

PHYSICOCHEMICAL, MICROBIAL AND SENSORY CHARACTERISTICS OF GREEN TEA BLEND WITH MORINGA, ORANGE PEEL, MINT AND JOBA FLOWER DURING STORAGE.

A PROJECT REPORT

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APRIL 2023

APPROVAL

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My humble statement is that it is my sincere declaration that the "Physicochemical, Microbial and Sensory characteristics of green tea blend with moringa, orange peel, mint and joba flower during storage". I have written is not a copy of any other students writing or any previous thesis report. I also express my legitimate authority to prove that fact that the thesis report mentioned above has never been used to meet the requirements of any other course and that I will not be provided to any other person or authority in the future.

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ACKNOWLEDGMENT

First of all, I would like to thank Almighty Allah for giving me stability and opportunity while working on my dissertation.

I would like to express my sincere gratitude to my supervisor **Dr. Nizam Uddin, Associate Professor and Head** of Department of Nutrition and Food Engineering at Daffodil International University for his kind guidance and support. I now thank him for his contribution. My deepest gratitude goes to Daffodil International University's entire NFE Department for giving me an opportunity that helped me not only with theoretical knowledge but also with proper practical implications.

At last, but not the least, I'd like to thank the entire NFE dept. lab assistant for helping me from moral to material needs as well as enlightening me with their knowledge to improve mine.

Author

ABSTRACT

The data presents the results of an experiment conducted to analyze the moisture content, pH, ash, and fat levels of green tea infused with natural ingredients over a period of 21 days. On day 0, all samples had moisture content ranging from 7.33to 8.33, with sample 2 having the highest pH level 6.76 and ash content 5.56. All samples had a fat content of 0. On day 7, the moisture content, pH, and ash levels of all samples remained largely unchanged, with sample 2 still having the highest pH level and ash content. During 14 days of storage, there was an increment of the moisture content of sample 1 to 8.66, while sample 2 had the highest moisture content of 9.66, along with the highest pH level 6.86 and a slightly higher ash content of 5.73. Sample 3 had the highest ash content of 5.83. On day 21, sample 2 had the highest moisture content of 10.66, along with the highest pH level of 6.86, while sample 3 had the highest ash content of 5.86. Sample 1 had a moisture content of 9.66, similar to that of sample 2. Overall, the data suggests that the natural ingredients used in the infusion of green tea may have an impact on the moisture content, pH, and ash levels of the tea over time, with sample 2 consistently showing the highest levels across all measures. However, the fat content remained unchanged always zero in all samples throughout the 21-day period. The TPC results showed that sample 1 had a TPC of 3 to 6×10^3 cfu/gm, sample 2 had a TPC of 6 to 8×10^3 cfu/gm, and sample 3 had a TPC of 11 to 12×10^3 cfu/gm. The sensory evaluation results indicated that sample 1 had an overall acceptability score of 7.4, with high scores for color 7.93, appearance 7.66, smell 7.96, and taste 7.23. Sample 2 had an overall acceptability score of 7.46, with high scores for color 7.96, appearance 7.33, smell 7.93, and taste 7.7. Sample 3 had an overall acceptability score of 6.53, with lower scores for color 7.93, appearance 6.26, smell 7.26, and taste 5.66.

In conclusion, the experiment analyzed the effects of natural ingredients on green tea blend moisture content, pH, ash, and fat levels over a 21-day period. The results showed that the sample 2 consistently showing the highest levels. However, the fat content remained unchanged. The TPC results indicated that sample 3 had a higher microbial load compared to samples 1 and 2. The sensory evaluation results showed that samples 1 and 2 were generally well-received, while sample 3 had lower scores in most attributes. The green tea blend infusions can affect its properties, and microbial load and sensory acceptability should be considered in product development.

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Chapter I

Introduction

1.1Introduction:

Moringa is a plant genus native to India and widely grown in many tropical and subtropical regions of the world. The most common species is Moringa oleifera, which is valued for its high nutritional content and medicinal properties. Moringa leaves are a good source of vitamins, minerals, and antioxidants, and have been traditionally used to treat various ailments such as diabetes, hypertension, and anemia. Recent scientific research has confirmed many of the traditional uses of Moringa and uncovered new potential applications. For example, Moringa extracts have been shown to have anti-inflammatory, anti-cancer, and anti-diabetic properties, among others. The plant also has promising potential as a sustainable source of biofuels and animal feed. (Siddhuraju&becker;2021) Mint is a genus of aromatic herbs that belongs to the family Lamiaceae. The most commonly cultivated species is peppermint (Mentha piperita), which is known for its distinctively refreshing flavor and aroma. Mint has been used for centuries in traditional medicine and culinary applications, and is now widely used in the food, beverage, and cosmetic industries. Mint contains several bioactive compounds such as menthol, menthone, and rosmarinic acid, which have been shown to have anti-inflammatory, antimicrobial, and antioxidant properties. As a result, mint is used in various therapeutic applications, including the treatment of digestive disorders, respiratory ailments, and skin conditions. (Elgayyar et al., 2001) Orange peel is the outermost layer of the citrus fruit, which includes oranges, lemons, and grapefruits. While the flesh of the fruit is consumed for its juice and nutritional benefits, the peel is often discarded. However, orange peel is rich in several bioactive compounds, including flavonoids, carotenoids, and essential oils, which have potential health benefits. Orange peel has been traditionally used in various cultures for its medicinal properties, including the treatment of digestive and respiratory ailments. Recent studies have confirmed the antioxidant and anti-inflammatory properties of orange peel extracts, which may have potential applications in the prevention and treatment of chronic diseases, such as cancer and cardiovascular disease. (Khan et al., 2021). Hibiscus rosa-sinensis, also known as Chinese hibiscus or shoe flower, is a species of flowering plant native to East Asia. It is widely cultivated for its large, showy flowers that come in a range of colors, including red, pink, orange, yellow, and white. Hibiscus rosa-sinensis has been used in traditional medicine for its antifungal, antibacterial, and anti-inflammatory properties. Recent scientific research has confirmed many of the traditional uses of Hibiscus rosa-sinensis and uncovered new potential applications. For example, Hibiscus extracts have been shown to have antioxidant and hypoglycemic properties, among others. The plant also has promising potential as a natural dye and as a source of bioactive compounds for the food, beverage, and cosmetic industries. (Chen et al., 2020) Green tea, a traditional beverage made from the leaves of the Camellia sinensis plant, has been consumed for centuries for its numerous health benefits (Chacko et al., 2010). Its polyphenol content, particularly catechins, has been shown to exhibit potent antioxidant, anti-inflammatory, and anti-carcinogenic properties (Singh & Rani,2015). Regular consumption of green tea has been linked to reduced risk of cardiovascular disease, metabolic disorders, and certain cancers (Tang et al, 2020). Despite its numerous health benefits, the flavor of green tea can be perceived as bitter and unappealing, which may deter some consumers from consuming it on a regular basis (Chen et al., 2019). In recent years, there has been a growing interest in developing value-added tea products that not only provide health benefits but also offer desirable sensory attributes. One approach to enhancing the sensory profile of green tea is to blend it with other natural ingredients. Mint, moringa, joba flower, and orange peel are natural ingredients that have been reported to exhibit various health benefits. Mint is known for its refreshing taste and aroma, and has been shown to have potential therapeutic effects on digestion, pain relief, and respiratory disorders (Babar et al., 2021). Moringa, also known as the "miracle tree", has gained attention for its high levels of antioxidants, anti-inflammatory compounds, and various vitamins and minerals (Anwar et al., 2020). Joba flower, a traditional herbal remedy, is commonly used for its analgesic and anti-inflammatory effects (Chen et al., 2014). Orange peel is a rich source of flavonoids, which have been associated with a reduced risk of cardiovascular disease (Manach et al., 2004). Combining these natural ingredients with green tea may lead to a value-added product with enhanced sensory attributes and potential health benefits.

