

## UTILIZATION OF BANABA (*Lagerstroemia speciosa* L.) LEAVES FOR POWDERED MILK-TISANE

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### ABSTRACT

Banaba (*Lagerstroemia speciosa* L.) tree was observed to be herbal tree inside the Philippines as well as in India, Malaysia, Vietnam and Indonesia. Banaba leaf tea is usually used as medicinal drinks which are known for its blood-sugar lowering mainly its efficiency in curing diabetes, and antioxidant. It is simply by using their leaves extracted with hot water. Although in locals, the Banaba leaves are used to treat diabetes and are taken as a medicine. On the quest for creating new product of local milk-tea, the studies include the physico-chemical analysis, its sensory evaluation, antioxidant potential. Analyzing the formulated milk tea with the commercial milk tea as controlled, with codes of 863 and 924, respectively, the appearance of the showed little difference of mean scores. In its aroma, given 924 as more acceptable while the color, taste, flavor and its overall acceptability shows 863 is higher. In the t-test; appearance, aroma, and color with P value of 0.89585, 0.257584, 0.49589 respectively, showed no significance while the taste, flavor and overall acceptability with 0.01448, 0.007409, 0.00347 respectively, showed significance between the two products. In the DPPH Assay, ascorbic acid as positive control in its absorbance has continuously decreasing compared to the sample that has retained in constant with less increase and decrease of absorbance. The scavenging activity between two sample as ascorbic acid its positive control has significantly in an increase in concentrations while the sample became as well in constant or no significant differences with an increase of dilution. In conclusion, the Banaba milk tea can potentially be considered for its sensory traits and its antioxidant activity.

### INTRODUCTION

Instant tea is an outstanding interest to the majority, medicinal and nutritional experts, and health and food technological knows how researchers to know the antioxidant capacity and constituents within the food we eat. Health advantages of diets rich in antioxidants encompass reducing the danger of cardiovascular diseases, positive cancers and natural degeneration of the body associated with the aging method. Bush tea and black tea are drinks wealthy in antioxidants, which make them exquisite sources for multiplied health advantages. Black tea is one of the most commonly fed on liquids inside the world for its suited aroma, taste and putative high-quality physiological capabilities. It is the most widely used tea in making iced tea and English tea. New flavor and flavors has created by means of blending black tea with chrysanthemums, plums, ginger, hibiscus or lemon. (Pandey, Manimehalai. 2014).

Furthermore, the tea has its chemical and biological activities that include their taste and aroma due to the molecules from carotenoids, glycosides, and lipids. These found in green, black, and oolong teas. (Chi-Tang, Xin, Shiming. 2015). Spices of indigenous have the possibility of a use as a flavor agent on green and black tea such as the ginger, lemongrass, kaffir lime and jasmine. It resulted in hiding the aroma and flavor of these drinks. (Supartono, Sukartiko, Yulianto, Kristani. 2015). According to Aprotosaie 2011, not only these spices produce aroma that can be volatile but also cocoas contain molecules that are volatile, which means that its gases readily vaporized at room temperature. It says that the polyphenols of its seeds are similarly to wine, tea, or vegetables making its taste to be sharp and bitter. (Aprotosaie, Luca, Miron. 2015).

As the worldwide consumption of tea is second only to water, the beneficial effects of tea represent important public health issues. However, up to now, it is not known whether addition of milk to tea, as widely practiced in the UK, influences these vasoprotective properties. According to Lorenz, Jochmann, Krosigk, Martus, Baumann, Stangl, Stangl, therefore investigated the outcomes of tea with or without milk on endothelial characteristic as a touchy parameter of vascular wall homeostasis. Many pathophysiological conditions inside the cardiovascular device characterize by means of reduce production of protecting vasoactive materials in endothelial cells, ensuing in a condition called endothelial disorder.

