

## ABSTRACT

The demand of a food preservative has been increasing due to foodborne diseases due to foodborne pathogens. Moreover, there is also an issue on the health risks caused chemical food preservatives, further increasing demand on a safer food preservative. Recently, *Pediococcus acidilactici* has been gaining interest as a potential natural food preservative due to its ability to produce pediocin, a small peptide that exhibit antimicrobial activity. The antimicrobial activity of *P. acidilactici* is attributed to the metabolites it produces, which can be found in its cell-free supernatant (CFS). However, the metabolites production is effected by the incubation period of *P. acidilactici*. This study aims to determine the optimum incubation period of *P. acidilactici* that will result in CFS exhibiting the highest antimicrobial activity. In this study, CFS at various incubation period is subjected to antimicrobial activity testing against two foodborne pathogens, *Listeria monocytogenes* and *Staphylococcus aureus*. Through agar-well diffusion method, it is found that the optimum incubation period that results in CFS with highest antimicrobial activity varies depending on the pathogens, 44 hours for *L. monocytogenes* and 36 hours for *S. aureus*. It is also found that *P. acidilactici* produces CFS exhibiting highest antimicrobial activity during the stationary phase. CFS at T44 and T36 are also subjected to time-kill assay and antimicrobial activity characterization. Through time-kill assay, it can be seen that the CFS is able to suppress the growth of pathogens, having bactericidal effect towards *L. monocytogenes* and bacteriostatic effect towards *S. aureus*. From the characterization, it is seen that both organic acid and protein compounds present in the CFS are responsible for the antimicrobial activity of *P. acidilactici* CFS. Further studies can be done in order to better understand how the CFS interacts towards the pathogens.

**Keywords:** *Pediococcus acidilactici* cell-free supernatant, optimum incubation period, antimicrobial activity, antimicrobial characterization