1.2: The objective of the Study:

- To standardize the formulation of green tea blend with mint, moringa, joba flower, and orange peel extracts.
- > To assess the nutritional properties of green tea.
- > To evaluate the consumer acceptance of green tea.

Chapter II

Literature Review

2.0: Literature Review

2.1Moringa: Moringa, also known as the "Miracle Tree," is a plant that is rich in nutrients and has been used for centuries in traditional medicine. The composition of moringa includes: Vitamins: Moringa is a rich source of vitamins A, B, and C. It also contains high levels of vitamin E. Minerals: Moringa is rich in minerals such as calcium, potassium, iron, and magnesium. Protein: Moringa leaves contain all nine essential amino acids, making it a complete protein source. Antioxidants: Moringa contains high levels of antioxidants, such as flavonoids and polyphenols, which help protect cells from damage caused by free radicals. Anti-inflammatory compounds: Moringa contains compounds such as quercetin and kaempferol, which have anti-inflammatory properties. Fiber: Moringa leaves are high in fiber, which can help regulate digestion and prevent constipation. Omega-3 fatty acids: Moringa seeds are a good source of omega-3 fatty acids, which are important for maintaining heart health. Overall, the composition of moringa makes it a highly nutritious plant with a wide range of potential health benefits. (Leone et al.,2016)

2.1.1Health benefits: Anti-inflammatory effects: Moringa has been shown to have antiinflammatory properties, which may help reduce inflammation in the body and prevent chronic diseases such as arthritis and heart disease. (Pal et al.,2013). Antioxidant effects: Moringa is rich in antioxidants, which can help protect cells from damage caused by free radicals and reduce the risk of chronic diseases such as cancer and diabetes. (Fahey,2005). Blood sugar control: Some studies have suggested that moringa may help regulate blood sugar levels in people with diabetes, possibly due to its high fiber content and anti-inflammatory properties. (Gupta et al.,2012) Blood pressure control: Moringa may also help lower blood pressure levels, possibly due to its high content of potassium and other beneficial nutrients. (Jaiswal.,2009)

2.2Mint: Mint is a popular herb that is commonly used for its refreshing flavor and aroma. The composition of mint includes: Essential oils: Mint contains a high amount of essential oils, which give it its characteristic flavor and aroma. The main components of mint essential oil are menthol, menthone, and limonene. Vitamins: Mint is a good source of vitamins A and C. Minerals: Mint contains minerals such as calcium, potassium, and magnesium. Antioxidants: Mint contains antioxidants such as rosmarinic acid, which help protect cells from damage caused by free radicals. Anti-inflammatory compounds: Mint contains compounds such as menthol and rosmarinic acid, which have anti-inflammatory properties. Fiber: Mint leaves are a good source of dietary fiber, which can help regulate digestion and prevent constipation. Overall, the composition of mint makes it a nutritious herb with potential health benefits. (Akhila & Shivanandappa,2014)

2.2.1Health benefits: Digestive health: Mint has been traditionally used to aid digestion and relieve symptoms such as bloating, gas, and nausea. Studies suggest that mint may have a relaxing effect on the digestive system, which can help relieve symptoms of irritable bowel syndrome (IBS) and other digestive disorders. (McKay& Blumberg,2006). Respiratory health: Menthol, a compound found in mint, has a cooling and soothing effect on the respiratory system. Menthol has been used to relieve symptoms of cough, congestion, and sore throat. (Eccles,1994). Anti-inflammatory effects: Mint contains anti-inflammatory compounds such as rosmarinic acid, which have been shown to reduce inflammation and relieve symptoms of conditions such as asthma and arthritis. (Harlev et al.,2013) Pain relief: Mint has a cooling and numbing effect on the skin, and can be used topically to relieve pain and discomfort from conditions such as headaches, muscle aches, and insect bites. (Gobel et al.,2016).

