MJFAS MALAYSIAN JOURNAL OF FUNDAMENTAL AND APPLIED SCIENCES
PRINT ISSN: 2289-5981 | ONLINE ISSN: 2289-599X

RESEARCH ARTICLE

Risk of acquiring listeriosis from consumption of chicken offal among high risk group

Ga Yarn Wai [a], John Yew Huat Tang [a], Nadiawati Alias [b], Chee Hao Kuan [c], Sur Guat Goh [d], Radu Son [e]

[a] *School of Food Industry, Faculty of Bioresources and Food Industry, Universiti Sultan Zainal Abidin (Besut Campus), 22200 Besut, Terengganu*
[b] *School of Agriculture and Biotechnology, Faculty of Bioresources and Food Industry, Universiti Sultan Zainal Abidin (Besut Campus), 22200 Besut, Terengganu*
[c] *School of Bioscience, Faculty of Health and Medical Sciences, Taylor's University Lakeside Campus, 47500 Subang Jaya, Selangor*
[d] *Food Division, National Public Health Laboratory, 47000 Sg. Buloh, Selangor*
[e] *Department of Food Science, Faculty of Food Science and Technology, Universiti Putra Malaysia, 43400 Serdang, Selangor*

* *Corresponding author: jyhtang@unisza.edu.my*

Article history
Received 8 Mac 2019
Revised 2 April 2019
Accepted 9 October 2019
Published Online 2 February 2020

Abstract

Listeria monocytogenes causes mild foodborne infection in healthy adults but serious complication in high risk group such as infant, pregnant women, immunocompromised and elderly. This study aims to estimate the probability of infection based on the survivability of *L. monocytogenes* in chicken offal, particularly chicken liver, gizzard and heart. A total of 20 µl of *L. monocytogenes* with inoculum size of 9.02 log CFU/ml was spiked on the chicken offal and stored at room temperature (28ºC) for 0, 1, 3, and 6 h. This experimental design was aimed to simulate the contaminated chicken offal stored at temperature danger zone. It was found that chicken offal supported the growth of *L. monocytogenes* that it significantly grows at 3 and 6 h incubation. Risk assessment was performed by combining this result with the data from our previous studies as well as data from government agencies and other studies. Dose-response model was used to estimate the probability of listeria infection per year. Immunocompromised was the highest risk group, followed by pregnant women, elderly and general population with probability of infection per year estimated at 3.78×10^{-3}, 1.76×10^{-4}, 5.68×10^{-8} and 5.75×10^{-9}, respectively. It can be concluded that high risk group requires extra caution in their food consumption in order to prevent listeriosis which can result in serious complication and death.

***Keywords*:** Risk assessment, *L. monocytogenes,* chicken offal, immunocompromised, pregnant women, elderly

INTRODUCTION

The genus Listeria includes six different species, *L. monocytogenes, L. ivanovii, L. innocua, L. welshimeri, L. seegligeri* and *L. grayi.* Both *L. monocytogenes* and *L. ivanovii* are pathogenic, but only *L. monocytogenes* is associated with humans and animal illness (Rodríguez-Lázaro *et al.*, 2004). *L. monocytogenes* has emerged as a foodborne pathogen and implicated with various food ranging from dairy products, fresh produce, meats and seafoods. High risk groups such as pregnant woman, immunocompromised individuals, young children and elderly are normally suffer from serious listeriosis infection through consumption of contaminated foods (EFSA, 2018; Norton and Braden, 2007). Cases of *L. monocytogenes* are rare as shown in the United States with estimated 0.26 cases per 100,000 individuals for the year of 2013 (Nastasijevic *et al.*, 2017). However, the severity of listeriosis accounted for about 30% more than other common pathogens which resulted in high hospitalization rates and mortality (Crim *et al.*, 2014). Listeria has also been implicated with outbreaks caused by contaminated ice cream (CDC, 2015a) and cheese (CDC, 2015b) and it caused the highest number of death in Europe in the period of 2008-2012 (EFSA and ECDC, 2014). Recently, *L. monocytogenes* has also implicated with outbreak related to pork products which cause four people hospitalized (CDC, 2018).