With the study on Black tea with addition of milk content, the results on the absorption of the tea's polyphenols were unaffected with the addition of milk. Associated significances such as the increase in plasma antioxidant capacity, and concentration of total phenols, catechins, and flavonols quercetin and kaempferol, in observance during the period of 80 minutes of infusion time. (Kyle, Morrice, McNeill, Duthie. 2007.). On another study of tea which is green tea, with regards to different amounts of additional milk-

protein on green tea did not have any additional effect on diet-induced thermogenesis. Thus, it explained the inhibitory effects of milk on the effect of the green tea. (Hursel, Westerterp-Plantenga. 2011.) But through all the studies, one suggested that it is not the protein content itself that causes the inhibition of the tea by the possibly the fat content. Catechins have a complex ring structure and are fat-soluble which causes the formation of complexes and between fat and polyphenols. (Van het Hof, Kivitis, Weststrate, Tijburg. 1998.).

Tea consumption practices as a tradition, has shown potential to improve human health. Maximal uptake of tea antioxidants and milk proteins without a negative impact on tea flavor is highly desired by consumers. There is a conflicting evidence of the effect of milk addition to tea on antioxidant activity. Differences in the type of tea, the composition, type and amount of milk, preparation method of tea-milk infusions, criticism used to measure antioxidant activity, and sampling size likely account for different findings. Interactions between tea polyphenols and milk proteins, especially between catechins and caseins, could account for a decrease in antioxidant activity, although other mechanisms are also possible, given the similar effects between soy and bovine milk. (Birch, Sun-Waterhouse, Everett. 2015).

Several studies have demonstrated that tea flavonoids protect cells and tissues against free radicals have implicates in the etiology of oxidative stress-related disease disorders. However, black tea is commonly consumed with additives that could otherwise affect the bioavailability of the active tea molecules. The biochemical parameters of Kenyan teas were determined and the effect of added milk and sweeteners on the antioxidant activity of Kenyan teas was investigated. The effect of tea antioxidants on glutathione (GSH) was also evaluated in vivo in a time series study using Swiss mice. The addition of milk, sugar and honey significantly decreased the antioxidant activity of tea in a concentration-dependent manner. (Korir, Wachira, Wanyoko, Ngure, Khalid. 2013).

For the addition of the sweetener, stevia (*Stevia rebaudiana* Bertoni), showed no significant influence on the antioxidant activity of tea and therefore can be recommended as a preferred sweetener for tea. Significant higher levels of GSH has observed in plasma than in other tissues. GSH levels were generally highest 2hrs after tea consumption, which indicates that, to fully maximize the potential health benefits of the tea being consumed, the drinkers should repeat the intake of the same type of tea every 2hrs. (Korir, Wachira, Wanyoko, Ngure, Khalid. 2013).

According from the Charite Hospital in Berlin, compared to the endothelial function in the brachial artery of 16 postmenopausal women, after drinking black tea with no milk; black tea with milk or hot water. They found that while tea increased the artery's ability to relax and expand to

accommodate increased blood flow compared to water; this effect can block when milk is added to the beverage. (Lorenz, Stangl. 2008).

In spray drying of instant tea powder. Extracts are prepared from the milled dried tea and then dissolves to a hot water with different temperature and ratio to water with 1:10 (w/v). According to Pandey and Manimehalai, it recommends brewing tea at 65 degrees Celsius for 50 minutes. During the feed flow at 20mL/min with the in-let temperature of 200 degrees Celsius does not affect the total polyphenols content. The TPC should be 13.19 mg/100gm. In a study according to Susantikarn and Donlao, they have studied the conditioning of the tea extract with different properties such as its water activity, solubility, hygroscopicity, color value. They have studied three values of the addition of maltodextrin, 3, 5, and 7% (w/v). It has resulted that having the 3% (w/v) value has produced good properties of the green tea powder respectively with the temperature in-let air temperature of the spray dryer at 210 degrees Celsius. (Susantikarn, Donlao. 2015).

Banaba (*Lagerstroemia speciosa* L.), came from a family Lythraceae. In which the name came from the queen of flower or the pride of India. In India, it is called as arjuna, bungur in Malaysia and Indonesia, Thailand is tabak, and Banaba in the Philippines. (Chan, Tan, Wong. 2014.). A deciduous herbal tree observed inside the Philippines, has been notably studied for its efficiency in curing diabetes. (Deocaris, Dela Ysla, Mojica, Aguinaldo, Ascencion, 2005).

