

CHAPTER 1 INTRODUCTION

1.1 Introduction

There are numerous worldwide incidences of food borne illnesses. All of these numbers combined were caused from more than two hundred fifty different food borne pathogens. For bacteria, there are five main species that contribute to all of these food borne outbreaks, starting with *Salmonella*, *Clostridium perfringens*, *Campylobacter*, *Escherichia coli*, and *Listeria monocytogenes* (Scallan et al., 2011). In all of these bacteria, *Listeria* inflicted the highest mortality, more than sixteen percent of hospitalized cases resulted in death. The reported listeria outbreaks showed that the main causes were from delay of detection and false negativity (Manolopolu et al., 2003; CBC news, 2008; Food poisoning attorney, 2011). These outbreaks can bring about serious economic and health damages. So to prevent these unfortunate situations, the isolation and detection of *L. monocytogenes* contamination were investigated. The idea is to identify the target microbe at the earlier stage of production as possible.

The same conventional method to isolate *Listeria* spp. that recommended by international organizations is composed of primary selective enrichment, secondary selective enrichment and selective agar plating. For the primary selective enrichment, there were several research groups reported the use of non selective enrichment media was able to improve *Listeria* recovering and detection (Kosonpisit et al., 2011; Sheridan et al., 1994b; Budu-Amoako et.al, 1992; Beumer et al., 1996; Walsh et al., 1998; Vaz-Velho et.al, 2001 and Nancy et.al, 2004). So, we eliminated the primary selective enrichment and replaced with non selective enrichment. Since there are a variety of non-selective enrichment media, the experiment was divided into conventional non-selective media (i.e., Nutrient Broth, Lactose Broth and Tryptic Soy Broth) and *Listeria*-specific

broth base media (i.e., Buffered *Listeria* Enrichment Broth base, Fraser Broth base and Palcam Broth base). Not only was the type of non-selective media explored, but also the optimal condition for the non-selective enrichment step was examined.

For the selective enrichment step, different organizations used different types and sequences of selective media to select *Listeria* spp. (Netten et al., 1989; IDF method, 1990; NMKL Method No.136, 2004). These selective media were able to cause the incidences of false negativity from the effect of their compositions (Patel and Beuchat, 1995; Donnelly, 2002). This research also emphasized on critical compositions of inhibitors in the conventional selective media. Improper concentration of selective agents is able to affect the growth of *Listeria* spp. in food samples. Interestingly, different selective media use different types and quantities of selective agents. So the interaction of each selective agent on the growth and selectivity of *Listeria* strain was investigated to optimize the combination of selective enrichment medium.

1.2 The Objectives

1. To propose alternative techniques to improve *Listeria* spp. amplification and isolation for enrichment step
2. To improve the effectiveness of enrichment media in fostering *Listeria* growth and selectivity
3. To understand the interactions of selective agents and their effect on *Listeria* growth and isolation

1.3 The Expected Outcomes

1. To develop effective enrichment protocol and improve *Listeria* growth and selectivity for industrial food application
2. To reduce enrichment period and cost by 10 fold comparing to the routine protocols
3. Also establish fundamental concept and knowledge of high throughput and rapid methods for industrial *Listeria* detection