative). However, there was a significantly higher increase for ambient and least refrigerated temperature.

4.5.2.5 Reducing sugars (%)

The data for reducing sugars given in Table 4.17 pertaining significant increasing trend in all three treatments of honey malta RTS, maximum reducing sugars 5.940 % was recorded in T3 (pasteurization with half preservative) during six month storage and minimum reducing sugars 5.150 % was recorded in T1(pasteurization 65 ^{o}C for 30 minutes) during 3 month storage. However, there was a significantly higher increase at ambient conditions than under refrigerated conditions.

Table: 4.17 Effect of different treatments, storage duration and conditions on reducing sugars (%) of honey malta RTS

Treatments	3 Months			6 Months		
	Ambient	**Refrigerated**	**Mean**	**Ambient**	**Refrigerated**	**Mean**
T1	5.447	5.150	**5.299**	5.857	5.753	**5.805**
T2	5.503	5.197	**5.350**	5.907	5.803	**5.855**
T3	5.487	5.430	**5.459**	5.940	5.887	**5.914**
Mean	**5.479**	**5.259**		**5.901**	**5.814**	
CD $_{0.05}$						
Time (A)		0.012	**AXB**		0.008	
Conditions (B)		0.012	**A x T**		0.010	
Treatments (T)		0.007	**B x T**		0.010	
			AxBxT		0.015	

4.5.2.6 Non- reducing sugars (%)

Table: 4.18 Effect of different treatments, storage duration and conditions on non-reducing sugars (%) of honey malta RTS

Treatments	3 Months			6 Months		
	Ambient	**Refrigerated**	**Mean**	**Ambient**	**Refrigerated**	**Mean**
T1	6.857	6.753	**6.805**	6.150	6.447	**6.299**
T2	6.907	6.803	**6.855**	6.197	6.503	**6.350**
T3	6.940	6.887	**6.914**	6.430	6.487	**6.459**
Mean	**6.901**	**6.814**		**6.259**	**6.479**	
CD $_{0.05}$						
Time (A)		0.006	**AXB**		0.008	
Conditions (B)		0.006	**A x T**		0.020	
Treatments (T)		0.007	**B x T**		0.010	
			AxBxT		0.015	

A perusal of data in Table 4.18 shows that there was a significant difference of non-reducing sugars for different treatments in honey malta RTS. The maximum non-reducing sugars 6.940 %was recorded in T3 (pasteurization with half preservative) and minimum non-reducing sugars 6.150 % was observed in (pasteurization 65 ^{o}C for 30 minutes).However, the decrease in non-reducing sugars irrespective of the storage conditions**.**

4.5.2.7 pH

Table: 4.19 Effect of different treatments, storage duration and conditions on pH of honey malta RTS

Treatments	**3 Months**			**6 Months**		
	Ambient	**Refrigerated**	**Mean**	**Ambient**	**Refrigerated**	**Mean**
T1	3.717	3.460	**3.589**	3.820	3.787	**3.804**
T2	3.707	3.490	**3.599**	3.850	3.827	**3.839**
T3	3.700	3.383	**3.542**	3.743	3.740	**3.742**
Mean	**3.708**	**3.444**		**3.804**	**3.785**	
CD $_{0.05}$						
Time (A)		0.007	**AXB**		0.009	
Conditions (B)		0.006	**AX T**		0.130	
Treatments (T)		0.008	**B X T**		0.011	
			AxBxT		0.015	

A perusal of data in Table 4.19 reveals that a significant difference was observed in the pH which was increased with the advancement of storage duration, irrespective of storage conditions. Which was ranging from 3.383 to 3.850. Among different preservative methods the highest changes was observed under ambient storage conditions and least was under refrigerated conditions.

4.5.2.8 Total phenols (mg/100mL)

The data of total phenols of honey malta RTS during different storage intervals and conditions is given in Table 4.20. There was a significant decrease was observed for different treatments in honey malta RTS. The maximum total phenol 7.740mg/100mL was recorded in T1 (pasteurization 65 ^{o}C for 30 minutes) during 3 month storage and minimum

6.860 mg/100mL in T3 (pasteurization with half preservative). However, the decrease in total phenol content was irrespective of the storage conditions**.**

Table: 4.20 Effect of different treatments, storage duration and conditions on total phenols of honey malta RTS

Treatments	3 Months			6 Months		
	Ambient	Refrigerated	Mean	Ambient	Refrigerated	Mean
T1	7.740	7.747	**7.744**	6.757	6.977	**6.867**
T2	7.263	7.467	**7.365**	6.577	6.960	**6.769**
T3	7.660	7.683	**7.672**	6.527	6.860	**6.694**
Mean	**7.554**	**7.632**		**6.620**	**6.932**	
CD $_{0.05}$						
Time (A)		0.007	**AXB**		0.009	
Conditions (B)		0.006	**A x T**		0.130	
Treatments (T)		0.008	**B x T**		0.011	
			AxBxT		0.015	

4.5.2.9 β-carotenoids (mg/100mL)

Table: 4.21 Effect of different treatments, storage duration and conditions on β-carotenoids of honey malta RTS

Treatments	3 Months			6 Months		
	Ambient	Refrigerated	Mean	Ambient	Refrigerated	Mean
T1	9.943	9.930	**9.937**	8.930	8.960	**8.945**
T2	9.147	9.257	**9.202**	8.760	8.763	**8.762**
T3	9.767	9.327	**9.547**	8.310	8.367	**8.339**
Mean	**9.619**	**9.505**		**8.667**	**8.697**	
CD $_{0.05}$						
Time (A)		0.011	**AXB**		0.015	
Conditions (B)		0.011	**A x T**		0.019	
Treatments (T)		0.013	**B x T**		0.019	
			AxBxT		0.027	

It is evident from the data presented in Table 4.21that the total β-carotenoids content decreasing during storage. The maximum β-carotenoids 9.943 mg/100mL were recorded in T1 (pasteurization 65 ^{o}C for 30minutes) during 3 month storage and minimum 8.367 mg/100mL was recorded in T3 (pasteurization with half preservative) during six month storage. However the decrease in the β-carotenoids content was higher under ambient temperature than refrigerated temperature.

4.5.2.10 Effect of different treatments, storage duration and conditions on sensory parameters of honey malta RTS

The sensory evaluation of honey malta RTS is graphically represented in fig 4.6. The sensory evaluation for colour, flavour, taste and overall acceptability of sugar malta RTS reveals that highest mean were after three and six months of storage, among different treatments, the sensory scores were higher T3 (pasteurization with half preservative) stored under refrigerated temperature.

4.5.3 Effect of different treatments, storage duration and conditions on quality characteristics of sugar malta iced tea

4.5.3.1Total Soluble Solids (^{0}B)

Table: 4.22 Effect of different treatments, storage duration and conditions on TSS (^{0}B) of sugar malta iced tea

Treatments	**3 Months**			**6 Months**		
	Ambient	**Refrigerated**	**Mean**	**Ambient**	**Refrigerated**	**Mean**
T1	12.837	12.243	**12.540**	13.660	13.450	**13.555**
T2	12.950	12.340	**12.645**	13.760	13.550	**13.655**
T3	12.920	12.807	**12.864**	13.823	13.720	**13.772**
Mean	**12.902**	**12.463**		**13.748**	**13.573**	
CD $_{0.05}$						
Time (A)		0.011	**AXB**		0.015	
Conditions (B)		0.011	**A x T**		0.019	
Treatments (T)		0.013	**B x T**		0.019	
			AxBxT		0.026	

A perusal of data in Table 4.22 reveals that a significant difference was observed in the total soluble solids content which was increased with a progressive increase in storage duration, irrespective of storage conditions. Maximum TSS 13.823°B was recorded in T3 (pasteurization with half preservative) during six month storage while minimum TSS 12.340°B was recorded in T2 (sodium benzoate 120 ppm) during three month storage .Among different preservative methods the highest changes was observed under ambient storage conditions and least was under refrigerated conditions.

4.5.3.2Titratable Acidity (%)

Table: 4.23 Effect of different treatments, storage duration and conditions on titratable acidity (%) of sugar malta iced tea

Treatments	**3 Months**			**6 Months**		
	Ambient	**Refrigerated**	**Mean**	**Ambient**	**Refrigerated**	**Mean**
T1	0.530	0.497	**0.514**	0.483	0.427	**0.455**
T2	0.560	0.537	**0.549**	0.527	0.413	**0.470**
T3	0.453	0.450	**0.452**	0.457	0.410	**0.434**
Mean	**0.514**	**0.495**		**0.489**	**0.417**	
CD $_{0.05}$						
Time (A)		0.006	**AXB**		0.009	
Conditions (B)		0.006	**A x T**		0.011	
Treatments (T)		0.008	**B x T**		0.011	
			AxBxT		0.015	

The titratable acidity was maintained at range of 0.410 % to 0.560 %. The maximum 0.560 % was recorded in T2 (sodium benzoate120 ppm) during three month storage and minimum acidity 0.410 % was recorded in T3 (pasteurization with half preservative) during six month storage. However, the data in Table 4.23 shows that there was a decreasing trend in the acid content during storage. It is evident from the data that the decrease reveals that advancement of storage duration, irrespective of storage conditions. Among different preservative methods the highest changes was observed under ambient storage conditions and least was under refrigerated conditions.

4.5.3.3 Ascorbic acid (mg/100mL)

The data of ascorbic acid content of sugar malta iced tea during different stages of storage are presented in Table 4.24. However, significantly higher ascorbic acid content in T2 (sodium benzoate 120 ppm) 20.283 mg/100 mL followed by T1 (pasteurization 65 ^{o}C for 30 minutes and least in T3 (pasteurization with half preservative) in sugar malta iced tea. However, there was a significantly higher decrease at ambient conditions than under refrigerated conditions.

Table: 4.24 Effect of different treatments, storage duration and conditions on ascorbic acid (mg/100mL) of sugar malta iced tea

Treatments	**3 Months**			**6 Months**		
	Ambient	**Refrigerated**	**Mean**	**Ambient**	**Refrigerated**	**Mean**
T1	17.700	18.617	**18.159**	10.583	11.617	**11.100**
T2	20.283	20.183	**20.233**	10.583	11.633	**11.108**
T3	18.800	18.750	**18.775**	10.583	11.517	**11.050**
Mean	**18.928**	**19.183**		**10.583**	**11.589**	
CD $_{0.05}$						
Time (A)		0.006	**AXB**		0.009	
Conditions (B)		0.006	**A x T**		0.011	
Treatments (T)		0.008	**B x T**		0.011	
			AxBxT		0.015	

4.5.3.4 Total sugars (%)

Table 4.25 shows the effect of storage on the total sugar content of sugar malta iced tea. The total sugars increased with a progressive increase in storage duration, with less change under refrigerated temperature than under ambient temperature. Which ranges between 10.577 % in T1 (pasteurization 65 ^{o}C for 30 minutes) to 11.453 % in T2 (sodium benzoate 120 ppm) and 10.577 % in T1 (pasteurization 65 ^{o}C for 30 minutes) to 11.440 % in T3 (pasteurization with half preservative) for ambient and refrigerated temperature respectively. However, their interactions were found to be non-significant.