2.3Orange peel: Orange peel is the outer, colored layer of the orange fruit, which is commonly used in cooking and baking. The composition of orange peel includes: Essential oils: Orange peel contains a high amount of essential oils, which are responsible for its distinctive aroma and flavor. The main components of orange peel essential oil are limonene, myrcene, and alphapinene. Flavonoids: Orange peel contains flavonoids such as hesperidin, which have antioxidant properties and may help lower cholesterol levels. Fiber: Orange peel is a good source of dietary fiber, which can help regulate digestion and prevent constipation. Vitamin C: Orange peel contains vitamin C, which is important for immune system function and collagen production. Minerals: Orange peel contains minerals such as calcium, potassium, and magnesium. (Jin et al.,2013).

2.3.1Health benefits: Digestive health: Orange peel contains compounds that stimulate digestion, help relieve constipation and bloating, and reduce the risk of gastric ulcers.(Liu et al.,2009).Immune system support: Orange peel contains vitamin C and other antioxidants that help boost the immune system and protect against infections. (Hamauzu et al.,2008)Skin health: Orange peel contains compounds that help protect against UV-induced skin damage, reduce the appearance of wrinkles, and improve skin texture.(Kim et al.,2011)Anti-inflammatory effects: Orange peel contains compounds that have anti-inflammatory properties and can help reduce inflammation throughout the body (Lee et al.,2010).

2.4Joba flower: Hibiscus rosa-sinensis L. is a species of flowering plant that belongs to the family Malvaceae. The plant is known for its ornamental value as well as its potential medicinal properties. Here is the composition of Hibiscus rosa-sinensis L., along with a reference: Flavonoids: Hibiscus rosa-sinensis L. contains flavonoids such as kaempferol, quercetin, and rut in. These compounds have antioxidant properties and may help protect the body against cellular damage caused by free radicals. Organic acids: Hibiscus rosa-sinensis L. contains organic acids such as citric acid, malic acid, and tartaric acid. These acids give the plant its characteristic tart taste and may also have health benefits. Polysaccharides: Hibiscus rosa-sinensis L. contains polysaccharides such as pectin and hemicellulose. These compounds have been shown to have potential immunomodulatory and anticancer properties. Anthocyanins: Hibiscus rosa-sinensis L.

contains anthocyanins, which are water-soluble pigments that give the flowers their vibrant colors. These compounds have antioxidant properties and may also have anti-inflammatory effects. (Yang et al, 2009).

2.4.1Health benefits: Hibiscus rosa-sinensis L. has been traditionally used in various parts of the world for its medicinal properties. Here are some of the potential health benefits of Hibiscus rosa-sinensis L. Hypotensive effects: Hibiscus rosa-sinensis L. has been shown to have hypotensive effects, meaning it can help lower blood pressure. A study conducted on hypertensive patients found that hibiscus tea consumption for 12 days significantly reduced both systolic and diastolic blood pressure. (Haji & Haji,1999) Antioxidant effects: Hibiscus rosa-sinensis L. contains antioxidants such as flavonoids and anthocyanins, which can help scavenge free radicals and protect the body against oxidative stress. A study conducted on rats found that Hibiscus rosa-sinensis L. leaf extract had potent antioxidant activity. (Bhat et al.,2011)Anti-inflammatory effects: Hibiscus rosa-sinensis L. has been shown to have anti-inflammatory effects, which may help reduce inflammation in the body. A study conducted on rats found that Hibiscus rosa-sinensis L. leaf extract had potent anti-inflammatory activity. (Ajayi et al.,2007)

2.5Value of this product: The combination of moringa, mint, joba flower, and orange peel can potentially add nutritional and functional value to a variety of products, such as tea blends, herbal supplements, and health foods. Here are some potential benefits of using these raw materials in products:

2.5.1Nutritional value: Moringa, mint, and orange peel are all rich in antioxidants, vitamins, and minerals, which can contribute to the overall nutritional value of a product. For example, a tea blend made with these ingredients can provide a natural source of vitamins C and A, iron, and calcium. Flavor and aroma: Mint and orange peel can add a refreshing and citrusy flavor to products, while joba flower can add a floral note. These natural flavors can help enhance the taste and aroma of products without the need for artificial additives.

2.5.2Health benefits: Each of these ingredients has potential health benefits, such as antiinflammatory, anti-diabetic, and anti-cancer properties, which can appeal to consumers who are looking for natural and holistic remedies for various health conditions. Marketing appeal: The use of natural and exotic ingredients can make products stand out in a crowded marketplace and appeal to consumers who are interested in health and wellness products. Overall, the use of these raw materials in products can potentially add value in terms of nutritional, functional, and marketing benefits. However, it's important to ensure that these ingredients are sourced from reputable suppliers and used in safe and appropriate doses to avoid potential adverse effects.

2.6Green Tea: Green tea is a type of tea that is made from Camellia sinensis leaves and buds. Here is the composition of green tea, along with a reference: Catechins: Green tea contains catechins such as epicatechin, epicatechin gallate, epigallocatechin, and epigallocatechin gallate (EGCG). EGCG is the most abundant and well-studied catechin in green tea, and it has been shown to have various health benefits. Caffeine: Green tea contains caffeine, a natural stimulant

that can help improve mental alertness and concentration. Theanine: Green tea contains theanine, an amino acid that has been shown to have a relaxing effect on the body and may help reduce stress and anxiety. Flavonoids: Green tea contains flavonoids such as quercetin and kaempferol, which have antioxidant properties and may help protect the body against cellular damage caused by free radicals. (Sartorius et al.,2015)

2.6.1Health benefits: Antioxidant properties: Green tea contains a high level of catechins, which are potent antioxidants that can help protect cells from damage caused by free radicals. (Khan &Mukhtar,2007). Improved brain function: Green tea contains caffeine, which can improve brain function and mood. It also contains the amino acid L-theanine, which can have a calming effect and enhance focus and attention. (Dietz & Dekker,2017). Lower risk of cardiovascular disease: Drinking green tea has been associated with a lower risk of cardiovascular disease, as it can help lower blood pressure and improve cholesterol levels. (Yang et al.,2004). Lower risk of certain cancers: Green tea has been studied for its potential anti-cancer properties, particularly in reducing the risk of breast, prostate, and colorectal cancers. (Yang et al.,2011). Weight loss and metabolism: Some studies suggest that green tea can help boost metabolism and aid in weight loss, although the evidence is mixed. (Hursel et al.,2009)

Chapter-III

Material and Methods

3.1 Materials:

3.1.1 Sample collection and sample preparation: Among the ingredients used in green tea, moringa, joba flowers are collected from ashulia bazar and orange peel, mint leaves are purchased from the khagan bazar.

