



UNIVERSITI PUTRA MALAYSIA

**PREVALENCE, RISK AND ANTIBIOGRAM OF *Listeria*
Monocytogenes IN RAW CHICKEN MEAT**

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**PREVALENCE, RISK AND ANTIBIOGRAM OF *Listeria monocytogenes*
IN RAW CHICKEN MEAT**

By

GOH SUR GUAT

**Thesis Submitted to the School of Graduate Studies, Universiti Putra
Malaysia, in Fulfilment of the Requirements for the Degree of Master of
Science**

October 2013

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**Dedicated to my beloved parents, siblings and friends for their eternity
love and endless support**

Abstract of thesis presented to the Senate of Universiti Putra Malaysia in
fulfilment of the requirement for the degree of Master of Science

**PREVALENCE, RISK AND ANTIBIOGRAM OF *Listeria monocytogenes*
IN RAW CHICKEN MEAT**

By

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October 2013

Chairman: Professor Son Radu, PhD

Faculty: Food Science and Technology

Chicken is a popular food in Malaysia. Consumption of poultry products has been increasing in the past decades. However, the contamination levels of *Listeria monocytogenes* (*L. monocytogenes*) in retailed chicken meat remain uncertain. This research was conducted to study the prevalence and antibiotic resistance of *L. monocytogenes* in raw chicken meat and effects of clove extracts against *L. monocytogenes* isolates. This study also aimed to determine transmission of *L. monocytogenes* from raw to cooked chicken meat via cutting boards as well as the risk of acquiring listeriosis through consumption of raw chicken meat.

A total of 210 samples were purchased from three hypermarkets and three wet markets in Selangor from September 2011 to January 2012. Most Probable Number (MPN) method had been used together with Polymerase Chain Reaction (PCR) and conventional plating for detection of *L.*

monocytogenes in raw chicken meat. Prevalence of *L. monocytogenes* in raw chicken meat was found to be 20%, in which the prevalence of *L. monocytogenes* in samples purchased from hypermarkets (25.71%) was higher than those of wet markets (14.29%). *L. monocytogenes* was detected in chicken breasts, drumsticks and thigh at prevalences of 42.03%, 11.27% and 7.14% respectively. MPN-PCR showed higher sensitivity compared to MPN-plating. By using MPN-PCR, the prevalence of *L. monocytogenes* in chicken meat was 20%, while MPN-plating could only detect 9.52%. The density of *L. monocytogenes* found in the samples ranged from <3.0 to 16 MPNg⁻¹.

Twenty-three isolates of *L. monocytogenes* were recovered from raw chicken meat. All isolates showed resistance to at least one antibiotic tested. Eleven isolates (47.83%) were resistant to at least 8 antibiotics. The isolates showed highest resistance against chloramphenicol (69.57%) followed by tetracycline (60.87%). Resistance of *L. monocytogenes* isolates towards antibiotic demonstrates the importance of identifying novel antimicrobial agent to inactivate *L. monocytogenes* in food before consumption. Thus, clove extracts was used to inhibit the *L. monocytogenes* isolates. Clove extracts showed inhibitory effect on *L. monocytogenes* isolates. MIC (Minimal inhibitory concentration) and MBC (Minimal bactericidal concentration) of clove extracts ranged from 156.25 ± 0.00 to 1041.67 ± 360.84 and 520.83 ± 180.42 to 4166.67 ± 1443.38 µg/ml, respectively.

A simulation study was conducted to determine the transmission of *L. monocytogenes* from raw to cooked chicken meat via polyethylene and wooden cutting boards. *L. monocytogenes* was found to contaminate cooked chicken meat regardless of the temperature of the chicken meat (room temperature or boiling temperature) and the material of the cutting boards when it is used within 30 min of contamination.

Stepwise risk assessment was used to determine the risk of acquiring listeriosis by consumption of raw chicken meat. An estimate of worst case scenario showed that the risk of acquiring listeriosis for healthy population was 0.008 per 100,000 populations annually. The risk estimates were higher for AIDS patients (6.670 per 100,000), Diabetes Mellitus Type II patients (0.190 per 100,000) and elderly (0.060 per 100,000).