Table: 4.25 Effect of different treatments, storage duration and conditions on total sugar (%) of sugar malta iced tea

Treatments	3 Months			6 Months		
	Ambient	**Refrigerated**	**Mean**	**Ambient**	**Refrigerated**	**Mean**
T1	10.783	10.577	**10.680**	11.367	11.077	**11.222**
T2	10.883	10.677	**10.780**	11.453	11.173	**11.313**
T3	10.953	10.843	**10.898**	11.330	11.440	**11.385**
Mean	**10.873**	**10.699**		**11.383**	**11.230**	
CD $_{0.05}$						
Time (A)		0.008	**AXB**		N/S	
Conditions (B)		0.008	**A x T**		0.014	
Treatments (T)		0.01	**B x T**		0.014	
			AxBxT		0.020	

4.5.3.5 Reducing sugars (%)

Table: 4.26 Effect of different treatments, storage duration and conditions on reducing sugars (%) of sugar malta iced tea

Treatments	3 Months			6 Months		
	Ambient	**Refrigerated**	**Mean**	**Ambient**	**Refrigerated**	**Mean**
T1	4.723	4.827	**4.775**	4.120	4.417	**4.269**
T2	4.773	4.877	**4.825**	4.167	4.473	**4.320**
T3	4.857	4.910	**4.884**	4.400	4.457	**4.429**
Mean	**4.784**	**4.871**		**4.229**	**4.449**	
CD $_{0.05}$						
Time (A)		0.006	**AXB**		0.008	
Conditions (B)		0.006	**A x T**		0.01	
Treatments (T)		0.007	**B x T**		0.01	
			AxBxT		N/S	

The data in Table 4.26 pertaining increasing trend in all three treatments of sugar malta iced tea, however the interaction was found non-significant. Maximum reducing sugars 4.910 % was recorded in T3 (pasteurization with half preservative) during three month storage and minimum reducing sugars 4.120 % was recorded in T1 (pasteurization 65 ^{o}C for 30 minutes) during six month storage. However, there was a significantly higher increase at ambient conditions than under refrigerated conditions.

4.5.3.6 Non- reducing sugars (%)

Table: 4.27 Effect of different treatments, storage duration and condition on non-reducing sugars (%) of sugar malta iced tea

Treatments	3 Months			6 Months		
	Ambient	**Refrigerated**	**Mean**	**Ambient**	**Refrigerated**	**Mean**
T1	5.723	5.827	**5.775**	4.120	4.417	**4.269**
T2	5.773	5.877	**5.825**	4.167	4.473	**4.320**
T3	5.857	5.910	**5.884**	4.400	4.457	**4.429**
Mean	**5.784**	**5.871**		**4.229**	**4.449**	
CD $_{0.05}$						
Time (A)		0.006	**AXB**		0.008	
Conditions (B)		0.006	**A x T**		0.010	
Treatments (T)		0.007	**B x T**		0.010	
			AxBxT		0.015	

A perusal of the data showed significant difference of non-reducing sugars presented in Table 4.27 which indicates that there was a gradual decline in non-reducing sugars content of sugar malta iced tea every successive storage interval in all treatments. The maximum non reducing sugars 5.910 % T3 (pasteurization with half preservative) and minimum 4.120 % was recorded in T1 (pasteurization 65 ^{o}C for 30 minutes) However, the decrease in non-reducing sugars irrespective of the storage conditions**.**

4.5.3.7 pH

A perusal of data in Table 4.28 reveals that a significant difference was observed in the pH which was increased with the advancement of storage duration, irrespective of

storage conditions. Which was ranging from 3.410 in T3 (pasteurization with half preservative) to 3.753 in T2 (sodium benzoate 120 ppm).Among different preservative methods the highest changes was observed under ambient storage conditions and least was under refrigerated conditions.

Table: 4.28 Effect of different treatments, storage duration and conditions on pH of sugar malta iced tea

Treatments	3 Months			6 Months		
	Ambient	Refrigerated	Mean	Ambient	Refrigerated	Mean
T1	3.657	3.413	**3.535**	3.713	3.513	**3.613**
T2	3.613	3.417	**3.515**	3.753	3.563	**3.658**
T3	3.567	3.410	**3.489**	3.713	3.573	**3.643**
Mean	**3.612**	**3.413**		**3.726**	**3.550**	
CD $_{0.05}$						
Time (A)		0.005	**AXB**		0.007	
Conditions (B)		0.005	**A x T**		0.009	
Treatments (T)		0.006	**B x T**		0.009	
			AxBxT		0.013	

4.5.3.8 Total phenols (mg/100mL)

Table: 4.29 Effect of different treatments, storage duration and conditions on total phenols of sugar malta iced tea

Treatments	3 Months			6 Months		
	Ambient	Refrigerated	Mean	Ambient	Refrigerated	Mean
T1	7.747	8.100	**7.924**	6.757	6.977	**6.867**
T2	7.467	7.623	**7.545**	6.577	6.960	**6.769**
T3	7.683	8.020	**7.852**	6.527	6.860	**6.694**
Mean	**7.632**	**7.914**		**6.620**	**6.932**	
CD $_{0.05}$						
Time (A)		0.017	**AXB**		0.008	
Conditions (B)		N/S	**A x T**		0.029	
Treatments (T)		0.021	**B x T**		0.029	
			AxBxT		0.041	

The data of total phenols of sugar malta iced tea during different storage intervals and conditions is given in Table 4.29. There was a significant decrease was observed for different treatments in sugar malta iced tea. The maximum total phenol 8.100 mg/100mL was recorded in in T1 (pasteurization 65 ^{o}C for 30 minutes) and minimum 6.527 mg/100mL was recorded in T3 (pasteurization with half preservative). However, the decrease in total phenol content was irrespective of the storage conditions**.**

4.5.3.9 β-carotenoids (mg/100mL)

Table: 4.30 Effect of different treatments, storage duration and conditions on β-carotenoids of sugar malta iced tea

Treatments	**3 Months**			**6 Months**		
	Ambient	**Refrigerated**	**Mean**	**Ambient**	**Refrigerated**	**Mean**
T1	7.747	8.100	**7.924**	6.757	6.977	**6.867**
T2	7.467	7.623	**7.545**	6.577	6.960	**6.769**
T3	7.683	8.020	**7.852**	6.527	6.860	**6.694**
Mean	**7.632**	**7.914**		**6.620**	**6.932**	
CD $_{0.05}$						
Time (A)		0.011	**AXB**		0.016	
Conditions (B)		0.011	**A x T**		0.019	
treatments (T)		0.014	**B x T**		0.019	
			AxBxT		0.027	

It is evident from the data presented in Table 4.30 that the total β-carotenoid content decreasing during storage. The maximum β-carotenoids 8.100 mg/100mL were recorded in T1 (pasteurization 65 ^{o}C for 30minutes) during 3 month storage and minimum 6.527 mg/100mL was recorded in T3 (pasteurization with half preservative). However the decrease in the β- carotenoids content was higher under ambient temperature than under refrigerated temperature.

4.5.3.10 Effect of different treatments, storage duration and conditions on sensory parameters of sugar malta iced tea

The sensory evaluation of sugar malta iced tea is graphically represented in fig 4. (C). The sensory evaluation for colour, flavour, taste and overall acceptability of sugar malta RTS reveals that highest mean were after three and six months of storage, among different treatments, the sensory scores were higher T3 (pasteurization with half preservative) stored under refrigerated temperature.

4.5.4 Effect of different treatments, storage duration and conditions on quality characteristics of honey malta iced tea

4.5.4.1Total Soluble Solids (^{0}B)

Table: 4.31 Effect of different treatments, storage duration and conditions on TSS (^{0}B) of honey malta iced tea

Treatments	3 Months			6 Months		
	Ambient	Refrigerated	Mean	Ambient	Refrigerated	Mean
T1	14.437	14.367	**14.402**	14.470	14.427	**14.449**
T2	14.477	14.353	**14.415**	14.500	14.467	**14.484**
T3	14.390	14.350	**14.370**	14.393	14.403	**14.398**
Mean	**14.435**	**14.357**		**14.454**	**14.432**	
CD $_{0.05}$						
Time (A)		0.006	AXB		0.009	
Conditions (B)		0.006	A x T		0.011	
Treatments (T)		0.007	B x T		0.011	
			AxBxT		0.016	

A perusal of data in Table 4.31 reveals that a significant difference was observed in the total soluble solids content which was increased with a progressive increase in storage duration, irrespective of storage conditions. Maximum TSS 14.500 ^{o}B was recorded in T2 (sodium benzoate 120 ppm) and minimum TSS 14.350 ^{o}B was recorded in T3 (pasteurization with half preservative). Among different preservative methods the highest changes was observed under ambient storage conditions and least was under refrigerated conditions.

4.5.4.2Titratable Acidity (%)

The titratable acidity was maintained at range of 0.440 % to 0.590 %. Maximum titratable acidity 0.590 % was recorded in T2 (sodium benzoate 120 ppm) during 3 month storage and minimum 0.440 % was recorded in T3 (pasteurization with half preservative). However, the data in Table 4.32 shows that there was a decreasing trend in the acid content during storage. It is evident from the data that the decrease reveals that advancement of storage duration, irrespective of storage conditions. Among different preservative methods the highest changes was observed under ambient storage conditions and least was under refrigerated conditions.

Table: 4.32 Effect of different treatments, storage duration and condition on titratable acidity (%) of honey malta iced tea

Treatments	3 Months			6 Months		
	Ambient	**Refrigerated**	**Mean**	**Ambient**	**Refrigerated**	**Mean**
T1	0.560	0.527	**0.544**	0.513	0.457	**0.485**
T2	0.590	0.567	**0.579**	0.557	0.443	**0.500**
T3	0.483	0.480	**0.482**	0.487	0.440	**0.464**
Mean	**0.544**	**0.525**		**0.519**	**0.447**	
CD $_{0.05}$						
Time (A)		0.006	**AXB**		0.009	
Conditions (B)		0.006	**A x T**		0.011	
Treatments (T)		0.008	**B x T**		0.011	
			AxBxT		0.015	

4.5.4.3 Ascorbic acid (mg/100mL)

The data of ascorbic acid content of honey malta iced tea during different stages of storage are presented in Table 4.33. However, significantly higher ascorbic acid content in T2 (sodium benzoate 120 ppm) 22.183 mg/100 ml followed by T1 (pasteurization 65 ^{o}C for 30 minutes and least in T3 (pasteurization with half preservative) in honey malta iced tea. However, there was a significantly higher decrease at ambient conditions than under refrigerated conditions.