3.1.2 Process for formulation of green tea with others blend: All the ingredients were washed well, dried in the oven, and grind them by hand. The preparation process has shown in the figure 1.

| Ingredient | Sample 1 Amount (g) | Sample 1 Percentage (%) | Sample 2 Amount (g) | Sample 2 Percentage (%) | Sample 3 Amount (g) | Sample 3 Percentage (%) |
|----------------|---------------------------|-------------------------------|---------------------------|-------------------------------|---------------------------|-------------------------------|
| Moringa | 0.6 | 60 | 0.8 | 40 | 0.7 | 35 |
| Orange peel | 01 | 10 | 0.7 | 35 | 0.4 | 20 |
| Mint | 0.2 | 20 | 0.3 | 15 | 0.7 | 35 |
| Joba flower | 0.2 | 20 | 0.2 | 10 | 0.2 | 10 |
| Total | 2.0 | 100 | 2.0 | 100 | 2.0 | 100 |

3.1.3Formulation of green tea blend

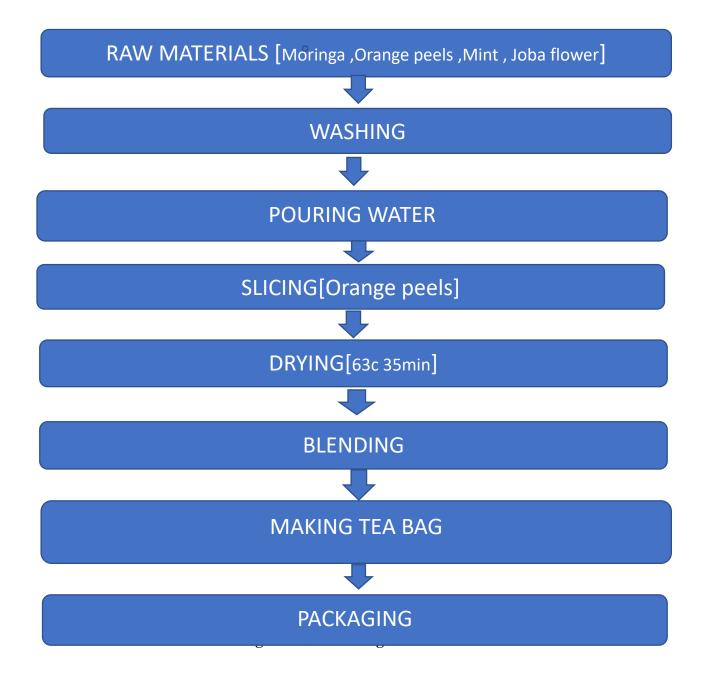


Fig 1: Process diagram.



Fig 2: Sample of Final Product.

3.2Method

3.2.1Determination of pH

Procedure:

Turn on the light first. Take a sample and put it in a beaker. Make sure the temperature sensor and pH electrode are dry and clean. Calibrate meter using buffers 1 and 2.4. Collect sample and place the pH in beaker. Insert the pH meter into the measuring cup. Read after correcting the digits in meters.

3.2.2 Determination of Moisture:

Place 2gm of sample in the moisture analyzer, turn it on, and wait around 5 minutes. The moisture analyzer then displays the results on the screen.

3.2.3 Determination of Ash:

Ash percentage of green tea blend was determined according to the method reported by t Nielsen (2010) with few modifications. Weigh the empty crucible and record its weight as W1. Weigh a representative sample of the substance to be tested and record its weight as W2. Place the sample into the crucible and heat it to a temperature of 500-600°C for 4-6 hours until all organic matter is burned off. After cooling the crucible to room temperature in a desiccator, weigh it with the ash residue and record the weight as W3. Calculate the weight of the ash by subtracting the weight of the empty crucible(W1) from the weight of the crucible with the ash residue (W3 - W1 = W4).

Calculation:

Ash % =
$$\frac{w4}{w^2} \times 100$$

3.2.4 Determination of Fat:

Fat percentage of green tea blend was determined according to the method reported by the AOAC, (2016) with few modifications. Weigh a clean, dry and empty thimble of Soxhlet apparatus and record its weight (W1). Transfer 5gm of the green tea blend sample into the thimble and weigh it accurately. Record the weight (W2). Place the thimble with the sample into the Soxhlet apparatus and add a sufficient amount of solvent, such as petroleum ether or hexane, to cover the sample completely. Heat the apparatus, and the solvent will evaporate and condense in the condenser, creating a cyclical process that will extract fat from the sample. Allow the extraction process to continue 6 hours until the solvent level in the flask reaches a steady state. This will ensure that the fat in the sample has been completely extract. After the extraction is complete, remove the thimble from the Soxhlet apparatus and allow it to dry at room temperature for a few minutes .Weigh the thimble with the extracted fat and record the weight (W3).Place the thimble with the extracted fat into a drying oven and dry it at a constant temperature of around 105°C to remove any residual solvent. After the fat has been completely dried, remove the thimble from the ocol at room temperature, and weigh it again (W4).

