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PENGARUH PENAMBAHAN EKSTRAK KAYU SECANG (*Caesalpinia sappan L.*) TERHADAP PERTUMBUHAN KULTUR, KUALITAS ORGANOLEPTIK DAN AKTIVITAS ANTIOKSIDAN *YOGHURT DRINK*

The Effect of Addition With The Wood Extract of Secang (Caesalpinia sappan L.) on Culture Growth, Organoleptic Quality, and Antioxidant Activity of Drink Yoghurt

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ABSTRACT

The objective of research was to find the effect of addition wood extract of secang (*Caesalpinia sappan L.*) on culture growth, organoleptical quality, and antioxidant activity of yoghurt drink. Materials of research include yoghurt plain. Method of research was laboratory experiment using a design of Complete Random Planning involving four treatments and four replications. Treatments of this research consist of: P0 (Yoghurt drink without addition of secang wood extract), P1 (Yoghurt drink with addition of 10% secang wood extract), P2 (Yoghurt drink with addition of 20% secang wood extract), and P3 (Yoghurt Drink with addition of 30% secang wood extract). The obtained data are analyzed with Analysis of Variance (ANOVA). If different results are obtained, then the analysis is continued with Duncan Multiple Interval Test. It was concluded that higher concentration of secang wood extract showed the wider preventive zone as inhibitation bacterial growth. This finding was supported by the result of diffusion test using disc paper. In the other hand, wider preventive zone is related with the increasing level of antioxidant activity, pH value, and water content of yoghurt drink, although the total level of acidity and viscosity is declining. The increasing concentration of secang wood extract does not deliver obvious difference ($P>0.05$) in its effect on the total level of lactate acid bacteria (LAB). Secang concentration at 30% does not influence taste and aroma of yoghurt drink but it gives orange color on yoghurt drink.

Keywords : yoghurt drink, antimicrobial activity, number LAB by TPC, organoleptical quality, antioxidant activity.

INTRODUCTION

Yoghurt is a food material derived from cattle milk. It is a result of milk fermentation by which the milk would change into a form like porridge or ice cream with little acid taste caused by the presence of fermentation-inducing bacteria. Few lactate acid bacteria are utilized in this fermentation such as: *Lactobacillus bulgaricus* and *Streptococcus thermophilus*. The latter, *Streptococcus thermophilus*, proliferates faster, and produces acid and CO_2. The method of yoghurt production has evolved in centuries. The naturally contaminated milk becomes sour at high temperature, precisely around 40-50^{0}C. The acid taste and quite viscous texture are too unpleasant for consumers. Therefore, it needs product diversification, and it is done by producing yoghurt with less acid

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taste (by adding natural essence) and less viscosity (diluted) to facilitate consumption. This product is called yoghurt drink. Diversification of yoghurt drink is made by adding secang wood extract into yoghurt preparation. Secang is selected because secang wood contains natural red color agent, antioxidant source, and antimicrobial substance. Yoghurt drink is expected to consume as *functional food*.

Secang (*Caesalpinia sappan L.*) is a plant living on the land in the altitude of 1,000 meters above the sea. The wood part of secang, after boiled, can produce ivory pink color which is considered as attractive coloring agent for paints and plaits, and also useful to be the ornamental color for cake and drink, and the content of ink (Arisandi and Andriani, 2008). Antioxidant substance in the secang wood shows a sign that this natural substance is quite potential to produce antioxidant activity. Stem or wood parts of secang have contained with tanin, galate acid, resin, brasilein, dalpha-phelandrene, oscimene, volatile oil, reorsin, and brazilin. Basilin/brazilin is a compound with red color agent at secang wood. Brazilin is also an antioxidant substance that its chemical structure contains cathecol. The quantity of cathecol has increased from 32,340 tons in 2015 to 33,052 tons in 2016. Hariana (2006) adds chemical substances of secang wood into the study because these are considered as having antimicrobial activity. These substances are brazilin (estimated as having antibacterial activity against *Staphylococcus aureus* and *Escherichia coli*), tanin, polyphenol groups, and galate acid.

The objective of research is to understand the effect of secang wood extract on yoghurt culture growth, LAB total, pH, organoleptical quality, viscosity, color, antioxidant activity, water content and acid total.