Foodborne diseases cause significant social and economic burdens from productivity losses, cost of medical treatment and hospitalization (Jeyaletchumi *et al.*, 2010). Risk analysis is an important model to improve food safety by determining the estimated risk of infection in a particular community exposed to the pathogen (Marvin *et al.*, 2009; Jeyaletchumi *et al*, 2010). Foodborne illness can occur from poor hygiene practices by food handlers as well as consumer. Those who do not practice proper food handling are potentially transmitted pathogens to food due to cross contamination from food contact surfaces (Soon, 2019). Study shown food handlers were not practicing the food safety knowledge they have acquired (Zanin *et al.*, 2017).

Chicken has been the major source of protein in Malaysia compared to beef, pork, duck and lamb. The per capita consumption of chicken has been steadily increasing from 38.59 kg in the year 2007 to 50.32 kg in year 2016 (DVS, 2018). Besides chicken meat, chicken offal also serves an important part of Malaysian food. Our previous studies have shown *L. monocytogenes* present in both chicken meat and chicken offal at an average of 20% and 24 %, respectively (Goh *et al.*, 2012; Kuan *et al.*, 2013). The results show a significant percentage of chicken meat and chicken offal were contaminated and posed risk of listeriosis

in a susceptible individual. In the present study, a risk assessment model was developed to determine the risk of acquiring listeriosis based on the survivability of *L. monocytogenes* on chicken offal.

METHODOLOGY

Statement of purpose

The goal of this study was to estimate the probability of listeria infection from chicken offal consumption based on the *L. monocytogenes* survivability on chicken offal. The calculation of risk estimate was based on data from this study, previous study (Goh *et al.*, 2014), and assumptions based on the data from other studies and government data. All the assumptions and uncertainties surrounding inputs were clearly stated. Risk assessment model was developed based on hazard identification, hazard characterization, exposure assessment and risk characterization. The probability of listeriosis from chicken offal consumption was calculated.

Hazard identification

L. monocytogenes can be found in a diverse environment, food products, animals and human (Buchanan *et al.*, 2017). Almost all cases of human listeriosis are foodborne which are contaminated with *L. monocytogenes* (Cossart and Bierne, 2001). Studies have found *L. monocytogenes* present in chicken offal, such liver and gizzard averaging from 26 to 63 (Arumugaswamy et al., 1994; Kuan *et al.*, 2013). Previous risk assessment study concluded that most listeriosis cases happened to the susceptible groups after the consumption of food that supports growth stored at inappropriate temperature for an extended period of time (US FDA/FSIS, 2003). Based on the prevalence study, chicken offal consumption has been reported to pose risk of listeriosis among high risk group (Kuan *et al.*, 2015).

Hazard characterization

L. monocytogenes is the bacteria species that are predominantly associated with human illnesses such as meningitis, encephalitis, and sepsis (Schlech and Acheson, 2000). *Listeriosis* normally causes serious complication among susceptible groups from neonates, infants, young children, pregnant women, elderly and immunocompromised patients (Buchanan *et al.* 2017, Jeyaletchumi *et al.*, 2010). A large number of *L. monocytogenes* ranging from 10 to 100 million of viable cells are required to cause illness in a healthy person. However, a relatively small number of the bacteria (0.1 – 10 million viable cells) is capable of causing illness in high risk groups (Bortolussi, 2008; Tauxe, 2001).

L. monocytogenes infections are generally non-invasive that resulted in mild, febrile illness. The non-invasive infection generally required ingestion of a large number of *L. monocytogenes* cells. Infected individuals will develop "flu-like" symptoms as well as fever and diarrhea (Aureli *et al.*, 2000). However, it can cause invasive listeriosis which resulted in severe symptoms and fatality. The cases of invasive listeriosis were found to be more prevalent among high risk groups (Buchanan *et al.*, 2017). Thus, the current risk assessment focused on invasive listeriosis among high risk groups. Dose-response relations model used to describe dose interaction, infectivity and the probability of infection from the exposure to the *L. monocytogenes* was adapted from FAO/WHO (2004). In this model, each microbial cell is assumed to have an equal chance of causing infection. Walls (2006) highlighted the need to dose-response model which help to address the need of human being exposed to the hazard in order to calculate the probability.

Dose response model FAO/WHO (2004) used for estimating the probability of listeriosis in the study was as follow:

$P_{illness} = 1- \exp^{-r*N}$

where r is a variable that describes the dose/response relationship and N is the amount of microbes ingested.