Table: 4.33 Effect of different treatments, storage duration and conditions on ascorbic acid (mg/100mL) of honey malta iced tea

Treatments	3 Months			6 Months		
	Ambient	Refrigerated	Mean	Ambient	Refrigerated	Mean
T1	16.700	20.617	**18.659**	9.583	9.460	**9.521**
T2	19.283	22.183	**20.733**	9.613	9.713	**9.663**
T3	17.800	20.750	**19.275**	9.583	9.683	**9.633**
Mean	**17.928**	**21.183**		**9.583**	**9.619**	
CD $_{0.05}$						
Time (A)		0.118	**AXB**		0.168	
Conditions (B)		0.118	**A x T**		0.205	
Treatments (T)		0.145	**B x T**		0.205	
			AxBxT		0.29	

4.5.4.4 Total sugars (%)

Table: 4.34 Effect of different treatments, storage duration and conditions on total sugar (%) of honey malta iced tea

Treatments	3 Months			6 Months		
	Ambient	Refrigerated	Mean	Ambient	Refrigerated	Mean
T1	11.350	11.250	**11.300**	11.470	11.440	**11.597**
T2	11.550	11.340	**11.445**	11.393	11.550	**11.700**
T3	11.610	11.480	**11.545**	11.860	11.610	**11.917**
Mean	**12.629**	**12.802**		**11.518**	**11.958**	
CD $_{0.05}$						
Time (A)		0.012	**AXB**		0.017	
Conditions (B)		0.012	**A x T**		0.021	
Treatments (T)		0.015	**B x T**		0.021	
			AxBxT		0.029	

Table 4.34 shows the effect of storage on the total sugar content of honey malta iced tea. The significant decrease in total sugar during storage, with less change under refrigerated temperature than under ambient temperature. Which ranges between 11.300 % in T1 (pasteurization 65 ^{o}C for 30 minutes) to 12.773 % in T3 (pasteurization with half

preservative) and 11.893 % in T1 (pasteurization 65 °C for 30 minutes) to12.880 % in T3 (pasteurization with half preservative) for ambient and refrigerated temperature respectively.

4.5.4.5 Reducing sugars (%)

Table: 4.35 Effect of different treatments, storage duration and conditions on reducing sugars (%) of honey malta iced tea

Treatments	3 Months			6 Months		
	Ambient	Refrigerated	Mean	Ambient	Refrigerated	Mean
T1	5.877	5.580	**5.729**	6.287	6.183	**6.235**
T2	5.933	5.627	**5.780**	6.337	6.233	**6.285**
T3	5.917	5.860	**5.889**	6.370	6.317	**6.344**
Mean	**5.909**	**5.689**		**6.331**	**6.244**	
$CD_{0.05}$						
Time (A)		0.006	AXB		0.008	
Conditions (B)		0.006	A x T		0.010	
Treatments (T)		0.007	B x T		0.010	
			AxBxT		0.015	

The data in Table 4.35 pertaining significant increasing trend in all three treatments of honey malta iced tea, maximum reducing sugars 6.370 % was recorded in T3 (pasteurization with half preservative) during six month storage and minimum reducing sugars 5.627 % was recorded in T2 (sodium benzoate 120 ppm) during three month storage. However, there was a significantly higher increase at ambient conditions than refrigerated conditions.

4.5.4.6 Non- reducing sugars (%)

A perusal of data in Table 4.36 shows that there was a significant difference of non-reducing sugars for different treatments in honey malta iced tea. The maximum non-reducing sugars 6.940 % was recorded in T2 (sodium benzoate 120 ppm) and minimum non-reducing sugars 6.150 %was recorded in T2 (sodium benzoate 120 ppm). However, the decrease in non-reducing sugars irrespective of the storage conditions**.**

Table: 4.36 Effect of different treatments, storage duration and conditions on non-reducing sugars (%) of honey malta iced tea

Treatments	3 Months			6 Months		
	Ambient	Refrigerated	Mean	Ambient	Refrigerated	Mean
T1	5.573	5.677	**5.625**	5.190	5.267	**5.229**
T2	5.623	5.727	**5.675**	5.017	5.323	**5.170**
T3	5.707	5.627	**5.660**	5.250	5.307	**5.279**
Mean	**5.634**	**5.721**		**5.079**	**5.299**	
$CD_{0.05}$						
Time (A)		0.006	AXB		0.008	
Conditions (B)		0.006	A x T		0.01	
Treatments (T)		0.007	B x T		0.01	
			AxBxT		0.015	

4.5.4.7 pH

Table: 4.37 Effect of different treatments, storage duration and conditions on pH of honey malta iced tea

Treatments	3 Months			6 Months		
	Ambient	Refrigerated	Mean	Ambient	Refrigerated	Mean
T1	3.437	3.367	**3.402**	3.460	3.440	**3.430**
T2	3.477	3.357	**3.417**	3.500	3.360	**3.430**
T3	3.390	3.350	**3.370**	3.420	3.400	**3.410**
Mean	**3.358**	**3.435**		**3.454**	**3.347**	
$CD_{0.05}$						
Time (A)		0.006	AXB		0.009	
Conditions (B)		0.006	A x T		0.011	
treatments (T)		0.008	B x T		0.011	
			AxBxT		0.015	

A perusal of data in Table 4.37 reveals that a significant difference was observed in the pH which was ranging from 3.357 to 3.500 and pH increased with the advancement of storage duration, irrespective of storage conditions. Among different preservative methods the highest changes was observed under ambient storage conditions and least was under refrigerated conditions.

4.5.4.8 Total phenols (mg/100mL)

Table: 4.38 Effect of different treatments, storage duration and conditions on total phenols of honey malta iced tea

Treatments	3 Months			6 Months		
	Ambient	**Refrigerated**	**Mean**	**Ambient**	**Refrigerated**	**Mean**
T1	7.553	7.830	**7.692**	6.547	6.750	**6.649**
T2	7.753	7.930	**7.842**	6.580	6.667	**6.624**
T3	7.640	7.927	**7.784**	6.653	6.740	**6.697**
Mean	**7.649**	**7.896**		**6.593**	**6.719**	
CD $_{0.05}$						
Time (A)		0.010	**AXB**		0.014	
Conditions (B)		0.010	**A x T**		0.017	
treatments (T)		0.012	**B x T**		0.017	
			AxBxT		0.024	

The data of total phenols of honey malta iced tea during different storage intervals and conditions is given in Table 4.38 There was a significant decrease was observed for different treatments in honey malta iced tea. The maximum total phenols 7.930 mg/100mL was recorded in T2 (sodium benzoate 120 ppm) during3 month storage and minimum total phenols 6.547 mg/100mL was recorded in T1 during 6 month storage. However, the decrease in total phenols content was irrespective of the storage conditions**.**

4.5.4.9 β-carotenoids (mg/100mL)

It is evident from the data presented in Table 4.39 that the total carotenoids content decreasing during storage. The maximum β-carotenoids 7.943 mg/100mL was recorded in T1 during 3 month storage and minimum β-carotenoids 6.310 mg/100mL was recorded in T3 (pasteurization with half preservative) during six month storage However the decrease in the β-carotenoids content was higher under ambient temperature than under refrigerated temperature.

Table: 4.39 Effect of different treatments, storage duration and conditions on β-carotenoids of honey malta iced tea

Treatments	3 Months			6 Months		
	Ambient	**Refrigerated**	**Mean**	**Ambient**	**Refrigerated**	**Mean**
T1	7.930	7.943	**7.937**	6.930	6.960	**6.945**
T2	7.147	7.257	**7.202**	6.760	6.763	**6.762**
T3	7.767	7.327	**7.547**	6.310	6.407	**6.359**
Mean	**7.619**	**7.505**		**6.667**		
CD $_{0.05}$						
Time (A)		0.011	**AXB**		0.015	
Conditions (B)		0.011	**A x T**		0.019	
Treatments (T)		0.013	**B x T**		0.019	
			AxBxT		0.027	

4.5.4.10 Effect of different treatments, storage duration and conditions on sensory parameters of honey malta iced tea

The sensory evaluation of honey malta iced tea is graphically represented in fig 4.8. The sensory evaluation for colour, flavour, taste and overall acceptability of honey malta iced tea reveals that highest mean were after three and six months of storage, among different treatments, the sensory scores were higher T3 (pasteurization with half preservative) stored under refrigerated temperature.

4.6 Effect of different treatments, storage duration and conditions on microbial quality characteristics of malta products

During storage the all treatment of prepared malta products viz; sugar malta RTS, honey malta RTS, sugar malta iced tea, and honey malta iced tea stored under both ambient and refrigerated storage conditions were subjected for total microbial count at initial (0 month) three and six month. All the treatments in relation to storage conditions were found to be free from microbes up to six month of storage.

4.7 Cost of production of malta fruit beverages:

The cost incurred in preparation of malta beverages was calculated by taking into consideration the cost of all inputs used and cost involved during the preparation during

the preparation of all beverages. The processing and other expenses including depreciation were added to total expenditure. The comparative cost of honey and sugar based malta RTS has been given in Table 4.40 and the comparative cost of honey and sugar based malta iced tea has been given in Table 4.41. The sale price per bottle of products was calculated after adding 25% profit margin. The cost of production of sugar malta RTS packed in bottles of 500 mL capacity was calculated as Rs. 30.00 on the other hand the cost of production of honey malt RTS packed in bottles of 500 mL capacity was calculated as Rs. 63.00. The cost of production of sugar malta iced tea was calculated as Rs.35.00 and Rs.65.00 for honey malta iced tea.

Table 4.40 Cost of production of Sugar malta RTS and honey malta RTS

S. No.	Items	Sugar malta RTS			Honey malta RTS		
		Quantity	Rate (Rs/Kg)	Amount	Quantity	Rate (Rs/Kg)	Amount
1	Cost of fruits	720 ml	100	72	840 ml	100	84
2	Cost of sugar/ honey	720 g	50	36	840 ml	250	210
3	Cost of sodium benzoate	1 g	500	0.50	1 g	500	0.50
4	Bottles required(200ml)	12 Nos.	5/bottle	60	12 Nos.	5/bottle	60
5	Crown corks	12 Nos.	2Rs/cork	24	12 Nos.	2Rs/cork	24
6	Total			192.5			378.5
7	Overhead charges @ 20 % (office)			38.5			75.7
8	Processing charges @ 20 %			38.5			75.7
9	Additional charges @10 %			19.25			37.85
10	Depreciation of machinery and equipment @ 10 %			19.25			37.85
11	Total			288.75			605.6
12	Profit margins 25 %			72.18			151.4
13	Grand total			360.93			757
14	Cost per unit bottle (500ml)			30.07 ≈ 30.00			63.08≈63

Table 4.41 Cost of production of Sugar malta iced tea and honey malta iced tea

S. No.	Items	Sugar malta Iced Tea			Honey malta Iced Tea		
		Quantity	Rate (Rs/Kg)	Amount	Quantity	Rate (Rs/Kg)	Amount
1	Cost of fruit juice	720 ml	100	72	840 ml	100	84
2	Cost of sugar/ honey	720 g	50	36	840 ml	250	210
3	Tea leaves(Tata premium)	60 g	250	15	60g	250	15
4	Cost of sodium benzoate	1 g	500	0.50	1 g	500	0.50
5	Bottles required(200ml)	12 Nos.	5/bottle	60	12 Nos.	5/bottle	60.00
6	Crown corks	12 Nos.	2Rs/cork	24	12 Nos.	2Rs/cork	24.00
	Total			207.50			393.50
7	Overhead charges @ 20 % (office)			41.50			78.70
8	Processing charges @ 20 %			41.50			78.70
9	Additional charges @ 10 %			20.75			39.35
10	Depreciation of machinery and equipment @ 10 %			20.75			39.35
11	Total			332.00			629.60
12	Profit margins 25 %			83.00			157.40
13	Grand total			415.00			787.00
14	Cost per unit bottle (200ml)			34.58≈ 35.00			65.58≈ 65.00

