Host Range of *Listeria* Prophages Induced from Lysogenic *Listeria* Isolates from Foods and Food-related Environments in Thailand

Hue Thi Kim Vu[1,2], Soottawat Benjakul[1], Varaporn Vuddhakul[3], and Kitiya Vongkamjan[1*]

[1]Department of Food Technology, Faculty of Agro-Industry, Prince of Songkla University, Hat Yai 90110, Thailand
[2]Department of Veterinary Hygiene and Food Safety, National Institute of Veterinary Research (NIVR), Dong Da, Hanoi, Vietnam
[3]Department of Microbiology, Faculty of Science, Prince of Songkla University, Hat Yai 90110, Thailand

**Corresponding author. E-mail: kitiya.v@psu.ac.th*

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ABSTRACT

Prophages have been shown to be associated with improving the survival and fitness of Listeria spp., especially Listeria monocytogenes-a fatal foodborne pathogen. Bioinformatics has revealed the presence of many prophages in Listeria genomes. However, understanding the distribution of prophages in Listeria isolates is limited to those that have no sequences available on the database. In this study, Mitomycin C induction was used to obtain prophages in free-floating form. Among 236 isolates from various sources, prophages were induced from 13/108 (12%) isolates of L. monocytogenes and 10/128 (7.8%) isolates of Listeria spp. Of 39 induced phages obtained, most phages were originated from foods. Phenotypic characterization was performed by the host range determination against 17 hosts representing 9 major serotypes of L. monocytogenes and 4 other species of Listeria. Induced phages were classified into three groups. The majority of phages (groups A and C) were host-specific-phages with the ability to lyse one to seven (<42%) hosts. However, five phages (group B) showed broader host range than phages in other groups, which could lyse 8–10 (47–59%) hosts. Both induced and isolated Listeria phages showed

a high ability to lyse strains of L.monocytogenes serotype 4, while the induced phages showed narrow host range compared to the isolated phages. Host range data allows the prediction of particular L. monocytogenes subtypes or Listeria species that could be affected by the induced phages, leading to gene transfer upon phage infection. Information obtained here is useful to understand the diversity and role of prophages in Listeria genomes.

Keywords: *Listeria* spp., *Listeria* prophage, Food processing, Phage host range

INTRODUCTION

Listeria monocytogenes is an important foodborne pathogen causing potentially fatal listeriosis infections with a high mortality rate of up to 30% (Swaminathan and Gerner-Smidt, 2007). The genus of *Listeria* comprises 17 species, including two pathogenic species *L. monocytogenes* and *L. ivanovii*. Previous studies reported that various types of foods could be contaminated with *L. monocytogenes* (Vongkamjan et al., 2015, 2016). This pathogen has been shown the ability to be well adapted and survive longer in specific environments (Palumbo and Williams, 1991; Tolvanen et al., 2008; Burgess et al., 2016). Prophages have been shown the linkage on providing greater survival and fitness of *Listeria* spp., especially *L. monocytogenes* (Orsi et al., 2008; Verghese et al., 2011).

Prophage is a phage-related sequence which has integrated into bacterial chromosome and become part of the bacterial genome. Prophages have been reported to be commonly present in the *Listeria* genomes (Nelson et al., 2004; Kuenne et al., 2013), for example, *L. monocytogenes* strains F6854, L99, HCC23, J0161 (Kuenne et al., 2013; den Bakker et al., 2013). *L. innocua* strain CLIP11262 harbored up to six prophage (-like) elements, including 5 prophages and 1 monocin (Nelson et al., 2004). Previous studies mostly applied bioinformatics analysis to search for prophage regions in the genome of *Listeria*. This approach restricts information on prophage diversity to those lysogens (prophage-carrying *Listeria*) that have no sequences available on the database.

Induction is a mechanism by which prophage can be induced to escape a dying host and enter the lytic cycle, producing a free-floating form called induced phage. In previous studies, the occurrence of prophages has been measured by induction from the lysogenic isolates (Jiang and Paul, 1996; Chen et al., 2006). Some antibiotics, UV radiation, sunlight, temperature and pressure have been previously reported as common inducing-agents for prophage induction (Jiang and Paul, 1996; López et al., 2014). However, mitomycin C has been regarded as the most popular agent to induce prophages and produce infective phages (Loessner et al., 1991; Verghese et al., 2011).