Chapter 5

Conclusion, Original contributions and Recommendation

5.1 Conclusion

The effect of starter cultures including *P. pentosaceous*, *P. acidilactici*, *W. cibaria*, *L. plantarum*, *L. pentosus*, and *L. sakei* on the production of Thai fermented sausage. The use of starter cultures influenced the considered biochemical and sensory characteristics of the fermented sausage. It can be concluded as follows:

1. The results of microbiological analysis indicated that the dominance of lactic acid bacteria (LAB) could inhibit the growth of pathogens and spoilage. Sausage inoculated with *P. pentosaceus* was the significant highest population of LAB. Moreover, this batch also showed the lowest population of staphylococci/micrococci and yeast and mold.
2. Sausages inoculated with starters exhibited lower pH than the uninoculated sausage about 1.04 times. The decrease of pH was attributed to organic acid, mainly lactic acid production by LAB. The initial rate of lactic acid production and lactic acid yield in sausages inoculated with starter cultures was approximately 1.23 times higher than the control. Sausage inoculated with *P. acidilactici* showed the highest initial rate of lactic acid production and lactic acid yield.
3. The percentage of total heme pigments in fermented sausage inoculated with *P. acidilactici* presented the highest conversion ($p < 0.05$) to cured meat pigments during fermentation.
4. The use of LAB as starter cultures could accelerate proteolysis on Thai fermented sausage. Proteolysis was observed during fermentation by the reduction of myofibrillar and sarcoplasmic proteins and the increase in non-protein nitrogen (NPN) and total free amino acid. The highest increase in concentration of NPN and total free amino acid was obtained from sausages inoculated with *L. sakei* and *L. plantarum*, respectively. The result of SDS-PAGE profiles of sarcoplasmic proteins showed a similar

pattern of proteolysis in all sausages, while that of the inoculated sausages with *L. plantarum*, *L. pentsus* and *L. sakei* exhibited the most intense degradation of myofibrillar proteins, especially myosin and actin.

5. LAB had influence on flavour formation in fermented sausages. The addition of LAB to fermented sausages could enhance the formation of volatile compounds resulting in flavour improvement of the product. The concentration of volatile compound, 3-methyl-butanic acid, from Leu was higher in LAB inoculated samples than in control samples and was the principal flavor enhancing compound.
6. The sensory evaluations indicated that Thai fermented sausage inoculated with *P. acidilactici* had better scores for color, flavour, taste, texture, sourness, saltiness and overall preference than inoculation with other starter cultures. Therefore, *P. acidilactici* should be considered as a starter culture for fermented sausage in order to improve sensory characteristic. Comparing between sausage inoculated with *P. acidilactici* and commercial fermented sausage named S. Khonkhan, the acceptability found no significant difference between two batches.
7. Mathematical models were developed to fit the experimental data including the growth of LAB, the consumption of protein and glucose, and the production of NPN, lactic acid and formic acid concentration. The goodness of fit was evaluated non-parametrically by estimating the χ^2 statistic at 95% confidence interval. It signifies the quality of agreement between predicted and observed results of naturally fermented sausage (control) and sausage inoculated with *P. acidilactici*.

5.2 Original contributions

The most significant original contributions that were achieved in this study are summarized as follows:

1. Thai fermented sausage production could be improved by the addition of starter culture. The inoculation of Thai fermented sausages with LAB caused rapid decrease of pH, inhibition of growth of contaminant pathogen, and acceleration of proteolytic activity. In addition, it yielded higher concentration of end-products, i.e., cured meat pigment, flavour compounds, and lactic acid.
2. Agreement with chemical analysis, the sensory evaluation found that Thai fermented sausage inoculated with LAB, especially, *P. acidilactici* had better scores for colour, flavour, taste, texture, and sourness than uninoculated sausage (control).
3. The proposed kinetic models were validated by experimental data derived directly from Thai fermented sausage in its natural state. In previous studies of mathematical modeling of growth of LAB in food systems, the experimental data was obtained from growth of LAB in synthetic liquid media or food extracts under specific conditions due to complicated systems of meat. Moreover, several models have been suggested to describe the growth of microorganisms accompanied by sugar utilization. Despite that fact that there are proteins in meat product which can be consumed by microorganisms. Therefore, the proposed unstructured kinetic model was the first study relating to prediction of the growth of LAB, the consumption of protein and glucose, and the production of NPN, lactic acid and formic acid concentration during fermentation of Thai fermented sausage. The unstructured models could be applicable to sausage inoculated with *P. acidilactici* and control batches.

5.3 Recommendation

The following recommendations are offered for future study:

1. The future studies are to assess the mixture of functional starter cultures for the improvement of Thai sausage fermentation. The use of functional starter cultures represents a potential technique in improving and optimizing the sausage fermentation process. The fermented sausages also achieve tastier, safer and healthier products. Microorganisms involved in sausage fermentation use for a tasty product such as lactic acid bacteria, *Staphylococcus xylosus* and *Staph. carnosus*.
2. The use of bacteriocin-producing starter cultures, LAB, may contribute to food safety, prevention of food spoilage and reduction of undesirable toxic compounds such as biogenic amines. *L. fermentum* could be applied as new starter culture due to curing agents and colour formation. In addition, some strains of *Lactobacillus* such as *L. rhamnosus*, *L. sakei* and *P. acidilactici* are good candidates as probiotic cultures because they are beneficial to health. Therefore, it should be emphasized that the performance of a selected starter culture for quality characteristics.
3. Additionally, freeze drying is a well known procedure for the concentration and conservation of lactic acid bacteria. The most important criteria in developing a meat starter cultures are concentrated and preserved before used by freeze drying. The practical use of frozen cultures is the convenience of the actual handling of the cultures.
4. The modeled results of total protein concentration were slightly compared with the observed results. This may be due to protein measurement. The experimental data of total protein concentration verified with the proposed total protein model equation was determined by Bradford method. Bovine serum albumin (BSA) was used as a standard protein to establish a standard curve. Evidently, BSA cannot represent muscle proteins, myofibrillar and sarcoplasmic protein. Thus, the method of total protein measurement in meat product should be improved.