This model makes assumption that every ingested pathogen will have equal probability (variable "r") to cause infection within a particular population. The numbers of *L. monocytogenes* cells used to calculate the probability of infection among high risk group was around 5 log CFU per serving based on the average dose capable of causing illness in high risk groups (Bortolussi, 2008; Tauxe, 2001). This model predicts the likelihood of getting foodborne illness based on the amount of pathogens ingested. Thus, the likelihood of listeriosis will remain the same regardless of the amount of cells ingested once the infective dose is achieved. The dose-response relationship values for different high risk group were summarized in Table 1.

Table 1 Dose-response relationship for *L. monocytogenes* for different high-risk groups

Group	Dose-response relationship r-value	Reference
General population	8.5×10^{-16}	FAO/WHO (2004)
Immunocompromised	5.6×10^{-10}	Lindqvist and Westwoo (2000)
Pregnant women	2.6×10^{-11}	FDA/FSIS (2003)
Elderly, above 60 years of age	8.4×10^{-15}	FDA/FSIS (2003)

It was assumed that there were no seasonality and consumption pattern changes throughout the year (52 weeks). The probability of illness per year was calculated based on the formula below (Robertson *et al.*, 2005):

Probability of illness per year = $1 – (1 – P_{inf}\text{per individual})^{52}$

Exposure assessment

Exposure assessment requires data such as prevalence or concentration of microbes found in a specific food per serving (Walls, 2006). The exposure assessments for estimating the risk of *L. monocytogenes* infection were based on the survivability of *L. monocytogenes* on chicken liver, gizzard and heart performed in this study with reference to the previous study on the transmission of the bacteria from raw to cooked samples after contact with food contact surfaces (Goh *et al.*, 2014).

Survivability of L. monocytogenes on chicken liver, gizzard and heart

L. monocytogenes growth on chicken liver, gizzard and heart were determined to simulate its survival patterns in cases of cross-contamination due to poor food handling. The level of *L. monocytogenes* on chicken offal will affect the amount being transfer to the cooked chicken offal via food contact surfaces (Goh *et al.*, 2014).

- *Preparation of L. monocytogenes inoculum*

L. monocytogenes ATCC 7944 was revived in 10 ml of Tryptic Soy Broth (TSB) (Merck, Germany) and incubated at 37°C for 24 h. The revived culture was streaked on PALCAM agar (Merck, Germany) and incubated at 37°C for 24 h to obtain a purified culture. Pure *L. monocytogenes* culture was grown in TSB (Merck) in a condition described earlier before the culture was harvested for inoculum preparation. The inoculum of *L. monocytogenes* was determined using spectrophometer at 600 nm wavelength to give OD 0.99 which corresponds to 9.20 log CFU/ml.

- *Survival determination of L. monocytogenes on chicken offal*

The survival determination study was carried out as described in our previous study with modification (Tang *et al.*, 2014). Three grams of chicken liver, gizzard and heart were placed in universal bottles meant for different holding time (0, 1, 3, and 6 h). A total of 20 µl of inoculum was spiked onto the chicken offal and incubated at 28°C. *L. monocytogenes* enumeration was performed using a spread plate method on PALCAM agar (Merck, Germany). The experiments were done in triplicates.

- *Data analysis*

Data collected during the experiment was analyzed using SPSS 17.0 software. The data were analyzed using oneway ANOVA. The significance level was set at $p < 0.05$.

Transfer of L. monocytogenes to cooked sample

The transfer data of *L. monocytogenes* from raw to cooked sample was obtained from our previous study (Goh *et al.*, 2014). Poor food handling which let to contaminated food has been recognized as one of the main factors which lead to the foodborne illness or outbreak (Soon, 2019). Goh *et al.* (2014) reported the transfer rate of *L. monocytogenes* to cooked sample ranging from 0 to 87% depending on the type of cutting boards and conditions of the cooked samples (hot or cooled). An average of 44% was used in this study to cover all the possible scenarios. The serving size of chicken offal consumption in Malaysia was based on data reported by Kuan *et al.* (2015) at 125 g per week.

