# Contamination of *Salmonella spp.* on Chicken Carcasses and their Risk Contributing Factors during Supply Chain in Jakarta

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# ABSTRACT

The consumption of chicken meat by consumers in Jakarta is very high, which is in line with the high supply of it. Salmonella spp. is pathogenic bacteria which is frequently found in chicken carcasses and can cause food borne disease. The aims of this study were to determine the prevalence of Salmonella spp contamination on chicken carcasses and critical factors during distribution chain in Jakarta. The study was conducted based on the risk based approach compiled surveillance data, quantitative analysis of Salmonella spp., survey of good handling, constructed risk ranking and analysis sensitivity. Quantitative analysis determined by plate count method with using selective media XLT4 agar as Salmonella presumptive. Risk profile was involved risk-ranking and sensitivity analysis using Monte Carlo simulation of 100,000 iterations from Oracle Crystal Ball®. The highest prevalence of Salmonella in chicken carcasses is showed from traditional markets (41.86%) with amount was 1.09  $\pm$  0.12 log CFU/g; then is followed from TPHU distribution (17.65%) with amount was 1.92 $\pm$  1.11 log CFU/g. The risk contribution from TPHU distribution (17.65%) with amount was 1.92 $\pm$  1.11 log CFU/g. The risk contribution and the unproper equipment's cleaning method in traditional market (33.2% contribution). These were the most important aspects to be controlled.

Keywords: Chicken carcasses, critical point, risk profile, Salmonella spp., sensitivity analysis

#### ABSTRAK

Konsumsi daging ayam oleh konsumen di Jakarta sangat tinggi, sejalan dengan tingginya pasokan daging ayam. *Salmonella spp.* merupakan bakteri patogen yang sering ditemukan pada karkas ayam dan dapat menyebabkan penyakit bawaan makanan. Tujuan dari penelitian ini adalah untuk mengetahui prevalensi kontaminasi *Salmonella spp.* pada karkas ayam dan faktor kritis selama rantai distribusi di Jakarta. Penelitian dilakukan berdasarkan pendekatan berbasis risiko dengan menghimpun data surveilans, analisis kuantitatif *Salmonella spp.*, survei penanganan yang baik, menyusun peringkat risiko dan analisis sensitivitas. Analisis kuantitatif ditentukan dengan metode plate count dengan menggunakan media selektif agar XLT4 sebagai presumptive Salmonella. Profil risiko melibatkan peringkat risiko dan analisis sensitivitas menggunakan simulasi Monte Carlo dengan iterasi 100.000 dari Oracle Crystal Ball®. Prevalensi Salmonella tertinggi pada karkas ayam ditunjukkan dari pasar tradisional (41.86%) dengan jumlah 1.09  $\pm$  0.12 log CFU/g; Kemudian diikuti oleh distribusi TPHU (17.65%) dengan jumlah 1.92 $\pm$ 1.11 log CFU/g. Faktor risiko yang berkontribusi adalah tidak terpenuhinya suhu pengangkutan dari TPHU (kontribusi 12.3%) dan metode pembersihan peralatan yang tidak tepat di pasar tradisional (kontribusi 33.2%). Faktor risiko tersebut merupakan faktor risiko terpenting yang harus dikendalikan.

Kata kunci: Karkas ayam, titik kritis, profil risiko, Salmonella spp., analisis sensitivitas

# INTRODUCTION

The protein consumption rate in Jakarta, especially for broiler chicken meat, was 10,198 kg per capita per year (KEMENTAN 2022). Salmonella spp. are pathogenic bacteria that often contaminate chicken carcasses and cause foodborne diseases (Zelpina et al. 2020). Based on National Standard of Indonesia (SNI) 3924:2009, Salmonella spp contaminant in chicken carcasses must be negative for every 25 g. However, many studies show that poultry and its derivative product became the source of Salmonella spp. contamination (Sipayung et al. 2023). The occurrence of Salmonella spp. may be caused by improper handling through slaughterhouses, distribution, selling process, and food preparation (Novera et al. 2020). Salmonella spp. infection can caused illness called Salmonellosis that can cause nausea, vomiting, abdominal cramps, fever, dehydrated, bloody diarrhoea and may cause death especially to vulnerable population such as young infant or elderly (Ehuwa et al. 2021)