MATERIAL AND METHOD

Material used in this research is yoghurt plain that is processed into yoghurt drink by adding secang wood extract. The milk is obtained from Koperasi Mitra Bhakti Makmur "Junrejo", Junrejo District, Batu City. The quantity of yoghurt drink is 750 ml. Secang wood is extracted using aquades solvent. Secang wood itself is obtained from Blimbing Market, Malang City.

The analysis devices include: pH meter (Hanna HI2002-edge), Brookfield DVII viscometer, color reader (Minolta), incubator (INB-200 Memmert), UV-Vis spectrophotometer, centrifuge (Gemmy PLC-038 HOLE).

Method of research is experiment which is conducted at the laboratory using the design of Complete Random Planning (CRP) involving 4 treatments and 4 replications. Treatment by adding secang wood extract into yoghurt drink is performed by referring to preliminary study.

P_0 = Yoghurt drink without addition of secang wood extract.

P_1 = Yoghurt drink with addition of 10% secang wood extract.

P_2 = Yoghurt drink with addition of 20% secang wood extract.

P_3 = Yoghurt drink with addition of 30% secang wood extract.

Variable of Research

The variable of this research is plentiful and how to measure them is explained as following. Determine pH value principle is the method of ph based on measurements of ion hydrogen activity in potentiometri/electrometric using ph meters. The pH value is measured by pH meter (Hanna HI2002-edge) with the procedure suggested by Jannah et al., (2014).

Determine water content principle is a sample dried in the oven with 100°C until gained weight fixed. Water content has been observed using oven method by following AOAC procedure (1984).

Determine acid total principle is titration by the use of alkaline solutions as NAOH. Acid total is measured with titration method by referring to AOAC procedure (1984).

Determine viscosity value principle is the spindle was rotating quickly so the viscosity is dilute, if the spindle slower rotating so viscosity is viscous. Viscosity is estimated used (Brookfield DVII Viscometer) following the procedure of Widagdha and Fithri (2015).

Determine organoleptical quality is assessed by 5 trained panelists.

Determine colour reader principle is exposure color system by using CIE system with three color namely receptors L, a, b hunter. L indicate the level of brightness based on white color, a* shows reddish or greenish, and b* indicate yellowish or bluish. Color is made certain using color reader (Minolta) with the procedure of Widagdha and Fithri (2015).

Determine antimicrobial activity principle is the antimicrobial activity can be seen based on the culture inhibition of bacteria, the area around the establishment of a clear antimicrobial substance. But in testing activity antimicrobial extra wood secang on yogurt hopefully do not formed the clear. When formed the indicates that there are clear inhibition of the growth of culture as a starter yogurt additional extra secang the wood. Antimicrobial activity against culture growth of yoghurt is understood using disc paper with procedure suggested by Ruhimat (2015).

Determine number Lactic Acid Bacterial (LAB**)** is measured with a method called TPC (Total Plate Count) which involves implantation procedure involving *spread plate* (spreading method) with the procedure of Widodo et al., (2015).

Determine antioxidant activity principle is the free radicals stable which DPPH who get mixed with compound antioxidant with the ability to donate hydrogen, so free radicals can be stifled. Antioxidant activity is examined using DPPH (*Di-Phenyl-Picryl-Hydrazyl*) based on procedure of Widowati (2011).

Data Analysis

Data of research are subjected to statistic analysis. The analysis technique is Analysis of Variance (ANOVA). If the treatment shows obvious effect, the analysis continues with Duncan Multiple Interval Test (DMIT).

RESULT AND DISCUSSION

The Effect of Addition with The Wood Extract of Secang (*Caesalpinia sappan L.*) on Chemical and Physical of Yoghurt Drink

The analysis of variance on treatment with secang wood extract at various levels of concentration, including 0%, 10%, 20% and 30%, has given some results. Each concentration has a very obvious different effect (P<0.01) on water content . However, each concentration does not have obvious different effect (P>0.05) on pH, acid total, and viscosity. The mean score of pH, water content, acid total and viscosity in yoghurt drink can be seen in Table 1.