The Banaba is cultivated in Manila for its beautiful vegetation. Within the Philippines, Banaba is a famous folk medicinal plant. It is commonly found planted alongside of the roads, gardens and in park. On a study of phytochemistry, its specie has been known to be a product of anti-diabetic drugs. On its leaves itself, triterpenes, tannins, ellagic acids, glycosides, and flavones are to be found. (Chan, Tan, Wong. 2014.). Also, in the Philippines, it is prepared and taken like tea. It is then consumed as herbal tea for its blood-sugar lowering level and reducing body weight. (Park, Lee. 2011.) In the tradition of the people in Southeast Asia, its leaves, roots and barks are all used for different illnesses and ailments for remedy. (Ragasa, Ngo, Rideout. 2005.). Banaba is located within the Bataan Islands and from northern Luzon to Palawan, Mindanao and the Sulu Archipelago. It was discovered in maximum or all islands and provinces, mainly in secondary forests at low and medium places.

An antioxidant capacity of any product or raw materials is used to be a parameter in characterizing for its activity, potentiality, and power). Several methods are used to identify and DPPH Assay is the commonly wide used method. It is to test its ability whether the product can act as a free radical scavengers or a hydrogen donor. (Pekal, Pyrzynska. 2012). In a study of Farooq and Sehgal, they have compared different type of green tea such as the loose leaf, the bagged, and the powdered match in their scavenging activity. The IC50 values of these three shows no significance except for the bagged tea with faintly higher scavenging potentials. But, in general, the green teas are still stronger than the black and herbal teas. (Chan, Soh, Tie, Law. 2011).

The antioxidative activity of water extracts of Banaba has the content of 37% of tannin in dry weight. It has also showed strong anti-oxidative activity in a linoleic acid autoxidation system. It also founded potentials on a radical scavenging action on DPPH radicals and superoxide radicals. (Anil, Manish, Garvendra, Vijay, Tarachand. 2010.). Moreover, in an in vitro study the Banaba water extract also inhibited lipid peroxidation in a rat liver homogenate. With regards to the anti-oxidative action of Banaba extract, was suggested that the tannins (polyphenolic compounds) were the mostly attributed to this which plays an important role on the activity, also the absorption of (-)-epigallocatechin gallate (a major polyphenol found in green tea). (Unno, Sakane, Masumizu, Kohno, Kakuda. 1997). The antioxidant activities of *L. speciosa* tea were compared to green tea, oolong tea, and black tea. The results have shown that the *L. speciosa* has obtained a greater antioxidant property and phenolic content compared to black and oolong tea according to Chan, Lim, Chong, Tan, Wong. 2010 and Chan, Lye, Tan. 2011. According to Anil, Manish, Garvendra, Vijay, and Tarachand, 2010. It was found that in their study of the Banaba leaf powdered extract (*Lagerstroemia speciosa* L.) can be a source of antioxidant activity, which it helps to inhibit against oxidative stress diseases. The extract helps to maintain regular level

of enzymatic and non-enzymatic antioxidants, because Banaba plant extract can be a source of antioxidative property which could improve the action of pathological alterations (Saumya and Basha, 2011).

The study aims in utilizing the Banaba-tisane in to formulate a milk tea. The study explore the consumer's preference between the new formulation made from a local tea and the commercial milk tea using a standard test. It also examine the effects of the antioxidants present in the product of Banaba milk-tisane. The study will only cover the use of dried Banaba leaf in producing the milk-tisane.

#### Research Design

The study conducts an exploratory design to focus on studying the development of Banaba leaves to a milk tea and its effects on the anti-oxidative activity with the addition of milk.

## MATERIALS AND METHODS

### Materials Preparation

#### Banaba Leaf

The fresh banaba leaves was purchased from a local plantation in Tabang, Bulacan, and the specimen was brought to the Bureau of Plant Industry (BPI) for authentication.

