

**Pengaruh Bakteriosin sebagai Pengawetan Daging pada Penyimpanan Suhu 4°C****The Influence of Bacteriocin as a Preservation of Beef to Extend Storage at 4°C****<sup>1</sup>Dewi Sartika Ariyani, <sup>2</sup>Nenny Harijani, <sup>2</sup>Mas'ud Hariadi**<sup>1</sup>Student, Veterinary Public Health Department<sup>2</sup>Faculty of Veterinary Medicine Airlangga University

Kampuc C Unair, Jl. Mulyorejo Surabaya – 60115

Telp. 031-5992785, Fax. 031-5993014

Email : vetunair@telkom.net

**Abstract**

The aim of this research was to determine the influence of addition of bacteriocin as the preservation of beef to extend storage at 4°C. 500 grams of fresh beef from Karangmenjangan market and 500 grams fresh beef from Pucang market were used and divided into two groups for each market, after dipped in bacteriocin for 10 minutes and stored in refrigerator with temperature of 4°C. Every beef sample divided into two group, first was control group and the other was treatment group with the addition of bacteriocin. The replication for each sample has been done for 5 times in 5 days consecutively. Eber test was used to determine the early spoilage process in beef. The result showed that dipping in bacteriocin for 10 minutes was effective to extend the storage span of beef up to the minimum day of day 4 for samples from both market.

**Keywords:** Bacteriocin, Preservation, Beef, 4°C.**Introduction**

The meat has long been considered a highly desirable and nutritious food, but unfortunately it is also perishable food because it provides the nutrients needed to support the growth of many types of microorganisms (Kolalou *et al.*, 2004). Due to its unique biological and chemical nature, the meat undergoes progressive deterioration from the time of slaughter until consumption under certain aerobic conditions, or generally introduced during certain processing, leads to the manifestation of changes or spoilage of meat (Nychas *et al.*, 2008).

It has been recognized that Lactate Acid Bacteria (LAB) are capable of producing inhibitory substances other than organic acids (lactate and acetate) that are antagonistic toward other microorganisms (Ahmed *et al.*, 2004) where they serve preservative role (Stiles and Holzapfel, 1997) for the food. Lactic acid bacteria (LAB) are characterized as Gram-positive cocci or rods, aerotolerant, able to ferment carbohydrates for energy and lactic acid production.

The growing need for naturally safe and healthy food has led to increased interest in the use of bacteriocin-producing from LAB in possibility of introducing bacteriocin as a factor in the "barrier" approach to food protection has attracted scientists in the field of food safety (Moracanin, 2012). Bacteriocin application as biocontrol on preservation of food products from animal origin, such as fresh meat and pasteurized milk, can directly use a bacteriocin supernatants derived from *Pediococcus pentosaceus* culture (Harijani, 2008).

Ray (2004) wrote that any microorganism that can multiply in a food to reach a high level (spoilage detection level) is capable of causing food to spoil. Some of the detectable parameters associated with spoilage of different types of foods are changes in color, odor, and texture; formation of slime; accumulation of gas (or foam); and accumulation of liquid (exudate, purge). Spoilage by microbial growth occurs much faster than spoilage by microbial extra- or intracellular enzymes in the absence of viable microbial cells.

Food preservation technic by cooling method is accepted well by the people. Cooling method can be combined by controlling the temperature level during the storage in order to prolong the storage span (Estiasih *et al.*, 2010). Chilling involves reducing food temperatures, but only to approximately -1°C. Refrigerators for cold storage/chilling are normally used at 0°C to +8°C for preservation of a wide variety of food products and also chilling can slows the growth of most bacteria (Estiasih *et al.*, 2010).

Based on that explanation, the utilization of natural preservation is needed to prevent the early process of beef spoilage that is related to storage extension of beef. This research was to determine the influence by the addition of bacteriocin as the preservation of beef to extend storage at 4°C.

**Materials and Methods**

The research has been carried out in the Laboratory of Veterinary Public Health faculty of Veterinary Medicine, Universitas Airlangga Surabaya on February 2013.

This research has used 500 grams of fresh beef from Karangmenjangan market and 500 grams fresh beef from Pucang market, bacteriocin, Eber Reagent, spiritus. The equipment in this research required Erlenmeyer tube, Beaker glass, bunsen burner, measuring cup, weigher, pinset, scissors, sealable plastic, styrofoam box, alumunium foil, refrigerator with 4°C temperature.

