Milk quality improvement through the application of plantaricin IIA-1A5 as biopreservatives on fresh milk in dairy cow farm

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Abstract. Foods are better being consumed raw and fresh, however, fresh milk has different case. In fact, drinking fresh milk still became a debate until this day. Some claims said unprocessed fresh milk contains more nutrition compared with pasteurized milk, although milking process do not guarantee protection from bacteria and dirt contamination. One of example bacteria which can cause damage on dairy products are \(S.\) aureus. \(S.\) aureus is a bacterium that is often found in highly-contained protein foods, especially livestock products such as meat, egg, milk and their processed products. These bacteria can cause food poisoning because it can produce enterotoxins which is resistant to heat and still active after heated at 100ºC for 30 minutes. These bacteria are not expected to be present in food products, \(S.\) aureus growth can be prevented by giving bacteriocin to the product.

Bacteriocin is a component which is synthetized ribosomally and being produced by bacteria in order to inhibit the growth of other bacteria. Bacteriocin sometimes can be considered as an antibiotic, however, bacteriocin are different with antibiotics in general because bacteriocin synthesis occurs in ribosomes, cell of host is not being affected by bacteriocin. The difference between bacteriocin and antibiotic are bacteriocins only able to fight closely related bacteria because it has limited inhibition. \textit{Lactobacillus plantarum} are the most known lactic acid bacteria which has been studied in genetics, ecology, and physiology. \textit{Lactobacillus plantarum} has ability to produce antimicrobial peptide called

1. Introduction
Foods are better being consumed raw and fresh, however, fresh milk has different case. In fact, drinking fresh milk still became a debate until this day. Some claims said unprocessed fresh milk contains more nutrition compared with pasteurized milk, although milking process do not guarantee protection from bacteria and dirt contamination. One of example bacteria which can cause damage on dairy products are \(S.\) aureus. \(S.\) aureus is a bacterium that is often found in highly-contained protein foods, especially livestock products such as meat, egg, milk and their processed products. These bacteria can cause food poisoning because it can produce enterotoxins which is resistant to heat and still active after heated at 100ºC for 30 minutes. These bacteria are not expected to be present in food products, \(S.\) aureus growth can be prevented by giving bacteriocin to the product.

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bacteriocin, which is useful to protect radical free oxygen with superoxide-mediated non enzymatic reducing agent manganese [1]. One type of bacteriocin known is plantaricin IIA-1A5, this bacteriocin has role as natural biopreservative because it contains antimicrobial compound which expected to be able to damage and kill pathogenic bacteria such as *S. aureus* by inhibiting other bacteria growth with damaging its cell membranes. Therefore research is needed to analyze the application of bacteriocin on fresh dairy milk.

2. Method
The study was conducted at dairy farm Kawasan Usaha Peternakan (KUNAK), Cibungbulang, Bogor from June to October 2020. This research trial used fresh milk from dairy farms in Kunak. Treatments were separated into 3 treatments with 3 repetition. The treatments were control, plantaricin 5%, and synthetic antibacterial penicillin were used on the milk. Milk with treatment were stored at room temperature for 6 hours and were observed every 2 hours (0 hour, 2 hours, 4 hours, 6 hours). Physical tests, microbiological tests, and physicochemical tests were carried out at Integrated Lab Animal Technology Production Division, Faculty of Animal Science and Technology, Bogor Agricultural University, Bogor.

This study data analysis was conducted using randomized block design (RDB) with 3 repetitions. Treatment design consisted of control, plantaricin (11.02 mL), and penicillin (7.5 mL). Tukey test will be performed if different results were obtained. RBD systematic model according to Steel and Torie (1991) as following equation [2]:

\[ Y_{ij} = \mu + T_i + \beta_j + \varepsilon_{ij} \]

- \( Y_{ij} \): The results of the response variable
- \( \mu \): Common middle value
- \( T_i \): Effect of treatment (bacteriocin 0% control, bacteriocin and synthetic antibiotic)
- \( \beta_j \): Effect of the block (week 1, 2, and 3)
- \( \varepsilon_{ij} \): Error due to the treatment in the test

3. Result and discussion
Total plate count (TPC) of fresh dairy milk were analysed from 3 different dairy cows. The test were divided into 2 treatment, no treatment (control), and milk with *S. aureus* (Table 1). Cow 1 has least amount of TPC, which is 1.38 x 10^5, and cow 3 has the most TPC in the milk, which is 4.8 x 10^5.

| Content | Type of Cow (cfu mL^-1) |
|---------|------------------------|
|         | Cow 1 | Cow 2 | Cow 3 |
| TPC     | 3.15x10^5 | 1.38x10^5 | 4.8x10^5 |
| *S. aureus* | <1.0x10^1 | <1.0x10^1 | <1.0x10^1 |

These amount of TPC in the milk are dependent on the milking processing, which includes handling, hygiene during milking, transportation, and milk products processing. Maximum limit of TPC amount which can exist in colony are 1 x 10^6, while *S. aureus* are 1 x 10^2 [3] (SNI 7388:2009). The number of TPC in the table were lower compared to SNI. *S. aureus* were also confirmed are less than 1x10^1 which are also lesser than. There are some factors that affects bacteria growth, namely heat, time, humidity, oxygen, pH, and light availability [4].

Milk physicochemical were also analyzed by lactoscan, which the results are displayed in table 2. Milk fat amount from all 3 cows were surpassed the amount of minimum required, however, protein amount from cow A and cow B are slightly lesser than standard. Protein amount minimum in milk are 2.8%, while fat are 3.0, SNI 3141.1, 2011 [5].
Table 2. Mean of physicochemical characteristics fresh milk

| Content  | A     | B     | C     |
|----------|-------|-------|-------|
| Fat      | 3.03  | 3.2   | 3.06  |
| SNF      | 7.78  | 7.81  | 7.79  |
| Protein  | 2.77  | 2.78  | 2.8   |

SNF minimum amount in milk are 7.5% [6] (SNI 01-3951:1995), results in table 2 clearly shows that SNF amount of cow A, cow B, and cow C are slightly higher than minimum. Lastly, Lowry Protein Assay test of plantaricin obtained from 22 liter whey as its growth media were carried out. According to table 3, protein yield of 1 mL plantaricin are 1086.604, and the total amount of entire plantaricin from plantaricin production, which is 89 mL are 96707.77. According to Soenarno (2019), protein concentration amount for 1 mL plantaricin was 242.29 μg/mL, which is different from the results obtained from plantaricin production. Protein amount from the media used to made plantaricin may affect the result of protein concentration after plantaricin production [7].

Table 3. Protein yield obtained from crude plantaricin according to Lowry Protein Assay Method.

| Plantaricin obtained (mL) | Protein Yield (mL⁻¹) | Protein Yield (89 mL) |
|---------------------------|----------------------|-----------------------|
| 89                        | 3.03                 | 3.2                   |

4. Conclusion

Plantaricin obtained from 22 liters of cheese whey is 89 mL. Lactoscan results and microbiological characteristics of fresh milk have met SNI 3141.1 2011.

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