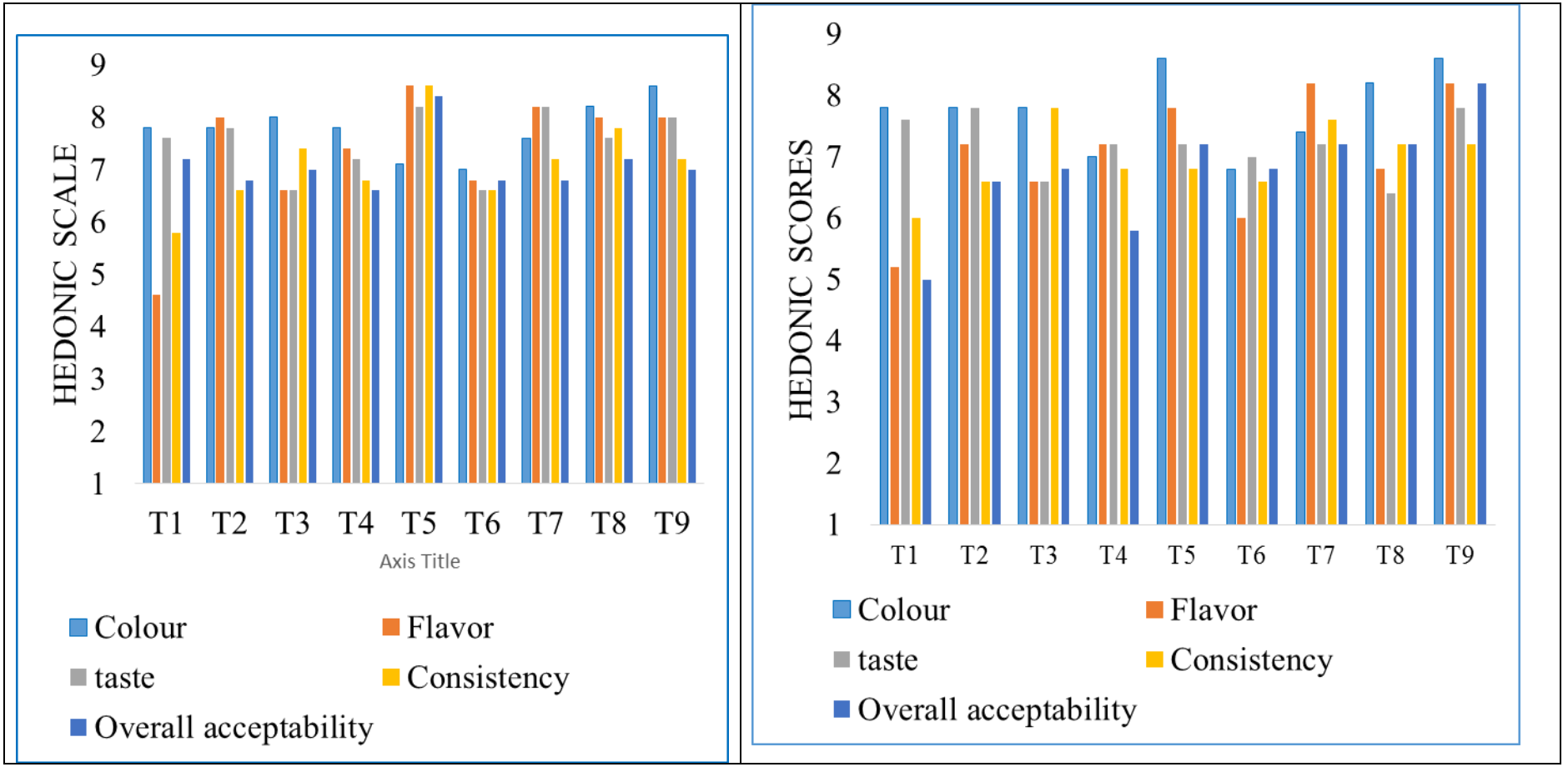
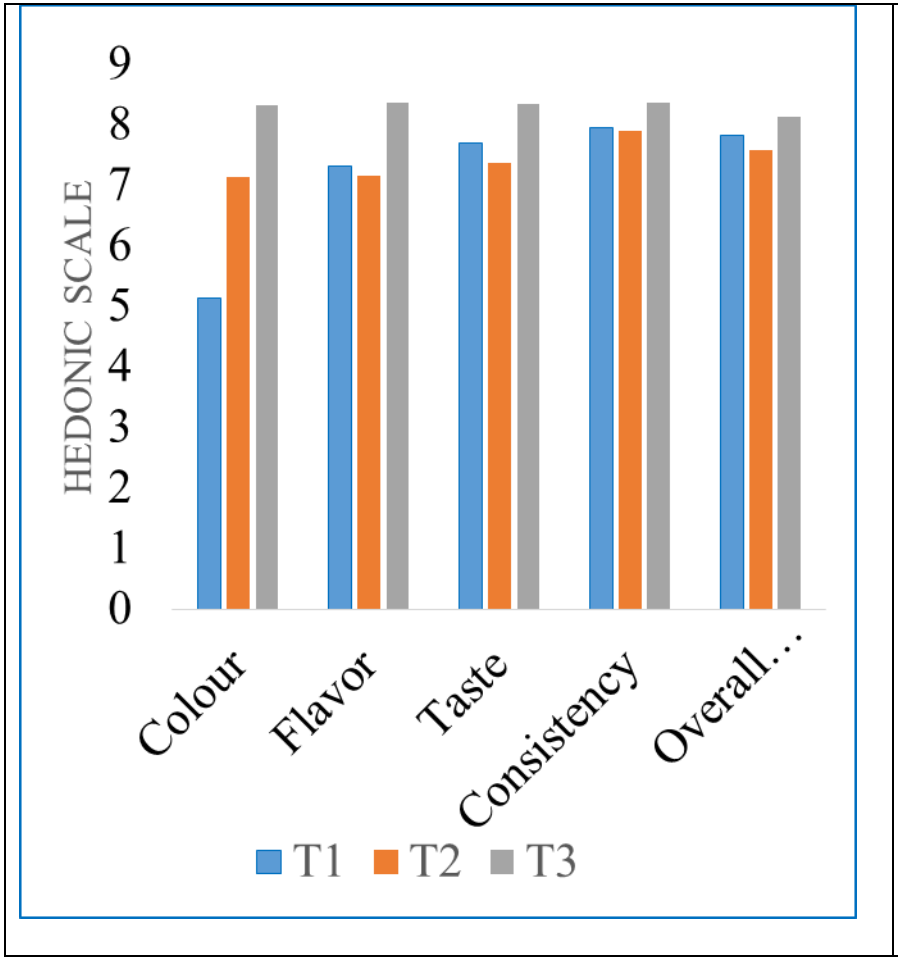