First sample of fresh beef were from Karangmenjangan market (A). Second sample of fresh beef were from Pucang market (B). Samples of each market was taken 100 grams of fresh beef each day, then every sample was put in an styrofoam box and immediately transported to the laboratory. The replication for control and treatment as the sample has done for 5 times in 5 days consecutively. Each sample was inspected everyday until the early process of spoilage in the beef was occurred.

100 grams of fresh beef was cut into 20 parts. Then put those parts into 20 sealable plastic as a placing sample, then samples were divided into two groups, (I) group as a control or without the addition of bacteriocin as much as 10 samples and (II) group as a treatment that was added by bacteriocin for 10 minutes as much as 10 samples.

**Data Processing**

The data in this research was processed descriptively by determined the minimum day of the early day spoilage occurrence on beef to determine the influence of bacteriocin of beef that has been added the bacteriocin compared to the fresh beef that has not been added the bacteriocin.

**Result and Discussion**

The result of research showed that the addition of bacteriocin to the beef that has been dip in it for 10 minutes was very effective in increasing the storage span at 4°C from the sample from Karangmenjangan market (A) and Pucang market (B) were up to the minimum day on day 4 for both market. The result of bacteriocin in influenced the early spoilage process on the fresh beef during daily inspection based on the result of Eber test can be seen on Table 1.

Table 1. The early spoilage process on the beef that has been dip in the bacteriocin on daily inspection

Replication	Samples			
	A Market		B Market	
	Control	Treatment	Control	Treatment
1	2	7	2	4
2	2	6	2	5
3	2	6	2	6
4	2	5	2	6
5	2	4	2	7

Note : Control : The sample without the addition of bacteriocin

Treatment : The sample with the addition of bacteriocin

Control samples (I) which was not added by bacteriocin to the fresh beef from A and B market for all 5 replications showed that the early spoilage process occurred on day 2. Treatment samples (II) with the addition of bacteriocin to the fresh beef that has been dip in it for 10 minutes for the samples from A market on replication 1, 2, 3, 4, and 5 examined daily showed that the early spoilage process on the beef occurred on days 7, 6, 6, 5, and 4 respectively, while the fresh beef for the sample from B market occurred on replication 1, 2, 3, 4, and 5 examined daily showed that the early spoilage process on the beef occurred on days 4, 5, 6, 6, and 7 respectively. It can be drawn the result from the samples that the minimum day of early spoilage process was occurred on day 4 for both market.

Sample of (I) group that was not added by bacteriocin on day 2 from A market and B market has undergone the early spoilage process with the formation of  $\text{NH}_4\text{Cl}$  mist which was happened a reaction in which the binding of  $\text{NH}_3$  gas produced from the beef reacted with HCl in the Eber reagent that indicate the early spoilage process. This was happened due to the beef samples that was not added by bacteriocin (I) has undergone the growth process of bacteria based on the growth curve on the beef that consist of lag phase, log phase, stationary phase, and decline or death phase in 24 hour that include the intrinsic factor such as nutritional value of meat, water conditions, pH, oxidation-reduction potential and the presence or absence of substance barrier or obstacle and the extrinsic factor such as temperature, relative humidity, presence or absence of oxygen and the shape or condition of the meat (Fardiaz, 1992) so that it lead to the spoilage process and also the spoilage process has changed the structure of beef became mushy consistency, pale colored, and the smell turned fishy.

Day 1 inspection of samples from (II) group that added by bacteriocin has not been spoiled due to the bacteriocin work process that happened quickly that has been cause the growth of other undesirable microbes may be prevented, and also the giving of bacteriocin derived from LAB metabolites can also lower the pH of food, so that it can be slower the growth of spoilage microorganisms (Buckle *et al.*, 1987).

The result of (II) group from A market, the minimum day of early spoilage process occurred on day 4 while from B market also on day 4 of inspection day and the maximum day of early spoilage process occurred on day 7. This was happened due to the relation between the general extrinsic and intrinsic quality of those beef samples from each market was almost same and also there was the process of decreasing ability of bacteriocin in influencing the growth of spoilage microorganisms on beef, so that it was no longer affected the storage span of beef more than 7 days.

Based on the result, both sample from A and B market can be seen the ability of bacteriocin as the natural preservation was depend on the intrinsic and extrinsic from the beef that was used as the sample which is related to production of  $\text{NH}_3$  gas that showing the early spoilage process of the beef that binding with Eber reagent.