Population of the high- risk groups

Based on the Malaysia population in year 2018 (Department of Statistics Malaysia, 2019), it was estimated 127, 900 live births as of the year 2018 and it was assumed that each live birth was from one pregnant woman (Department of Statistics Malaysia, 2019). Thus, it was assumed there were 127, 900 pregnant women as of year 2018. Due to no data available for immunocompromised population in Malaysia, it is assumed 3.6% of the total population are immunocompromised which give around 1.2 million people fall within this group based on the estimation made by Laura (2008). Table 2 shows the population for each high- risk group in Malaysia.

Table 2 Population of high- risk groups in Malaysia.

Group	Year	Number of people	Reference
Total population	2018	32,600,000	Department of Statistics Malaysia (2019)
Immuno-compromised	NA	1,200,000	Laura (2008)
Pregnant women	2018	[a]127,900	Department of Statistics Malaysia (2019)
Elderly, above 60 years of age	2020	[b]2,347,200	Department of Statistics Malaysia (2006)

[a]It was assumed 3.6% of the total population are immunocompromised based on assumption made by Laura (2008).
[b]It was projected 7.2% of the total population are elderly based on the projection made by Department of Statistics Malaysia (2006).

RESULTS AND DISCUSSION

The growth of *L. monocytogenes* on chicken offal stored at room temperature (28ºC) over 6 h period was summarized in Table 3. *L. monocytogenes* were found to grow significantly in chicken liver, gizzard and heart at 3 and 6 hours. There was no significant difference at 1 h. The survival pattern of *L. monocytogenes* was similar among chicken liver, gizzard and heart. The amount of *L. monocytogenes* at cooked chicken offal was calculated based on the average bacteria level at 6 h and transfer rate of 44% which corresponds to 3.02 log CFU/g or 1.04×10^3 CFU/g.

The calculated amount of *L. monocytogenes* (1.04×10^3 CFU/g) was used to calculate the probability of infection as shown in Table 4. Based on the dose-response model (FAO/WHO, 2004), it was found the immunocompromised group has the highest probability of infection per year at 3.78×10^{-3} cases. This is followed by pregnant women 1.76×10^{-4}, elderly 5.68×10^{-8} and general population 5.75×10^{-9}.

It is generally recognized that *L. monocytogenes* is capable of growing at chilled or refrigerator temperature (Omac, Moreira and Castell-Perez, 2018; Millilo et al., 2012). *L. monocytogenes* was found to grow on chicken liver, gizzard and heart after 3 h storage at room temperature (28ºC) even though it is known to be psychophilic bacteria. The finding is line with other studies that *L. monocytogenes* was capable of growing in various food stored at 25ºC. *L. monocytogenes* was reported to grow 4 – 6 log CFU/g on celery and chicken salad stored at 25ºC over 4 days period (Sahu et al., 2017). Studies have found significant *L. monocytogenes* contamination in chicken offal (Arumugaswamy et al., 1994; Kuan et al., 2013). Thus, the growth of *L. monocytogenes* on chicken offal at room temperature will increase the risk of cross contamination in the kitchen environment. Storage of chicken offal at temperature danger zone or under cooking may allow *L. monocytogenes* to survive and increase in number (Lin et al., 2004).

Table 3 Growth of *Listeria monocytogenes* on chicken liver, gizzard and heart over 6 h incubation at room temperature.

Samples	Level of *Listeria monocytogenes* (log CFU/g)			
	0 h	1h	3h	6h
Liver	$6.08 \pm 0.19^{a,A}$	$6.24 \pm 0.06^{a,A}$	$6.61 \pm 0.18^{b,A}$	$7.16 \pm 0.08^{c,A}$
Gizzard	$5.97 \pm 0.46^{a,A}$	$6.17 \pm 0.37^{a,A}$	$6.44 \pm 0.16^{b,A}$	$7.23 \pm 0.48^{c,A}$
Heart	$5.21 \pm 0.04^{a,B}$	$5.31 \pm 0.08^{a,B}$	$5.56 \pm 0.09^{b,B}$	$6.18 \pm 0.07^{c,B}$

Data represent mean ± standard deviation of three replications.
a, b, c Data in the same row with different letters is different significantly ($p < 0.05$).
A, B, C Data in the same column with different letters is different significantly ($p < 0.05$).

Table 4 Risk assessment of *Listeria monocytogenes* in chicken offal consumption by high risk groups in Malaysia.