Presence of *L. monocytogenes* in raw chicken meat is unavoidable, thus, consumers should be aware of the significance of *L. monocytogenes* to human health and the ways to prevent its contamination.

Abstrak tesis yang dikemukakan kepada Senat Universiti Putra Malaysia
sebagai memenuhi keperluan untuk ijazah Master Sains

**PREVALENS, RISIKO DAN ANTIBIOGRAM *Listeria monocytogenes*
PADA AYAM MENTAH**

Oleh

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Ayam adalah makanan popular di Malaysia. Pengambilan produk daging ayam telah meningkat pada dekad lalu. Walau bagaimanapun, tahap pencemaran *Listeria monocytogenes* (*L. monocytogenes*) dalam daging ayam masih tidak diketahui. Matlamat kajian ini adalah untuk menentukan prevalen dan rintangan *L. monocytogenes* dalam daging ayam mentah terhadap antibiotik serta kesan ekstrak pencilan-pencilan *L. monocytogenes* terhadap cengkih. Kajian ini juga mengkaji pemindahan *L. monocytogenes* melalui papan pemotong polietilena dan kayu, dan juga risiko dijangkiti wabak listeriosis daripada pengambilan daging ayam mentah.

Sejumlah 210 sampel dibeli dari tiga pasaraya dan tiga pasar di kawasan Selangor dari September 2011 sehingga Januari 2012. Kaedah Jumlah Paling Mungkin (MPN) telah digunakan bersama dengan Tindak Balas Berantai Polimerasi (PCR) dan piring untuk mengesan *L. monocytogenes* dalam daging

ayam mentah. Kekerapan kehadiran *L. monocytogenes* dalam daging ayam ialah 20%, di mana kekerapan *L. monocytogenes* dalam daging ayam dibeli dari pasaraya adalah lebih tinggi (25.71%) daripada yang dibeli dari pasar basah (14.29%). *L. monocytogenes* dikesan pada dada ayam, betis ayam dan paha ayam masing-masing pada kekerapan 42.03%, 11.27% dan 7.14%. MPN-PCR menunjukkan kesensitifan yang lebih tinggi berbanding dengan piring MPN. Dengan menggunakan kaedah MPN-PCR, kekerapan *L. monocytogenes* dalam daging ayam adalah 20%, manakala hanya 9.52% dikesan sekiranya piring MPN digunakan. Kepadatan *L. monocytogenes* yang dikesan dalam daging ayam berada pada julat <3.0 hingga 16 MPNg⁻¹.

Dua puluh tiga pencilan *L. monocytogenes* telah dipencilkan daripada daging ayam mentah. Semua pencilan tersebut menunjukkan kerintangan terhadap sekurang-kurangnya sejenis antibiotik yang diuji. Sebelas (47.83%) pencilan menunjukkan kerintangan terhadap lapan antibiotik yang diuji. Pencilan-pencilan ini menunjukkan tahap kerintangan tertinggi terhadap kloramfenicol (69.57%), diikuti dengan tetrasiklin (60.87%). Rintangan pencilan-pencilan *L. monocytogenes* terhadap antibiotik menunjukkan kepentingan untuk menyelidik agen antimikrob baru untuk menyahaktif *L. monocytogenes* dalam makanan sebelum pengambilan. Oleh itu, kesan rintangan ekstrak cengkih terhadap pencilan-pencilan *L. monocytogenes* telah dikaji. Ekstrak cengkih telah menunjukkan kesan rintangan kepada pencilan-pencilan *L. monocytogenes*. Kepekatan merencat minimum (MIC) dan

kepekatan bakteriasid minimum (MBC) ekstrak cengkih berada dalam julat 156.25 ± 0.00 ke 1041.67 ± 360.84 dan 520.83 ± 180.42 ke 4166.67 ± 1443.38 $\mu\text{g/ml}$, masing-masing.

