Detection of Contamination *Salmonella sp*. of Beef in Banyuwangi Traditional Market

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Abstract

Salmonella sp. is one of the bacteria from Food-Borne Disease agents, can contaminate meat and cause *salmonellosis*. According to National Indonesian Standard 2009, meat is safe for consumption if it has a negative result of salmonella in 25 grams of meat. The purpose of this study was to determine whether there was contamination of *Salmonella sp.* on beef sold in the traditional market of Banyuwangi District. A total of 18 samples used in this study were taken according to the criteria for isolation and identification. All samples were enriched with the pre-enrichment stage using Lactose Broth media, and the enrichment medium using Tetrathionate Broth. Furthermore, in the isolation stage using Salmonella Shigella Agar (SSA) media. Then proceed with the identification stage using the Triple Sugar Iron Agar (TSIA) test, the Sulfide Indole Motility (SIM) test, and the Urease test. The results showed that 5 out of 18 samples tested positive for Salmonella.

Keywords: Beef, *Salmonella sp.*, Banyuwangi Market

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INTRODUCTION

Salmonella sp is one of the Food-Borne Disease agents that can cause Salmonellosis (Moekti *et al*., 2020). Salmonellosis can infect humans through *Salmonella sp* contaminated animal products (Sartika *et al*., 2016). Meat is the most common transmission medium for *Salmonella sp* (Husna *et al*., 2020). The bacterial contamination can occur anywhere from slaughterhouse into the ready-to-consume phase. *Salmonella sp.* can contaminate animal food products through air, water, soil, environment, human excrement traces, or animal diet (Arifah, 2010).

According to Indonesia National Standard (SNI) (2009), contaminated meat can be caused by an unhygienic location that is a mixture of various good vendors. According to the Minister of Agriculture's Decree No.:413/Kpts/TN.310/7/1992, the area for meat vendors in markets should be apart from other commodities vendors. The traditional markets are among the most plausible contamination due to the minimum sanitation and contiguous of various vendors selling different products. This situation can expose the meat to microbe contamination. Selling meat on an open platform can also entice buyers to browse through the products by touching them, exposing the meat with microbe contamination risks. This situation led to change in texture and reduce meat quality (Ratnawati *et al*., 2014).

High-quality products should require the Safe, Healthy, Whole, and Halal criteria by having microbe contamination limit that must not be above the determined standard (Susanto *et al*., 2013). According to Indonesia National Standard (SNI) 7388:2009, the maximum *Salmonella sp* contaminant in fresh, frozen meat, and minced meat is negative/25g. In 2018, Banyuwangi district had around 1.832 beef cattle population. In 2014, there were 2.037 beef cattle slaughtered for consumption in the same district. This data indicates that the highest consumption of meat comes from the Banyuwangi district (BPS Banyuwangi, 2019).

This research aimed to identify the existence of *Salmonella sp.* contaminants in beef products sold in Pasar Tradisional Kecamatan Banyuwangi.

MATERIAL AND METHOD

Sample Collection

The 18 samples of raw beef were obtained from two traditional markets: Blambangan market and Banyuwangi market. Samples were collected in the morning at 5 am. Each sample were taken 25 grams especially those that displayed openly on a table without any cover and located in beef vendors area. Samples were labelled as BA (samples that taken from Banyuwangi market) and BL (samples that taken from Blambangan market). The collected samples then secured into a sterile zipped plastic and transported to the laboratory using cool box for *Salmonella sp*. evaluation.

Agar Media Preparation

The media used for pre-enrichment was Lactose Broth (Merck®). Selective enrichment media used for isolating *Salmonella sp* was Tethrathionate Broth (Merck®). Salmonella Shigella Agar (SSA) (Oxoid®) then used for *Salmonella sp* isolation. Further test for *Salmonella sp* identification were using media including Sulfide Indol Motility (SIM) agar (Merck®), Triple Sugar Iron Agar (TSI-A) (Oxoid®) and Urea Broth (HIMEDIA®).

Salmonella sp. Evaluation

First step for isolation and identification of *Salmonella sp* was conducted by Pre-enrichment technique. Pre-enrichment was done by transferring 25 g samples into 225 ml of Lactose Broth (Merck®) in Erlenmeyer flasks and slowly stirred until homogeneous. The suspension then incubated at 35ºC for 24 hours. The enrichment method then conducted by inoculating 1 ml suspension from Lactose Broth (Merck®) into 10 ml Tetrathionate Broth (Merck®) and incubated at 35ºC for 24 hours (SNI, 2008). Thereafter, one loopful of suspension was streaked into Salmonella Shigella Agar (SSA) (Oxoid®). The plate was incubated at 37ºC for 24 – 48 hours. *Salmonella sp.* appeared as a colorless colony with centered black spot (SNI, 2897:2008).