Determining critical control points requires understanding contamination sources that significantly contribute to the prevalence of Salmonella spp. (Akil & Anwar 2019). Salmonella contaminated poultry meat in Indonesia since the stage of farm production, slaughtering house and distribution chain (Sipayung et al. 2023; Nissa et al. 2023). Therefore, it is crucial to evaluate the most significant interfence that could be done throughout distribution chain from various sampling location. Strategies to reduce Salmonella spp. in the supply chain include risk assessments, hazard identification, hazard characterization, and risk characterization (Attrey et al. 2017). Implementation of good practices in slaughterhouses, new technology, and personnel training advised in some studies to reduce contamination risk (Nurjanah et al. 2020; Hernandez-Jover et al. 2021). The Indonesian government has released regulations in order to maintain food safety for animal-origin products through Government Regulations authorized by the Indonesia Ministry of Agriculture (KEMENTAN 2020). The application of good handling practices is also regulated by Indonesia National Agency for Food and Drugs Control (BPOM 2021). The next challenge is to determine control measurements throughout the chicken carcass supply chain that most significantly reduce potential contamination of Salmonella spp. in the final product and public health risks (Xiao et al. 2022). Risk ranking and sensitivity analysis could be used as tools that aim at enhancing monitoring effectiveness, reducing inspection expenses, and assisting governmental prioritization efforts (Van der Fels-Klerx et al. 2018). Sensitivity analysis is used in many aspects, especially for quality assurance as a tool to quantify the contributions of data input and the uncertainty in the model output (Saltelli et al. 2019).

The aim of this study were to determine *Salmonella spp.* prevalence in chicken carcasses and critical control points based on risk profile in Jakarta distribution chain. Risk profiling identified the contribution of GDP (Good Distribution Practices) and GHP (Good Handling Practices) implementation to contamination rate by combined risk

ranking and sensitivity analysis. The results of this study were expected to be used as considerations for making priority development of food safety control for chicken carcasses distributed in Jakarta province and Indonesia.

### MATERIAL AND METHODS

### Quantitative analysis of Salmonella spp.

The materials used in this study were fresh broiler chicken carcasses obtained from TPHU (traditional slaugthering house), traditional markets, and modern retail outlets in Jakarta province. Salmonella spp. contaminant levels were determined by spread plate count method with selective media xylose lysine tergitol-4 agar base (XLT4) dan Supplement XLT4 agar (Tergitol4) (Djordjević et al. 2018). Chicken carcasses were cut into 25g and placed into 225 ml of buffered peptone water (BPW). Vortex (Thermo Fisher Scientific at speeds of 200-300 rpm) was used to dissolved the sample in 30 seconds. Selective media growth initiated by inoculating the suspension into XLT4 agar with spread plate method with by glass spreader. The plates incubated in a Memmert 55 incubator at a temperature of 37 °C for 48 hours. Typical Salmonella spp. colonies grown on agar media have a black center and a reddish-transparent zone. The grown population is reported as Log CFU/g presumptive Salmonella spp. Analysis results compared with positive controls. Prevalence positive contamination counted from primary samples and secondary data from government agencies by using Microsoft Excel 2010.

#### Survey

The survey was undertaken regarding the implementation methods of GDP for TPHU (Table 1) and GHP for traditional markets as well as modern retail outlets (Table 2). Samples were collected from TPHUs, traditional markets, and modern retail outlets from well-known brand from Jakarta Province area. Purposive sampling for traditional market and modern market were determine based on amount of outlet in Jakarta province with ratio 1:10 and slovin formula as follows:

$$=\frac{N}{1+N(e^2)}$$

Note :

(n) = number of sampel

(N) = ratio outlet of modern market or traditional market

(e) =error tolerance criteria (0.05)

n

The questionnaire refers to the Safety and Security Guidelines for the Transportation and Distribution of Meat, Poultry, and Egg Products, USDA (FSIS 2005), and Indonesia regulation for processed food safety management systems in the distribution chain (BPOM 2021). Survey results described potential sources of contamination related to improper GDP or GHP implementation. Survey assessment was counted as conformity percentage (CP) based on compliance score with requirements. Score of compliance from each aspects compared to the total maximum score of compliance with the following formula: Conformity Percentage (CP) (%) = Maximum Compliance Score Compliance Score - x100

The scores as follow: (0) if not conformed, (1) if partially conformed, and (2) if fully conformed. Maximum score was come from total question multiplied by 2 as the highest score.