Table 1.The mean score of pH, water content, acid total and viscosity in yoghurt drink

Treatment	pH	Water Content (%)	Acid Total (%)	Viscosity (cP)
P0	3.74±0.01	83.63±0.24[a]	0.60±0.03	64.00±3.27
P1	3.75±0.03	86.86±0.43[b]	0.58±0.02	58.25±3.30
P2	3.76±0.01	86.98±0.25[b]	0.55±0.04	58.25±1.71
P3	3.78±0.02	87.28±0.27[b]	0.54±0.03	52.00±1.41

Note : Different superscripts in the same column are showing very obvious difference (P<0.01).

pH Value of Yoghurt Drink

Result of research indicates that the mean score of pH value of yoghurt drink with addition of secang wood extract is ranging from 3.74 to 3.78. Adding secang wood extract at higher level would increase pH value of yoghurt drink. It is evident because secang wood extract has pH 6.32, and therefore, its addition into yoghurt drink would increase pH.

According to SNI, pH value of yoghurt drink should be in the range 4-4.5. However, yoghurt drink's pH value is lower than SNI, precisely at 3.7. Widodo (2002) mentions that pH value that should be achieved by yoghurt is 4.5. The low pH value in yoghurt drink is caused by three kinds of *strater*, including *Streptococcus thermophilus*, *Lactobacillus bulgaricus*, and *Lactobacillus acidophilus*. It is consistent with Jannah et.al., (2014) who state that probiotic bacteria, mainly *L. acidophilus*, when combined with other, such as *S. thermophilus* and *L. bulgaricus*, can produce yoghurt at lower pH because it can ferment sugar into lactate acid in faster ways. This finding is supported by Yildiz (2010) who states that yoghurt prepared with *L. achidophilus* can increase acidity faster and produce pH value below 4. Shah (2000) adds that the growth of *L. achidophilus* stops at pH below 4.0. Chairunnisa (2009) indicates that during fermentation, the growth of *Streptococcus thermophilus* is faster than *Lactobacillus bulgaricus* and *Lactobacillus acidophilus*.

Water Content of Yoghurt Drink

Result of variance analysis in Table 1 shows that the addition of secang wood extract at different percentages of concentration into yoghurt drink indicates very obvious different effect (P>0.01) on water content of yoghurt drink. It is estimated so because the solvents used to prepare secang wood extract are liquid. At fresh mik, water presents, and thus, less surprisingly, yoghurt drink water content increases after the addition of secang wood extract. As said by Mirdhayati et.al., (2008), water content of the milk is very high, counted for 87.25% in average.

The principle of water content determination in dry method is by drying out or by evaporizing water in a material (sample) in such way that it leaves out the solid material freed of water. The determination of water content is indeed necessary because it can affect material storability. Higher water content of a material may accelerate decomposition. Water content is highly affected by storage method or harvest length until the material is processed into a product. Storability of a material can be prolonged by removing part of waters in the material until it reaches water content at certain level.

Acid Total of Yoghurt Drink

Variance analysis in Table 1 indicates that the addition with the wood extract of secang (*Caesalpinia sappan L.*) at different percentages of concentration into yoghurt drink does not have very obvious different effect (P>0.05) on acid total of yoghurt drink. It is caused by a fact that pH value of yoghurt drink increases with the addition of secang wood extract. Lactate acid is a product of lactate acid bacteria during sugar fermentation activity. Therefore, lactate acid rate in the yoghurt drink is affected by total level of yoghurt probiotic bacteria. The greater is the quantity of probiotic bacteria, the more results of metabolism of lactate acid can be obtained. The increase of lactate acid rate in milk fermentation is always perpendicularly related with the reduction of pH value of yoghurt. It means that the higher level of lactate acid produced during fermentation is always followed by the reduction of yoghurt pH value. This reduction of pH value induces acid taste (Kusumaningrum, 2011).

Acid total value in the yoghurt drink product ranges between 0.54%-0.595%. Acid total of yoghurt drink is still in compliance with SNI (2009), precisely at range 0.5-2.0%. According to Askar and Sugiarto (2005), the acid in yoghurt is the main product that characterizes the taste of yoghurt. This acid is derived from milk carbohydrate (lactose) fermented by the bacteria. The resultant acid is called lactate acid. Bacteria utilize lactose as their source of energy and carbon for growth. Acid total value obtained from yoghurt product is ranged between 0.73%-1.92%. The good yoghurt contains acidity 0.85-0.95% of acid total.