Fat content for all sample were calculated using the following formula:

Fat content (%) = $[(W3 - W1) / (W2 - W1)] \times 100$

3.2.5 Total Plate count:

The total plate count of green tea blend was determined according to the method reported by Rana et al., (2020) with few modifications. Weigh or measure an appropriate amount of the sample and add it to a sterile diluent (such as 0.1% peptone water) in a sterile container. Mix the sample and diluent thoroughly to make a homogenous mixture. Prepare a series of dilutions by transferring a known volume of the sample/diluent mixture to a new container containing fresh diluent, and mixing thoroughly. Typically, a 10-fold dilution series is used. Pipette a suitable volume (usually 0.1 ml or 1 ml) of each diluted sample onto the surface of a sterile agar plate (such as Plate Count Agar). Spread the sample evenly over the surface of the agar using a sterile spreader. Incubate the plates at an appropriate temperature (usually 30-35°C for 48 hours) to allow bacterial colonies to grow. Count the number of bacterial colonies on each plate and calculate the total number of viable bacteria in the sample.

Calculation:

Total Plate Count $\left(\frac{CFU}{ml}\right) = \frac{No. \ of \ Colonies \times Dilution \ factor}{ml \ aliquote \ taken}$

3.2.6 Sensory Evaluation form of green tea blend.

Sensory characteristics of green tea blend sample was carried out on 21 days of storage. Semitrained thirty panel members were selected from the University community. Randomly, the samples were presented to the panel members. Green tea blend samples were evaluated organoleptically for color, smell, appearance, taste and overall acceptability, according to the hedonic scale of nine points (9-likeextremely to 1- dislike extremely) as reported by Roy et al. (2018).

Chapter IV

Result and Discussion

| Days | Sample | Moisture Content | рН | Ash | Fat |
|------|--------|---------------------|-----------------|-----------------|-----|
| | 1 | 7.33±0.57 | 6.66±0.05 | 5.46±0.05 | 0 |
| 0 | 2 | 8.33 <u>+</u> 0.57 | 6.76±0.05 | 5.56±0.05 | 0 |
| | 3 | 8.33 <u>+</u> 0.57 | 6.76±0.05 | 5.73±0.05 | 0 |
| | 1 | 7.33 <u>+</u> 0.57 | 6.66±0.05 | 5.46±0.05 | 0 |
| 7 | 2 | 8.33 <u>+</u> 0.57 | 6.76±0.05 | 5.56±0.05 | 0 |
| | 3 | 8.33 <u>+</u> 0.57 | 6.76 ± 0.05 | 5.73±0.05 | 0 |
| | 1 | 8.66 ± 0.57 | 6.76 ± 0.05 | 5.76 ± 0.05 | 0 |
| 14 | 2 | 9.66±0.57 | 6.86 ± 0.05 | 5.73±0.1 | 0 |
| | 3 | 9.66±0.57 | 6.8±0.1 | 5.83 ± 0.05 | 0 |
| | 1 | 9.66±0.57 | 6.8±0.1 | 5.83 ± 0.05 | 0 |
| 21 | 2 | 10.66 ± 0.57 | 6.86 ± 0.05 | 5.73±0.05 | 0 |
| | 3 | 11.66±0.57 | 6.83±0.05 | 5.86 ± 0.05 | 0 |

 Table4.1: The proximate composition of green tea blend

The table 4.1 shows the physico-chemical characteristics of green tea blend room temperature condition with 21 days. The moisture content of the mixed tea will depend on various factors such as the initial moisture content of the individual ingredients, the drying method, and the storage conditions. However, as a rough estimate, assuming that the individual ingredients are dried to a standard moisture content, the moisture content of the mixed tea could be around 5% to 10%. At Day 0 and Day 7, all samples had a similar moisture content, with Sample 1 and Sample 2 having a moisture content of 7.33±0.57 and 8.33±0.57, respectively, and Sample 3 having a moisture content of 8.33±0.57.By Day 14, the moisture content of all samples had increased, with Sample 1 having a moisture content of 8.66±0.57, Sample 2 having a moisture content of 9.66±0.57, and Sample 3 having a moisture content of 9.66±0.57.By Day 21, the moisture content had increased further for all samples, with Sample 1 having a moisture content of 9.66±0.57, Sample 2 having a moisture content of 10.66±0.57, and Sample 3 having the highest moisture content of 11.66±0.57.At Day 0 and Day 7, all samples had a similar ash content, with all samples having an ash content between 5.46±0.05 and 5.56±0.05.By Day 14, the ash content had increased for all samples, with Sample 1 having an ash content of 5.76 ± 0.05 , Sample 2 having an ash content of 5.73 ± 0.1 , and Sample 3 having the highest ash content of 5.76±0.05.By Day 21, the ash content had increased further for Sample 3, which had an ash content of 5.86 ± 0.05 , while the ash content for Sample 1 and Sample 2 had decreased slightly to 5.73±0.05 and 5.86±0.05, respectively. The amount of ash present in green tea can vary depending on various factors such as the soil composition, processing method, and storage conditions. However, as per the Indian Tea Board, the maximum permissible limit of total ash content in green tea is 8.5% (by weight) and the minimum limit is 4.5% (by weight) (as of August 2021). The pH values for all samples seem to be within a relatively narrow range, with only small variations from sample to sample and day to day. The pH values for all samples fall within the range of 6.66 to 6.86, which suggests that the tea is slightly acidic. There are no clear trends indicating that any one sample consistently has higher or lower pH values across all four time points. However, it is worth noting that Sample 1 has the lowest pH value on Day 14 (6.76) and the highest pH value on Day 21 (6.8 ± 0.1), while Sample 3 has the highest pH value on Day 14 (6.8 ± 0.1) and the lowest pH value on Day 21 (6.8 ± 0.1), while Sample 3 has the highest pH value on Day 14 (6.8 ± 0.1) and the lowest pH value on Day 21 (6.8 ± 0.1), while Sample 3 has the highest pH value on Day 14 (6.8 ± 0.1) and the lowest pH value on Day 21 (6.8 ± 0.1), while Sample 3 has the highest pH value on Day 14 (6.8 ± 0.1) and the lowest pH value on Day 21 (6.8 ± 0.1), while Sample 3 has the highest pH value on Day 14 (6.8 ± 0.1) and the lowest pH value on Day 21 (6.8 ± 0.1). The pH of green tea can vary depending on various factors such as the type of tea, the water used for brewing, and the steeping time. Generally, the pH of green tea is slightly acidic, ranging from 6 to 7.5. A study published in the Journal of Food Composition and Analysis measured the pH of different types of green teas and found that the pH ranged from 6.35 to 7.54, with an average pH of 7.05. Another study published (Komes et al.,2010) found that the pH of brewed green tea ranged from 6.41 to 7.42, depending on the brewing conditions. (Ruan & Chen,2008). It is important to note that the pH of green tea can affect the taste and color of the tea, and different brewing methods can result in different pH levels. However, in general, green tea is considered to be a slightly acidic beverage.

