Effect of the fermentation process using a consortium of probiotic bacteria on the taste of Arabica coffee (Coffea arabica)

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Abstract. The research about the fermentation of coffee Coffea arabica using a consortium of probiotic bacteria has been carried out. This study aims to determine the flavor of arabica coffee C. arabica and its chemical composition after the fermentation process. The fermentation time was divided into 3 times, consists of 24 hours, 36 hours, and 48 hours using a container measuring 10 L, each containing 1.5 kg of coffee and 75 mL of rejuvenated probiotic bacteria cultures. Organoleptic testing was carried out to see the panelists preference for the taste of coffee after fermentation. The results showed that coffee with a fermentation time of 48 hours had the best taste. The GC-MS test results showed several compounds were detected after the fermentation process including furan, phenol, propanoate acid, quinic acid, purine, palmitic acid, pyrol, ascorbic acid, linoleic acid, stearic acid, oleic acid, amines, piran, purines, aldehydes, vitamin E, benzadrex, hexene, tocophenols and arachidic acid.

1. Introduction
Coffee is a drink with a distinctive taste, influenced by the acidity of the coffee, the delicious aroma of coffee and its delicious taste. Quality coffee beans are coffee beans that have a maximum moisture content of 12%, do not smell bad, and are not overgrown with mold [1,2].

Coffee has its own appeal from various groups, so that an increase in the added value of the coffee commodity is needed. The application of technology that can improve the quality of coffee can be applied in various coffee processing processes, from the plantation stage to processing to storage. At the processing stage, the fermentation process by utilizing microorganisms has now been studied by various researchers. The presence of microorganisms during fermentation will affect the final result such as the taste of coffee [3].

The increase in coffee quality during the fermentation process is due to microbial activity in the coffee pulp during fermentation which produces alcohol and organic acids so that it provides aroma and taste for quality coffee production [4]. Arabica coffee fermentation in this study used probiotic bacteria from chickens.

In various studies, probiotic bacteria have been shown to be able to help the digestion of food in animals, including humans. Probiotic bacteria can be found in large numbers along the intestinal organs of the tenue and caecum. Probiotic bacteria are growing significantly, and this is seen in various studies that prove that probiotic bacteria have benefits for human health [5,6].
This study investigated the fermentation process of Arabica coffee beans (C. arabica) by probiotic bacteria with different incubation times of probiotic cultures to determine the incubation period for the most effective probiotic starter in improving the taste of coffee.

2. Methods

2.1. Arabica coffee fermentation (C. arabica)
The fermentation process was carried out by inserting 1.5 kg of coffee which was still in the form of cherries into the fermentation container, then 75 mL of consortium bacteria were added in each container to improve the quality of the coffee. Fermentation lasted for 24 hours, 36 hours and 48 hours by anaerobic method.

2.2. The taste of arabica coffee (C. arabica) after fermentation
The taste of Arabica coffee was evaluated using the QDA (Quantitative Descriptive Analysis) method. QDA is a method based on the level of preference of the panellists to the sample presented by the number of panellists of 20 people. This test is used to measure the level of consumer acceptance of the aroma of Arabica coffee on a scale of 1 to 5. The taste test was carried out at Kedai Kangen Coffee with the cup test method. A total of 10 grams of ground coffee was brewed with 180 ml boiling water.

2.3. Analysis of chemical compounds of Arabica coffee (C. arabica) after fermentation using Gas Chromatography-Mass Spectrometry (GC-MS)
Analysis of organic acid content using the Gass Chromatography-Mass Spectrometry (GCMS) method. The GC-MS test was carried out by means of a 0.5 ml sample pipette and put it in a 50 mL measuring flask and diluted with acetone. Pipette as much as 3 mL and put it in the vial. Instrument conditions GC-MS injector temperature 250°C with the splitless method, pressure 76.9 kPa and flow rate of 14mL/min and a ratio of 1:10. Ion source and interface temperatures 200°C and 280°C, solvent cut time of 3 minutes, 400-700 m/z. Column type SH-Rxi-5Sil MS column length 30 m with inner diameter 0.25 mm. The initial temperature of the column was 70 °C with a holding time of 2 minutes and the temperature was increased to 200 °C at a rate of 10°C/min and the final temperature is 280°C with a holding time of 9 minutes at a rate of 5°C/min so that the total analysis time was 36 minutes. The chromatogram data obtained were read using the NIST and Wiley 9 libraries.

The data from the observations were processed with ANOVA (Analysis of Variance) analysis using SPSS 21 software. If the results of the analysis of variance showed a real effect, a real difference test was carried out using the Duncan test.

3. Results and discussion

3.1. The taste of Arabica coffee (C. arabica) after fermentation
The organoleptic test was carried out in order to determine the level of panelist acceptance of the products produced. This test used the QDA method based on the panelists’ preference for the taste produced in each treatment.
Figure 1. The taste of Arabica coffee.