	General population	Immuno-compromised	Pregnant women	Elderly
Concentration				
Mean of *L. monocytogenes* (CFU/g)	1.04×10^3	1.04×10^3	1.04×10^3	1.04×10^3
Dose				
Serving size (g)	125	125	125	125
Dose (log CFU/serving)	1.30×10^5	1.30×10^5	1.30×10^5	1.30×10^5
Probability of infection per serving				
r-value	8.5×10^{-16}	5.6×10^{-10}	2.6×10^{-11}	8.4×10^{-15}
Probability of infection per serving	1.11×10^{-10}	7.28×10^{-5}	3.38×10^{-6}	1.09×10^{-9}
Probability of listeriosis cases per 100, 000 population				
Population	32,600,000	1,200,000	127,900	2,347,200
Probability per 100 000 population	2.87×10^{-4}	188.92	8.79	2.84×10^{-3}
Probability of infection per year				
Number of servings	52	52	52	52
Probability of infection per year	5.75×10^{-9}	3.78×10^{-3}	1.76×10^{-4}	5.68×10^{-8}

Chicken offal is not generally consumed raw but chicken liver tend to be undercooked in order to retain its juiciness. Undercooking and contamination due to poor handling practices will contribute to the risk of listeriosis among high risk groups. Poor food preparation practices have been identified as the contributing factor to these contamination (Soon, 2019).

Immunocompromised patients cover a variety of diseases that affect different components of the immune system. Immunocompromised patients can be categorized into congenital and acquired immunodeficiencies with the former is primary immunodeficiency diseases (PIDs) and the later cover range of disease from HIV/AIDs, cancers, hematologic and lymphatic malignancy, and others. Infection has been recognized as the main factor causing illness and death that required medical treatment (Meidani et al., 2014). From the risk estimates, it was found that this group is the most high risk group to get listeriosis. As such, this population group requires more vigilant in the selection of the uncontaminated food for consumption as low number of *L. monocytogenes* might cause serious illness. Brain infection cases due to *L. monocytogenes* has been reported in multiple sclerosis and stem cell transplant patients (Barocci et al., 2015; Tecellioglu et al., 2019).

Listeriosis is recognized as the most dangerous bacteria that cause death for fetuses and infants (Xu et al., 2017). A study reported women experiencing down-regulation of the immune system during pregnancy due to hormonal changes. Such changes increase the risk of foodborne illness among pregnant women (Smith, 1999). Pregnant women are generally 20 times greater risk of acquiring listeriosis than general population (Cook et al., 2018). Listeriosis can cause serious complication during pregnancy such as miscarriage, stillbirth, preterm labor, or death to the newborn (Xu et al., 2017). Listeriosis during pregnancy frequently occurs during the third trimester of pregnancy (Cook et al., 2018). This study found pregnant women were of higher risk of acquiring listeriosis than the elderly group was in agreement with a study done by Silk et al. (2012).

This risk assessment study proved high risk group should pay extra attention to this foodborne pathogen which can cause serious illness or even death. The incidence of listeriosis might be underreported due to mild clinical symptoms such as flulike fever and diarrhea. Microbiological examination was rarely performed for mild clinical symptoms and early pregnancy loses are generally not being investigated microbiologically (McLauchilin et al., 2004).

CONCLUSION

L. monocytogenes ability to grow in chicken offal stored at room temperature posed a significant risk of foodborne illness from inadequate cooking or contamination of cooked food. An immunocompromised group has the highest risk of listeriosis followed by pregnant women, elderly and general population. Risk assessment study based on the data from multiple sources from farm to table is important to provide information on the probability of illness resulted from contaminated food consumption by the high risk groups.

ACKNOWLEDGEMENT

The authors would like to thank the fund provider International Foundation of Sciences, Sweden (E/5237-2F). Special thanks to Universiti Putra Malaysia and Universiti Sultan Zainal Abidin, Malaysia for providing the facilities.

REFERENCES

Arumugaswamy, R. K., Rahmat Ali, G. R., Hamid, S. N. 1994. Prevalence of *Listeria monocytogenes* in foods in Malaysia. *International Journal of Food Microbiology* 23, 117–121.