All samples had a fat content of 0, so there is no highest or lowest value for this parameter. Green tea typically contains very low amounts of fat, usually less than 0.1 gram per serving. The fat content of green tea is primarily due to the small amount of fatty acids present in the tea leaves. According to the United States Department of Agriculture (USDA), 1 cup (240 ml) of brewed green tea contains approximately 0.02 grams of total fat, which is negligible. Green tea is generally considered to be a low-calorie and low-fat beverage that can be a good addition to a healthy diet. However, it is important to note that some commercially available green tea products may contain added ingredients, such as milk or sugar, which can significantly increase the fat content.

| Days | Sample | No. of colonies (cfu/ml) |
|------|--------|-----------------------------|
| 0 | 1 | 3 to 4×10^3 |
| | 2 | 4 to 5×10^{3} |
| | 3 | 5 to 6×10^3 |
| 7 | 1 | 3 to 4×10^3 |
| | 2 | 4 to 5×10^3 |
| | 3 | 5 to 6×10^3 |
| 14 | 1 | 6 to 7×10^3 |
| | 2 | 7 to 8×10^3 |
| | 3 | 7 to 8×10^{3} |
| 21 | 1 | 11 to 12×10^3 |
| | 2 | 11 to 12×10^3 |
| | 3 | 11 to 12×10^3 |

Table 4.2: Total Plate count for green samples blend

The total bacterial count in green tea can vary depending on a number of factors, such as the origin and processing of the tea leaves, storage conditions, and brewing methods. In general, high-quality green tea is processed in a way that minimizes bacterial contamination and has low levels of bacteria. According to a study published in the Journal (Kumar et al.,2016), the total bacterial count in green tea samples ranged from less than 10 colony-forming units (CFU) per gram to over 10,000 CFU per gram. However, the majority of samples had bacterial counts below 1,000 CFU per gram. It's important to note that some of the bacteria present in green tea can be beneficial, such as certain strains of probiotics

| Sample | Color | Appearance | Smell | Taste | Overall acceptability |
|--------|-----------|------------|-----------|-----------|--------------------------|
| 1 | 7.93±0.77 | 7.66±1.15 | 7.96±0.66 | 7.23±0.85 | 7.4±0.56 |
| 2 | 7.96±0.66 | 7.33±0.88 | 7.93±0.78 | 7.7±1.11 | 7.46±0.5 |
| 3 | 7.93±0.78 | 6.26±0.94 | 7.26±0.78 | 5.66±0.71 | 6.53±0.5 |

 Table 4.3: Sensory characteristics of green tea blend

The results an overall acceptability score of 7.46, followed by Sample 1 with a score of 7.4, and Sample 3 with a score of 6.53. It's worth noting that these scores are relatively close together, so it's possible that there may not be a significant difference between the samples in terms of overall acceptability.

Looking at the individual attributes, Sample 1 had the highest scores for color (7.93) and smell (7.96), which suggests that this sample was visually appealing and had a pleasant aroma. Sample 2 had the highest score for appearance (7.33), which may suggest that it had an attractive texture or was well-presented. Sample 3 had the lowest scores across all attributes, indicating that this sample was the least well-liked by the panel of testers. In terms of taste, Sample 2 had the highest score (7.7), followed by Sample 1 (7.23), and Sample 3 (5.66). This suggests that Sample 2 had the most desirable taste profile, while Sample 3 had the least desirable taste. It's important to note that the standard deviations for each attribute are relatively small, which suggests that the panel of testers had fairly consistent opinions about the samples. However, without additional information about the testing methodology, it's difficult to assess the reliability of the results. Overall, based on the data provided, it appears that Sample 2 was the most well-liked by the panel of testers in terms of taste and appearance, while Sample 1 had the highest scores for color and smell. Sample 3 was the least well-liked across all attributes. However, it's important to keep in mind that there may be other factors that could influence consumer preferences.

Chapter V

Conclusion

Conclusion:

The green tea was analyzed at 0, 7, 14, and 21 days to determine changes in the aforementioned parameters. The results indicated that there was no significant change in moisture content, pH, ash, or fat content during the 21-day period. The TPC of the food product was found to be within the acceptable range of TPC 3 to 12×10^3 cfu/ml throughout the study period. Sensory evaluation was conducted to determine the acceptability of the food product, and it was found that the color, appearance, smell, taste, and overall acceptability of the product remained fairly consistent throughout the 21-day period. In conclusion, the green tea analyzed in this study was found to be stable in terms of its chemical composition and microbial growth over a period of 21 days. The sensory evaluation results indicate that the product was acceptable in terms of its sensory characteristics throughout the study period. These findings suggest that the food product has a good shelf life and can be stored for up to 21 days without significant changes in quality.