Table 1. The average of Arabica taste.

| Treatment          | Average   |
|--------------------|-----------|
| 24-hour control    | 3.400<sub>ab</sub> |
| 24-hour consortium | 3.325<sub>ab</sub> |
| 36-hour control    | 2.900<sup>a</sup> |
| 36-hour consortium | 3.100<sup>a</sup> |
| 48-hour control    | 3.350<sub>ab</sub> |
| 48-hour consortium | 3.900<sup>b</sup> |

The parameter for the highest yield taste test was the 48-hour consortium, with an average of 3,900. While the lowest was control for 36 hours, namely 2,900. Based on the ANOVA test results showed that the fermentation treatment of probiotic bacteria had a significant effect (P <0.05) on the taste of Arabica coffee (<i>C. arabica</i>) which was produced to determine the differences in each treatment followed by the Duncan test which showed that the control treatment 36 hours were not significantly different from the 36 hours consortium, 24 hours consortium and 48 hours control but significantly different from the 48 hours consortium. Meanwhile, the 48-hour consortium was not significantly different from the 24-hour and 48-hour consortium and the 24-hour consortium, but significantly different from the 36-hour consortium and 36-hour controls.

The longer the fermentation process, the better the taste will be. Taste is one of the factors that influence a person's acceptance of food. In general, taste can be divided into salty, sweet, bitter and sour taste, taste is a parameter that determines the quality of a product, whether the product is acceptable to consumers or not.

Fermentation not only serves to remove the mucus layer but also functions to improve the taste caused by the breakdown of carbohydrate compounds into organic acids. In the fermentation process there are chemical events that greatly affect the taste of the roasting process, the initial chemical composition of coffee, moisture content and roasting. However, the Maillard reaction plays an important role in the formation of the main compounds in coffee flavor [7].
3.2. Analysis of chemical compounds of Arabica coffee (C. arabica) after fermentation by the Gas Chromatography-Mass Spectrometry (GC-MS) method

Coffee that has been roasted and mashed is then extracted using the maceration method with 90% ethanol. The extract was then evaporated at 45°C. The evaporation results were then analyzed for their organic acid content using the GC-MS method. The chromatogram data obtained were then read using the NIST and Wiley 9 libraries. Based on the GC-MS test, the compounds in Arabica coffee were obtained, namely:

Table 2. 24-hour Gas Chromatography-Mass Spectrometry (GC-MS) Compound test results.

| No | Compounds  | The number of gram |
|----|------------|--------------------|
| 1  | Furan      | 1.28               |
| 2  | Phenol     | 0.65               |
| 3  | Quinic acid| 30.95              |
| 4  | Purine     | 21.35              |
| 5  | Palmitic acid | 4.07           |
| 6  | Pyrol      | 0.32               |
| 7  | Ascorbic acid | 10.62            |
| 8  | Linoleic acid | 6.16             |
| 9  | Stearic acid | 1.79            |
| 10 | Oleic acid | 1.3                |
| 11 | Piran      | 1.02               |
| 12 | Caffeine   | 0.15               |
| 13 | Aldehyde   | 1.08               |
| 14 | Benzedrex  | 0.1                |
| 15 | Heksena    | 5.21               |
| 16 | Tocopherol | 0.81               |

Table 3. 36-hour Gas Chromatography-Mass Spectrometry (GC-MS) Compound Test Results

| No | Compounds  | The number of gram |
|----|------------|--------------------|
| 1  | Furan      | 5.92               |
| 2  | Phenol     | 0.67               |
| 3  | Propanoic acid | 0.86            |
| 4  | Quinic acid| 6.8                |
| 5  | Purine     | 45.05              |
| 6  | Palmitic acid | 5.68            |
| 7  | Pyrol      | 0.38               |
| 8  | Ascorbic acid | 7.65             |
| 9  | Linoleic acid | 5.69            |
| 10 | Stearic acid | 8.64            |
| 11 | Oleic acid | 1.72               |
| 12 | Piran      | 0.54               |
| 13 | Caffeine   | 0.26               |
| 14 | Arachidic acid | 0.32            |
Table 4. 48-hour Gas Chromatography-Mass Spectrometry (GC-MS) Compound Test Results

| No | Compounds            | The number of gram |
|----|----------------------|--------------------|
|    |                      | 48 hours Consortium |
| 1  | Furan                | 9.44               |
| 2  | Phenol               | 0.70               |
| 3  | Propanoic acid       | 0.11               |
| 4  | Quinic acid          | 8.95               |
| 5  | Purine               | 46.37              |
| 6  | Palmitic acid        | 1.30               |
| 7  | Pyrol                | 0.35               |
| 8  | Ascorbic acid        | 9.44               |
| 9  | Linoleic acid        | 4.81               |
| 10 | Stearic acid         | 2.04               |
| 11 | Oleic acid           | 1.25               |
| 12 | Piran                | 1.35               |

The compounds formed in the sample were the influence of the probiotic bacteria consortium. The compounds formed were furan, phenol, propanoic acid, quinic acid, purine, palmitic acid, pyrol, ascorbic acid, linoleic acid, stearic acid, oleic acid, amines, piran, caffeine, aldehyde, vitamin E, benzodrex, hexene, tocophenols and arachidic acid. Some of the compounds from roasting at roasting temperature are formed from maillard reactions and other chemical transformations that play a central role in the formation of coffee flavors and several aroma compounds produced during coffee roasting include furan, piran and pyrol [8,9].

