## Analytic approach for endometrial polymorphonuclear cells cytology threshold for the definition of cytological endometritis in primiparous and multiparous dairy cows

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Cytological endometritis (CEM) is usually diagnosed based on proportion of polymorphonuclear cells (PMN) in endometrial cytology. Incidence of CEM varies greatly among studies, ranging from 12 - 45%, presumably reflecting diversity among herds, but potentially due to diagnostic criteria, including parity, threshold of PMN proportion, as well as interval from calving to cytology sample collection. Our objective was to examine whether primiparous and multiparous cows should be diagnosed for CEM by different criteria and timing, using a combination of selected reproductive performance outcomes [interval to first service and to pregnancy, pregnancy rate at 180 days-in-milk (DIM), first service conception rate, and number of AIs (artificial inseminations) to pregnancy]. Two endometrial cytobrush cytology samples were collected from Holstein dairy cows (n = 213; 133 multiparous, 80 primiparous), at 30 - 40 and at 60 - 70 DIM; slides prepared and stained with Diff-Quick and PMN proportions evaluated blindly. PMN proportion thresholds were set at > 1to  $\geq 10, \geq 15$ , and  $\geq 20\%$ , for diagnosis of CEM. Data were analyzed by Cox's proportional hazard model for time to first service and time to pregnancy (from calving and from first service), controlling for the effect of farm. Cytological endometritis in primiparous cows were best identified at 30 - 40 DIM, whereas in multiparous cows, CEM was best diagnosed at 60 - 70 DIM. In primiparous cows, a threshold of  $\geq$  7% at 30 - 40 DIM was associated with significant reductions in the majority of reproductive performance parameters analyzed (time to first service and to pregnancy, pregnancy rate at 180 DIM, first service conception rate, and number of AIs to pregnancy), with specificity, sensitivity and positive predictive value of 85.2, 41.2, and 59.2%, respectively. However, at 60 - 70 DIM, none of the PMN thresholds were associated with reduced reproductive performance in primiparous cows with CEM. In contrast, in multiparous cows, a threshold of  $\geq$  3% at 60 - 70 DIM was associated with significant reductions in the majority of the reproductive performance parameters analyzed, with specificity, sensitivity and positive predictive value of 83.1, 39.1, and 62%, respectively. However, at 30 - 40 DIM, none of the PMN thresholds were associated with reduced reproductive performance in multiparous cows with CEM. Differences between primiparous and multiparous cows could be related to metabolism, immune function, and uterine involution. In conclusion, diagnosis of CEM in primiparous and multiparous cows should be made using different PMN threshold criteria and at different intervals after calving.

Keywords: Dairy cows, endometritis, cytobrush, PMN threshold