Viscosity Rate of Yoghurt Drink

Result of variance analysis as shown in Table 1 indicates that the addition with the wood extract of secang (*Caesalpinia sappan L.*) at different concentration into yoghurt drink does not show obvious different effect (P>0.05) on viscosity rate. Indeed, viscosity rate tends to decrease with the rate in control treatment is the biggest. It happens because viscosity rate has close relationship with pH value, although the trend of pH value is increasing. It aligns with Sutedjo and Nisa (2015) who state that the lower pH value is followed by higher viscosity rate. It is said so because at lower pH value, isoelectric point is activated, and it is a place where protein of material will coagulate and it must increase yoghurt viscosity. It is supported by Kartikasari and Nisa (2014) who assert that the reduction of pH value in yoghurt may disturb the balance of casein in milk protein and force pH value to reach its isoelectric point at 4.6 where casein indeed coagulates and forms a coagulum which represents a semi-solid structure. When pH value stays below isoelectric pH value, then casein bond increases. However, the excess of casein will shrink protein that forces protein to release water which in turn reduces gel strength. The reduction of gel strength can decrease texture quality (Manab, 2008).

Table 2. The Mean Score of Organoleptical Quality and Color in Yoghurt Drink

Treatment	Average ± Deviation Standard				
	Organoleptical Quality		Color		
	Aroma	Taste	L	a*	b*
P0	3.25±0.44	3.20±0.41	61.85±0.59^{a}	9.38±0.10^{a}	9.10±0.18^{a}
P1	3.6±0.82	3.95±0.94	59.98±0.39^{b}	9.88±0.10^{b}	19.25±0.64^{ab}
P2	3.3±1.17	3.25±0.97	59.58±0.66^{b}	10.83±0.26^{bc}	23.25±0.51^{b}
P3	3.85±1.31	3.50±1.47	57.50±0.50^{c}	11.00±0.27^{c}	24.90±0.34^{b}

Note : Different superscripts in the same column and the same row are showing very obvious effect (P<0.01).

Organoleptical Quality of Yoghurt Drink (Aroma and Taste)

Result of variance analysis in Table 2 indicates that the addition of the wood extract of secang (Caesalpinia sappan L.) at different percentages of concentration into yoghurt drink does not have very obvious different effect (P>0.05) on aroma. It is found so because the unique aroma of yoghurt is more dominant and can disguise the aroma of secang. The unique flavor of acid in yoghurt emerges because lactose is converted into lactate acid by lactate acid bacteria during fermentation. It is supported by Winarno and Fernandez (2007) who state that during fermentation of milk, lactate acid bacteria will ferment almost all milk lactose contents into lactate acid, and give a distinctive aroma to yoghurt through diacetyl and acetyldehyde. This unique aroma is established by the presence of acetyldehyde, diacetyl, acetate acid, and other acid groups that are produced during fermentation by lactate acid bacteria such as *Lactobacillus bulgaricus*, *Streptococcus thermophillus*, *Bifidobacterium*, and *Lactobacillus acidophillus* (Triyono, 2010).

Result of variance analysis in Table 2 indicates that the addition of the wood extract of secang (Caesalpinia sappan L.) at different percentages of concentration into yoghurt drink does not have very obvious different effect (P>0.05) on taste. Yoghurt drink's taste is mostly affected by the activity of lactate acid bacteria during fermentation process and the dominant taste is acid. It is consistent with Winarno and Fernandez (2007) who explain that the image of uniqe acid in yoghurt drink is produced by several compounds such as lactate acid, dehyde acetate, acetate acid, and other easily vaporized substances. The growth of Streptococcus thermophillus may produce few acid substances in the milk, and then, *Lactobacillus bulgaricus* would give more powerfulacid taste. According to Hidayat et.al., (2006), *Lactobacillus bulgaricus* plays more significant role in establishing aroma, while Streptococcus thermophillus has more contributing role to the establishment of yoghurt taste.

Color (L, a*, and b*) of Yoghurt Drink

Result of variance analysis in Table 2 indicates that the addition of the wood extract of secang (*Caesalpinia sappan L.*) in different percentages of concentration into yoghurt drink has produced very obvious different effect (P<0.01) on brightness rate. Data of research show that the mean score of brightness rate (L) of yoghurt drink with the addition of the wood extract of secang (*Caesalpinia sappan L.*) is ranged between 61.85 and 57.5. The highest brightness rate (L) is shown by control treatment without the addition of the wood extract of secang (*Caesalpinia sappan L.*). It happens because brazilin compound in the wood extract of secang (*Caesalpinia sappan L.*) experiences degradation and causes the reduction of intensity of brightness. Factors affecting color stability include pH value, temperature, and oxygen (Miksusanti et.al., 2012).