Fig 4.1 Sensory evaluation of sugar malta RTS

Fig 4.2 Sensory evaluation of sugar malta RTS



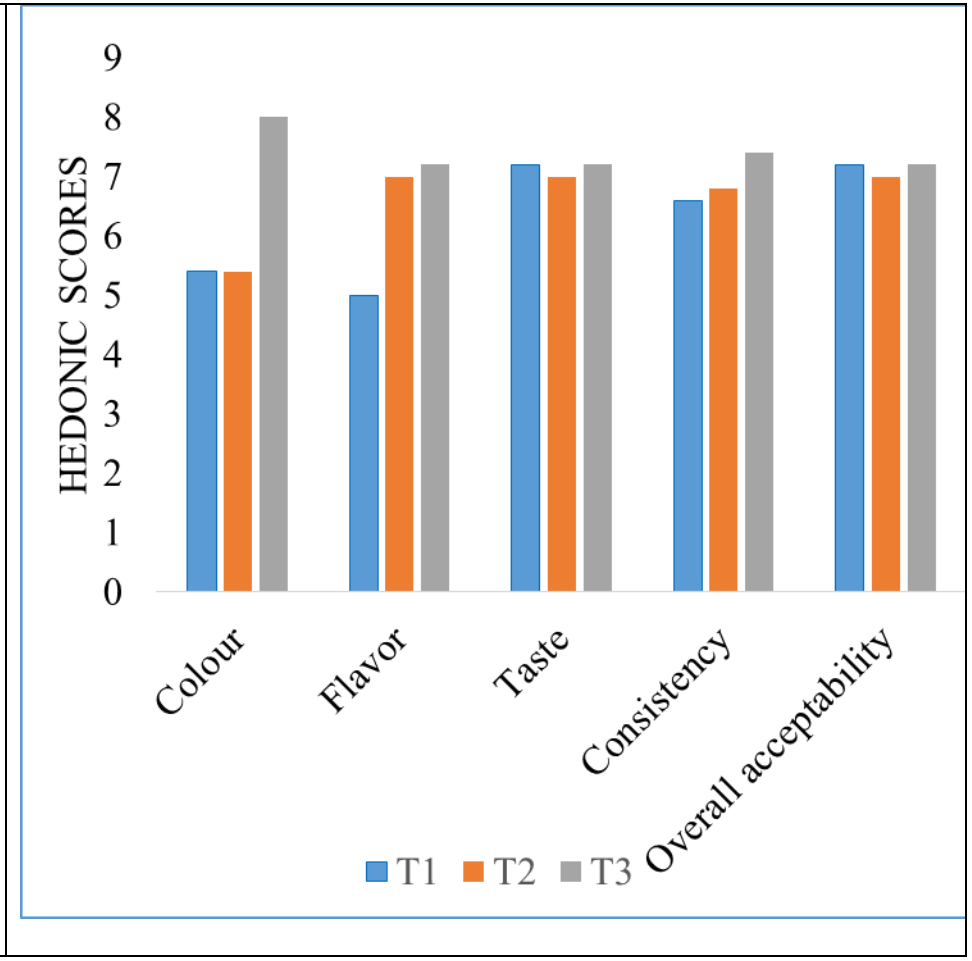


Fig 4.3 Sensory evaluation of sugar malta iced tea

Fig 4.4 Sensory evaluation of honey malta iced tea

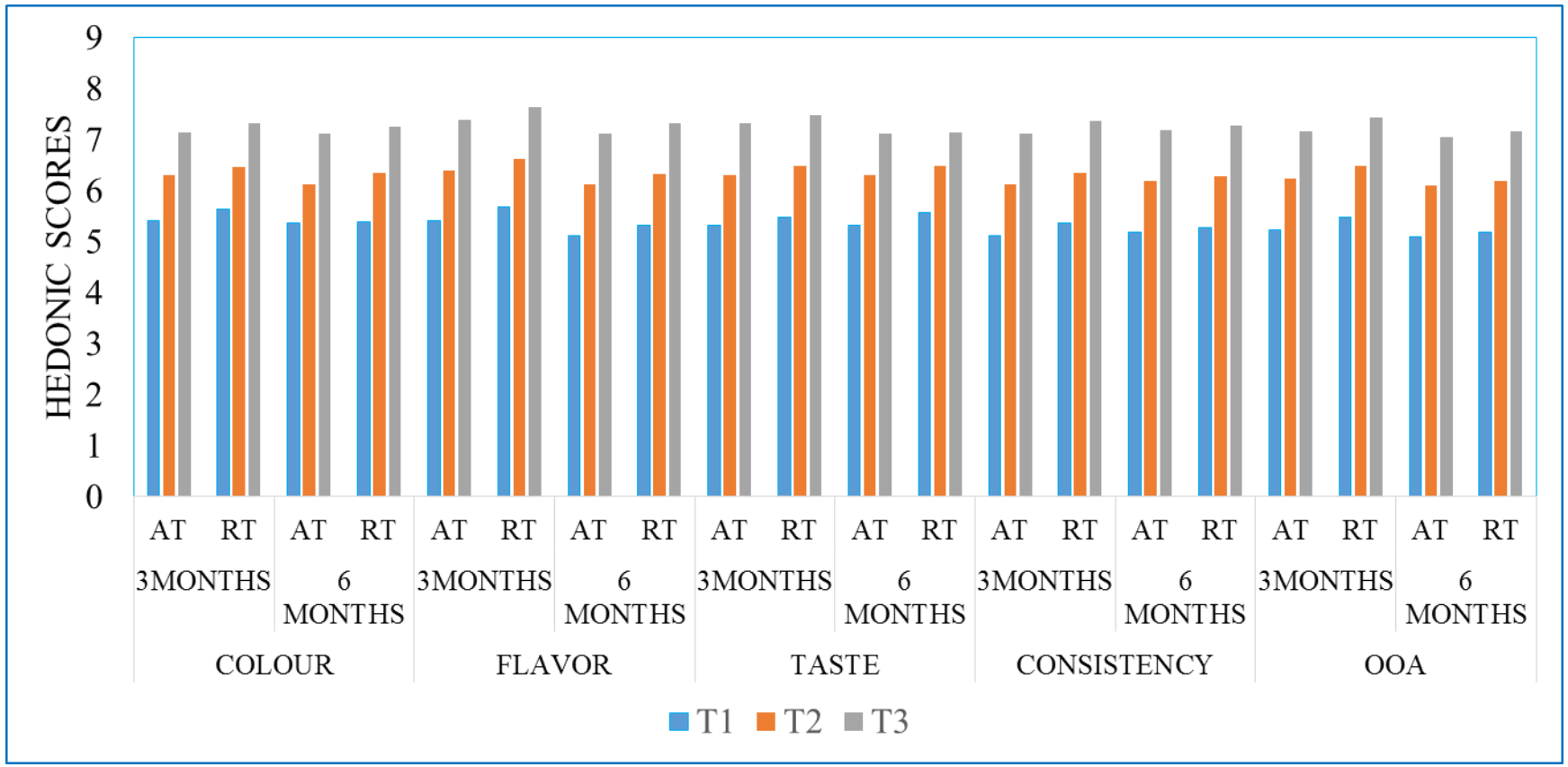


Fig 4.5: Effect of different treatments, storage duration and storage condition on sensory parameters of sugar malta RTS

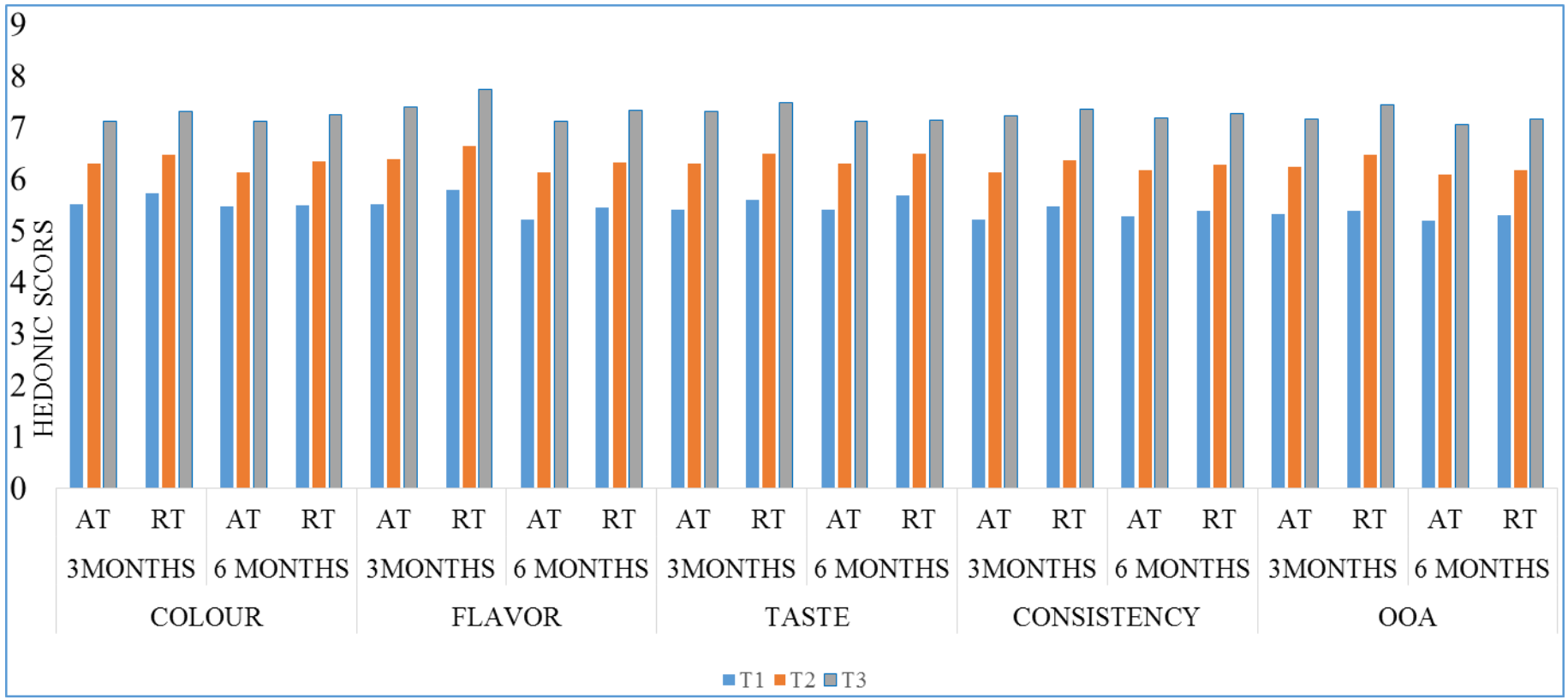


Fig 4.6: Effect of different treatments, storage duration and storage condition on sensory parameters of honey malta RTS

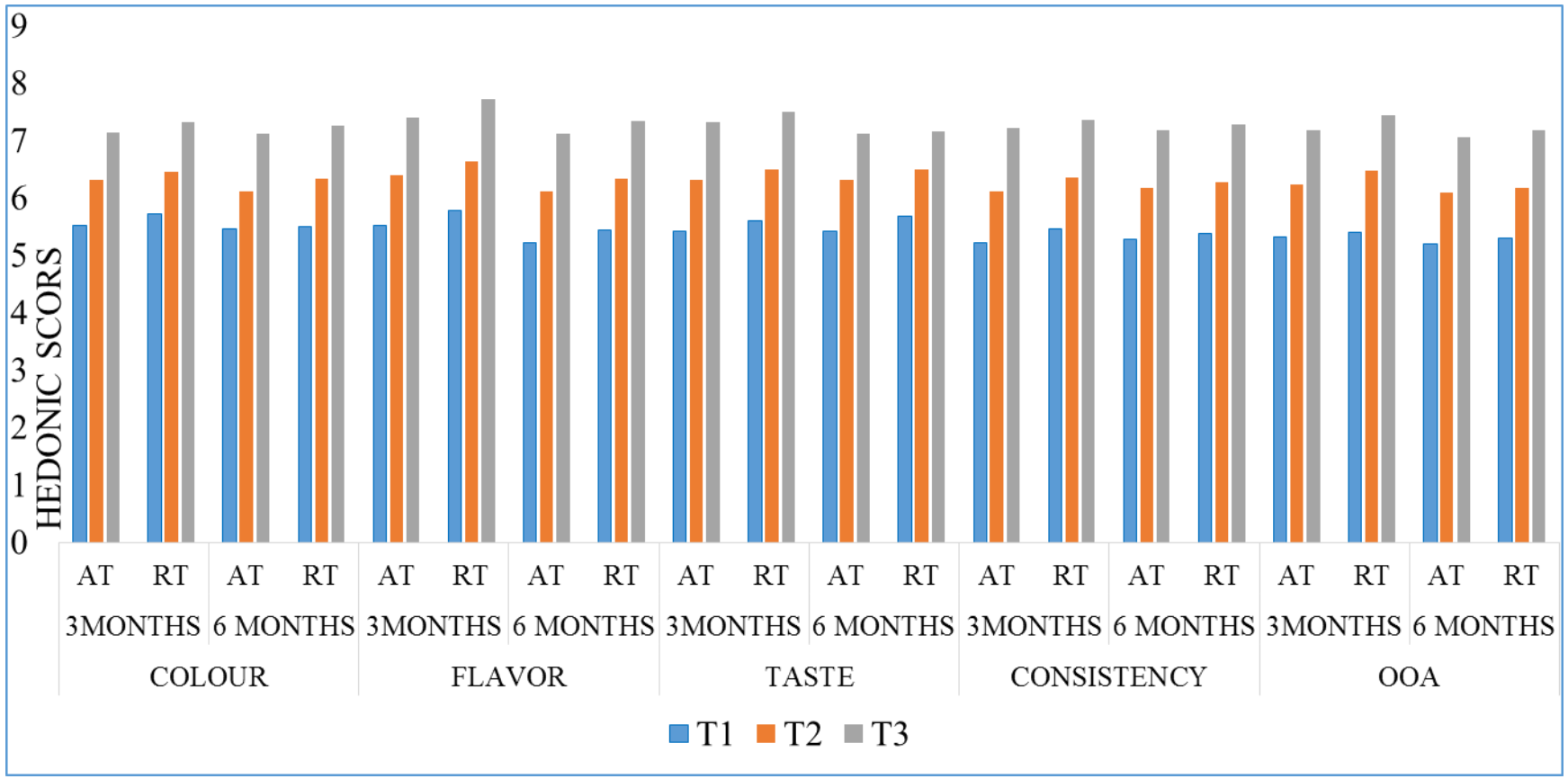


Fig 4.7: Effect of different treatments, storage duration and storage condition on sensory parameters of sugar malta iced tea

