ABSTRACTS

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THE EFFECT OF THE STAGE OF THE SIMULATED OESTROUS CYCLE AND THE PRESENCE OF UROPATHOGENIC VIRULENCE FACTOR GENES ON THE ADHESION OF ESCHERICHIA COLI TO THE CANINE UTERINE EPITHELIUM IN VITRO

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Introduction - The adhesion of bacteria to host epithelial cells is an essential step in the establishment of a mucosal infection (4). Uropathogenic virulence factors (UVF) promote the attachment of Escherichia coli to the urinary tract epithelium in humans and dogs (1,3,7). The role of UVF in the adherence of E. coli to the uterine epithelium in bitches is not known. The stage of the oestrous cycle, however, has been shown to influence the attachment of E. coli to the canine uterine epithelium (5,6).

Aims - The aim of this study was to determine the effect of UVF genes and the stage of the simulated oestrous cycle on the adhesion of E. coli to the canine uterine epithelium in vitro.

Materials and Methods - Three strains of E. coli - P3, F8 and DH5α, were used. The P3 strain was isolated from a clinical case of pyometra and carried five uropathogenic virulence factor genes (pap, sfa, hlyA, cnf1 and fim) as determined by the polymerase chain reaction. The F8 strain was isolated from the faeces of a clinically healthy dog and did not carry any known UVF genes. The DH5α strain was a laboratory strain carrying no UVF genes and no plasmids. Twelve ovariectomised Greyhound bitches were allocated randomly into three equal groups (Groups A, B and C). Exogenous oestradiol benzoate and megestrol acetate were administered to induce simulated oestrus in bitches in Group A and simulated dioestrus in bitches in Group B (2). Group C bitches did not receive any exogenous hormone treatment and remained in simulated anoestrus. Uterine tissues were collected at necropsy from Group A bitches on day 4 of simulated oestrus, Group B bitches on day 10 of simulated dioestrus, and Group C bitches in simulated anoestrus. The samples from each of the uteri were randomly allocated to one of the four treatment groups (T1 (strain P3), T2 (strain F8), T3 (strain DH5α) and T4 (Hank’s balanced salt solution, control buffer)) and incubated at 38.5 °C for 60 minutes. The samples were then processed for histological examination. Light microscopy was used to determine the percentage of surface uterine epithelial cells with bound bacteria (percentage binding).

Results - Three hundred cells were counted in each of 5-7 tissue sections per treatment per animal. Strain P3 bound to a greater percentage of uterine epithelial cells than did strain F8 on day 10 of simulated dioestrus (6.73 ± 0.24 vs 0.83 ± 0.13, P<0.001) and in simulated anoestrus (1.11 ± 0.28 vs 0.23 ± 0.10, P<0.05). The binding of strain P3 and F8 on day 4 of simulated oestrus was similar (0.85 ± 0.09, 0.43 ± 0.05, P>0.05). The adherence of strain P3 was influenced by the stage of the simulated oestrous cycle (P<0.0001), being maximal (6.73%) during simulated dioestrus. The differences in percentage binding amongst various stages of the simulated oestrous cycle were not due to physiological changes in surface size of epithelial cells, as confirmed by histomorphometry. The results demonstrated that the possession of UVF genes significantly enhanced the attachment of E. coli to the canine uterine epithelium in vitro during simulated dioestrus and simulated anoestrus. This suggests these UVF are important in the pathogenesis of CEH/pyometra complex in the bitch.
References