

**TITLE:** *E. coli* DIVERSITY PRESENT IN THE DAIRY CATTLE ENVIRONMENT BY VIRULENCE GENES DETECTION AND PFGE TYPING TECHNIQUE.

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## **ABSTRACT**

*Escherichia coli* has been described as prevalent and highly pathogenic in environmental mastitis etiology. Molecular typing is a powerful tool that can provide information about genetic characteristics of microorganism responsible for this disease. The objective of this work was to evaluate the genetic diversity of *E. coli* present in the milk production environment through the molecular typing technique *Pulsed Field Gel Electrophoresis* (PFGE). Circulating clones causing bovine mastitis present in the milk and feces of the bovine as well as in the water used in the management of these animals was determined. 282 milk samples were collected from 94 lactating cows in three consecutive weeks in summer, winter, spring and autumn. Also were collected 94 samples of fecal material from these animals and water samples from nineteen different points related to the milk production line. Thus, 152 strains of *E. coli* were obtained through the phenotypic and proteomic analysis. The strains were investigated by virulence genes such as Intimin (*eaeA*), Shiga toxin (*stxI* and *stxII*), thermostable (*LT*) and thermostable (*ST*) enterotoxins, invasiveness (*ial*) and enteroaggregative *E. coli* (*eagg*). Furthermore, genes associated with adherence were analyzed as fimbria F1 (*fimH*), fimbria curli (*csgA*) and antigen 43 (*flu*). According to the genes evaluated, the *fimH*, *csgA* and *flu* genes were prevalent in strains isolated within the milk production environment being *fimH* and *csgA* the most detected genes in the 72.2% and 55.5% of the strains respectively. The *flu* gene was not detected in water samples. On the other hand, percentage of genes related to the production of toxins *eaeA*, *LT* and *stxI* were detected in the 11.1%. None of isolates presented *stxII*, *ST*, *ial*, *eagg* genes. Eighteen profiles based on the virulence pattern were determined and 18 strains were selected to PFGE technique, performing 16 different patterns. Two clusters with 100% homology, with 2 strains each other, were found, one of them clustered two strains isolated from milk and water that share the same virulence genes. The present study describes different genotypes in *E. coli*, with pili genes domination and shows that there is no predominant PFGE pattern and any association between these virulence genes and PFGE patterns.

**Keywords:** bovine mastitis, circulating clones, diversity, *Pulsed Field Gel Electrophoresis*

**Development Agency:** CAPES, FAPERJ