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Phytochemical and Organoleptic Tests of Combined Extracts of Turmeric, Black Tea, and Ginger

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ABSTRACT

Phytochemical screening to conduct a preliminary evaluation of the chemical content of black tea (Camellia sinensis), Turmeric (Curcuma longa), and Ginger (Zingiber officinale). In addition, the content of secondary metabolites in black tea, turmeric and ginger can be used as an indication of the quality of these plants. The need for a combination of black tea extract, turmeric, and ginger from various sources so that it can be liked by the public. This study aims to test the phytochemical extracts of black tea, turmeric, and ginger, as well as conduct organoleptic tests on the most optimal combination for α-glucosidase enzyme inhibitory activity. Phytochemical screening showed that black tea and ginger extract contains alkaloids, flavonoids, tannins, saponins, terpenoids, quinones, glycosides, and phenolics, while turmeric extract contains alkaloids, flavonoids, tannins, terpenoids, quinones, glycosides, and phenolics. The organoleptic test of the F7 combination, which includes black tea extract, turmeric, and ginger, showed the highest level of preference by P3 and P2 in terms of color, aroma, consistency, and aftertaste, while taste was most preferred by P3.

Keywords: black tea, ginger, organoleptic, phytochemical screening, turmeric.

1. INTRODUCTION

Tea (*Camellia sinensis* L. Kuntze) is a plantation product which is generally consumed in the form of a beverage. The effect of tea on health is caused by the presence of phenolic compounds in the form of catechins which act as antioxidants (Halim *et al.* 2015). Black tea is a type of tea that is fully fermented. Fermentation does not use microbes as a source of enzymes but uses polyphenol oxidases contained in the tea leaves themselves. Enzyme activity plays an important role in the formation of theaflavin and thearubigin pigments. This process produces tea leaves that are brown or black in color (Friskilla & Rahmawati 2018).

Turmeric (*Curcuma longa* L.) is a tropical plant in the Asian continent which is extensively used as a coloring agent and food fragrance. Turmeric is a type of plant that is used as a spice ingredient by giving it a bright yellow color. Besides that, turmeric rhizome is also useful as an anti-inflammatory, antioxidant, antimicrobial, anti-senile, cancer prevention, antitumor, lowers blood fat and cholesterol levels, and blood purifier (Juswono *et al.* 2013). Curcumin is the main component found in turmeric. These polyphenolic compounds in the flavonoid group have various properties, including anti-proliferative, anti-inflammatory, antioxidant and anti-angiogenic properties. Some of the potential curcumin compounds can be utilized in efforts to prevent diabetes in patients with diabetes mellitus (Simorangkir 2020).

Ginger (*Zingiber officinaIe* Rosc.) is an annual herbaceous plant with leaves in pairs of sword-shaped leaves with a horn-like rhizome and aromatic. Ginger as a flavor and aroma enhancer in the process of making food ingredients containing flavanoids, polyphenols and essential oils. These compounds make the aroma of ginger distinctive with a refreshing spicy taste. Ginger contains phytochemical components such as alkaloids, tannins, glycosides, saponins, polyphenols (flavonoids), and terpenoids. These components have health benefits such as anti-inflammatory, antioxidant, hepatoprotective, and antidiabetic (Afandi 2011).

Domestic preparations of black tea, turmeric, and ginger are often in the form of dried leaves and rhizomes, while pharmaceuticals are in the form of extracts. According to Suryono *et al*. (2018) considers it necessary to carry out organoleptic trials of a product preparation, as well as the importance of carrying out hedonic tests on several types of food which can strengthen consumer acceptance of the food product under study.

These three ingredients have the potential to be used as herbal beverage preparations that inhibit the action of the α-glucosidase enzyme, but there is no information on which combinations are preferred by the public. Therefore, this study aims to test the phytochemical extracts of black tea, turmeric, and ginger, as well as conduct organoleptic tests on the most optimal combination for α-glucosidase enzyme inhibitory activity.

2. METHODOLOGY

Materials

The materials used in this study were black tea water extract, 70% ethanol extract of turmeric, 70% ethanol extract of ginger (extract from PT. Haldin Pacific Semsesta), distilled water, chloroform, ammonia, H_2SO_4 2 M, Dragendorf reagent, Meyer, Wagner, 96% ethanol, Mg powder, concentrated HCl, amyl alcohol, $FeCl_3$, diethyl ether, 70% ethanol, NaOH, methanol, and anhydrous acetic acid.