### **Risk Ranking**

Risk ranking was used to determine the riskiest points in each question aspect based on survey results. Risk values are calculated in a matrix based on the probability of occurrence and severity level. The severity level was determined based on compliance with GDP or GHP aspects. Values for severity levels (I) minor, (II) moderate, and (III) serious. Values for probability levels (I) rare, (II) possible, and (III) almost certain. Risk level classification according to Kovačevic et al. (2019) can be seen in Table 3.

Table 1. GDP Aspect quistionnare for implementation in slaughtering house (TPHU)

| Code                      | Good Distribution Practices | Question Number |  |  |
|---------------------------|-----------------------------|-----------------|--|--|
| Α                         | Storage and loading         | A1- A5          |  |  |
| В                         | Transport carrier           | B1-B10          |  |  |
| С                         | Documentation               | C1              |  |  |
| Modified from FSIS (2005) |                             |                 |  |  |

fied from FSIS (2005)

Table 2. GHP Aspect quistionnare for implementation in traditional market and modern retail

| Code                      | Good Handling Practices         | Question Number |  |
|---------------------------|---------------------------------|-----------------|--|
| D                         | Personel hygiene                | D1- D9          |  |
| Е                         | Product handling                | E1-E6           |  |
| F                         | Handwash and hygiene facilities | F1-F4           |  |
| G                         | Waste handling                  | G1-G2           |  |
| Modified from PDOM (2021) |                                 |                 |  |

Modified from BPOM (2021)

Table 3. Risk ranking classification

| Probability          | Severity   |           |             |  |
|----------------------|------------|-----------|-------------|--|
|                      | Minor      | Moderate  | Serious     |  |
|                      | (I)        | (II)      | (III)       |  |
| Rare (I)             | Low risk   | Low risk  | Medium risk |  |
|                      | (II)       | (II)      | (III)       |  |
| Possible (II)        | Low risk   | Medium    | High risk   |  |
|                      | (II)       | risk (IV) | (VI)        |  |
| Almost Certain (III) | Medium     | High risk | High risk   |  |
|                      | risk (III) | (IV)      | (IX)        |  |

Modified from Kovačevic et al. (2019)

### **Sensitivity Analysis**

Sensitivity analysis aimed to quantitatively measure the contribution of GDP or GHP aspects to the risk of Salmonella spp. contamination in the distribution chain. Monte Carlo simulation was used to represent the variability distribution and simulated repeatedly by selecting random values for each variable from its probability distribution based on available data (Chen et al. 2021). Input data was obtained from the median values of contamination

concentration and the percentage chance at the highest risk for each question number. Sensitivity analysis processed with Microsoft Excel 2010 add-in Oracle Crystal Ball® (Version 11.1.3.3.0). Fit - Distribution tools assisted probability distribution determination based on data at the highest risk. Triangular distribution was selected if the data is limited and model distribution determination cannot be done using the software. Further data was subjected to Monte Carlo simulation with 100,000 iterations. The contribution of each aspect was determined by squaring the Spearman rank correlation coefficient and then normalizing it to 100% (Chou 2019). Sensitivity values range from 0% to 100%, where higher values indicate greater influence of the input variable on the output variable (Chen et al. 2021). Critical control points were determined based on the risk contribution level to contamination shown in the sensitivity graph.

#### **RESULTS AND DISCUSSION**

The distribution chain of chicken carcasses in Jakarta province is displayed in Figure 1. Consumers in Jakarta province prefer labeled fresh poultry that is mostly supplied by modern retail companies as quality became the reason (Indrawan et al. 2021). Supply chain management was intended to ensure stability of quantity, cost, and quality of chicken carcasses from upstream to downstream.

#### Prevalence of Salmonella spp

Salmonella spp contamination could arise from multiple points along the food chain involved in the breeding site, process, preparation and cooking stages (Ehuwa et al. 2021). Common issue observed in Indonesia for both traditional and modern markets was the deterioration in chicken carcass quality which could influence consumer perceptions and purchasing decisions regarding chicken products (Utari et al. 2016).

Salmonella spp. contamination has been presence since chicken carcasses were distributed in TPHU Jakarta with a prevalence rate of 17.65% (n=51) (Table 4). The absence of separation between clean and dirty areas can be a potential source of pathogenic bacterial contamination in TPHU (Nurjanah et al. 2020).

The highest prevalence of Salmonella spp. contamination was found in chicken carcasses distributed through traditional markets in Jakarta, with a prevalence rate of 41.86% (n=43). The lowest prevalence rate of 0% was observed in samples distributed through modern retail outlets. Differences prevalence values can influence by several factor such as initial level of contaminants and higiene sanitation practices (Safitri et al. 2019; Regalado-Pineda et al. 2020; Ehuwa et al. 2021).