Aureli, P., Fiorucci, G. C., Caroli, D., Marchiaro, B., Novara, O., Leone, L., Salmoso, S. 2000. An outbreak of febrile gastroenteritis associated with corn contaminated by *Listeria monocytogenes*. *New England Journal of Medicine* 342, 1236-1241.

Barocci, S., Mancini, A., Canovari, B., Petrelli, E., Sbriscia-Fioretti, E., Licci, A., D'Addesa, S., Petrini, G., Giacomini, M., Renzi, A., Migali, A., and Briscolini, S. 2015. *Listeria monocytogenes* meningitis in an immunocompromised patient. *New Microbiologica* 38(1), 113-118.

Bortolussi, R. 2008. Listeriosis: A Primer. *Canadian Medical Association Journal* 179(8), 795-797.

Buchanan, R. L., Gorris, L. G. M., Hayman, M. M., Jackson, T. C., Whiting, R. C. 2017. A review of *Listeria monocytogenes*: An update on outbreaks, virulence, dose-response, ecology, and risk assessments. *Food Control* 75, 1-13.

CDC (Centers for Disease Control and Prevention). 2015a. Multistate Outbreak of *Listeriosis* Linked to Blue Bell Creameries Products. http://www.cdc.gov/listeria/outbreaks/ice-cream-03-15 Accessed on 15 January 2019

CDC (Centers for Disease Control and Prevention). 2015b. Multistate Outbreak of *Listeriosis* Linked to Soft Cheeses (Final Update). http://www.cdc.gov/listeria/outbreaks/soft-cheeses-09-15/index.html Accessed on 15 January 2019

CDC (Centers for Disease Control and Prevention). 2018. Outbreak of *Listeria* linked to pork products. https://www.cdc.gov/listeria/outbreaks/porkproducts-11-18/index.html Assessed on 15 January 2019

Cook, J. L., Graves, L., Kirkham, C. 2018. Listeriosis in pregnancy: Practitioners' food safety counselling practices to pregnant women. *Journal of Obstetrics and Gynaecology Canada* 40(9), 1139-1147.

Cossart, P., Bierne, H. 2001. The use of host cell machinery in the pathogenesis of *Listeria monocytogenes*. *Current Opinion in Immunology 13*(1), 96-103.

Crim, S. M., Iwamoto, M., Huang, J. Y., Griffin, P. M., Gilliss, D., Cronquist, A. B. et al. 2014. Incidence and trends of infection with pathogens transmitted commonly through food. Foodborne Diseases Active Surveillance Network, 10 U.S. sites, 2006- 2013. *Morbidity and Mortality Weekly Report* 63(15), 328-32.

Department of Statistics Malaysia. 2016. Population projection (revised), Malaysia, 2010 – 2040. https://www.dosm.gov.my/v1/index.php?r=column/cthemeByCat&cat=118&bul_id=Y3kwU2tSNVFDOWp1YmtZYnhUeVBEdz09&menu_id=L0pheU43NWJwRWVSZklWdzQ4TlhUUT09 Assessed on 15 February 2019.

Department of Statistics Malaysia. 2019. Population and Demography. https://www.dosm.gov.my/v1/index.php?r=column/ctwoByCat&parent_id=115&menu_id=L0pheU43NWJwRWVSZklWdzQ4TlhUUT09 Assessed on 15 February 2019.

DVS (Department of Veterinary Services). 2018. Malaysia: Per Capita Consumption of Livestock Products, 2007-2016. http://www.dvs.gov.my/dvs/resources/user_1/DVS%20pdf/Perangkaan%202015%202016/page_10.pdf Assessed on 15 January 2019

EFSA (European Food Safety Authority) and ECDC (European Centre for Disease Prevention and Control). 2014. The European Union summary report on trends and sources of zoonoses, zoonotic agents and foodborne outbreaks in 2012. EFSA Journal 12 (2): 3547. http://www.efsa.europa.eu/en/efsajournal/doc/3547.pdf Assessed on 15 January 2019

EFSA (European Food Safety Authority). 2018. Listeria infection increases in vulnerable groups. https://www.efsa.europa.eu/en/press/news/180124 Assessed on 15 January 2019

FAO/WHO. 2004. Risk assessment of *Listeria monocytogenes* in ready-to-eat foods. Technical Report. http://www.fao.org/3/a-y5394e.pdf Assessed on 15 February 2019.