Phytochemical Screening

Alkaloids. Based on Mardhiyah (2019) as much as 0.1 g of extract was added to 5 mL of chloroform and 3 drops of ammonia. The chloroform fraction was separated and acidified with 30 drops of H_2 SO_4 2 M. The acid layer which was located at the top of the mixture was separated, then 6 drops of Dragendorff, Meyer, and Wagner reagent were added. The presence of alkaloids was indicated by the formation of an orange precipitate by Dragendorf's reagent, a white precipitate by Meyer's reagent and a brown precipitate by Wagner's reagent. Galangal as a positive control and fructose as a negative control.

Flavonoids. Based on Mardhiyah (2019) as much as 0.1 g of extract was added to 10 drops of 96% ethanol then stirred until completely dissolved. Then 5 mL of distilled water was added and then heated at 100℃ for 5 minutes. The filtered filtrate was added with 0.01 g of Mg powder and 5 drops of concentrated HCl then vortexed. 1 mL of amyl alcohol was added. Formation of red, yellow or orange in the amyl alcohol layer indicates the presence of flavonoids. Galangal as a positive control and fructose as a negative control.

Tannins. Based on Mardhiyah (2019) as much as 0.1 g of extract was added to 2 mL of distilled water and then boiled for 5 minutes. The solution was filtered and the filtrate was added with 5 drops of 1% (w/v) $FeCl_3$. Dark blue or greenish-black color indicates the presence of tannins. Black tea as a positive control and fructose as a negative control.

Saponins. Based on Mardhiyah (2019) 0.1 g of extract was added to 5 mL of distilled water and boiled for 5 minutes after which it was filtered. The filtrate in the test tube was shaken for 10 seconds and then left for 10 minutes. The presence of saponins was indicated by the formation of stable foam. Galangal as a positive control and fructose as a negative control.

Triterpenoids and Steroids. Based on Mardhiyah (2019) 0.1 g of extract was added to 2 mL of 70% ethanol then boiled for 5 minutes and filtered. The filtrate was evaporated then added 1 mL of diethyl ether and homogenized. Lieberman Burchard reagent (6 drops of anhydrous acetic acid and 6 drops of concentrated H_2SO_4 was added to the mixture. Red color indicates the presence of triterpenoids while green color indicates the presence of steroids. Galangal as triterpenoid positive control, bay leaf as steroid positive control, and fructose as negative control.

Quinone. Based on Simaremare (2014) 0.1 g of extract was added to 1 mL of distilled water, then added 2 mL of methanol, then filtered. The filtrate was added with 2 drops of 1 M NaOH. A positive result was indicated by the formation of a red color. Turmeric as a positive control and fructose as a negative control.

Glycosides. Based on Syafitri *et al*. (2014) took 0.1 g of sample and added 1 mL of distilled water and then added 5 drops of Fehling A and 5 drops of Fehling B to the mixture. The mixture was boiled until a brick/orange yellow color indicated the presence of glycosides. Curcuma as a positive control and table salt as a negative control.

Phenolic. Based on Susilawati (2020) 0.1 g of the extract was dissolved in 5 mL of technical methanol and then boiled for 5 minutes. Add 10 drops of 1 N NaOH to the mixture. The presence of phenolic compounds is indicated by the formation of a yellow precipitate in the mixture. Curcuma as a positive control and fructose as a negative control.

Organoleptic Test

The organoleptic test is a modification of the method by Widyawati *et al*. (2018) carried out in this study was a hedonic test (favorability test) by 30 student panelists on the best combination . The combination F7 (K-TH-J) is from a previous study by Hasan *et al.* (2022) with the addition of fructose in a ratio of 0% (P0), 5% (P1), 10% (P2) and 15% (P3) and dissolved in hot water (85-90^{o}C). The combination F7 for K is turmeric, TH is black tea, and J is ginger. The organoleptic test that was carried out was an acceptance test, namely that each panelist was required to submit a response about the product presented. The purpose of this test is to find out which product is preferred. The acceptance test used is the hedonic test. The hedonic scale used is with 4 numerical scales, namely very like (4), likes (3), mediocre (2), and dislikes (1).

Data Analysis

Data were processed using Microsoft Excel 2010 and SPSS version 25 according to the procedures in Yuningtyas & Artanti (2015).