Similar study was held in Medan city, prevalence of Salmonella spp. in slaughtered chicken meat in traditional markets, modern markets, and modern retail (well-known brands) were respectively 75% (n=8), 50% (n=8), and 0% (n=8) (Nissa et al. 2023). The prevalence of Salmonella spp. contamination in samples collected from traditional markets in Lampung, Surabaya, Pangkal Pinang, and Mataram showed prevalence rates of 100% (n=36), 6.7% (n=30),

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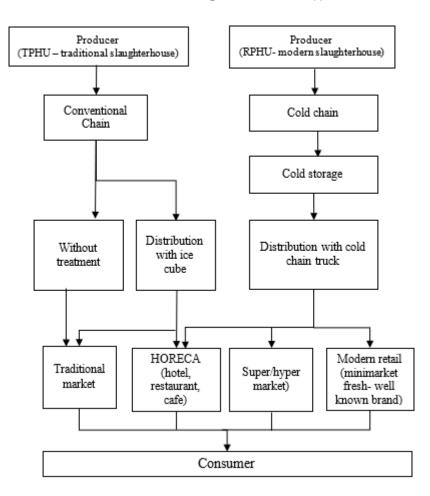


Figure 1. Distribution chain for chicken carcasses in Jakarta

| Year               | Prevalence positive | Presumtive Salmonella             | Source   | Method                   |
|--------------------|---------------------|-----------------------------------|----------|--------------------------|
|                    |                     | (Log cfu/g)                       |          |                          |
| TPHU               |                     |                                   |          |                          |
| 2019-2022          | 2,44% (n=41)        | Not Available                     |          | SNI 2897:2008            |
| 2023               | 80% (n=10)          | 1,09 ±0,12 Log cfu/g              | Kesmavet | Plate count              |
|                    |                     |                                   |          | (Djordjević et al. 2018) |
| Total              | 17,65% (n=51)       |                                   |          |                          |
| Traditional market |                     |                                   |          |                          |
| 2020-2022          | 7,41% (n=27)        | Not Available                     | Kesmavet | SNI 2897:2008            |
| 2023               | 100% (n=16)         | $1,92 \pm 1,11 \text{ Log cfu/g}$ |          | Plate count              |
|                    |                     |                                   |          | (Djordjević et al. 2018) |
| Total              | 41,86% (n=43)       |                                   |          |                          |
| Modern retail      |                     |                                   |          |                          |
| 2020-2022          | 0% (n=85)           | Not Available                     | Kesmavet | SNI 2897:2008            |
| 2023               | 0% (n=16)           | No growth bacteria                |          | Plate count              |
|                    |                     |                                   |          | (Djordjević et al. 2018) |
| Total              | 0% (n=101)          |                                   |          |                          |

32.14% (n=28), and 20% (n=5) respectively (Sartika *et al.* 2016; Safitri *et al.* 2019; Karisma *et al.* 2021; Shofia *et al.* 2023). Otherwise, research in Mexico indicated that carcasses distributed through supermarkets had a higher

prevalence of contamination compared to those distributed through markets (Regalado-Pineda *et al.* 2020).

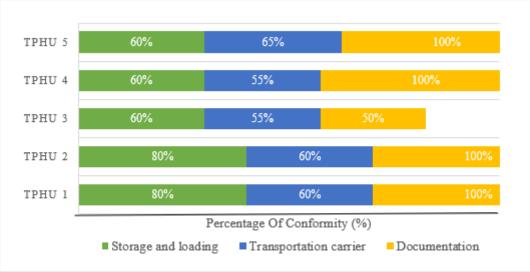
Contamination concentration can be influenced by the type of packaging and transportation methods to the market. The packaging itself should protect quality and safety of the product (Hosseinnezhad *et al.* 2018). Modified atmosphere packaging (MAP) and vacuum packaging had better microbiology protection compare to aerobic packaging (Nauman *et al.* 2022). Several recommended methods to control *Salmonella spp.* contamination from upstream to downstream include identifying contamination sources, food safety practices (GFP, GMP, GDP, GHP and HACCP), government monitoring-surveillance programs, and regulatory standards development (Nidaullah *et al.* 2017; Zelpina *et al.* 2020; Nurjanah *et al.* 2020). Personnel plays a crucial role in determining the contamination levels in products in the distribution chain (Ehuwa *et al.* 2021).