FDA/FSIS. 2003. Quantitative assessment of the relative risk to public health from foodborne Listeria monocytogenes among selected categories of ready-to-eat foods. https://www.fda.gov/downloads/Food/FoodScienceResearch/UCM197330.pdf Assessed on 15 February 2019.

Goh, S. G., Kuan, C. H., Loo, Y. Y., Chang, W. S., Soopna, P., Tang, J. Y. H., Nakaguchi, Y., Nishibuchi, M., Afsah-Hejri, L., Son R. 2012. *Listeria monocytogenes* in retailed raw chicken meat in Malaysia. *Poultry Science* 91, 2686-2690.

Goh, S. G., Afsah-Hejri, L., Kuan, C. H., Loo, Y. Y., Lye, Y. L., Chang, W. S., Soopna, P., Shahril, N., Tang, J. Y. H., Yaya, R., Nakaguchi, Y., Nishibuchi, M., Son R. 2014. Transmission of *Listeria monocytogenes* from raw chicken meat to cooked chicken meat through cutting boards. *Food Control* 37, 51-55.

Jeyaletchumi, P., Tunung, R., Margaret, S. P., Chai, L. C., Son, R., Farinazleen, M. G., Cheah, Y. K., Nishibuchi, M., Nakaguchi, Y., Pradeep, K. M. 2010. Assessing the risk of acquiring listeriosis from consumption of minimally processed vegetables using a step-wise risk assessment. *Asian Journal of Food and Agro-Industry* 3(6), 587-596.

Kuan, C. H., Goh, S.G., Loo, Y. Y., Chang, W. S., Puspanadan, S., Tang, J. Y. H., Nakaguchi, Y., Nishibuchi, M., Mahyudin, N. A., Radu, S. 2013. Prevalence and quantification of *Listeria monocytogenes* in chicken offal at the retail level in Malaysia. *Poultry Science* 92(6), 1664-1669.

Kuan, C. H., Goh, S.G., Loo, Y. Y., Chang, W. S., Lye, Y. L., Puspanadan, S., Tang, J. Y. H., Mahyudin, N. A., Nishibuchi, M., Nakaguchi, Y., Son, R. 2015. Risk assessment of acquiring listeriosis from consumption of chicken offal in Selangor, Malaysia. *International Food Research Journal* 22(4), 1711-1718.

Laura, H. K. 2008. The growing number of immunocompromised. Bulletin of the Atomic Scientists. https://thebulletin.org/2008/01/the-growing-number-of-immunocompromised/ Accessed on 15 February 2019.

Lin, Y. T., Labbe, R. G., Shetty, K. 2004. Inhibition of *Listeria monocytogenes* in fish and meat systems using oregano and cranberry synergies. *Applied and Environmental Microbiology* 70, 5672–5678.

Lindqvist, R., Westoo, A. (2000). Quantitative risk assessment for *Listeria monocytogenes* in smoked or gravid salmon and rainbow trout in Sweden. *International Journal of Food Microbiology* 58, 181-196.

Marvin, H. J. P., Kleter, G. A., Frewer, L. J., Cope, S., Wentholt, M. T. A. and Rowe, G. 2009. A working procedure for identifying emerging food safety issues at an early stage: Implications for European and international risk management practices. *Food Control* 20, 345-356.

McLauchilin, J., Mitchell, R. T., Smerdon, W. J., and Jewell, K. 2004. *Listeria monocytogenes* and listeriosis: a review of hazard characterization for use in microbiological risk assessment of foods. *International Journal of Food Microbiology* 92, 15-33.

Meidani, M., Naeini, A. E., Rostami, M., Sherkat, R., and Tayeri, K. 2014. Immunocompromised patients: Review of the most common infections happened in 446 hospitalized patients. *Journal of Research in Medical Sciences* 19(Supp 1), S71-S73.

Milillo, S. R., Friedly, E. C., Saldivar, J. C., Muthaiyan, A., O'bryan, C., Crandall, P. G., Johnson, M. G., Ricke, S. C. 2012. A review of the ecology, genomics, and stress response of Listeria innocua and Listeria monocytogenes. *Critical Review in Food Science and Nutrition* 52 (8), 712-725.

