CHAPTER 3

OBJECTIVES

Microbial proteases are one of the essential virulence factors of pathogenic bacteria for the progression of a disease. SPATEs, which belong to the subfamily of autotransporters are secreted serine proteases of *Enterobacteriaceae* and are known to be strongly associated with diverse pathogenic bacterial isolates. Several studies have evidenced the distribution of SPATEs among the diarrheagenic *E. coli*. Sat and other subtypes of SPATEs were reported from clinical isolates of DAEC and EAEC. The distribution of SPATEs is also well studied among various pathotypes of ExPEC isolates. However, no study has yet been undertaken to show the association of SPATEs among the *E. coli* isolates causing neonatal septicaemia.

The objectives of our study are

(I) Phylogrouping and genotypic characterization of several virulence factors of *E. coli* strains from three distinct sources (a) blood of septicemic neonates (b) feces of healthy neonates and (c) ground water resources.

(II) Determination of the genotypic distribution of SPATEs with the generic primer pair for SPATEs and different subtypes of SPATEs among septicemic, fecal and environmental *E. coli* isolates. To compare the prevalence of SPATEs among the three different groups of *E. coli* isolates and to compare the presence of SPATEs and the other virulence factors among the three groups of *E. coli* isolates.

(III) Determination of the clonality of septicemic isolates to understand whether the occurrence of SPATEs among septicemic strains is independent of the clonal relatedness of the strains.

(IV) To study the association of SPATEs with *E. coli* isolates causing neonatal septicaemia in a suckling mouse model.

(V) Purification, identification and characterization of a protease from a clinical isolate of neonatal septicemic *E. coli* (NSEC).