3. RESULT

Phytochemical Screening

Phytochemical analysis aimed to identify secondary metabolites in extracts that are capable of having an effect on the bioactivity test of medicinal plants. The results of the phytochemical tests in Table 1 show that the water extract of black tea contains almost all of the tested secondary metabolites, namely alkaloids, flavonoids, tannins, saponins, terpenoids, quinones, glycosides, and phenolics, but does not contain steroids.

Meanwhile, the ethanol extract of turmeric contains alkaloids, flavonoids, tannins, terpenoids, quinones, glycosides, and phenolics and does not contain saponins and steroids. In addition, the ethanol extract of ginger contained almost all of the tested secondary metabolites (alkaloids, flavonoids, tannins, saponins, terpenoids, quinones, glycosides, and phenolics) except for steroids.

The results of alkaloid testing with Dragendorff, Mayer, and Wagner reagents were indicated by a change in red color, brown precipitate, and brown color. Flavonoids contained in the extract are marked with red, yellow or orange on the top layer, namely the amyl alcohol layer, while tannins are marked with the formation of dark blue or green-black color. Saponins in the extract were characterized by the formation of stable foam, terpenoids and quinones by the formation of a red color, glycosides by the formation of dark red, brick red and yellow, and phenolics by the formation of a yellow precipitate.

Table 1 Phytochemical screening results

No	Test	Black tea water extract	Turmeric ethanol extract	Ginger ethanol extract
1	Alkaloids			
	Dragendorf	+++	+++	+++
	Meyer	-	-	+++
	Wagner	+++	+++	+++
2	Flavonoids	+++	+++	+++
3	tannins	++++	++	++
4	Saponins	+++	-	+++
5	terpenoids	++++	+++	+
6	Steroids	-	-	-
7	Quinone	++++	++++	+
8	Glycosides	++++	++++	+++
9	Phenolic	++++	++++	++++

Description: (++++) Very strong, (+++) Strong, (++) Strong enough, (+) Weak, (-) No compound

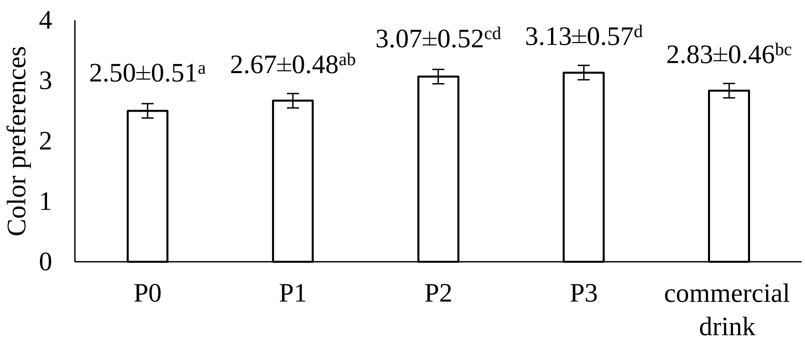

Figure 1. The results of the hedonic test for color attributes of the F7
Note: Different lowercase letters show statistically significant differences (α= 0.05)

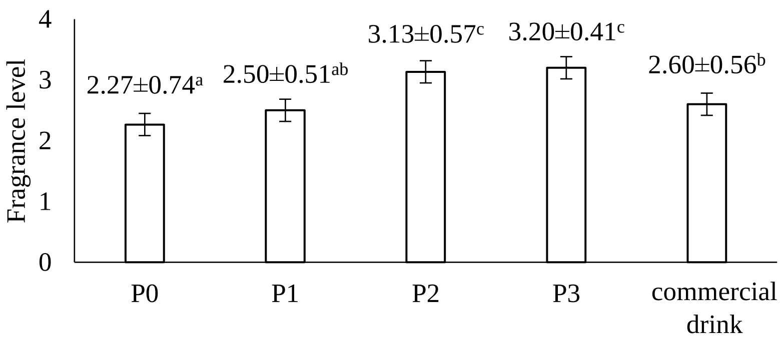

Figure 2. The results of the hedonic test for aroma attributes of the F7
Note: Different lowercase letters show statistically significant differences (α= 0.05)