Nastasijevic, I., Milanov, D., Velebit, B., Djordjevic, V., Swift, C., Painset, A., Lakicevic, B. 2017. Tracking of *Listeria monocytogenes* in meat establishment using Whole Genome Sequencing as a food safety management tool: A proof of concept. *International Journal Food Microbiology* 257, 157-164.

Norton, D. M., Braden, C. R. 2007. Foodborne listeriosis. In E. T. Ryser, & E. H. Marth (Eds.), Listeria, listeriosis, and food safety. New York: CRC Press. 305-356.

Omac, B., Moreira, R. G., Castell-Perez, E. 2018. Quantifying growth of cold-adapted *Listeria monocytogenes* and *Listeria innocua* on fresh spinach leaves at refrigeration temperatures. *Journal of Food Engineering* 224, 17-26.

Robertson, L. J., Greig, J. D., Gjerde, B., Fazil, A. 2005. The potential for acquiring cryptosporidiosis or giardiasis from consumption of mung bean sprouts in Norway: a preliminary step-wise risk assessment. *International Journal of Food Microbiology* 98, 291-300.

Rodríguez-Lázaro, D., Hernández, M., Pla, M. 2004. Simultaneous quantitative detection of *Listeria* spp. and *Listeria monocytogenes* using a duplex real-time PCR-based assay. *FEMS Microbiology Letters* 233, 257–267.

Sahu, S. N., Kim, B., Ferguson, M. S., Zink, D. L., Datta, A. R. 2017. Growth potential of *Listeria monocytogenes* in artificially contaminated celery and chicken salad. *Food Control* 73, 1229-1236.

Schlech, W. F. III, Acheson, D. 2000. Foodborne listeriosis. *Clinical Infectious Disease* 31(3), 770-775.

Silk, B. J., Date, K. A., Jackson, K. A., Pouillot, R., Holt, K. G., Graves, L. M., et al. 2012. Invasive listeriosis in the Foodborne Diseases Active Surveillance Network (FoodNet), 2004-2009: Further targeted prevention needed for higher-risk groups. *Clinical Infectious Diseases* 54(suppl. 5), S396-S404.

Smith, J. L. 1999. Foodborne infections during pregnancy. *Journal of Food Protection* 62, 818-829.

Soon, J. M. 2019. Finger licking good? An observational study of hand hygiene practices of fast food restaurant employees and consumers. *British Food Journal* https://doi.org/10.1108/BFJ-07-2018-0420

Tang, J. Y. H., Mohd-Noor, N. H., Mazlan, N., Yeo, C. C., Abu-Bakar, C. A., and Radu, S. 2014. Survival of *Vibrio cholerae* O1 and *Vibrio parahaemolyticus* in fried and boiled Malaysian fish sausage. *Food Control* 41, 102-105.

Tauxe. R. V. 2001. Food Safety and Irradiation: Protecting the Public from Foodborne Infections. *Emerging Infectious Diseases* 7(3), 516-521.

Tecellioglu, M., Kamisli, O., Kamisli, S., Erdogmus, U. A., and Özcan, C. 2019. *Listeria monocytogenes* rhombencephalitis in a patient with multiple sclerosis during fingolimod therapy. *Multiple Sclerosis and Related Disorders* 27, 409-411.

U. S. Department of Health and Human Services, Center for Food Safety and Applied Nutrition, Food and Drug Administration and U.S. Department of Agriculture Food Safety and Inspection Service (U.S. FDA/FSIS). (2003). Quantitative assessment of relative risk to public health from foodborne *Listeria monocytogenes* among selected categories of ready-to-eat foods. http:// www.fda.gov/downloads/Food/FoodScienceResearch/UCM197330.pdf Accessed 10 February 2019

Walls, I. 2006. Role of quantitative risk assessment and food safety objectives in managing *Listeria monocytogenes* on ready-to-eat meats. *Meat Science* 74, 66-75.

Xu, W., Cater, M., Gaitan, A., Drewery, M., Gravois, R., Lammi-Keefe, C. J. 2017. Awareness of *Listeria* and high-risk food consumption behavior among pregnant women in Louisiana. *Food Control* 76, 62-65.

Zanin, L. M., de Cunham, D. T., de Rosso, V. V., Capriles, V. D., Stedefeldt, E. 2017. "Knowledge, attitudes and practices of food handlers in food safety: an integrative review", *Food Research International* 100(1), 53-62.