Result of variance analysis in Table 2 has also shown that the addition of the wood extract of secang (*Caesalpinia sappan L.*) at different percentages of concentration into yoghurt drink has very obvious different effect (P<0.01) on reddish color (a*). It is evident because the higher concentration of the wood extract of secang (*Caesalpinia sappan L.*) in yoghurt culture will increase the quantity of brazilin compound that has red color character. The wood extract of secang (*Caesalpinia sappan L.*) contains brazilin compound and the prominent nature of this compound is its red color (Farhana et.al., 2015). The color produced by the wood extract of secang (*Caesalpinia sappan L.*) is still determined by pH value and the received heat condition. At pH value of 2-5, brazilin compound is yellowish, while at pH value of 6-7, it becomes red but at pH value above 8, it is purplish red (Umami and Diana, 2015).

Result of variance analysis in Table 2 also reveals a fact that the addition of the wood extract of secang (*Caesalpinia sappan L.*) at different percentages of concentration into yoghurt drink has very obvious different effect (P<0.01) on yellowish color (b*). The red-colored brazilin compound from the wood extract of secang (*Caesalpinia sappan L.*) is tarnishing after added into yoghurt drink. Yoghurt is a processing product with acid pH, and therefore, when the wood extract of secang (*Caesalpinia sappan L.*) is added into it, yoghurt will have a color of yellowish white or orange (Umami and Diana, 2015). This result is consistent with Riska (2008) who explains that the color of acidity in brazilin compound is always yellow. The intensity of red color from the wood extract of secang (*Caesalpinia sappan L.*) has very small effect or has no effect at all. Yellow color has higher intensity, especially to yoghurt product stored at 5^0C.

The Effect of Addition with The Wood Extract of Secang (*Caesalpinia sappan L.*) on Microbial Quality and Antioxidant Activity of Yoghurt Drink

The analysis of variance on treatment with secang wood extract at various levels of concentration, including 0%, 10%, 20% and 30%, has given some results. Each concentration has a very obvious different effect (P<0.01) on antimicrobial and antioxidant activity in yoghurt drink.. However, each concentration does not have obvious different effect (P>0.05) on number Lactic Acid Bacterial (LAB) in yoghurt drink. The mean score of antimicrobial activity, number Lactic Acid Bacterial (LAB) and antioxidant activity in yoghurt drink can be seen in Table 3.

Table 3.The mean score of antimicrobial activity, number Lactic Acid Bacterial (LAB) and antioxidant activity in yoghurt drink

Treatment	Preventive Zone Diameter		LAB Total (Log CFU/ml)	Antioxidant Activity (ppm)
	(*Streptococcus thermophilus*) (mm)	(*Lactobacillus bulgaricus*) (mm)		
P0	0.00±0.00[a]	0.00±0.00[a]	7.60±0.75	131.77±4.97[b]
P1	0.37±0.03[b]	0.30±0.03[b]	7.65±0.13	72.45±2.03[ab]
P2	0.40±0.02[b]	0.33±0.03[b]	7.85±0.25	49.56±1.79[a]
P3	0.42±0.03[b]	0.33±0.03[b]	7.63±0.67	24.15±2.42[a]

Note : Different superscripts in the same column and the same row are showing very obvious effect (P<0.01).

Culture Growth (Antimicrobial Activity) of Yoghurt Drink

Result of research shows that in average, preventive zone on *Streptococcus thermophilus* is ranged at 0.00-0.37 mm whereas against *Lactobacillus bulgaricus*, it ranges about 0.00–0.33 mm. The larger preventive zone in the yoghurt drink culture is caused by *Streptococcus thermophilus* and *Lactobacillus bulgaricus*. Both bacteria are used in test, and both are genuine bacteria estimated that their growth is easily distracted when they have direct contact with antimicrobial compounds. These bacteria do not have a protective mechanism during the test. Yoghurt culture contains gram-positive bacteria which their growth is easily disturbed by the presence of antimicrobial compunds. The addition with secang wood extract at higher concentration should enlarge preventive zone.