#### Low-fat Milk powder

The Low-fat Milk powder was locally purchased from Rigels Food Corporation in the Philippines.

#### Pure Stevia Powder

Pure Stevia powder was locally purchased from Sweet and Fit stevia on April.

#### Carrier Material

The main carrier material used is the maltodextrin, it was collected in Neco Food Corporation Manila Philippines. The required type used for the spray drying method is the 5-50% matrix type load with a particle size of 10-400 micrometer (Nicolas and Shimoni, 2010).

#### Tea Preparation

For the preparation of Banaba tea, the leaves were washed then it was infused with 200g of its leaves and 4 liters acidified water (pH was controlled at 5.0 using citric acid at 90 degrees Celsius for 40 minutes. The solids were filtered using a 5 mm mesh from the extract. The extracted liquid was mixed with maltodextrin with the ratio of 3% (w/v). Then it was stored at 4 degrees Celsius until needed. (Susantikarn, P., Donlao, N. 2015).

#### Spray-drying Method

The prepared Banaba tea was dried by using spray-drying method with a two-fluid nozzle at 50 degrees Celsius, the temperature of the air-inlet controlled in between 150 up to 220 degrees Celsius including its flow rate, feed rate, feed temperature, and the evaporative cooling. The maltodextrin was the carrier material that will help to sustain the properties of the extracted liquid while being dried in the spray drier. The droplet temperature should not exceed than 100 degrees Celsius (Nicolas and Shimoni, 2010). After spray dried, the powdered Banaba-tisane was package in a transparent plastic bag.

#### Physico-Chemical Analysis

The pH analysis of the powdered milk-tea was based on the AOAC International, 20<sup>th</sup> edition using Electrometry method Overall moisture content based on AOAC International, 20<sup>th</sup> edition using Gravimetry method.

### Microbiological Analysis

Yeast count in solid (total) based on Bacteriological Analytical Manual using Pour Plate. Mold counting was based on Bacteriological Analytical Manual using Pour Plate.

Table 1. Formulation of the Banaba milk-tisane

Ingredients	Trial 1
Banaba	75%
Stevia (sweetener)	10%
Milk	15%

The table shows the computation of formulation of the 100% formulated ingredients which equivalent into 12 g per 250mL of cold water. The trials presented on the table are evaluated accordingly to sensorial tests. The trial that was preferred will be furthered formulate.

### Panelists and Sensory Evaluation

#### The subject preparation and sample preparation

The 80 consumer panelists composed of residents who are living in Manila. The two samples contain a cold 30-mL freshly served in 50-mL plastic cups. The samples were coded with a random three-digit code.

#### Evaluation Procedure

Tea was evaluated by its properties such as appearance, flavor, taste, color and overall acceptability by scoring method using 9-point hedonic scale (Adnan, Ahmad, Ahmed, Khalid, Hayat, and Ahmed, 2013). Each consumer panelists are served with different samples and codes. Before the evaluation began, it is explained to the consumer panelists about the tasting procedure and the rinsing procedure. It is also explained on how to evaluate the samples

Using the t-test, each consumer panelists are served with two samples with sample codes. One sample for the control and the other is the formulated milk tea. Before the test began, it was explained to the consumer panelists about the tasting procedure and the rinsing procedure. It was also explained on how to evaluate the samples using the paired preference test. In the sensory evaluation test, 9-point hedonic scale was used to determine the acceptability of Banaba Milk-tisane and the Commercial product. 9-point hedonic scale was used to know which the two samples is much acceptable by the panelist, and it is convenient and easy to use because of the categorized choices and information of the two products (Wichchukit and Mahony, 2014). Eighty (80) panelists were asked to rate the two milk "tea" attributes in different codes. The student researchers do not indicate the percentage of milk "tea" to avoid bias in panelists.

To interpret the 9-point hedonic scale and its choices used are: 1= Extremely Unacceptable, 2= Very Much Unacceptable, 3= Moderately Unacceptable, 4= Slightly Unacceptable, 5= Neither Acceptable or Unacceptable, 6= Slightly Acceptable, 7= Moderately Unacceptable, 8= Very Much Unacceptable and 9= Extremely Acceptable (Stone, Sidel 1993; Hough, 2005).

