



Original Research

Equine Infectious Endometritis—Clinical and Subclinical Cases



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ABSTRACT

Equine endometritis continues to be a major cause of subfertility and infertility in mares, and recently, the subclinical form of endometritis has been in focus. The purposes of this study were to investigate the prevalence of subclinical infectious endometritis, defined as mares without clinical symptoms indicating endometritis, but with bacterial growth and positive cytology from an endometrial sample, and the relation to clinical parameters before and after artificial insemination (AI). A total of 76 Standardbred mares submitted for AI were included in the study, in which a uterine sample was collected before (biopsy) and after AI (swab) for bacterial culture, cytology, and histology. The results showed that 28.6% of clinically normal mares, submitted for breeding with AI, had subclinical infectious endometritis before breeding. The endometrial edema score early in estrus was a strong diagnostic indicator of a subclinical infection with *Streptococcus equi* subsp. *zooepidemicus* (odds ratio, 5.48; $P < .0001$). An altered endometrial edema pattern in a mare showing increased edema could therefore imply a subclinical uterine infection, and therefore, further examination of the mare should be conducted to rule out infectious endometritis. Sensitivity, specificity, positive predictive value, and negative predictive value of the cytology smear compared with the presence of polymorphonuclear neutrophils (PMNs) on histology were 0.78, 0.84, 0.72, and 0.87, respectively. A moderate agreement ($k = 0.60$) was found between the cytologic response of the smear and the presence of PMNs on histology.

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1. Introduction

Optimal management of the mare around the time of breeding is important to increase the chance for a successful outcome, that is, a pregnant mare. Infectious

endometritis before or after breeding and persistent breeding-induced endometritis are major causes of infertility. The incidence of bacterial uterine infections has been reported to be 25%–60% in mares [1–3], and endometritis was the third most common medical problem ranked by equine practitioners [4].

The concept of resistance and susceptibility of mares to endometritis is well established in equine reproductive science [5–7]. Retrospective studies, primarily in Thoroughbred mares bred by natural cover, have summarized the prevalence of different uterine bacterial pathogens from either a random population of mares or a population of known problem mares. In these studies, β -hemolytic

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streptococci and *Escherichia coli* are the most prevalent bacteria isolated from the uterus [1,2].

Recently, more interest has been focused on the presence of subclinical endometritis as an underlying cause for susceptibility and infertility of mares [8–10]. Subclinical endometritis is only vaguely defined in the literature. Overbeck et al [9] and Buczkowska et al [10] evaluated the ability of common diagnostic methods, for example endometrial biopsy, cytobrush, and uterine swab, to detect subclinical endometritis in mares. They defined subclinical endometritis as mares without clinical signs of endometritis (intrauterine fluid) but with positive cytology and/or bacteriology. Furthermore, Buczkowska et al [10] pre-selected the mares based on poor reproductive performance, for example bred three or more times unsuccessfully or a history of 1 year of reproductive failure. LeBlanc and Causey [8] defined subclinical endometritis as mares that do not display the typically expected clinical symptoms such as intrauterine fluid and heavy infiltration of inflammatory cells in the uterine lumen after breeding. Furthermore, LeBlanc and Causey [8] stated that the clinical symptoms of subclinical endometritis can be very subtle and only present during certain phases of the estrus cycle. Subclinical endometritis can therefore be very difficult to diagnose by traditional methods. The lack of efficient procedures and diagnostic tools to establish a correct diagnosis of mares with subclinical endometritis impede the initiation of relevant treatment, leading to suboptimal management of the mare [8]. Petersen et al [11] showed by fluorescence in situ hybridization that *Streptococcus zooepidemicus* resides deep in the endometrium in chronically infected mares. The deep localization of *S. zooepidemicus* in the endometrium could be one explanation as to why mares with subclinical endometritis fail to display clinical symptoms and are difficult to diagnose. An endometrial biopsy, which samples deep in the endometrium, was demonstrated by Nielsen [12] to be a better diagnostic test than an endometrial swab, which only samples the endometrial surface, especially in detecting infectious endometritis caused by *S. zooepidemicus*. The prevalence of subclinically infected brood mares, which might go undetected by the veterinarian because of the previously mentioned, are not known but may be substantial.

Limited information is available about the clinical, bacteriologic, and cytologic findings before and after breeding in a population of mares using artificial insemination (AI). The few studies that have been done on mares bred by AI in a commercial setup have primarily focused on describing the prevalence of the bacterial pathogens before breeding and their antimicrobial susceptibility or the effect on pregnancy rates [2,13,14]. Most studies have been done in Thoroughbred mares where only natural cover is allowed [1,15]. During natural cover, contamination of the reproductive tract inevitably occurs due to the stallion's penis entering the vagina, whereas a more strict hygienic procedure can be practiced during AI. Furthermore, most semen extenders include antibiotics [16].

The purpose of this study was to investigate the prevalence of infectious endometritis and subclinical endometritis and the relation to clinical parameters of the mare before and after AI. Mares with subclinical endometritis

were in the present study defined as foaling mares and barren mares that had been barren for ≤ 1 year, without clinical signs of endometritis (intrauterine fluid) but with bacterial growth and inflammation of endometrium.

2. Materials and Methods

2.1. Study Design

The study was performed during May–July 2010 at a stud farm in Denmark serving approximately 150 mares by AI per breeding season. An in-house veterinarian performed the daily examinations of the broodmares. The in-house veterinarian decided time of insemination, relevant treatment if indicated, and other relevant activities regarding broodmare breeding management. The study was performed blinded for the in-house veterinarian, meaning that the person was not informed about the results of the study during the sampling and treatment periods. The in-house veterinarian of this study could independently request an endometrial swab for bacteriologic culture and cytology and initiate treatment according to the results. The author C.D.R. observed all examinations of the mares and recorded the information independently of the in-house veterinarian. All samples were collected and processed by C.D.R.

2.2. Animals

A total of 76 Standardbred mares submitted for AI at the stud farm were included in the study. For each mare, the following background information was recorded: age, parity, and reproductive status (maiden, barren, foal at foot). If the mare was barren, the number of years barren was recorded.

2.3. Clinical Examination and Sample Collection

Mares were observed for estrous behavior when exposed to a teaser stallion. Mares in natural estrus and mares that had induced estrus by prostaglandin analog (0.25 mg cloprostenol intramuscular; Estrumat Vet; Intervet, Ballerup, Denmark) were included. On the first day of estrus, the mare was examined by transrectal palpation and ultrasonography and then every other day until the dominant follicle was greater than 30 mm. When the dominant follicle exceeded 30 mm, the mares were examined daily until detection of ovulation.

2.3.1. Pre-AI Sample

The pre-AI sample was collected at the first estrus after arrival at the stud farm. When initial estrous behavior and a dominant follicle greater than 30 mm were detected, the mare was included in the study. The following parameters were recorded: presence and maximal height (cm) of intrauterine fluid measured in the uterine body, endometrial edema (score: 0: no edema, 1: mild edema, 2: moderate edema, 3: strong edema, and 4: excessive edema [modified scoring system based on Samper [17]]; Fig. 1), and number and size (mm) of dominant follicle(s).

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