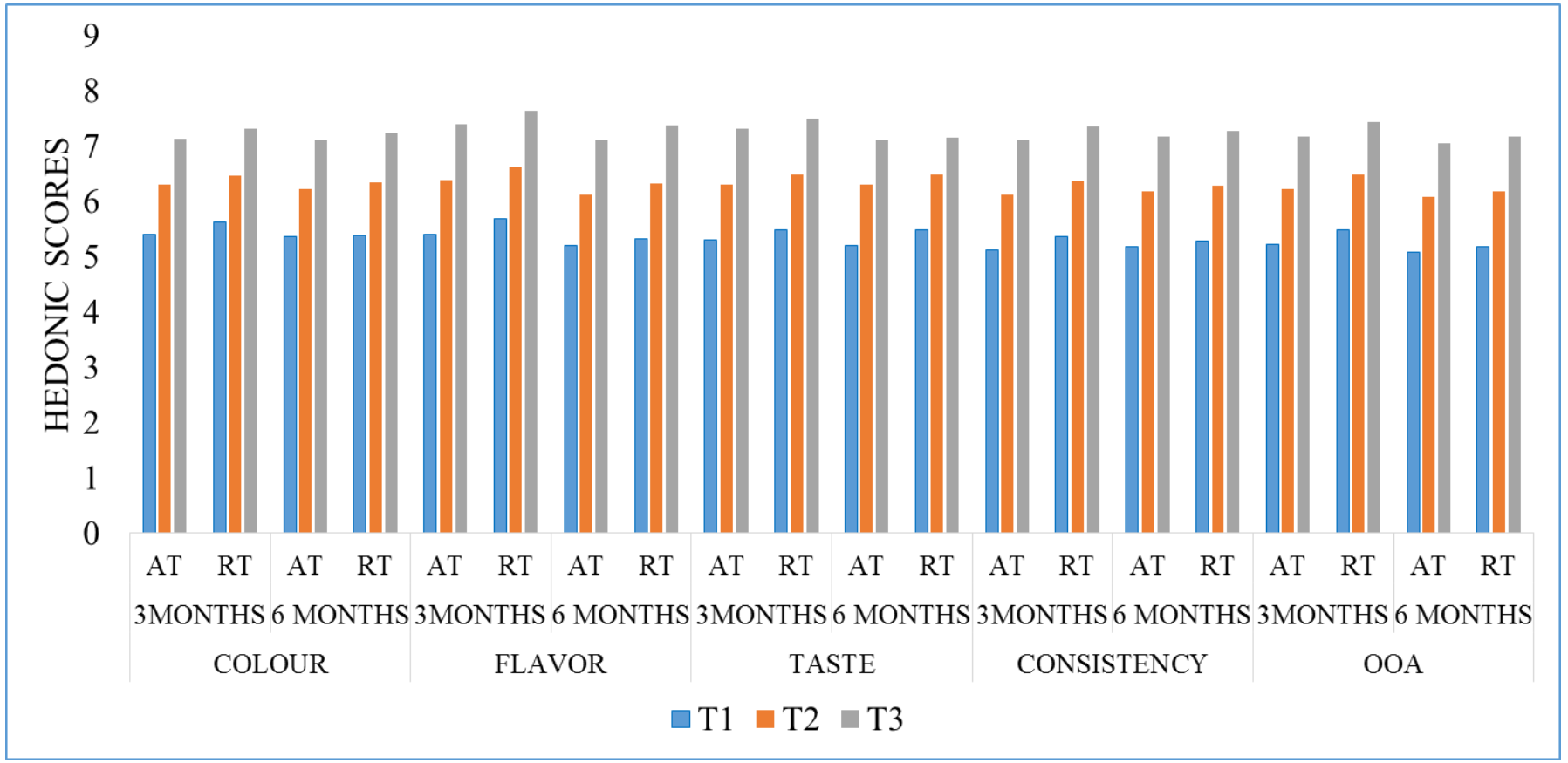


Fig: 4.8 Effect of different treatments, storage duration and conditions on sensory parameters of honey malta iced tea

A: Honey malta iced tea

B: Sugar malta iced tea

Plate: 1 Malta iced tea with different sweetening agents

A: Honey malta RTS

B: Sugar malta RTS

Plate: 2 Malta RTS with different sweetening agents

DISCUSSION

CHAPTER-5
DISCUSSION

The present investigations entitled "Studies on the preparation and evaluation of functionally enriched value added product from malta fruit (*Citrus sinensis* L Osbeck)*"* were carried out in the Department of Food Science and Technology, College of Horticulture, VCSGUUHF, Bharsar (Pauri Garhwal). The results obtained are discussed in this chapter under the following heads:

5.1Physico-chemical characteristics of fresh malta fruits.

5.2 Physico-chemical characteristics of Honey

5.3 Development of honey and sugar based products and changes their quality during storage.

5.4 Effect of storage on microbial quality

5.5 Economic evaluation of malta fruit beverages

5.1 Physico-chemical characteristics of fresh malta fruits.

The malta fruit had an average fruit weight, and width as 149.62g and 6.80±0.78 cm, respectively. The average number of ten fruit segments and number of seed/fruit was observed to be 10.00±0.00 and 31.33±1.53, respectively. The width and weight peeled fruits was 5.26±0.35 cm and 130.99±17.30g, respectively. The average weight of peel was found 40.89±3.01 g and thickness was 0.54±0.11 cm. The total soluble solids and pH of malta fruit juice was10.33±1.15°B and 2.97±0.02. The result were closer to Syed *et al.,* (2012), who reported the *citrus sinensis* consist of average fruit weight, diameter, thickness and weight of peel, weight of fruit without peel, number of seeds and segments per fruit, 199.0g, 84.06mm, 2.34mm, 47.10g, 167.25g, 17 seeds and 10 segments per fruit, TSS, pH, acidity and ascorbic acid of fruit as 10^{o}B, 3.7, 0.41% and 43.00mg/100g. The total phenols was recorded 7.98 mg/100g which was lower than 23.07mg/100g studied by Kumar *et al.* (2013). The total sugars were recorded 8.82% which was closer to 9.35% studied by Etebu *et al.* (2014).

5.2 Physico - chemical characteristics of honey

The total soluble solids content recorded for honey was 81.1^{o} B which was more than the values observed by Lakhanpal (2010). The total acids of honey was 4.28 % as

citric acid, which were slightly higher as recorded by Singh *et al.* (1988) and Kaushik *et al.* (1993). Lakhanpal (2010), Mishra (1995) and Phadke (1967) recorded the total sugars and non-reducing sugars of different types of honey, which were near to the values recorded in the investigation. The total phenols was 18.30 mg/100g which was slightly lower as recorded by Al-Mamary *et al.* (2002).

5.3 Development of honey and sugar based products and changes their quality during storage.

The changes in physico-chemical characteristics during storage and other quality characteristics of different beverages i.e. ready-to-serve malta drink and malta iced tea sweetened with different sweetening agents (honey and sugar) were studied. The beverages were packed in 500 ml glass bottles and after using different preservation methods viz; pasteurization (65^{o}C for 30 minutes), sodium benzoate 120ppm and pasteurization with half preservative (65^{o}C for 30 minutes and 60 ppm sodium benzoate) and stored under different storage conditions *i.e.* ambient temperature (13.2 to 27^{o}C) and refrigerated temperature (4 to 7^{o}C). These beverages were then analysed at periodic intervals of 0, 3 and 6 months. Lakhanpal (2010) prepared honey based products with mango, guava and kiwifruit with honey as sweetening agents.

5.3.1 Total soluble solids

During storage a marginal increase in total soluble solids was recorded for all the fruit beverages sweetened with honey and sugar during storage. Lanjhiyana *et al*. (2010) has also been recorded an increase in TSS in lime-ginger RTS/blended RTS, during storage by which was attributed to the conversion of polysaccharides like pectin, cellulose, starch etc into simple sugars. Das (2009) in jamun beverages. Deka (2004) reported an increasing trend in total soluble solids during storage at ambient and low temperature in lime-aonla and mango-pineapple spiced RTS beverages. The increase in TSS might also be due to the formation of pectic substances from protopectin and mono-saccharides from disaccharides *i.e.* degradation of sucrose into glucose and fructose. Similar results have been reported by Sarolia and Mukherjee (2002) in their studies.Kaushik *et al.* (1993) reported the increase in TSS in honey from 81.50 to 82.28 per cent after four months storage. The increase in TSS of honey based beverages may be because of this reason. Deka *et al*. (2005) recorded less increase in total soluble solids at low temperature as compared to ambient temperature. Jawanda *et al.,* (1978) reported the the increase in TSS

might be due to solublization of pulp constituents during storage and degradation of starch into simple sugars due to hydrolysis of polysaccharides.

Saravanan *et al*. (2004) reported the increase in TSS might be attributed to breakdown of the complex carbohydrates into simple soluble carbohydrates. A slight increase in total soluble solids of fruit beverages i.e. papaya, guava and bael were recorded by Similar results have been reported by Sarolia and Mukherjee (2002) in their studies on lime juice.

These results were in conformity with the findings of Tripathi *et al*. (1992); Attri *et al.* (1998) and Pandey and Singh (1999). The increase in total soluble solids in honey could be correlated with the increase in total soluble solids of fruit beverages during storage.This slight increase in total soluble solids is considered as a result of hydrolysis of polysaccharides into monosaccharide. The same increasing trend of total soluble solids was observed by Balaswamy *et al.* (2011), during storage period in different types of carbonated and non-carbonated RTS beverages.

5.3.2 Titratable acidity

Titratable acidity plays a vital role in the shelf-life of any type of beverage or drinks and provides unfavorable conditions for the multiplication of microorganisms. It also helps to ensure some chemical changes during processing and storage.

Changes occurring in fruit based beverages sweetened with sugar and honey at different storage temperatures during storage showed a significant decline in titratable acidity. The rate of decrease in acidity was higher at ambient storage as compare to refrigerated temperature. Khurdiya (1980) also reported a slight decrease in acidity of kinnow juice stored after preserving with sulphur dioxide. The decrease in acidity might be due to chemical reactions taking place between organic acids and pigments by the action of enzymes and temperature (Kannan and Thirumaran, 2001). Khurdiya (1980) noticed a slight decrease in acidity of dried ber beverage and lime squash during storage. Ahire *et al*. (2010) observed a slight decrease in acidity of pomegranate juice. They observed the the rate of decrease in acidity was higher at ambient storage. Bawa and Saini (1987) observed the the decrease in acidity was higher at ambient temperature in carrot juice. Similar findings were recorded in this study. Mehta *et al.* (1983) reported decline in acidity during storage of citrus juice. Titratable acidity decrease in juice could be attributed to chemical interaction between organic constituent of juice induced by temperature and action of enzymes.