Satu kajian simulasi telah dijalankan untuk menilai pemindahan *L. monocytogenes* dari daging ayam mentah ke daging ayam masak melalui papan pemotong polietilena dan kayu. Didapati *L. monocytogenes* boleh mengkontaminasi daging ayam masak tanpa mengira suhu (suhu bilik atau suhu mendidih) dan jenis bahan papan pemotong yang digunakan dalam tempoh 30 min selepas ia dikontaminasi.

Penilaian risiko berperingkat (stepwise risk assessment) telah digunakan untuk menentukan risiko dijangkiti wabak listeriosis dengan pengambilan daging ayam. Taksiran bagi senario paling buruk menunjukkan risiko dijangkiti wabak listeriosis untuk populasi yang sihat adalah 0.008 per 100,000 populasi setiap tahun. Anggaran risiko adalah lebih tinggi bagi pesakit AIDS (6.670 per 100,000), Diabetes Melitus jenis II (0.190 per 100,000) dan golongan tua (0.060 per 100,000).

Kehadiran *L. monocytogenes* dalam daging ayam mentah tidak dapat dielakkan, oleh itu, pengguna harus menyedari kepentingan *L. monocytogenes* terhadap kesihatan manusia dan cara-cara untuk mengelakkan kontaminasinya.

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APPROVAL

I certify that a Thesis Examination Committee has met on 23 October 2013 to conduct the final examination of GOH SUR GUAT on her thesis entitled “Prevalence, Risk and Antibigram of *Listeria monocytogenes* in Raw Chicken Meat” in accordance with the Universities and University Colleges Act 1971 and the Constitution of the Universiti Putra Malaysia [P.U.(A) 106] 15 March 1998. The Committee recommends that the student be awarded the degree of Master of Science.

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DECLARATION

I declare that the thesis is my original work except for quotations and citations which have been duly acknowledged. I also declare that it has not been previously, and is nor concurrently, submitted for any other degree at Universiti Putra Malaysia or at any other institution.

GOH SUR GUAT

Date: 23 October 2013

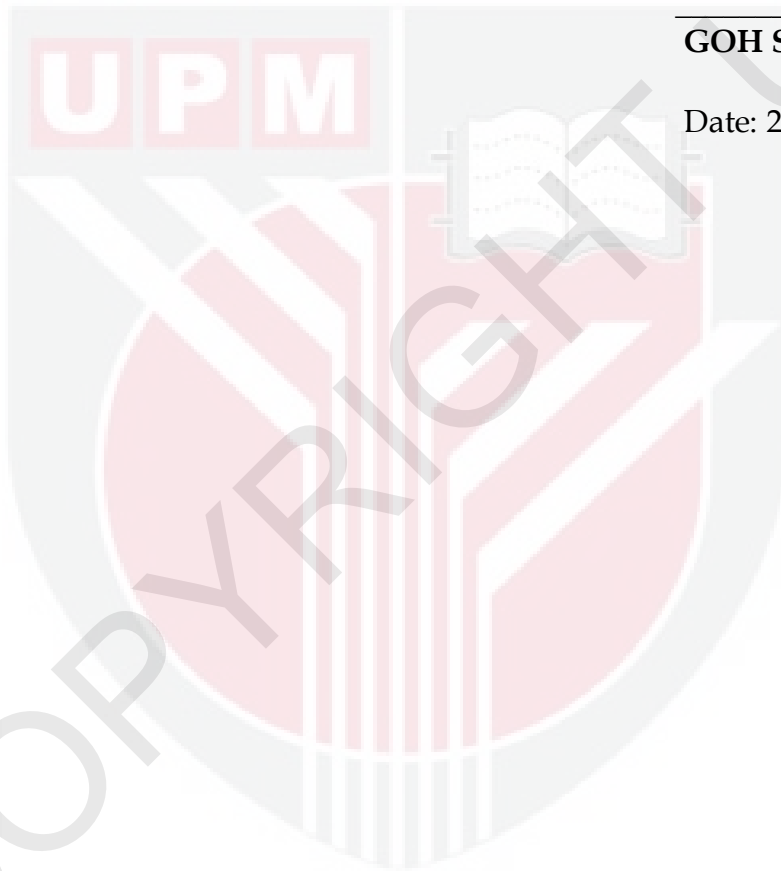


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