CHAPTER 4

CONCLUSION

The present study tried to highlight the importance of bacteriocins, peptides produced by lactic acid bacteria, and their potential as antimicrobial agents in food products. The work carried out may be summarised as follows:

- Various fermented food sources were screened for bacteriocin producing lactic acid bacterial strains. A novel lactic acid bacterial strain, identified as *Lactococcus garvieae* (LG1), was isolated from a fermented vegetable source producing a bacteriocin.

- The bacteriocin was purified by ultrafiltration, followed by preparative thin layer chromatography and RP-HPLC, which was a completely different method of purification compared to the conventional methods of purifying bacteriocins. This method proved to be more economical, less laborious and most of all loss of protein was minimised.

- N-terminal sequencing of the bacteriocin was carried out and it was concluded that the bacteriocin produced by LG1 might be a lantibiotic.
• Initial standardisation studies were carried out using one-dimensional single substitution method to evaluate the effect of various nutrients on the biomass and bacteriocin production.

• Response surface methodology (RSM) was used to determine the optimum response of the cells for the synthesis of a bacteriocin produced LG1 and biomass, under a range of nutrient conditions. The design of experiments chosen for this study was full factorial central composite design (Box- Wilson, 1951) for three independent variables to obtain the combination of values, which optimised the response.

• The effect of physical parameters such as pH and oxygen was studied on the bacteriocin and biomass production. Enhanced bacteriocin production and biomass was achieved by fed batch cultivation.

• The commercial value of this bacteriocin in the food industry was evaluated. The antimicrobial activity of nisin and the bacteriocin isolated from LG1 to increase the shelf life of paneer, a type of cottage cheese, was assessed. The bacteriocin produced by the isolate proved to be highly effective against Listeria monocytogenes and Staphylococcus aureus. This study showed that this bacteriocin was much more effective than the commercially available bacteriocin, nisin.
Possible directions for future work

- Structural elucidation and mode of action of the bacteriocin are two important areas to be investigated.

- On a more molecular level, ‘gene shuffling’ would prove to be a very interesting research field. Application of this technique could result in strains of greater efficacy and stability.

- Widening the antimicrobial spectrum and decreasing the susceptibility to proteolytic enzymes would make the bacteriocin therapeutically indispensable.

- Optimisation of a cost effective medium and developing a suitable purification method for the bacteriocin on an industrial scale, need to be delved into.