5.3.3 Ascorbic acid

A significant decrease in ascorbic acid content was observed in all the beverages during storage. The decrease of ascorbic acid was more at ambient temperature than under refrigerated temperature. Deka *et al.,* (2004) also observed loss in ascorbic acid it might be due to the oxidation or irreversible conversion of L-ascorbic acid into dehydroascorbic acid caused by trapped or residual oxygen in the glass bottles. De Man (1980) reported the out of four important enzymes (ascorbic acid oxidase, phenolase, cytochrome oxidase and peroxidase) catalyzing the decomposition of ascorbic acid, only ascorbic acid oxidase involves a direct reaction between enzyme, substrate and molecular oxygen while other enzymes oxidize the vitamins indirectly. Hussain *et al*. (2011) who reported a decrease in ascorbic acid of the processed products during the storage. They recorded the ascorbic acid content in strawberry pulp was affected by treatments *viz*., freezing, heating and accelerated storage. These losses of ascorbic acid were attributed to the effect of processing, storage time and exposure to light. Majumdar *et al*. (2011) also found remarkable loss of vitamin C (74%) during 6 month storage of cucumber-litchi-lemon juice at room temperature (28±2^{o}C). Tiwari (2000) reported 26.47% loss of vitamin C during 6 month storage of guava and papaya beverage at room temperature. Das (2009) observed a significant reduction in ascorbic acid content of jamun beverages during storage this decrease may be due to the oxidation of ascorbic acid into dehyroascorbic acid by trapped oxygen. Ahire *et al.* (2010) recorded higher decrease in ascorbic acid content in juices at ambient storage conditions. Viberg *et al.* (1999) also reported a decrease in ascorbic acid during storage. They recorded the ascorbic acid content in strawberry pulp was affected by treatments *viz*., freezing, heating and accelerated storage. These losses of ascorbic acid were attributed to the effect of processing, storage time and exposure to light. Reduction was higher at ambient temperature as compared to low temperature storage. Results were in accordance to Cortes *et al.* (2005) who reported 4.1 per cent loss of vitamin C during 132 day storage of orange-carrot juice at 40°C and noticed lower losses of vitamin C at refrigerated temperature during storage.

5.3.4 Sugars

The increase in total sugars and reducing sugars in beverages was less at low temperature as compared to beverages stored at room temperature during six months storage. The increase in reducing sugars content can be ascribed to the inversion of sucrose

to glucose and fructose by the acid of the beverages. Similar results were observed in fruit based nectars sweetened with honey by Lakhanpal (2010), in lime-ginger cocktail by Sethi (1992) and Krishnaveni *et al.* (2001) in jackfruit squash during storage. Lee and Nagy (1988) reported the the inversion of sucrose into reducing sugars during storage in presence of acidic environment might have increased the reducing sugars content of fruit nectars in grape juice. Madan and Dhawan (2005) observed a gradual increase in the total sugars content of carrot juice. They also reported the rate of increase in total sugars of carrot juice was recorded to be higher at ambient storage as compared to low temperature conditions. These findings are close agreement of present findings. Sood *et al*. (2009) recorded a significant increase in total and reducing sugars content of mango squash during storage and concluded the hydrolysis of non-reducing sugars into reducing sugars resulted in increasing the reducing sugars during storage. Similar results have been reported by Lanjhiyana *et al*. (2010) in lime-ginger squash. They related this variation in the different fraction of sugars to the hydrolysis of complex polysaccharides like starch and pectin into monosaccharides.

However, non-reducing sugars decreased throughout the storage interval. This slight decrease in non-reducing sugars during storage might be attributed to the involvement of sugars in browning reactions and formation of hydroxymethylfurfural (HMF). The findings of current research are well supported by Singh et al. (2009). Increase in storage time non-reducing sugars decreased. The results are also in lime with the findings of the Chowdhury et al. (2008) who studied the six month storage effect on the shelf life of mixed juice and found significant decrease in non- reducing sugars due to breakdown of sucrose with the reaction of acids. Din *et al.* (2014) observed significant increase in total sugars and reducing sugars and decrease in non -reducing sugars which findings are close agreement with the present research studies.

5.3.5 pH

pH is inversely proportional to acidity of any medium. The present study for malta beverages also depicted the same changing pattern of pH parameter. pH values affected by various treatments, storage intervals and storage conditions pH value showed significant increase in present study. Similar results were also observed Pawar *et al.* (2011) and concluded the acidity of beverage decreased with increase in storage period. Results were similar to the finding of Thamilsevi *et al*. (2015) also observed similar results in his study; they observed a marginal increase in pH of all beverages and the increase in pH was due to

the decrease of the products acidity. Similarly Saradha *et al.* (2004) have studied about the changes in pH of whey based fruit beverage. Archana and Laxman (2014) evaluate similar findings for storage of tamarind squash and concluded reason of corresponding decrease in acidity. Similar observation was observed by Nath *et al.* (2005) in kinnow mandarin-ginger squash.

5.3.6 Total phenols

Polyphenols are the most important chemical constituent they influences the taste and flavor in tea. Present study showed a significant decrease in total phenol content of malta ready-to-serve beverage and iced tea was observed during storage. Losses were higher at ambient storage as compared to low temperature storage. These results are almost similar to earlier study of total phenolic contents of Indian herbal teas, studied y Zhang *et al*. (2008) who recorded no significant changes in total phenolics content of apple juice upto five days storage which was found to be decreased significantly after five days storage at ambient temperature. Miller *et al*. (1995) also reported the same findings for apple juice stored at 4^{o}C over 10 days. Raj *et al*. (2011) reported a significant decrease in the phenolic content of sand pear and apple juice blends during six months storage. During the processing of food, various transformations of phenolics occur to produce yellowish to brownish pigments (Clifford, 2000). The decline in the phenol contents during storage has also been reported earlier by Duda-Chodak *et al.* (2008).

5.3.7 β- Carotenoids

The decrease in carotenoids was found during storage of all malta beverages under both the storage conditions. Decrease was more at ambient than the refrigerated temperature. Similarly, Srivastava (1998) recorded a slight decrease in total carotenoids of mango RTS beverage during six month of storage. The total carotenoids content has been found to decrease over a period of six month in peach nectar by Deka *et al.* (2005) who observed 87.53 to 90.24 per cent retention of total carotenoids after six month of storage. Whereas, Lavelli *et al*. (2009) observed no changes in β-carotene content of mango products. Many researchers have reported no or only a minor degradation of carrot carotenoids during cold storage (Kopas-Lane and Warthesen, 1995 and Howard *et al*., 1999). Apart from isomerization and oxidation of high carotenoids containing fruits and

vegetables, carotenoids level increases during processing (Chandler and Shwartz, 1988). Thermal processing has been reported to increase the amount of carotenoids in products.

5.3.8 Sensory characteristics

Sensory score of all the products exhibited decreasing trend with the passage of time. The decrease in score for colour, flavour, taste, consistency and overall acceptability was less in refrigerated temperature storage as compare to ambient temperature. Minimal reduction in colour score of all the products at low temperature in comparison to ambient storage might be due to the minimum degradation of carotenoids, at low temperature. The gradual loss in flavour scores over the entire storage period might be due to changes in volatile compounds of the products due to time, temperature and duration of storage. Flavour deterioration in beverage products has also been reported by Jain *et al*. (2003). According to Sistrunk and Morris (1985) the blend of apple and grape juices were highly acceptable in quality and retained acceptable flavour and colour during storage at 24°C for 12 month. The loss of flavour and taste may be due to the degradation of ascorbic acid into furfural during storage (Shimoda and Osajima, 1981). Balaswamy *et al*. (2011) reported that a minimal loss of visual colour was found in blended beverages with phalsa, but scored well in terms of overall acceptability after 4 month storage period. Ahmed *et al.* (1976) reported no significant effect of storage on sensory quality of orange squash stored for five month.

5.4 Effect of storage on microbial quality

During microbial examination of different honey and sugar based beverages, no apparent spoilage was observed. None of the isolates i.e. bacteria, yeasts and moulds were found in nutrient agar, potato dextrose agar and yeast extract malt agar medium which shows the all the products were safe for consumption and maintained good microbial quality during the storage period of 6 months. Similar results have been reported by Lakhanpal (2010) in honey based mango nectar.

Deka (2000) reported negligible growth of moulds and yeasts in lime-aonla and mango-pineapple spiced RTS beverages, which got further reduced during storage due to inhibitory effect and antioxidative properties of spices. Deka and Sethi (2001) reported the no bacterial growth was observed in the spiced mixed fruit juice and RTS beverages.

Bhardwaj and Mukherjee (2011) observed the Kinnow: Aonla: Ginger blend samples were contaminated with a large variety of bacterial, fungal and mould species but within the acceptable limit.

5.7 Cost of production

The cost incurred in preparation of juice blends was calculated by taking into consideration the cost of all inputs used and appropriate cost involved during the preparation of value added products. The electricity and other expenses including depreciation were added to the total expenditure. The sale price per litre of product was calculated after adding 20% profit margin. The cost of production of different products ranged in between Rs.30 .00 (sugar malta RTS) to Rs.63.00 (honey malta RTS) for 500 mL glass bottle whereas, for iced tea it was in between Rs. 35.00 (sugar malta iced tea) to Rs 65 (honey malta iced tea) for 500 mL glass bottle. The cost of production of was within the range of natural ready to serve (RTS) beverages being sold in the market.

SUMMARY AND CONCLUSION

CHAPTER -6

SUMMERY AND CONCLUSION

Present investigation on the "Studies on the preparation and evaluation of functionally enriched value added product from malta fruit (*Citrus sinensis* L Osbeck)" were carried out: i) To standardize the optimum quantity of sweetening agents for the preparation of malta RTS and Malta Iced tea. ii) To maintain the quality of standardized beverages by the application of different preservative techniques. iii) To evaluate the effect of storage on quality of standardized malta beverages.

6.1 Composition of fresh malta fruits

The composition of fresh malta fruit (*Citrus sinensis* L Osbeck) revealed that it contains 10.33 ± 1.15^{o}B, 8.82 ± 0.65%, 4.83 ± 0.22%, 3.99 ± 0.75% total soluble solids, total sugars, reducing sugars and non-reducing sugars respectively. In addition, it also contained 2.18 % titratable acidity, 41.3mg/100mL, ascorbic acid, total phenol 7.98 ± 0.74 mg/100mL and β- carotene 8.45 ± 7.38 mg/100mL.

6.2 Composition of honey

The composition of honey reveals that honey consist 81.1 ±1.15°B total soluble solids. The chemical analysis of honey contained total titratable acidity (as citric acid) 4.28±0.13%, reducing sugars, non-reducing sugars and total sugars were found to be 65.54 ± 0.22% 8.82 ± 0.75%, and 74.77 ± 0.65 respectively. Ascorbic acid was observed 38.74 ± 0.13 mg/100mL. Carotene and phenol content of Honey was recorded to be 89.47 ± 7.38 mg/100mL and 18.30 ± 2.46 mg/100mL.

6.3 Standardization of beverages

Among different concentration of pulp (%) and TSS (^{o}B) highest hedonic scores for colour flavour, taste, consistency and overall acceptability were found for sugar malta RTS which was prepared by using 12% pulp and 12^{o} B TSS on the other hand for honey malta RTS the highest sensory scores were observed in 14% pulp and 14^{o} B TSS. When the best combination of both the sweetening agents were further treated with different concentration of tea leaves the highest sensory scores were observed when both beverages were treated with 1% concentration of tea leaves.