### Statistical Analysis

A bar-graph analysis was performed to show the mean of the two samples preferred by the consumers according to its properties. Statistical analysis was conducted using Microsoft Excel for Windows.

#### Antioxidant Test

The Antioxidant Test was determined by using DPPH Radical Scavenging Method (Shekhar and Goyal, 2014).

### Method Flowchart

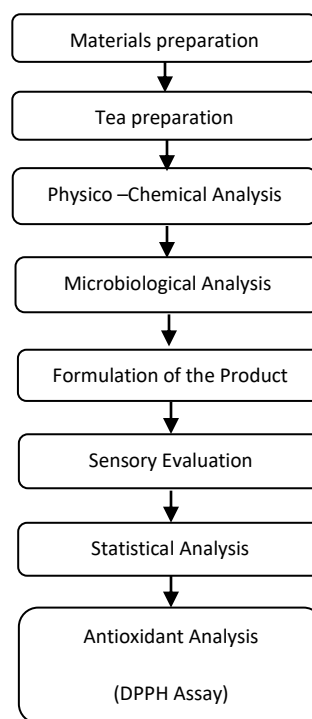


Figure 1. Method

### RESULTS AND DISCUSSION

Table 2 shows the %yield of the liquid extract after spray drying. It has shown that for 8 liters of liquid extract there is 2 kilograms of solid ingredient. There is about 25% of yield for powdered Banaba-tisane.

Table 2. % Yield of powdered Banaba-tisane

Total amount of solid	Amount of extracted liquid (volume)	%yield
2 kilograms	8 liters	25%

#### Physico-Chemical Test of Powdered Banaba Tisane

Table 3 shows the results in physicochemical properties of Powdered Banaba Tea. Using of Gravimetry Methodology of analysis, the moisture of Banaba Powdered Tea obtained a result of 4.7. To get the pH results of the Banaba Powdered Tea, the methodology used was Electrometry. It was shown on the table the obtained result of the pH of Banaba Powdered Tea was 4.74 at 25 degrees Celsius. The storage of Banaba Powdered Tea is one of the factors that there's a moderately high on its moisture.

Table 3. Physicochemical properties of Powdered Banaba Tisane

Parameters	Unit	Standard Unit
Moisture g/100g	4.7	3%-4% up to 15%
pH(10%w/v), pH units	4.74@ 25.6degrees Celsius	3.72to 4.11

According to studies, the properties of ready to drink commercial teas on pH. The pH of the ready-to-drink samples were analyzed and said to have a slightly acidic value ratio from 3.72 to 4.11. (Martínez, Urías-Orona, Hernández-García. 2018). For the moisture content of teas, specifically in black tea, in order not to take up spoilage, at its initial drying must have at least 3% to 4% only of moisture content. But it only requires up to 15% of its moisture to accumulate bacteria and mold. (Baird-Parker, Gould. 2000).

#### Microbiological Test of Powdered Banaba Tisane

Table 4 shows the results in microbiological properties of Powdered Banaba Tea. To get the results of the Yeast Count and Mold Count, it was determined by using a Pour Plate Method. The result obtained in Yeast Count was at <10est, while the Mold Count has a <100est. The presence of the microorganisms in Powdered Banaba Tea was grown during its storage condition on a room temperature in a transparent packaging plastic for 15 days.

Table 4. Result of Microbiological Test of Powdered Banaba Tisane

Parameters	Unit	Standard Unit
Yeast Count, CFU/g	<10est	$\leq 10^4$ /g
Mold Count, CFU/g	<100est	$\leq 10^5$ /g

#### Sensory Evaluation Test for Powdered Banaba Milk-Tisane

Figure 2 below shows an interpretation of results in sensory properties of two milk tea samples. The Banaba milk-tisane shows a red bar while the commercial milk tea shows the blue bar. For the appearance, there is a little difference of mean scores in between samples while the aroma and color has a huge difference between the two samples. Banaba-tisane is very aromatic and has contributed to the outcome of the sample. As you observed, the sensory properties of two samples for the taste, flavor, and overall acceptability, has a huge gap in their mean differences.