Moreover, *Salmonella sp* colonies was transferred into object glass for Gram staining. Identification of *Salmonella sp.* was established by microscopic examination using a monocular microscope at 1000 magnification (Mukhtaruddin *et al*., 2018). *Salmonella sp* colonies from SSA was taken by ose and streaked into Triple Sugar Iron Agar (TSI-A) (Oxoid®) and incubated at 35ºC for 24 – 48 hours (SNI, 2008). Further step is biochemical testing using urease and indole test. Urease test was conducted by inoculating positive *Salmonella sp* colony from TSI-A using ose into Urea Broth (HIMEDIA®) and incubated at 35ºC for 24-48 hours. Whereas, the indole test was done by transferring one loopful of positive colony into Sulfide Indol Motility (SIM) agar (Merck®) and 0,2 - 0,3 ml of Kovac's reagent was added (SNI, 2008).

Data Analysis

The data obtained were analyzed descriptively and presented as table.

RESULT AND DISCUSSION

The results showed that 5 samples (27,77%) were tested positive and 13 samples (72,23%) were tested negative for *Salmonella sp*. The positive samples were labeled as DS.2.BA, DS.3.BA, DS.2.BL, DS.7.BL, DS.10.BL (Table 1).

Table 1. *Salmonella sp.* detection in raw beef from traditional markets in Banyuwangi

Sample Code	Variables			
	SSA	TSIA	SIM	Urease
DS.2.BA	+	+	+	+
DS.3.BA	+	+	+	+
DS.2.BL	+	+	+	+
DS.7.BL	+	+	+	+
DS.10.BL	+	+	+	+

These results obtained are supported by research of (Ramadhani *et al*., 2017) which found 3 of 9 samples of beef obtained from traditional markets in the city of Banda Aceh were contain *Salmonella sp.*, together with the research of (Indri *et al*., 2018) which was found that 1 of 32 samples of cattle obtained from a slaughterhouse in Banyuwangi sub-district were tested positive for *Salmonella sp*. Research by (Sugiyoto *et*

al.,2015) also showed that there were 3 out of 17 beef samples taken from traditional markets in the city of Bandar Lampung showed that amount of microbial contamination were exceeded the Indonesian National Standard (SNI).

Beef is a farm commodity popularly used in various dishes, whether it is served fresh or made into various cooked dishes (Rohmah *et al*., 2018). Beef is rich in protein, fat, minerals, and other nutrition needed. The protein in beef contains complete amino acid structure and the overall macro and micro nutrition in it is relatively complete - which makes it an efficient growing medium for bacteria (Ernawati *et al*., 2018).

Factors like storage temperature, storage time, oxygen rate, and moisture balance affect the bacteria growth and activities (Hajrawati *et al*., 2016). This environment provides great microorganism growth because of the high moisture (around 68-75%), nitrogen-rich, fermentable substances, high pH of 5,3-6,5; and rich in minerals (Sangaji *et al*., 2019). Water become a transport medium between fiber and meat, which makes moisture level a crucial living factor for microorganisms. A newly slaughtered cattle usually contains 10^2-10^4 mesophillic bacteria per inch that comes from the digestion system and the outer skin of the animal (Indriyani *et al*., 2018).

Salmonellosis commonly occurs on unhygienic food (Wahyuningsih *et al*., 2019). *Salmonella sp*. is one of the pathogenic bacteria that can cause diseases to humans. The contamination of *Salmonella sp*. is still a significant problem in the world (Momani *et al*,. 2019). The contamination of *Salmonella sp.* in the beef sold in Banyuwangi and Blambangan traditional market is due to the open selling environment, unwashed knife, the unhygienic water, untreated meat products, the distance of the slaughterhouse to the market, in addition the consumer's keep touching the meat before purchase it. Open selling platforms, untreated beef, as well as consumer's habit of touching the meat can speed up the growth of bacterial contaminants (Sa'idah *et al*., 2011).