Secang wood has antimicrobial compunds such as flavonoid, tanin, galate acid, dan phenol. The preparation of secang wood extract is using aquades at pH±7. This condition has activated antimicrobial activity in the compounds, including flavonoid, phenol and tanin. This finding is supported by Karlina et.al., (2016) at pH 7. Result of phytochemical filterization against secang wood extract has found substances of flavonoid and alkaloid as majority. Other minor chemical substances include tanin and phenol (Srinivasan et al., 2012).

Suerni et.al., (2013) explain that flavonoid is actually a compound of phenolic substances. It is disinfectant in nature and very effective to prevent the growth of gram-positive bacteria. It is said so because flavonoid has a polar character which facilitates flavonoid to penetrate gram-positive bacteria' peptidoglycan

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layers that are also polar. These layers are easily permeable than lipid layers that are non-polar. Moreover, cellular wall of gram-positive bacteria contains a polysaccharide (trichoate acid) which represents a water dissoluble polymer that is functional in transferring positive ion into or exit from cellular walls. This dissolution character means that the wall of gram-positive cells is always more polar. After flavonoid succesfully enters cellular wall, it works by destroying bacteria by denaturing bacterial proteins . The consequence is that cellular metabolism activity in the bacteria may stop because it has been catalyzed by protein-based enzyme.

Tanin has antioxidant activity. In general, the mechanism is that tanin toxicity can destroy bacterial cell membrane. Astringent compound in tanin can induce the establishment of adhesive compound complex that adhere to enzyme or microbial substrate (Suerni et.al., 2013).

On testing of antimicrobial activity against yoghurt culture, the resultant preventive zone is very small. It means that yoghurt culture still retains antimicrobial activity of the substances in secang wood extract. This finding is supported by Indu et.al., (2006). They assign preventive zone diameter into several groups, such as criteria of high (>16 mm), medium (12-16 mm), and no antibacterial activity (< 12 mm). Result of observation on antibacterial activity of secang wood against E. coli and S. typhi indicates that mineral water extract at pH 7 with concentration 10% does not have preventive zone, while concentration 20% has produced preventive zone of 8.07 and 9.4 mm in Aspergillus niger and Candida albicans (Karlina et.al., 2016).

Number Lactic Acid Bacterial (LAB) in Yoghurt Drink

The finding also shows that LAB total average in yoghurt drink is 7.60–7.85 log CFU/ml where LAB total has complied with SNI 2981:2009 which requires that minimum rate of LAB total in yoghurt must reach 107 CFU/ml. LAB total in this research, however, is still lower than the result obtained by Radiati et al. (2016) who have found that LAB total in yoghurt is 9.0±0.5 – 9.8±0.4 log CFU/g after the addition of fortified carrot juice. The explanation of this finding is that during fermentation process, S. thermophilus and L. bulgaricus may use sugar in the carrot as its nutrient source.

In the preparation of yoghurt drink, the addition of sugar, water and secang wood extract is given after incubation process. If the addition is made before incubation, it may disturb LAB growth During incubation, LAB uses only milk for its growth media. Milk contains lactose that would be the source of nutrient for growth and establishment. Kumalasari et.al., (2012) assert that the addition of simple sugar into coconut water cannot yet stimulate LAB growth. It is possible that LAB only utilizes milk-based sugar, or LAB may work maximally in milk media. This finding is supported by Sunarlim (2009) who states that the most important component during LAB fermentation is lactose. Lactose is used by LAB as the source of carbon and energy. The result of metabolism is lactate acid that may cause pH value of milk to decline. LAB cells can grow and cleave exponentially until its maximum level. Such proliferation is affected by environmental condition and nutrient stock in the media (Kumalasari et.al., 2012).

At P3, the addition of secang wood extract at concentration 30% into yoghurt culture can reduce LAB into 6.3 log CFU/ml. Total reduction of LAB is obvious because LAB is assumed as quite vulnerable to antimicrobial activity. Higher concentration of secang wood extract added means the higher also antimicrobial compounds in the yoghurt culture. Indeed, at the addition of 30% concentration, as shown in P3, the quantity of “died” LAB is quite great. Despite its reduction, the quantity still meets the condition of SNI

2981:2009 that sets the minimum rate of LAB total at 107 CFU/ml. This explanation is supported by Lindawati et.al., (2014) who state that LAB total at yoghurt preparation by addition of coconut water is 1.1×107 CFU/g.