For the Appearance the Banaba Milk-tisane and Commercial Milk tea has no significant difference at each other and same result obtained in aroma. When comes in color of the two milk "tea" samples, there is no significant difference between Banaba milk-tisane and commercialized milk tea. But when it comes to taste, there is a significant difference in the two samples as well as its flavor. Lastly the overall acceptability level of the two samples shows a significant difference. Refer to Appendix 3.

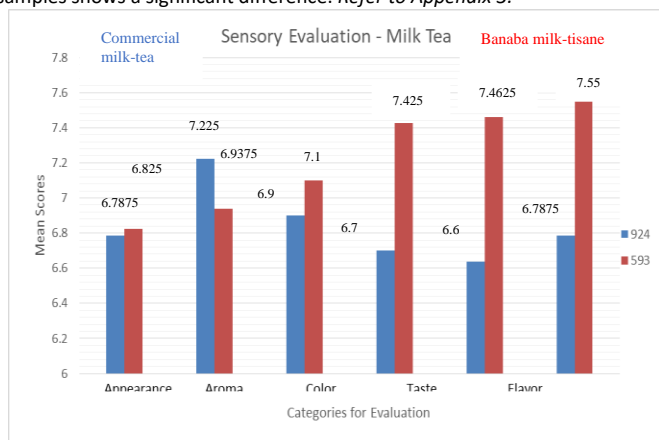


Figure 2. below shows an interpretation of results in sensory properties of two milk tea samples.

\*Legend = 9 – Extremely Acceptable, 8 – Much Acceptable, 7 – Moderately Acceptable, 6 – Slightly Acceptable, 5 – Neither Acceptable nor Unacceptable, 4 – Slightly Unacceptable, 3 – Moderately Unacceptable, 2 – Very Much Unacceptable, 1 – Extremely Unacceptable

#### Antioxidant Test (Dpph) for Powdered Banaba Milk-Tisane

The table 5 shows the result obtained of Banaba Milk-tisane its scavenging activity. As observed, the control that was used to compare the absorbance level of Banaba Milk tea was ascorbic acid. There are 5 different dilutions demonstrated the scavenging activity of the Ascorbic Acid and Banaba Milk tea. In column 1, with 5mcg/ml of dilution, the ascorbic acid got the absorbance of 35.404 and Banaba milk tea obtained a result of 29.052. With the 10mcg/ml, the ascorbic acid obtained a result of 51.258 and 27.439 scavenging activity was obtained in Banaba milk-tisane. For the dilution of 20mcg/ml ascorbic acid, it obtained a scavenging activity of 64.240 and the Banaba milk-tisane has 33.537.

When the positive control comes in 50mcg/ml of dilution it has obtained 96.761 scavenging activity while the Banaba milk-tisane has a 25.749 scavenging activity. The last column, dilution used was 100mcg/ml. As you have observed the ascorbic acid has 96.532 scavenging activity and for Banaba milk-tisane obtained 28.620 scavenging activity. The chemical components of the tea such as: total phenols and level of flavonoids can affect the scavenging activity of the tea, because higher level of flavonoids and total phenols can contribute for better scavenging activity of the tea (Pereira, Knor, Velloso, and Beltrame, 2014). It was found out in study, the way of processing of raw material with soluble components into infused tea plant conditions, and the type of plant specie has a possible effect in increased or decreased scavenging activity of the tea (Astill, Birch, Dacombe, Humphrey and Martin, 2001; Nikniaz, Mahdavi, Ghaemmaghami, Lotfi Yagin, Nikniaz, 2016).

