**Pathogenicity of *Escherichia coli* isolated from the equine uterus**

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*Escherichia coli* is one of the most common bacteria isolated from the equine uterus, representing 30-50% of all cases of infectious endometritis. *E. coli* isolates are recognized to have different virulence characteristics based on phylogenetic analysis. The goal of this study was to characterize *E. coli* isolates from the equine reproductive tract based on phylogenetic group, antibiotic susceptibility and *in vitro* biofilm production. The hypothesis was that the *E. coli* isolates could be classified into different phylogenetic groups that would correlate with their antibiotic resistance and biofilm propensity. *E. coli* isolates (n=43) were obtained from equine clinical cases in Lexington, Kentucky. Bacterial identification was first confirmed using MALDI-TOF. Antibiotic resistance was then determined using Kirby-Bauer disc diffusion for the following antibiotics: gentamicin, enrofloxacin, ceftiofur, amikacin, ampicillin, trimethoprim sulfamethoxazole, ticarcillin with clavulanic acid, and penicillin. *In vitro* biofilm production was evaluated by crystal violet staining and phylogenetic groups classified using multiplex PCR. Data were compared using Fisher’s exact test for antibiotic resistance and an unpaired t-test for biofilm formation. Results are reported as the mean ± SEM. Phylogenetic analysis showed that a majority of samples (p<0.05) could be classified into either B1 (n=22) or B2 (n=16) groups, with the remaining isolates belonging to group A (n=2) or were undetermined (n=3). There was a greater (p<0.05) percentage of isolates resistant to ticarcillin with clavulanic acid and ampicillin for group B2 (9 of 16 and 14 of 16, respectively) as compared to the B1 group (5 of 22 and 8 of 22, respectively). Biofilm formation was greater (p<0.05) in the B1 group (0.52 ± 0.09 OD₆₀₀) compared to the non-biofilm-forming B2 group (0.16 ± 0.01 OD₆₀₀). In summary, a majority of *E. coli* isolated from the equine reproductive tract were divided into two distinct groups (B1 and B2). Clinical B2 *E. coli* did not form a biofilm *in vitro*, but had greater antibiotic resistance as compared to the B1 group. Future studies will focus on differences in *in vivo* pathogenicity between B1 and B2 groups of *E. coli*.

**Keywords:** Equine, endometritis, *E. coli*, biofilm, antibiotic resistance