Pratiwi *et al*. (2014) stated that the increase of *Salmonella sp* contamination happens because the meat was cut using a traditional butchering method, improper hygiene, and the significant distance between the slaughterhouse and the meat vendors. After the slaughtering process, the meat was transported to two different traditional markets, which starts the contamination. *Salmonella sp* can contaminate beef because of the unhygienic butcher house and poor handling that cause it contact with Salmonella-infected feces (Suwito, 2011).

Unhygienic butchering processes leaves bacteria contamination on the cutting devices and transferred into the meat's surface (Arnia and Efrida, 2013). Cutting meat into smaller pieces will increase the surface area that can be contaminated with microbes because surface areas allow microbes to gain easier access to food, water, and oxygen which speeds up their growth and ruins the meat. Meat vendors' habit of not separating organs from meat can also increase the contamination of *Salmonella sp* (Harsojo and Darsono, 2013).

The lack of properly covered wet and dry trash bins and clean water supply will affect on the contamination of *Salmonella sp*. The vendors were also irresponsibly did not change the water that used for washing meat because of the lack of clean water supply. Unhygienic vendor's devices that used such as knives and cutting boards, along with their habits of drying their washed hands on a dirty rag are also additional factors to the *Salmonella sp*. contamination. This situation needs to be overcome with washing hands using soap and running water, and drying them using a dry and clean towel or a hand-drying machine (Hasanah *et al*., 2021).

In order to decrease the risks of *Salmonella sp* contamination, the whole process must be taken care properly from the farm to the table. Therefore, the safety of meat products becomes the collective responsibility of the provider, processor, distributor, consumer, and the government (Sukmawati, 2018). Beef is one of the food products that possess high nutritional value, which unfortunately can be a medium for enzyme activities and microorganism growth, establish it as the food products that have shortest shelf-life caused by microbe activity (Sarassati

and Kadek, 2015). Damaged meat caused by microbe contamination will induce change in smell, texture, taste, and color (Zahrarianti *et al*., 2012).

CONCLUSION

It can be concluded that 5 out of 18 beef samples sold in two traditional markets: Blambangan market and Banyuwangi market were tested positive for *Salmonella sp*.

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REFERENCES

Arifah, I. N. (2010). Microbiology analysis on food. Study of farm produces technology, Faculty of Agriculture, Universitas Sebelas Maret, Surakarta.

Arnia, & Efrida, W. (2013). Identifying the Contamination of Coliform Bacteria on Fresh Beef sold in Traditional Markets in Bandar Lampung. *Jurnal Majority*, 2(5).

Bureau of Planning and Development of Banyuwangi District. (2013). Banyuwangi District.

Hajrawati, H., Fadliah, M., Wahyuni, W., & Arief, I. I. (2016). Physical, microbiological, and organoleptic qualities of broiler meat in traditional markets in Bogor. *Jurnal Ilmu Produksi dan Teknologi Hasil Peternakan*, 4(3), 386-389.

Harsojo, & Darsono. (2016). Study of Heavy Metal Contaminant using Neutron Activation Analysis and Pathogenic Microbes in Beef and Organs. *Jurnal Ilmiah Aplikasi Isotop dan Radiasi*, 9(2), 129-137.

Hasanah, U., Teuku, R. F., Mahdi, A., Erina, N., & Cut, D. I. (2021). Detection of *Salmonella Sp.* in Meat Vendors' Hands and Prediction of Its Cause in Peunayong Traditional Market, Banda Aceh. *Jurnal Ilmiah Mahasiswa Veteriner*, 5(2), 100-107.

Husna, H., Nurlian, N., & Wiqayatun, K. (2020). Identification of Salmonella, Shigella, and *E. Coli* in the Processed-Meat Produce Sie Balu. *Jurnal Public Health*, 3(2), 88-94.

Indonesia National Standards (SNI) 2897-2008. Indonesia's Standards for Testing Methods and Contaminant Testing in Meat, Eggs, and Milk and Their Produces.

Indonesia National Standards (SNI) 7388-2009. Indonesia's Standards for Maximum Limit of Microbe Contamination in Food.

Indriyani, D. P., Wiwiek., T., & Ratih., N. P. (2019). Isolation and Identification of Salmonella in Beefs in Banyuwangi Butcher House. *Jurnal Medik Veteriner*, 2(2), 83-88.