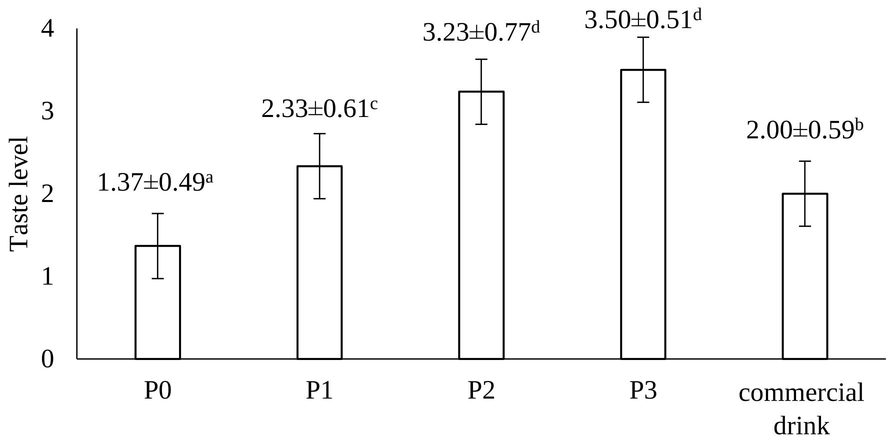

Figure 3. The results of the hedonic test for the taste attribute of the F7
Note: Different lowercase letters show statistically significant differences (α= 0.05)

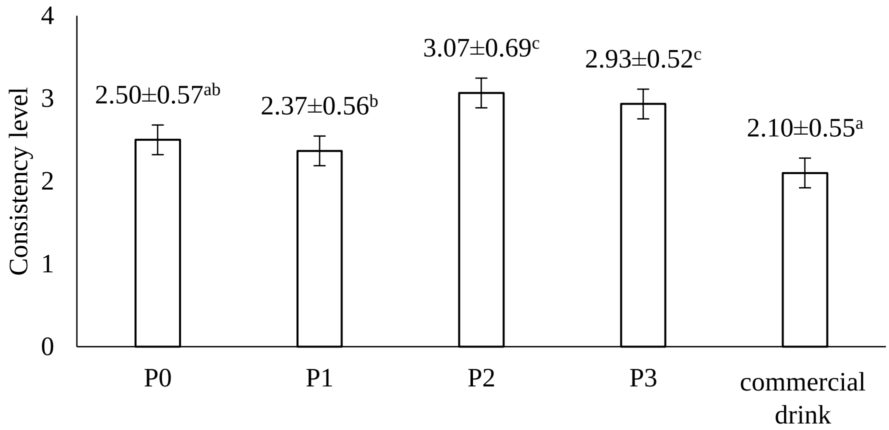

Figure 4. The results of the hedonic test for the consistency attribute of the F7
Note: Different lowercase letters show statistically significant differences (α= 0.05)

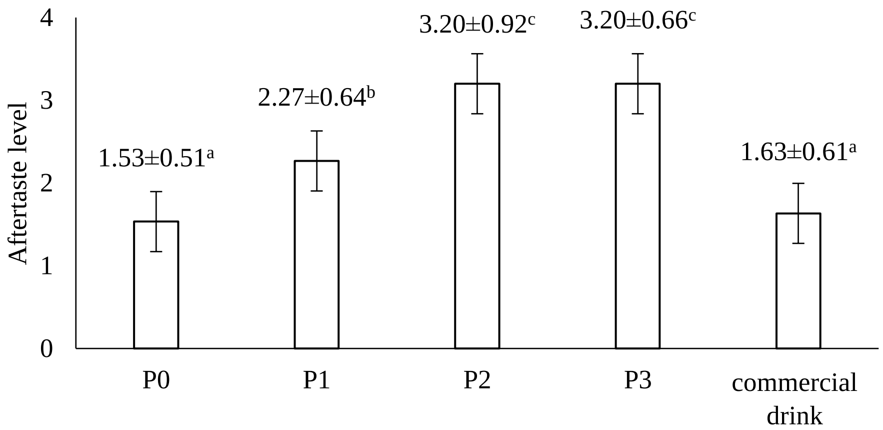

Figure 5. The results of the hedonic test for the aftertaste attribute of the F7
Note: Different lowercase letters show statistically significant differences (α= 0.05)

Organoleptic

The hedonic test was carried out using the category scale method (1 = dislike, 2 = normal, 3 = like, 4 = really like) where the panelists gave an assessment of the preference of the F7 drink (K-TH-J) on the attributes of color, aroma, taste , consistency, and aftertaste . P0 shows the combination of F7 (K-TH-J) with the addition of 0% fructose, P1 shows the combination of F7 (K-TH-J) with the addition of 5% fructose, P2 shows the combination of F7 (K-TH-J) with the addition of 10% fructose, and P3 indicates the combination of F7 (K-TH-J) with the addition of 15% fructose. The results of analysis of variance (ANOVA) showed that all attributes were significant ($p<0.05$), so a follow-up test (Duncan) was performed for all attributes.