6.4 Value added products from malta fruit

Among different concentration of TSS and juice% for honey malta beverage, concentration with 14% TSS and 14 % of malta fruit juice reached the highest hedonic scores. On the other hand for sugar malta beverages concentration with 12% TSS and 12 percent of malta fruit juice reached the highest hedonic scores.

6.4.1 Sugar malta ready-to-served drink

The physicochemical characteristics of sugar malta RTS was improved substantially by application of various treatments. There was a progressive and continuous increase in TSS, reducing sugars, total sugars and pH value with an increase in storage period upto six months. Simultaneously gradual declining trend in titratable acidity (%), Ascorbic acid (mg/100 mL), non-reducing sugars (%), β- carotene (mg/100 mL) and total phenol (mg/100 mL) was also observed. However minimum loss of Ascorbic acid was observed in T2 (sodium benzoate 120ppm). The sugar malta RTS was successfully stored for a period of six months.

6.4.2 Honey malta ready-to-served drink

The physicochemical characteristics of honey malta RTS was improved substantially by application of various treatments. There was a progressive and continuous increase in TSS, reducing sugars, total sugars and pH value with an increase in storage period upto six months. Simultaneously gradual declining trend was recorded for titratable acidity (%), Ascorbic acid (mg/100 mL), non-reducing sugars (%), β- carotene (mg/100 mL) and total phenol (mg/100 mL). However minimum loss of Ascorbic acid was observed in T2 (sodium benzoate 120ppm). The honey malta RTS was successfully stored for a period of six months.

6.4.3 Sugar malta iced tea

The physicochemical characteristics of sugar malta iced tea was improved substantially by application of various treatments. There was a progressive and continuous increase in TSS, reducing sugars, total sugars and pH value with an increase in storage period upto six months. Simultaneously gradual declining trend in titratable acidity (%), Ascorbic acid (mg/100 mL), non-reducing sugars (%), β- carotene (mg/100 mL) and total phenol (mg/100 mL) was also observed. However minimum loss of Ascorbic acid was observed

in T2 (sodium benzoate 120ppm). The sugar malta iced tea was successfully stored for a period of six months.

6.4.4 Honey malta iced tea

The physicochemical characteristics of honey malta iced tea was improved substantially by application of various treatments. There was a progressive and continuous increase in TSS, reducing sugars, total sugars and pH value with an increase in storage period upto six months. Simultaneously gradual declining trend in titratable acidity (%), Ascorbic acid (mg/100 mL), non-reducing sugars (%), β- carotene (mg/100 mL) and total phenol (mg/100 mL) was also observed. However minimum loss of Ascorbic acid was observed in T2 (sodium benzoate 120ppm). The honey malta iced tea was successfully stored for a period of six months.

6.5 Cost of production

The cost of production of different honey and sugar based malta beverages were ranged in between Rs.30.00 (sugar malta RTS) and Rs. 63.00 (honey malta RTS) for 500 mL glass bottle whereas, for malta iced tea it was in between Rs. 35.00 (sugar malta iced tea) and Rs 65.00 (honey malta iced tea) for 500 mL glass bottle.

Conclusion

Present investigation on the "Studies on the preparation and evaluation of functionally enriched value added product from malta fruit (*Citrus sinensis* L Osbeck)" has shown that a good quality honey malta beverages can be prepared by using 14°B TSS and 14% juice and sugar malta beverages can be prepared by using 12°B TSS and 12% juice, can be preserved by using preservative techniques viz; pasteurization 65°C for 30 minutes, sodium benzoate 120 ppm and pasteurization with half preservative (pasteurization with 60ppm sodium benzoate) which were effective in increasing the shelf life with the minimum changes in the quality characteristics of different value added products of malta fruit. The above information can be utilized in the good quality product development in future.

The above study gave the estimate of optimum quantity of sweetening agents preservation methods, storage temperature and storage condition for preparing beverages. This research will serve as a vital step in the development of quality product with

commercial potential. The storage life of sugar malta RTS, honey malta RTS, sugar malta iced tea and honey malta iced tea was successfully stored for six months with good sensory characteristics.

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CHAPTER -7
LITERATURE CITED

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APPENDICES

APPENDICES

Appendix 1: Anova table for changes in physico-chemical characteristics of sugar malta RTS during storage under different storage conditions

SSV	Mean sum of squares									
	DF	**TSS**	**Titratable acidity**	**Ascorbic acid**	**Total Sugars**	**Reducing Sugars**	**Non-reducing sugars**	**pH**	**Total phenols**	**β carotenoids**
Factor A	**1**	8.605	76.168	571.609	8.625	2.151	2.151	0.031	6.093	6.969
Factor B	**1**	0.847	0.019	1.266	0.853	0.212	0.212	0.013	0.362	0.016
A X B	**1**	0.157	0.024	3.578	0.156	0.039	0.039	0.013	0.112	0.047
Factor T	**2**	0.446	0.027	6.962	0.447	0.112	0.056	0.037	0.341	1.843
A X T	**2**	0.021	0.002	6.676	0.018	0.005	0.003	0.006	0.251	0.942
B X T	**2**	0.177	0.003	0.461	0.169	0.044	0.022	0.002	0.049	0.104
A X B X T	**2**	0.073	0.006	0.536	0.080	0.019	0.009	0.002	0.004	0.148
Error	**24**	0.006	0.002	0.130	0.007	0.001	0.001	0.002	0.008	0.006

Appendix 2: Anova table for changes in of physico-chemical characteristics of honey malta RTS during storage under different storage conditions

SSV	Mean sum of squares									
	DF	**TSS**	**Titratable acidity**	**Ascorbic acid**	**Total Sugars**	**Reducing Sugars**	**Non-reducing sugars**	**pH**	**Total phenols**	**β carotenoids**
Factor A	**1**	8.604	0.006	571.609	2.069	2.151	2.151	0.133	8.950	5.970
Factor B	**1**	0.846	0.019	1.267	0.242	0.212	0.212	0.181	0.002	0.016
A X B	**1**	0.158	0.024	3.577	0.001	0.040	0.039	0.429	0.795	0.047
Factor T	**2**	0.445	0.027	6.962	0.219	0.110	0.056	0.037	0.341	1.843
A X T	**2**	0.023	0.002	6.675	0.005	0.005	0.003	0.006	0.234	0.941
B X T	**2**	0.178	0.003	0.461	0.120	0.043	0.022	0.002	0.049	0.104
A X B X T	**2**	0.072	0.006	0.537	0.045	0.019	0.009	0.002	0.007	0.148
Error	**24**	0.006	0.002	0.130	0.003	0.002	0.001	0.002	0.014	0.006

Appendix 3: Anova table for changes in of physico-chemical characteristics of sugar malta iced tea during storage under different storage conditions

SSV	Mean sum of squares									
	DF	**TSS**	**Titratable acidity**	**Ascorbic acid**	**Total Sugars**	**Reducing Sugars**	**Non-reducing sugars**	**pH**	**Total phenols**	**β carotenoids**
Factor A	**1**	8.604	0.006	571.609	2.069	2.151	2.151	0.133	8.950	5.970
Factor B	**1**	0.846	0.019	1.267	0.242	0.212	0.212	0.181	0.002	0.016
A X B	**1**	0.158	0.024	3.577	0.001	0.040	0.039	0.429	0.795	0.047
Factor T	**2**	0.445	0.027	6.962	0.219	0.110	0.056	0.037	0.341	1.843
A X T	**2**	0.023	0.002	6.675	0.005	0.005	0.003	0.006	0.234	0.941
B X T	**2**	0.178	0.003	0.461	0.120	0.043	0.022	0.002	0.049	0.104
A X B X T	**2**	0.072	0.006	0.537	0.045	0.019	0.009	0.002	0.007	0.148
Error	**24**	0.006	0.002	0.130	0.003	0.002	0.001	0.002	0.014	0.006

Appendix 4: Anova table for changes in of physico-chemical characteristics of honey malta iced tea during storage under different storage conditions

SSV	Mean sum of squares									
	DF	**TSS**	**Titratable acidity**	**Ascorbic acid**	**Total Sugars**	**Reducing Sugars**	**Non-reducing sugars**	**pH**	**Total phenols**	**β carotenoids**
Factor A	**1**	0.021	0.006	891.819	8.605	2.151	2.151	0.031	6.434	6.969
Factor B	**1**	0.023	0.019	23.330	0.847	0.211	0.212	0.013	0.288	0.016
A X B	**1**	0.007	0.024	24.370	0.159	0.040	0.039	0.013	0.073	0.047
Factor T	**2**	0.027	0.014	5.972	0.441	0.110	0.056	0.037	0.254	1.843
A X T	**2**	0.002	0.002	7.711	0.020	0.005	0.003	0.006	0.283	0.942
B X T	**2**	0.006	0.003	0.372	0.174	0.044	0.022	0.003	0.092	0.104
A X B X T	**2**	0.001	0.006	0.671	0.077	0.019	0.009	0.002	0.014	0.148
Error	**24**	0.002	0.002	0.712	0.007	0.002	0.001	0.002	0.105	0.006

ABSTRACT

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Name : Beena Pathak Id. No : 13181

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Title of Thesis : "Studies on the preparation and evaluation of functionally enriched value added product from malta fruit *(Citrus sinensis* L Osbeck*)*"

The present investigations entitled **"**Studies on the preparation and evaluation of functionally enriched value added product from malta fruit (*Citrus* sinensis L Osbeck)" was carried out at VCSG UUHF, Bharsar during 2014-2015.

Malta fruits were collected from the local village of Pauri Garhwal. The fruits were sorted and washed with chlorinated water. After washing, the fruits were subjected to juice extraction by using screw type juice extractor. Freshly extracted juice was used for preparing value added products viz; Sugar malta RTS, Honey malta RTS, Sugar malta iced tea and honey malta iced tea were prepared on the basis of sensory evaluation best combination of TSS and pulp was evaluated for stored study upto six months. The prepared malta beverages were subjected to pasteurization, chemical treatment using 120ppm sodium benzoate and a combined treatment of both (pasteurization with 60 ppm sodium benzoate) and stored at ambient (16-25^{o}C) and refrigerated (4-7^{o}C) temperature for six months to assess the feasibility of different preservation methods. The prepared malta beverages were organoleptically evaluated by adopting 9 point hedonic scale.

Among different concentration of TSS and juice% for honey malta beverages, concentration with 14% TSS and 14 percent of malta fruit juice reached the highest hedonic scores and for sugar malta beverages, concentration with 12% TSS and 12 percent of malta fruit juice reached the highest hedonic scores. It was also found that prepared beverages could be successfully preserved by pasteurization, chemical treatment using 120ppm sodium benzoate and a combined treatment of both (pasteurization with 60 ppm sodium benzoate) at ambient and refrigerated temperature for the period of six months without significant changes in physicochemical quality profile.

Prof. B.P. Nautiyal
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