Table 5. Results of 2,2 Diphenyl-1-Picrylhydrazyl (DPPH) Assay in Scavenging Activity

Sample	Scavenging Activity				
	5 mcg/ml	10 mcg/ml	20 mcg/ml	50 mcg/ml	100 mcg/ml
Ascorbic Acid (Positive Control)	35.404	51.258	64.240	96.761	96.532
Banaba Milk Tea	29.052	27.439	33.537	25.749	28.620

Table 6 shows the indication of IC50 values of Ascorbic Acid and Banaba Powdered Milk Tea. The samples were the ascorbic acid as its positive control and the Banaba milk-tisane. It was shown in the table that the Banaba Powdered Milk-tisane has an immediate scavenging activity while the ascorbic acid has a high scavenging activity. The basis to interpret the results of the IC50 values in scavenging activity are 10-50 microgram/ mL marked as a strong antioxidant activity, 50-100 microgram/mL as the immediate antioxidant activity, and >100 microgram/ml marked as a weak antioxidant activity (Jadid, Hidayati, Hartanti, Arraniry, Rachman, and Wikanta, 2016).

Table 6. DPPH Radical Scavenging IC50 values of Banaba milk-tisane and ascorbic acid

Sample	IC <sub>50</sub> (mcg/ml)
Ascorbic Acid	5.79
Banaba Milk Tea	69.88

The graph shows a line graph of percentage of DPPH activity between the Banaba powdered milk-tisane sample and vitamin c (ascorbic acid). It was shown in the line graph that the Banaba powdered milk-tea has a low antioxidant activity while the vitamin c or also known as ascorbic acid has a high antioxidant activity. One of the factors why the Banaba powdered milk-tisane has a low antioxidant activity, because studies have found out that milk and sugars can contribute a multiple effect in the antioxidant capacity of the tea. (Wipatanawin, Phongsawanit, Maneeratprasert, Lertsiri and Deetae, 2014).

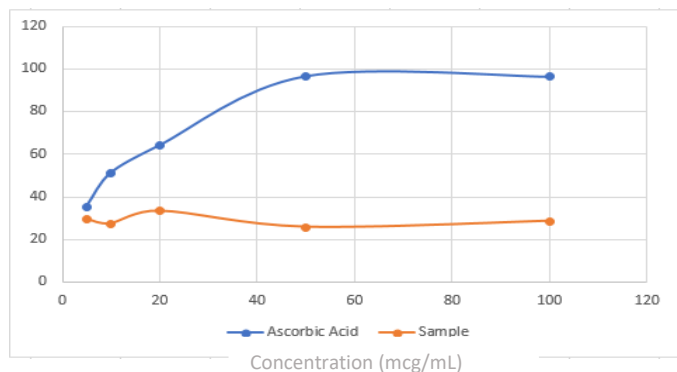


Figure 3. Line graph of Banaba milk-tisane and ascorbic acid in Scavenging Activity

## CONCLUSION

In conclusion of the research for the Banaba powdered milk-tisane, its sensory traits have shown no significance in its appearance, aroma, and color but with significance in its taste, flavor, and over acceptability to the commercial milk-tea. Given with the results of mean shown that the appearance, color, taste, flavor, and the over-all acceptability of the Banaba milk-tisane was higher compared to the commercial milk-tea except for its aroma. And for the antioxidant test, it was shown in results that the Banaba milk-tisane has an immediate antioxidant activity which can contribute the source of its antioxidant activity.

## RECOMMENDATION

The research has been limited on its test due to the availability of raw materials, time, and availability of test methods. For further studies, the researchers recommend on the following for best results of the product; the use of more efficient and suitable microencapsulating material such as the use of alginate, whey, and inulin for better support and improve its properties. Study on different air temperature and value of maltodextrin prior to spray drying to be able to know the yields in different values of temperature and its carrier material. Further examine on the differences of antioxidant specifically the powdered Banaba-tisane itself to be able to compare the activity to the powdered Banaba milk-tisane. Consider the dietary fiber and its glucosidic index and compare two samples; powdered Banaba-tisane and powdered Banaba milk-tisane to know whether it significantly differ from the Banaba tea itself. Storage, packaging of the raw materials must be provided and the shelf-life of the formulated milk-tisane.

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