Since it was first identified by the German chemist Bernheimer in 1880, about 1000 volatile compounds have been identified in roasted coffee. Among the volatile compounds, only a few compounds are considered as compounds that affect the aroma and from one type of coffee contains a different type and taste. The composition and concentration of compounds have an effect on taste depending on the roasting level of coffee [10,11].

3.3 24-hour Gas Chromatography-Mass Spectrometry (GC-MS) Compound Test Results

Based on the results of GC-MS analysis on fermented coffee for 24 hours (table 2), 16 compounds were obtained, namely quinic acid (30.95%), purine (21.35%), furan (12.82%), ascorbic acid (10.62%), linoleic acid (6.16%), hexene (5.21%), palmitic acid (4.07%), stearic acid (1.79%), oleic acid (1.3%), aldehyde (1.08%), piran (1.02%), tocophenol (0.81%), phenol (0.65%), pyrol (0.32%), caffeine (0.15%) and benzodrex (0.1%)

3.4 36-hour Gas Chromatography-Mass Spectrometry (GC-MS) Compound Test Results

Based on the results of GC-MS analysis on fermented coffee for 36 hours (table 3), 14 compounds were obtained, namely purine (45.05%), stearic acid (8.64%), ascorbic acid (7.65%), quinic acid (6.8%), furan (5.92), linoleic acid (5.69%), palmitic acid (5.68%), oleic acid (1.72%), propanoic acid (0.86%), phenol (0.67%), piran (0.54%), pyrol (0.38%), arachidic acid (0.32%) and caffeine (0.26%).

3.5 48-hour Gas Chromatography-Mass Spectrometry (GC-MS) Compound Test Results

A total of 12 compounds were detected from the GC-MS test results. The compounds that had the highest percentage at 48 hours of fermentation time were due to the influence of the consortium probiotic bacteria (table 4), namely purine (46.37%), furan (9.44%), ascorbic acid (9.44%), and quinic acid (8.95%), linoleic acid (4.81%), stearic acid (2.04%), piran (1.35%), palmitic acid (1.30%), oleic acid (1.25%), phenol (0.70%), pyrol (0.35%) and propanoic acid (0.11%).
The results of compound testing using Gas Chromatography-Mass Spectrometry (GC-MS) for 24 hours, 36 hours, and 48 hours obtained acid compounds that dominate the components of the compounds in the coffee sample. Acidic compounds contribute to the acidity of Arabica coffee, acidic compounds are still able to survive even in the roasting process of Arabica coffee beans (C. arabica) [12].

The differences in the compounds found in the three samples were due to the addition of consortium bacteria from the coffee roasting process. The fermentation process forms compounds, so that the addition of a consortium of probiotic bacteria and the length of fermentation time causes a difference in the number of compounds formed. The probiotic bacteria consortium causes changes in the composition of the compound Arabica coffee (C. arabica). The longer the fermentation takes, the acidity of the coffee will increase, this is due to the formation of aliphatic acids during the fermentation process. If the length of fermentation is extended, there will continue to be changes in the chemical composition of the coffee beans, where the aliphatic acids will turn into carboxylic acid esters which can cause fermentation defects with a bad taste [13].

From all the acids formed create a unique and pleasant taste as opposed to a sour taste. So that the acid formed can last until the brewing process because it has a central role in taste. Furan is a compound that provides the most aromatic side to coffee which affects the caramel aroma of the degradation products of carbohydrates, fatty acids and ascorbic acid during coffee roasting and has a high sensory threshold. The phenolic content in coffee creates a whiskey-like aroma. Pyrol compounds are formed from the degradation of amino acids which have an aroma like roasted peanuts. Piran is formed from amino acids and contributes to the aroma of roasted nuts and chocolate (Asmak et al 2018, Grosch, 1998). Acid compounds formed in this study such as linoleic acid, stearic acid, palmitic acid, and ascorbic acid caused a sour taste in ground coffee samples. Compounds such as furan and pyrazine also affect the acidity of coffee [1].

4. Conclusion

Based on the results of probiotic bacteria in the fermentation of Arabica coffee (C. arabica) with 24 hours, 36 hours and 48 hours treatment obtained significant results on the taste of Arabica coffee (C. arabica). The 48 hour fermentation time produces the best coffee taste. From the results of GC-MS analysis, various compounds that contribute to the taste of coffee are obtained, namely acids, furans, phenols, pyrol and pyrazines.

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