## Good Practice Implementation in the Distribution Chain

The prevalence value of contamination was closely related to food safety practices in the distribution chain. The highest GDP aspect fulfilled by the Jakarta TPHU was documentation aspect which can be seen in Figure 2. The record were contained date of arrival of live chickens, the number of slaughtering and the amount of product sent to consumers on that day. The record was also related to product traceability. Good records include location, date of activity and parties involved such as suppliers, consumers and distribution fleet (Zhang and Bhatt 2014). The gap between traditional markets and modern retail in GHP implementation was the product handling aspect (point E), which can be seen in Figure 3. The lack of refrigeration or freezing facilities could be a contamination factor in traditional markets (Safitri *et al.* 2019). Eventhough various standards for food safety practices had been created, implementation in traditional markets around the world still tends to be low therefore it was not effective in reducing the risk of food-borne illnesses (Lo *et al.* 2019).

## **Risk Profile**

Risk profile had a crucial role in risk management for food safety policy development. As the preliminary step in risk management, risk profile associated with state of knowledge, food safety policy and potential risk management options (Attrey *et al.* 2017). Risk profile was established through a risk assessment process that included risk estimation, risk ranking, and identification of control points (Nauta *et al.* 2012). Risk ranking determined the highestrisk factors by assessing their likelihood of occurrence and severity (Kovačević *et al.* 2019). Sensitivity analysis aimed to identify parameters that have the most significant impact on risk estimation, help in understanding the most important factors in determining risk and risk mitigation to be taken (Chen *et al.* 2021; Razavi *et al.* 2021). In this study, data input for sensitivity analysis included contamination



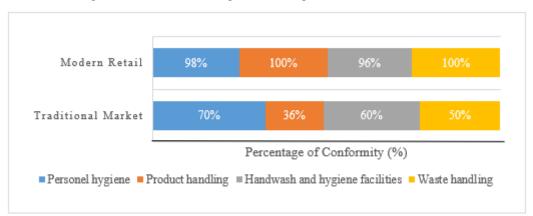
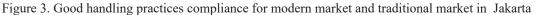


Figure 2. Good distribution practices compliance for TPHU in Jakarta



values of *Salmonella spp.* and probability of occurrence across various aspects. Monte Carlo simulation was used to iteratively generate random data based on limited data points, data distribution assumption and selected iteration values (Lien *et al.* 2023).

Risk ranking based on risk probability and severity of aspect GDP implementation showed that the highest risk occurred in 9 aspect points (Table 5). In order to determine handling priority, sensitivity analysis was carried out for each highest risk in GDP related questions. GDP aspects that most influenced the risk of contamination in TPHU were (B9) the non-fulfillment of temperature of transportation from TPHU (Figure 4). Transportation vehicle used ice cubes as a cooling agent to maintain low temperatures. Ice could be useful as a coolant but it potentially became the source of bacterial contaminants such as Salmonella spp (Liao et al. 2023). Unhigienic source of ice cube and slow thawing process could increase amount of Salmonella (Rahayu et al.2017). Recommendation vehicle temperature during transportation should be lower than 10-14 °C (Hosseinnezhad et al. 2018). Sensitivity analysis for risk contaminants in Taiwanese Salty Chicken showed that temperature gave 50.91% contribution to the risk level of contaminant (Lien et al. 2023). Sanitation in processing areas and transportation vehicles was needed to minimize biofilms that formed by Salmonella spp colonization (Nidaullah 2017; Mkangara 2023).

Risk ranking based on risk probability and severity of aspect GHP implementation in traditional market showed that highest risk occurred in 14 aspect point (Table 6). GHP aspect that most influenced risk of contamination in traditional market were (E4) the unproper equipment's cleaning method in traditional market (Figure 5). Study in Taiwan showed that unproper cleaning and sanitation of equipment gave contributed to contaminant risk about 20.11% for cutting board and 18.64% for knife (Lien *et al.* 2023). In order to reduce risk of contamination, display

Table 5. Risk ranking based on probability and severity on GDP aspect in TPHU Jakarta

| Probability             | Severity                |   |               |
|-------------------------|-------------------------|---|---------------|
|                         | Minor (I)               | Moderate (II)                           | Serious (III) |
| Rare (I)                |                         |   |               |
| Possible (II)           |                         | A1, A4, B2,<br>B5, C1 (#10-16)          |               |
| Almost<br>certain (III) | A2, A3, B10<br>(#10-16) | A5, B1, B3,<br>B4, B6, B8, B9<br>(#1-9) | B7 (#1-9)     |