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| Parameters | Replication | Sample 1% | Sample 2% | Sample 3% |
|--------------|-------------|-------------------|-------------------|-------------------|
| | 1 | 8 | 8 | 8 |
| Moisture | 2 | 7 | 8 | 8 |
| content | 3 | 7 | 9 | 9 |
| | 1 | 5.5 | 5.5 | 5.7 |
| Ash | 2 | 5.4 | 5.6 | 5.8 |
| | 3 | 5.5 | 5.6 | 5.7 |
| | 1 | 6.6 | 6.8 | 6.8 |
| pН | 2 | 6.7 | 6.7 | 6.7 |
| | 3 | 6.7 | 6.8 | 6.8 |
| | 1 | 0 | 0 | 0 |
| Fat | 2 | 0 | 0 | 0 |
| | 3 | 0 | 0 | 0 |
| | 1 | 3×10 ³ | 4×10 ³ | 5×10 ³ |
| TPC (cfu/ml) | 2 | 3×10 ³ | 5×10 ³ | 6×10^{3} |
| | 3 | 4×10 ³ | 5×10 ³ | 6×10 ³ |

Appendix-1: Moisture content, pH, Ash, Fat, Total plate count (TPC) of Moringa, Mint, Orange peel and Joba flower mixed green tea[0Days].

Appendix-2: Moisture content, pH, Ash, Fat, Total plate count (TPC) of Moringa, Mint, Orange peel and Joba flower mixed green tea[7Days].

| Parameters | Replication | Sample 1% | Sample 2% | Sample 3% |
|--------------|-------------|-------------------|--------------------|-------------------|
| | 1 | 8 | 8 | 8 |
| Moisture | 2 | 7 | 8 | 8 |
| content | 3 | 7 | 9 | 9 |
| | 1 | 5.5 | 5.5 | 5.7 |
| Ash | 2 | 5.4 | 5.6 | 5.8 |
| | 3 | 5.5 | 5.6 | 5.7 |
| | 1 | 6.6 | 6.8 | 6.8 |
| pН | 2 | 6.7 | 6.7 | 6.7 |
| | 3 | 6.7 | 6.8 | 6.8 |
| | 1 | 0 | 0 | 0 |
| Fat | 2 | 0 | 0 | 0 |
| | 3 | 0 | 0 | 0 |
| | 1 | 3×10 ³ | 4×10 ³ | 5×10 ³ |
| TPC (cfu/ml) | 2 | 3×10 ³ | 5-×10 ³ | 6×10 ³ |
| | 3 | 4×10 ³ | 5×10 ³ | 6×10 ³ |

| Parameters | Replication | Sample 1% | Sample 2% | Sample 3% |
|--------------|-------------|-------------------|-------------------|-------------------|
| Moisture | 1 | 8 | 9 | 10 |
| content | 2 | 9 | 10 | 10 |
| | 3 | 9 | 10 | 9 |
| Ash | 1 | 5.8 | 5.8 | 5.8 |
| | 2 | 5.8 | 5.8 | 5.8 |
| | 3 | 5.7 | 5.6 | 5.9 |
| pН | 1 | 6.8 | 6.8 | 6.9 |
| - | 2 | 6.7 | 6.9 | 6.7 |
| | 3 | 6.8 | 6.9 | 6.8 |
| Fat | 1 | 0 | 0 | 0 |
| | 2 | 0 | 0 | 0 |
| | 3 | 0 | 0 | 0 |
| TPC (cfu/ml) | 1 | 6×10 ³ | 7×10 ³ | 7×10 ³ |
| | 2 | 6×10 ³ | 8×10 ³ | 7×10 ³ |
| | 3 | 7×10 ³ | 7×10^{3} | 8×10 ³ |

Appendix-3: Moisture content, pH, Ash, Fat, Total plate count (TPC) of Moringa, Mint, Orange peel and Joba flower mixed green tea[14Days].

Appendix-4: Moisture content, pH, Ash, Fat, Total plate count (TPC) of Moringa, Mint, Orange peel and Joba flower mixed green tea[21Days].

| Parameters | Replication | Sample 1% | Sample 2% | Sample 3% |
|--------------|-------------|--------------------|--------------------|--------------------|
| Moisture | 1 | 10 | 11 | 11 |
| content | 2 | 10 | 11 | 12 |
| | 3 | 9 | 10 | 12 |
| Ash | 1 | 5.8 | 5.7 | 5.8 |
| | 2 | 5.8 | 5.8 | 5.9 |
| | 3 | 5.9 | 5.7 | 5.9 |
| pН | 1 | 6.9 | 6.9 | 6.8 |
| - | 2 | 6.7 | 6.9 | 6.8 |
| | 3 | 6.8 | 6.8 | 6.9 |
| Fat | 1 | 0 | 0 | 0 |
| | 2 | 0 | 0 | 0 |
| | 3 | 0 | 0 | 0 |
| TPC (cfu/ml) | 1 | 12×10 ³ | 11×10 ³ | 11×10 ³ |
| | 2 | 12×10 ³ | 12×10 ³ | 12×10 ³ |
| | 3 | 11×10 ³ | 12×10 ³ | 11×10 ³ |