4. DISCUSSION

Phytochemical Screening

Alkaloid compounds are semi-polar because they have a nitrogen atom that has a lone pair of electrons that will bond covalently with iodine ions in the reagent and have substituents such as phenol, amine, amide and methoxy. These compounds are important in the survival of plants against microorganisms, insects, herbivores and other plants (Mardhiyah 2019). Flavonoids are phenolic compounds found in vascular plants and are water soluble. There are about 10 types of flavonoids, namely antisoanins, proanthocyanidins, flavonols, flavones, glycoflavones, biflavonyls, chalcones, aurons, flavonones, and isoflavones. Flavonoid compounds found in all parts of the plant act as antifungals, heart stimulants, lower blood sugar levels, anti-inflammatory, and antioxidants (Mardhiyah 2019). According to Sasmita *et al*. (2017), flavonoids are substances that can regenerate pancreatic beta cells and help stimulate insulin secretion.

Tannin is a compound that has a phenol group and gives it an astringent taste. Method classic to identify tannin compounds with the formation of black-green or blue ink in the extract after adding $FeCl_3$ (Mardhiyah 2019). Erviani *et al.* (2019), stated that saponins function as antihyperglycemic by preventing gastric emptying and preventing increased glucose uptake in the brush border membrane in the intestine. Steroid compounds are not present in all extracts because extracts with water and ethanol solvents are polar, while steroid compounds are nonpolar, so they are not extracted completely in these solvents (Ergina *et al*. 2014). Terpenoids consist of volatile monoterpenes and sisquiterpenes, volatile diterpenes, and non-volatile triterpenes and sterols. Triterpenoids are active components in medicinal plants that can treat diabetes, liver damage and malaria. Plants themselves use triterpenoids as insecticides or antifungals (Khotimah 2016).

Glycosides are compounds that are hydrolyzed to form sugars (glycones) and other compounds (aglycones or genin) (Andriani 2011). Glycosides play a very important role in plants, namely they are involved in regulatory, protective, and sanitary functions (Martono & Setiyono 2014). Quinones are colored compounds and have basic chromophores. Benzoquinones (quinones with a chromophore consisting of 2 carbonyl groups conjugated with 2 carbon-carbon double bonds), naphtoquinones, anthraquinones are phenolic compounds which are generally hydroxylated, while isoprenoid quinones are involved in cell respiration (ubiquinone) and photosynthesis (plastoquinone) which are widely distributed. in plants (Andriani 2011; Sulasiyah *et al.* 2018). Phenolics in black tea are known to have a percentage of 5-27%. Phenolic compounds are known to have antioxidant activity. Phenolic compounds in black tea include flavanol compounds, namely catechins, catechin polymers (theaflavins and thearubigins), and other flavanol compounds kaempferol, myricetin, and quercetin (Paramita *et al.* 2020). Curcumin is a group of phenolic compounds contained in the rhizomes of the *Zingiberaceae* plant (Prihardini & Basuki, 2019). Ginger contain compound phenolic active like gingerol, shogaol, and curcumin (Susanti & Panunggal, 2015). Phenolic compounds have antioxidant activity that can reduce stress oxidative by preventing the chain reaction of converting superoxide into hydrogen superoxide by donating a hydrogen atom from the aromatic hydroxyl group (-OH) to bind to free radicals and remove them from the body through the excretory system (Andrie *et al*. 2014).

Compounds that were not found in the test extract may not have been present due to their low component levels, which prevented phytochemical screening from specifically detecting their presence (Rauf *et al*. 2011). According to Iswantini *et al*. (2011), variances in environmental factors may be to blame for the disparity in results. The kinds and concentrations of secondary metabolites that are present in a material can vary depending on the environmental circumstances. According to Widarta *et al*. (2013), various types and intensities of solvent polarity can result in various yields of extracts and bioactive components. According to Sompong *et al*. (2011), different species and environments produce various bioactive components.

