CHARACTERIZATION OF *LISTERIA MONOCYTOGENES* ISOLATED FROM FOODS

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AUGUST 2012

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2. Letter of Offer (Research Grant)

Surat Kami 600-RMI/ST/FRGS 5/3/Fst (20/2008)
Tarikh 10 Oktober 2008

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Tuan/Puan,

TAJUK PROJEK FRGS: CHARACTERIZATION OF LISTERIA MONOCYTOGENES ISOLATED FROM FOODS

Dengan segala hormatnya perkara di atas adalah dirujuk.

Sukacita dimaklumkan Jabatan Kementerian Pengajian Tinggi Malaysia telah meluluskan permohonan projek penyelidikan tuan/puan bagi FRGS Fasa 1/2008 seperti tajuk di atas dengan keputusan seperti berikut -

"Disokong dengan peruntukan RM87, 000.00".

Kelulusan ini juga tertakluk kepada syarat-syarat seperti berikut -

1. Tempoh projek penyelidikan ini ialah 2 tahun iaitu bermula 15 September 2008 sehingga **15 September 2010**.

2. Kos yang diluluskan ialah sebanyak **RM87, 000.00** sahaja. Sila kemukakan bajet yang baru mengikut kos yang diluluskan dengan menggunakan borang Bahagian E-Budget/Belanjawan. Perbelanjaan hendaklah mengikut butiran Belanjawan yang telah diluluskan.

3. Semua pembelian bahan/peralatan adalah diminta agar tuan/puan mematuhi prosedur perbendaharaan di mana pembelian melebihi RM500.00 hendaklah mengemukakan sebutharga.

4 Tuan/Puan dikehendaki mengemukakan laporan prestasi secara ringkas 2 kali setahun dan juga laporan prestasi projek tahunan secara terperinci dengan menggunakan format yang ditetapkan.

5 Tuan/Puan dikehendaki mengemukakan laporan prestasi kewangan tahunan secara terperinci dengan menggunakan format yang ditetapkan.

6 Laporan Akhir mesti dihantar dalam tempoh 3 bulan selepas projek penyelidikan disiapkan dengan menggunakan format yang ditetapkan dan hantar ke RMI sebanyak 2 naskah dan 1 "softcopy" dalam bentuk CD.

7 Adalah diminta supaya tuan/puan merujuk dan mematuhi syarat-syarat seperti dalam Garis Panduan Permohonan Geran Penyelidikan FRGS agar penyelidikan tuan/puan berjalan dengan lancar.

8 Tuan/Puan perlu menandatangani Borang Perjanjian Penyelidikan dengan kadar segera kerana penggunaan geran hanya dibenarkan setelah perjanjian ditandatangani.

9 Borang prestasi, borang laporan prestasi projek tahunan, borang prestasi kewangan tahunan dan borang perjanjian boleh diperolehi dari laman web RMI (http://www.rmi.uitm.edu.my)

Sekian, untuk makluman dan perhatian tuan/puan.

"SELAMAT MENJALANKAN PENYELIDIKAN"

Terima Kasih.

Yang benar,

PROF. DR AZNI ZAIN AHMED
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5.2 Enhanced Executive Summary

(Abstract of the research)

Listeria monocytogenes is one of the major food-borne pathogen with an opportunistic character that is able to cause severe human listeriosis worldwide. The development of a surveillance and detection system concerning food safety in this country needs to be improved. Therefore, this study was conducted to provide a general baseline on the contamination of *L. monocytogenes* in various foods in the local markets in Selangor. A total of 140 raw and ready-to-eat food samples were analyzed by plating on selective PALCAM medium and the suspected *L. monocytogenes* colonies were confirmed at molecular level by MPN-PCR technique using primers specific for this food-borne pathogen. All of the 23 isolated strains yield a PCR product of 938 bp and 701 bp for 16s rRNA gene and *hylA* virulence gene respectively. The confirmed isolates were characterized for their intra-species discrimination according to reaction between antigen-antisera by serotyping technique and patterns generated by RAPD bands. Two screened RAPD primers demonstrating potentially useful banding patterns were selected and examined against *L. monocytogenes* isolates. Positive isolates were evaluated for their susceptibility to eight commonly used antibiotic agents by using disc diffusion assay. *L. monocytogenes* was detected in 8.57% of collected food samples which 33.3%, 25% and 13.3% were detected in burger, minced meat and sausage samples respectively. The highest resistances (100%) of *L. monocytogenes* isolates were demonstrated against ampicillin and penicillin G antibiotics, whereas high susceptibility was showed toward streptomycin (100%). The molecular approaches used in this study provide highly sensitive and specific results which were very easy and fast to perform. These findings also suggested that the contamination of *L. monocytogenes* in foods is relatively low. However, it highlighted the emergence of antibiotics multi-resistant *Listeria* in the present environment.

5.3 Introduction

5.3.1. Background of study

Listeria monocytogenes (*L. monocytogenes*) is a non-spore forming, gram-positive bacteria that can be found in chains or as a single rod [1]. Despite its ability to grow in scarce oxygen and various conditions [2], this psychrophilic intracellular bacterium [3] is temperature dependent where it can grow best at 30 to 37°C [1]. Compared to other foodborne pathogens, this bacteria can also survive in a refrigerated temperature for some time [4] hence, developing listeriosis [5].

Listeriosis can be non-invasive or invasive [6]. Non-invasive listeriosis can cause gastrointestinal discomfort among healthy people and is less severe than invasive listeriosis Invasive listeriosis however can be fatal especially among newborns, pregnant women, elderlies and people with low immune system. The degree of severity however is very much dependent on the immune system of the individuals.

The advancement of the PCR methods such as duplex real-time PCR [7], quantitative PCR (qPCR) [8] and most probable number PCR (MPN-PCR) [9] makes the detection and identification processes much faster than the conventional methods. Introduction of the virulence genes specific to the molecular methods help in distinguising the L. monocytogenes from other Listeria spp. [10]. The development of the phenotypic and genotypic subtyping methods such as multilocus sequence typing (MLST) [11], ribotyping [12, 13], restriction enzyme analysis (REA) [14] and pulsed-field gel electrophoresis (PFGE) [15, 16] helps in characterizing this food-borne