Ministry of Agriculture. (1992). Decree No: 413/Kpts/TN.310 /7/1992, about Farm Animal Butchering and Handling of Meat and Its Other Produces. Jakarta

Moekti, B. S., Nabila, A. H., Mega, U. E. M., & Lailia, D. K. W. (2020). Prevention of Salmonellosis Disease Through Animated Educational Song Video for Elementary, Junior, and High School Students in Surabaya. *Jurnal Pengabdian Pada Masyarakat*, 2(1).

Momani, W. A., Janakat, S., & Khatatbeh, M. (2018). Bacterial Contamination of Table Eggs Sold In Jordania Markets. *Pakistan Journal of Nutrition*, 17(1), 15-20.

Mukhtaruddin, Fakhrurrazi, & Mahdi. A. (2018). Isolation and Identification of Salmonella Sp in Chicken's Intestines in Lampuja Village, Aceh Besar. *Jurnal Ilmiah Mahasiswa Veteriner*, 3(1), 24-36.

Pratiwi, F. S., Padaga, M. C., & Wuragil, D. K. (2014). Isolation and characterization of Salmonella Sp in the carcass and viscera of chicken sellers in Malang. *Jurnal Medika Veteriner*, 3(2).

Ramadhanii, D., Fakhrurazzi, & Mahdi, A. (2017). Isolasi dan Identifikasi Bakteri *Salmonella Enteritidis* Pada Daging Sapi Yang Dijual Di Pasar Tradisional Kota Banda Aceh. *Jurnal Ilmiah Mahasiswa Veteriner*, 1(4), 625-630.

Ratnawati, Nurliana, & Razal. (2014). Consumer Satisfaction Rate towards Quality and Price of Beef Sold in Banda Aceh. *Agripet*, 14(2), 125-131.

Rohmah, Mohammad, F. F. M., & Umi, P. (2018). Physical Analysis of Beef Affected by Concentrate and Soaking Duration of Kenikir (*Cosmos caudatus kunth*). *Agrointek*, 12(1), 51-54.

Sa'idah, F., Yusnita, S., & Herlinawati, I. (2011). Research results of microbe contamination in beef sold in the modern and traditional market. *Dilavet*, 21(2), 1-4.

Sangadji, I., Jurianto, & Muhammad, R. (2019). The Impact of Broiler Meat Storage Duration to Its Quality As Perceived from Its Protein and Total Bacteria. *BIOSEL (Biology Science and Education): Jurnal Penelitian Science dan Pendidikan*, 8(1), 47-58.

Sarassati, T., & Kadek, K. A. (2015). Quality of Wagyu Beef and Balinese Beef Stored at -19ºC. *Indonesia Medicus Veterinus*, 4(3), 178-185.

Sartika, D., Susilawati, & Gusman, A. (2016). Identifying *Salmonella Sp*. Contamination in Broiler Chicken by Quantifying Methods from Three Traditional and Two Modern Markets in Bandar Lampung. *Jurnal Teknologi & Industri Hasil Pertanian*, 21(2), 89-96.

Statistics Indonesia (BPS) Banyuwangi District. (2019). Banyuwangi District in Numbers.

Sugiyoto, Kusuma, A., & Veronica, W. (2015). Kandungan Mikroba pada Daging Sapi dari Beberapa Pasar Tradisional di Bandar Lampung. *Jurnal Ilmu Peternakan Terpadu*, 3(2), 27-30.

Sukmawati, S. (2018). Total Microbial Plates on Beef and Beef Offal. *Bioscience*, 2(1), 22-28.

Susanto, E. (2014). Standard Post-Harvest Handling of Fresh Meat. *Jurnal Ternak*, 5(1).

Suwito, W. (2011). Distribution of Salmonella serotype From Slaughterhouse (RPH) And Chicken Abattoir (TPA) In Bogor. *Widyariset*, 14(2), 361-366.

Wahyuningsih, E., Indah, S., & Musyarif, Z. (2019). Identifying *Salmonella sp*. In Chicken Eggs Sold in Wage Traditional Market Purwokerto as a Microbiology Teaching Material Development. *Bioedusiana: Jurnal Pendidikan Biologi*, 4(2), 79-83.

Zahrarianti, R., Kusmajadi, S., & Lilis, S. (2012). Effects of Soaking in Various Concentrate of Rosella Petal Extract (*Hibiscus Sabdariffa Linn*) to the Total Bacteria, Shelf Life, and Beef Color. *Students e-Journal*, 1(1), 15.

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