Organoleptic

The sensory evaluation of the best combination, namely the F7 combination based on previous research conducted by Hasan *et al*. (2022) which consisted of extracts of black tea, turmeric, and ginger was carried out using an organoleptic test (hedonic test). The dose used for black tea extract was 26.67 mg/kg, turmeric extract was 24 mg/kg, and ginger extract was 155.96 mg/kg so that the total extract used was 206.63 mg/kg dissolved in 200 mL. water in one drink. The concentration of fructose added to treatment P1 was 5% fructose by 10 grams, treatment P2 was 10% fructose by 20 grams, and treatment P3 was 15% fructose by 30 grams.

Organoleptic test is a test using the human senses as an instrument to measure product acceptance. Assessment of food ingredients is a property to determine whether a product is acceptable or not is called sensory properties. The senses used in assessing sensory properties are the senses of sight, touch, smell and taste. This study used an affective test based on preference or relative preference level measurements towards a product and requires a number of panelists who are often considered to represent certain consumer groups. The hedonic test is a test to express likes or dislikes for a product. This level of preference is called the hedonic scale, which is measured and written by the panelists in the questionnaire (Suryono *et al*. 2018).

The hedonic test was carried out on color, aroma and taste. The results of the analysis of variance showed that the comparison of the combination of F7 (K-TH-J) was significant ($p < 0.05$) to the color of the steeping using the hedonic test. Figure 1 shows the average sensory value by panelists ranging from 2 .50 ± 0.51 (likes) to 3.13 ± 0.57 (likes). The highest average value of panelists' preference for the color combination F7 (K-TH-J) was highest in treatment P3 (combination F7 (K-TH-J) with the addition of 15% fructose), namely by liking criteria while the lowest value was obtained in treatment P0 (combination of F7 (K-TH-J) with the addition of 0% fructose, namely with the criteria of liking, and commercial drinks (2 .83 ± 0.46), namely with the criteria of liking. Consumer acceptance of a product is highly dependent on physical characteristics such as color, shape and texture. Color is an important aspect because it relates to consumer acceptance of a product because of the first visual appearance in addition to several factors including taste, aroma and nutritional value (Savitri *et al*. 2019).

The results of the analysis of variance showed that the comparison of the combination of F7 (K-TH-J) was significant ($p < 0.05$) to the aroma of the brew using the hedonic test. Figure 2 shows the average sensory value by the panelists ranging from 2 .27 ± 0.74 (average) to 3.20 ± 0.41 (like). The highest average value of panelists' preference for the aroma of the combination F7 (K-TH-J) was highest in treatment P3 (combination F7 (K-TH-J) with the addition of 15% fructose) and was not significantly different from treatment P2 (combination F7 (K -TH-J) with the addition of 10% fructose, namely with the criteria of liking, while the lowest value was obtained in treatment P0 (combination F7 (K-TH-J) with the addition of 0% fructose), namely with the criteria of mediocre, as well as commercial drinks (2.60 ± 0.56), that is, with the like criterion.

The aroma of food arises due to the presence of volatile components which evaporate easily but the volatile components will be lost during the heating process. The distinctive aroma that arises is the result of a combination of black tea extract, turmeric and ginger. Smell assessment requires sensitivity in feeling and smelling, aroma exhibits sensory properties which are difficult to classify and describe because the variety is so large (Usman *et al.* 2015). A good aroma will also lead to good acceptance in consumers.

The results of the analysis of variance showed that the comparison of the combination of F7 (K-TH-J) had a significant ($p<0.05$) effect on the steeping taste using the hedonic test. Figure 3 shows the average sensory value by the panelists ranging from 1.37 ± 0.49 (dislike) to 3.50 ± 0.51 (very like). The panelist's preference for the highest taste combination F7 (K-TH-J) was obtained in treatment P3 (combination F7 (K-TH-J) with the addition of 15% fructose), namely with the criteria of really liking it while the lowest value was obtained in treatment P0 (combination F7 (K-TH-J) with the addition of 0% fructose) that is with the criteria of somewhat dislike, and commercial drinks (2.00 ± 0.59) that is with the criteria of mediocre. Taste is an important aspect in the assessment of a product. Taste assessment is carried out by the human sense of taste when food or drink is consumed (Savitri *et al*. 2019).