Note: Low risk, Medium risk, High risk, #Priority number

Table 6. Risk ranking based on probability and severity on GHP aspect in traditional market Jakarta

| Probability             | Severity  |   |                     |  |
|-------------------------|-----------|---|---------------------|--|
|                         | Minor (I) | Moderate (II)                                   | Serious (III)       |  |
| Rare (I)                |           |   |                     |  |
| Possible (II)           |           | D9,F2(#15-21)                                   |                     |  |
| Almost<br>certain (III) |           | D4,D7,E2,E3,E5,E<br>6,F1,F3,F4,G1,G2<br>(#1-14) | D6,E2,E4<br>(#1-14) |  |

Note: Low risk, Medium risk, High risk, #Priority number

facilities should combined with equipment that maintain temperature 40c or lower (Mkangara 2023). High occurrence of *Salmonella spp*. was found in poultry, contact surfaces of equipment and environmental not only in slaughterhouse processing plants but also wet market (Nidaullah *et al.* 2017). In order to protect product from cross contamination, it recommend to use plastic packaging while display the carcass in traditional market.

Risk mitigation to control *Salmonella* should involved many sectors such as government, private sector, small enterprise food handlers, consumers and academia. The stricter control system should be accompanied by

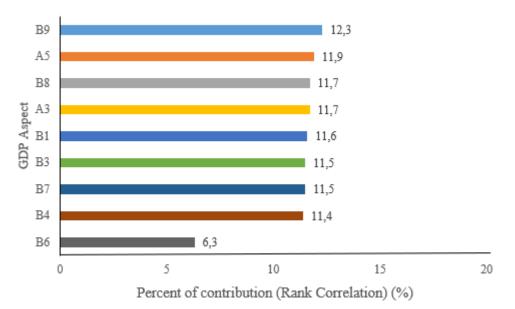
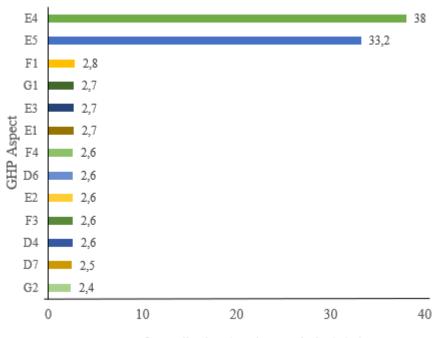


Figure 4. Sensitivity analysis for GDP aspect as critical control point in TPHU Jakarta

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Percent of contribution (Rank Correlation) (%)

Figure 5. Sensitivity analysis for GHP aspect as critical control point in traditional market Jakarta

periodic training for food handlers to communicate scientific information and raise awareness of salmonellosis to all interested parties (Ehuwa *et al.* 2021). Sensitivity analysis results could be referenced in choosing priority of risk mitigation (Jeong 2018). Sensitivity analysis described as rank correlation which the higher amount either negative or positive will indicate that those variables had bigger influence on the risk of contaminant (Lien *et al.* 2023). This study showed, that the utmost importance to protecting microbiology quality of chicken carcasses were cold chain distribution and sanitation of contacted surfaces that involved upstream to downstream. Educational programs for public about food safety and importance of cold chain distribution were needed to increase public acceptance of cold carcasses among Indonesian consumer.

# CONCLUSION

The highest prevalence of Salmonella spp. in chicken carcasses distributed across Jakarta province distribution chain occurred in the traditional market, TPHU, and modern retail, respectively. The highest GDP aspect fulfilled by the Jakarta TPHU was the documentation. The gap in GHP implementation between traditional markets and modern retail was product handling aspect. Risk ranking analysis for the high risk aspects showed TPHU had 9 GDP aspect points while the traditional market had 14 GHP aspect points that need to be controlled. In order to prioritize risk mitigation, a sensitivity analysis was carried out. Sensitivity analysis indicated the highest risk contributing factores during supply chain in Jakarta were were the non-fulfillment of temperature of transportation from TPHU (12.3% contribution) and the unproper equipment's cleaning method in traditional market (33.2% contribution).

Highest rank correlation shown that those aspects had high contribution to risk level of contaminant and could be the most critical aspects to be controlled.

### **CONFLICT OF INTEREST**

We certify that there is no conflict of interest with any financial, personal, or other relationships with other people or organization related to the material discussed in the manuscript.

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