Antioxidant Activity of Yoghurt Drink

Result of variance analysis indicates that the addition of secang wood extract at different concentration has contributed to very obvious difference (P<0.01). The highest value of antioxidant activity IC¬¬50 is found at P3 counted for 24.15±2.42. The lowest antioxidant activity rate is observed at P0 with 131.77±4.97. There is a fact that antioxidant activity rate is inversely related with the reading of IC50 value. Therefore, the highest antioxidant activity is obtained at the lowest IC50 value. Indeed, IC50 value is a parameter that determines the effective antioxidant activity that prevents 50% of the activity of a free radical, namely DPPH (1,1 Diphenyl-2-picrylhidrazyl).

The mean value of IC50 in the antioxidant activity at each treatment, in serial order, is 131.77 ppm, 72.45 ppm, 49.56 ppm, and 24.15 ppm. Sutedjo and Nisa (2015) admit that yoghurt has natural antioxidant content in the form of vitamin C or ascorbate acid. Result of data analysis has shown that the addition of secang wood extract can increase antioxidant activity rate in the yoghurt drink. Vitamin C is quite useful for growth, collagen synthesis, iron substance absorption, and also remains important being antioxidant (Radiati et.al., 2007). It is conceded so because as suggested by Widowati (2011), secang wood extract has a strong antioxidant activity, and it is assumed that this strength is related with very high phenol content and high flavonoid content during phytochemical test result. Phenol compound in plant has many biological activities, including also antioxidant activity. There is a positive relationship betwen antioxidant activity and phenol total rate. Antioxidant activity of phenol compound is quite apparent because it has higher capacity to embrace metals. This compound also has the clusters of hydroxil (OH) and carboxyl (COOH) that can bind heavy metals of Fe and Cu. There is a high correlation between phenol total and antioxidant activity, showing that phenol total plays greater role in contributing antioxidant activity from various vegetables. It agrees with Redha (2010) who asserts that antioxidant activity of flavonoid is evident in the cereal, vegetables, and fruits. Flavonoid is an antioxidant that works by donating hydrogen atom or by its capacity of bracing the metals. Flavonoid stays in the form of glucoside (containing side chain of glucose) and in a free form, called aglicon.

The analysis test on antioxidant activity is using IC50 value and DPPH. Method of DPPH (1,1 Diphenyl-2-picrylhidrazyl) is one method to determine antioxidant activity as free radical scavenger. Method of DPPH will provide information about reactivity of the compound by testing the compound with a stable free radical. This method works well when solvents of methanol or ethanol are involved. Both solvents do not influence reaction between tested sample as antioxidant and DPPH as free radical (Molyneux, 2004). DPPH provides strong absorption to wavelength 517 nm in dark violet color. Free radical scavenger causes electrons to match to each other which then reduces the color in comparison to the number of electrons taken (Sayuti and Yenrina, 2015).

CONCLUSION

The conclusion is disseminated as following. The higher concentration of secang wood extract will produce the wider preventive zone. It is confirmed by the result of diffusion test using disc paper. The wider preventive zone is related with the increasing level of antioxidant activity, pH value, and water content of yoghurt drink, but the total level of acidity and viscosity decreases. The increasing concentration of secang wood extract does not give obvious difference (P>0.05) in its effect on the total level of lactate acid bacteria (LAB). Secang concentration at 30% does not influence taste and aroma of yoghurt drink but it gives orange color on yoghurt drink.

SUGGESTION

The acceptable maximum concentration rate of secang wood extract added into yoghurt drink is 20%. In this concentration, LAB is highest, and the resultant yoghurt drink meets the condition of SNI. The effect of this concentration on pH value and preventive zone diameter is still in tolerance. Further research is needed to conduct hedonic scale test to understand the acceptance of yoghurt drink color after the addition of the wood extract of secang (Caesalpinia sappan L.). Next research may also examine the quality of chemical substances of yoghurt drink after the addition of secang wood extract. These potential researches above at least help the author to acknowledge which yoghurt drink that is the most acceptable to the consumer.

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