Tea leaves contain three main components, namely caffeine, polyphenols, and essential oils. These components influence the quality of tea. Black tea is brownish black in color and is produced through a fermentation process. The main compound of polyphenols in tea is tannin which can affect color and taste (Jayanto 2012). The tannins in black tea undergo a condensation reaction to form theaflavins and thearubigins. Theaflavins yellowish red color and slightly acidic which will affect the freshness and brightness. Thearubigin affects the color and thickness which is brownish red in color and contributes to the acidity and freshness of the tea (Ikrawan *et al*. 2019). Other polyphenol compounds are catechins which will give tea a bright color when catechin levels are high and taste bitter (Jayanto, 2012). The black tea which goes through an enzymatic oxidation process will produce the strongest aroma with a lighter taste (Savitri *et al.* 2019).

Turmeric contain curcumin and volatile essential oils give it a very characteristic aroma. Turmeric as a natural dye contains curcumin which can produce a yellow color. Curcumin causes a bitter taste while the essential oils that determine the aroma and taste of turmeric are essential oils. Compound The composition of essential oils in turmeric includes ketones, cis-quiterpenes, turmeron, zingiberen, felandren, sabinen, borneol, and sineil. Terpenoid compounds cause turmeric to have a pungent and bitter aroma (Mulyani *et al.* 2014).

Compound (6)-gingerol gives the sharpest taste in ginger. The drying process (rehydration or removal of water molecules) from fresh ginger to dry ginger changes gingerol homologs to shogaol homologs which have a lower sharpness of taste than gingerol. Among the gingerol homologs, (6)-gingerol has the highest sharpness of taste, followed by (8)-gingerol, and (10)-gingerol (Fathona 2011). The strong aroma of ginger is caused by the content of zingiberene and zingiberol compounds (Savitri *et al*. 2019)

The results of the analysis of variance showed that the combination of F7 (K-TH-J) was significantly ($p<0.05$) on the consistency of steeping using the hedonic test. Figure 4 shows the average sensory value by the panelists ranging from 2.10 ± 0.55 (average) to 3.07 ± 0.69 (like). The highest average value of panelists' preference for the consistency of the combination F7 (K-TH-J) was highest in treatment P3 (combination F7 (K-TH-J) with the addition of 15% fructose) and was not significantly different from treatment P2 (combination F7 (K -TH-J) with the addition of 10% fructose, namely with the criteria of liking, while the lowest value was obtained in treatment P0 (combination F7 (K-TH-J) with the addition of 0% fructose), namely with the criteria of mediocre, as well as commercial drinks (2.10 ± 0.55), that is, with normal criteria.

Consistency (thickness) is a physical property that must be considered in determining the quality of the F7 (K-TH-J) combination. Consistency is an important aspect for assessing the quality of food products. Consistency is included in one of the factors that influence consumer acceptance of food products. Component The greater solute in a solution will increase the viscosity. Viscosity can be observed absolutely by poise and relative f which is based on the amount of volume that can flow at a certain time. The level of consumer acceptance of the product depending on the consistency of the food product (Sawitri 2011).

The results of the analysis of variance showed that the combination of F7 (K-TH-J) was significantly ($p<0.05$) to *the aftertaste* of the brew using the hedonic test. Figure 5 shows the average sensory value by the panelists ranging from 1.53 ± 0.51 (average) to 3.20 ± 0.66 (like). The highest average value of panelist preference for *the aftertaste* combination of F7 (K-TH-J) was highest in treatment P3 (combination F7 (K-TH-J) with the addition of 15% fructose) and was not significantly different from treatment P2 (combination F7 (K -TH-J) with the addition of 10% fructose, namely with the criteria of liking, while the lowest value was obtained in treatment P0 (combination F7 (K-TH-J) with the addition of 0% fructose), namely with the criteria of mediocre, as well as commercial drinks (1.63 ± 0.61), that is, with normal criteria.

Aftertaste is an impression that is still felt after the sensing has been done. After effect duration is a state of excitatory substance that gives rise to the impression that it is easy or does not easily disappear after sensation is finished. The sweet taste has a lower or weaker lingering impression than the bitter taste. Aftertaste divided into several levels called the excitability threshold. Threshold stimuli, consisting of an absolute threshold , namely the smallest number of stimuli that have started to create an impression, the recognition threshold , namely the type of impression that has been recognized, the difference threshold , namely the smallest difference whose effect has been recognized, and the threshold (terminal threshold), which is the largest level of stimulation that can still be distinguished by the intensity of the effect (Ikrawan *et al*. 2019). The aftertaste of the F7 (K-TH-J) combination is felt as a result of the constituent components of each extract which have an astringent, bitter, hot, and spicy taste.

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