| Panelists | Color | Appearance | Smell | Taste | Overall |
|-----------|-------|------------|-------|-------|---------------|
| | | | | | acceptability |
| 1 | 8 | 7 | 7 | 6 | 6 |
| 2 | 7 | 6 | 8 | 7 | 7 |
| 3 | 8 | 8 | 8 | 6 | 7 |
| 4 | 9 | 8 | 8 | 8 | 8 |
| 5 | 8 | 9 | 7 | 8 | 8 |
| 6 | 9 | 8 | 9 | 7 | 8 |
| 7 | 8 | 9 | 9 | 7 | 8 |
| 8 | 7 | 8 | 8 | 7 | 7 |
| 9 | 7 | 9 | 7 | 6 | 7 |
| 10 | 8 | 6 | 8 | 8 | 7 |
| 11 | 7 | 6 | 7 | 8 | 7 |
| 12 | 8 | 7 | 8 | 8 | 7 |
| 13 | 7 | 7 | 8 | 6 | 7 |
| 14 | 9 | 8 | 9 | 6 | 8 |
| 15 | 9 | 8 | 7 | 8 | 8 |
| 16 | 8 | 9 | 8 | 9 | 8 |
| 17 | 7 | 6 | 8 | 7 | 7 |
| 18 | 8 | 8 | 8 | 6 | 7 |
| 19 | 9 | 8 | 8 | 8 | 8 |
| 20 | 8 | 9 | 7 | 8 | 8 |
| 21 | 9 | 8 | 9 | 7 | 8 |
| 22 | 8 | 9 | 9 | 7 | 8 |
| 23 | 7 | 7 | 8 | 7 | 7 |
| 24 | 7 | 9 | 7 | 6 | 7 |
| 25 | 8 | 6 | 8 | 8 | 7 |
| 26 | 7 | 6 | 7 | 8 | 7 |
| 27 | 8 | 7 | 7 | 8 | 7 |
| 28 | 7 | 7 | 8 | 7 | 7 |
| 29 | 9 | 8 | 9 | 7 | 8 |
| 30 | 9 | 9 | 8 | 9 | 8 |

Appendix-5: Sensory characteristics of Moringa, Mint, Orange peel and Joba flower mixed green tea [Sample 1].

| Panelists | Color | Appearance | Smell | Taste | Overall acceptability |
|-----------|-------|------------|-------|-------|--------------------------|
| 1 | 8 | 9 | 8 | 9 | 8 |
| 2 | 8 | 7 | 7 | 6 | 7 |
| 3 | 8 | 6 | 8 | 8 | 7 |
| 4 | 8 | 8 | 9 | 8 | 8 |
| 5 | 7 | 8 | 8 | 9 | 8 |
| 6 | 9 | 7 | 9 | 8 | 8 |
| 7 | 9 | 7 | 8 | 9 | 8 |
| 8 | 8 | 7 | 7 | 7 | 7 |
| 9 | 7 | 6 | 7 | 9 | 7 |
| 10 | 8 | 8 | 8 | 6 | 7 |
| 11 | 7 | 8 | 7 | 6 | 7 |
| 12 | 8 | 8 | 8 | 7 | 7 |
| 13 | 8 | 7 | 8 | 7 | 7 |
| 14 | 9 | 6 | 9 | 8 | 8 |
| 15 | 8 | 8 | 9 | 9 | 8 |
| 16 | 7 | 9 | 8 | 8 | 8 |
| 17 | 8 | 7 | 7 | 6 | 7 |
| 18 | 8 | 6 | 7 | 8 | 7 |
| 19 | 8 | 8 | 9 | 8 | 8 |
| 20 | 7 | 8 | 8 | 9 | 8 |
| 21 | 9 | 7 | 9 | 8 | 8 |
| 22 | 9 | 7 | 8 | 9 | 8 |
| 23 | 8 | 7 | 8 | 7 | 7 |
| 24 | 7 | 6 | 7 | 9 | 7 |
| 25 | 8 | 8 | 8 | 6 | 7 |
| 26 | 7 | 8 | 7 | 6 | 7 |
| 27 | 8 | 8 | 8 | 7 | 7 |
| 28 | 8 | 7 | 7 | 8 | 7 |
| 29 | 9 | 7 | 9 | 8 | 8 |
| 30 | 8 | 8 | 9 | 9 | 8 |
| | | | | | |

Appendix-6: Sensory characteristics of Moringa, Mint, Orange peel and Joba flower mixed green tea [Sample 2].

| Panelists | Color | Appearance | Smell | Taste | Overall acceptability |
|-----------|-------|------------|-------|-------|---------------------------------|
| 1 | 8 | 7 | 8 | 5 | 7 |
| 2 | 9 | 6 | 7 | 5 | 6 |
| 3 | 8 | 5 | 6 | 6 | 6 |
| 4 | 9 | 6 | 8 | 6 | 7 |
| 5 | 8 | 7 | 8 | 5 | 7 |
| 6 | 9 | 7 | 7 | 7 | 7 |
| 7 | 8 | 7 | 6 | 6 | 7 |
| 8 | 7 | 6 | 7 | 5 | 6 |
| 9 | 7 | 8 | 6 | 6 | 6 |
| 10 | 8 | 5 | 8 | 7 | 7 |
| 11 | 7 | 6 | 8 | 5 | 6 |
| 12 | 8 | 5 | 8 | 6 | 7 |
| 13 | 7 | 5 | 7 | 5 | 6 |
| 14 | 9 | 7 | 6 | 5 | 6 |
| 15 | 7 | 7 | 8 | 6 | 7 |
| 16 | 8 | 6 | 8 | 6 | 7 |
| 17 | 7 | 6 | 7 | 5 | 6 |
| 18 | 8 | 8 | 7 | 6 | 6 |
| 19 | 9 | 6 | 8 | 6 | 7 |
| 20 | 8 | 7 | 8 | 5 | 7 |
| 21 | 9 | 7 | 7 | 7 | 7 |
| 22 | 8 | 7 | 7 | 6 | 7 |
| 23 | 7 | 6 | 7 | 5 | 6 |
| 24 | 7 | 7 | 6 | 6 | 6 |
| 25 | 8 | 5 | 8 | 7 | 7 |
| 26 | 7 | 5 | 8 | 5 | 6 |
| 27 | 8 | 7 | 7 | 6 | 7 |
| 28 | 7 | 5 | 7 | 5 | 6 |
| 29 | 9 | 7 | 6 | 5 | 6 |
| 30 | 9 | 5 | 8 | 5 | 7 |

Appendix-7: Sensory characteristics of Moringa, Mint, Orange peel and Joba flower mixed green tea [Sample 3].