The purpose of this research was to determine the influence from the addition of bacteriocin as the preservation of beef to extend storage at  $4^\circ\text{C}$ . Checking method in this research was used Eber test. Eber test was used eber reagent that consists of Ether, Alcohol 96%, and HCl with the ratio 1:3:1 and placed in Erlenmeyer tube then put the cover lid tube that has been hanging a beef sample on the wire to be observed the process of  $\text{NH}_4\text{Cl}$  mist formation if it has been undergone the early spoilage process.

The storage of beef in the refrigerator at  $4^\circ\text{C}$  due to that temperature was the medium temperature that mostly available in any kind of refrigerator and not only the texture and structure of beef was not undergone the crystalization iced process but also there was not any much changes in beef and the bacteriocin could be well functioned at  $4^\circ\text{C}$ .

Bacteriocin is a substance that can kill the bacteria and can be used as the utilization of biological preservation or biopreservation on food, as the superior seed starter to increase the storage span and food safety (Sudirman *et al.*, 1995; Harijani, 1997). The advantages of bacteriocin as biological preservatives or biopreservation namely (i) it is not toxic and susceptible to degradation by proteolytic enzymes because it is a protein compound, (ii) it does not harm for intestinal microflora as easily digested by gastrointestinal enzymes, (iii) to reduce chemical use as a food preservative, and (iv) flexible to use and (v) stable to pH and temperature are sufficiently broad so it is resistant to treatment process involving acids and bases, as well as hot and cold conditions (Cleveland *et al.*, 2001). Instead of the advantages by using bacteriocin as preservation, the price for the bacteriocin is around Rp. 100.000,- for half litre and it can be used effectively for beef preservation.

### Conclusion

Based on the research about the influence of bacteriocin as a preservation of beef to extend storage at  $4^\circ\text{C}$  can be concluded that the addition of bacteriocin on the fresh beef as the samples that has been dip in it for 10 minute can be effective to increase the storage span at  $4^\circ\text{C}$  up to the minimum day on day 4 for both samples from Karangmenjangan and Pucang market.

## References

- Ahmed, T., R. Kanwal. 2004. Biochemical Characteristics Of Lactic Acid Producing Bacteria And Preparation Of Camel Milk Cheese By Using Starter Culture. *Pakistan Vet. J.* 2: 24.
- Buckle, K.A., R.A. Edwards, G.H. Fleet, M. Wooten. 1987. *Ilmu Pangan*. Translated by H. Purnomo dan Adiono. Universitas Indonesia Pres. Jakarta. 365 pages.
- Cleveland, J., T.J. Montville, I.F. Nes, M.L. Chikindas. 2001. Bacteriocins: Safe, Natural Antimicrobials for Food Preservation. *International J. of Food Microbiology* 71: 1–20.
- Estiasih, T., K.G.S. Ahmadi. 2010. *Teknologi Pengolahan Pangan*. Fak. Teknologi Pertanian. Univ. Brawijaya. Malang.
- Fardiaz, S. 1992. *Mikrobiologi Pangan 1*. Gramedia Pustaka Utama. Jakarta. 159, 327.
- Harijani, N., 2008. *Eksplorasi dan Aktifitas Bakteriosin dari Bakteri Asam Laktat sebagai Antimikroba dalam Proteksi Pembusukan Bahan Pangan Asal Hewan*. Bogor.
- Kolalou, I., M. Faid, A.T. Ahami. 2004. Extending the Shelf Life of Fresh Minced Camel Meat at Ambient Temperature by *Lactobacillus Delbruekii* Subsp. *Delbruekii*. *Electronic J. of Biotech.* 7: 246-251.
- Moracanin, S.V., L. Turubatovic, M. Skrinjar, D. Obradovic. 2012. Antilisterial Activity of Bacteriocin Isolated from *Leuconostoc mesenteroides* ssp.
- Nychas, G.J.E., P.N. Skandamis, C.C. Tassou, K.P. Koutsoumanis. 2008. Meat Spoilage During Distribution. *Meat Science* 78: 77–89.
- Ray, B. 2004. *Fundamental Food Microbiology* 3rd Edition. CRC Press. Florida.
- Stiles, M.E., W.H. Holzapfel. 1997. Lactic Acid Bacteria of Foods and Their Current Taxonomy. *Int. J. Food Microbiol.* 36: 1–29.