

Ultrasonographic and Cytological Diagnosis of Endometritis in the Mare

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Abstract: Our study aimed to demonstrate the importance of echographic and cytological diagnosis of endometritis in the mare. During the breeding season, 15 (12.6%) infertile mares out of 119 mares were monitored. Fertile mares were 104 (87.4%). Ultrasound and cytological examination were performed to determine the origin of this infertility. Among the fifteen infertile mares, seven had uterine fluid accumulation with scalloped appearance characteristic of endometritis. cytological examination confirmed our diagnosis by the presence of inflammatory cells and even the presence of fungi.

Key word: Infertility-endometritis • Cytology • Ultrasound • Mare

INTRODUCTION

Endometritis, or inflammation of the endometrium, has been recognized as the third medical condition commonly found in the equine species [1]. It is characterized by excess mucus, high leucocyte count, increased uterine blood flow and edema due to fluid accumulation [2], with no systematic clinical signs [3]. The presence of intrauterine fluids will hold the attention of the clinician, because the presence of moderate amounts of fluids during estrus is acceptable, whereas in significant quantities \pm 2cm of height [4] or during the di-estrus will be considered pathological.

Indeed, a relationship between the presence of intrauterine fluids and the shortening of inter-ovulatory intervals has been observed following premature regression of the corpus luteum following inflammation which causes the premature release of prostaglandins [5, 6].

It should be noted that echogenicity, the presence of hyperechoic particles and the diameter of these fluid areas, it will be important to differentiate between uterine secretions, inflammatory fluids or exudates [6]. In reality, there is a system of grades that classify the different types of fluid accumulations according to their volume and echogenicity (Related to the amount of debris or inflammatory cells). It is then possible to link this grade to the fertility of the mare [7].

On a transverse section of the uterine horn filled with secretions, the folds of the endometrium sometimes forms bulges projecting into the lumen. The line separating the secretions and the uterine wall is often wavy or scalloped. The 6-8 folds of the mucosa normally present in the mare are quite often recognized. As a result of dilation of the uterine cavity by secretions, the secretions penetrate between the folds and separate them, whereas the folds are closely contiguous when the uterus is empty [5].

The diagnosis of endometritis depended on the detection of excessive intrauterine fluid, which is the most important sign of endometritis, abnormal vaginal discharge and shortening of the estrous cycle. A positive endometrial cytology and / or culture positive endometrial biopsy confirmed with a positive diagnosis [8, 9].

MATERIALS AND METHOD

This study was carried out at the National Haras Chaouchaoua of Tiaret (Algeria) during the breeding season from February to June 2015.

Ultrasound Examination: ISCAN "DRAMINSKY" brand ultrasound system supplemented with an endorectal linear array real-time B-mode transducer of 7.5 MHz was used to scan all the mares for determining the state of their uteri at the different phases of the cycle (follicular and luteal).

Uterine Cytological Examination:

Principle of the Technique and Sampling Equipment:

Using a human uterine cyto-brush, inserted on an artificial insemination gun, the assembly is then placed in a plastic insemination sheath to stiffen the assembly. The gun is covered with an exploration glove to protect it from vaginal contamination.

Preparation of the Animal: The perineal region was washed with antiseptic soap three times and dried with paper towels.

Sampling Technique: The instrument is introduced through the vulva and guided vaginally to the external orifice of the cervix, we push the instrument to the body of the uterus and then we pierce the glove. The cyto-brush is pressed against the body wall in circular motions for at least 30 seconds. Finally, we remove the cyto-brush in his glove before removing the instrument to avoid vaginal contamination of the sample. Immediately after removal, smears are made by rolling the cyto-brush on a microscope glass slide. Three to four slides were prepared for each animal according to the richness of the cyto-brush by the mucus. Then the slides are fixed immediately by a spray cyto-fixator and then stained by Papanicolaou staining.

Smear Staining: Papanicolaou staining was used for staining the obtained cells which is usually used for staining the vaginal smear in women and is rarely used in mares.

RESULTS AND DISCUSSION

In this study, out of examined 119 mares, 104 were fertile (87.4%) and 15 were infertile (12.6%).

Of the fifteen infertile mares, nine mares had intrauterine fluid of which four mares had festooned-looking fluid during dioestrus (Figure 1and 2) and five mares had intrauterine fluid during estrus.

The cytological examination confirms the endometritis in four mares examined during diestrus and two mares with intrauterine secretions > 2 cm and in a mare with 5mm during estrus, the richness of the smears by the PNN (Polymorph nuclear neutrophils) and the lymphocyte cells (Figure 4 and 5.), in the mare 6 with the presence of fungi on the smear (Figure 3).

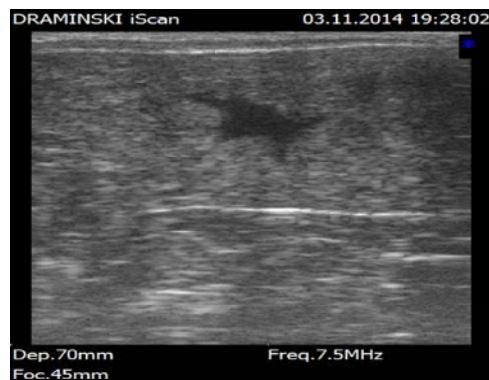


Fig. 1: Ultrasound image of the right horn of the mare 9.



Fig. 2: Ultrasound image of endometritis In the right horn of mare 2.

In the current study, out of 119 mares monitored, 104 were fertile (87.4%) and 15 were infertile (12.6%). The seven mares with endometritis confirmed by the diagnostic methods used in our study (5.88%). Thus, 46.66% of infertile mares had an endometritis problem, which indicate the importance of this pathology in equine herds. Similarly, Liu and Troedsson [9] reported that endometritis represents a major cause of infertility in the mare and which showed in 1999 that endometritis affects approximately 15% of thoroughbred mares.

It appeared from the current results that infertility problems increase with the age of the mare. Older mares ≥ 15 years old presented a decline of their fertility. As well as, by Davies Morel [2] and Betsch [10] recorded a gradually decrease in the fertility of mares from the age 12-13 accompanied with the increase of early embryonic mortality.

The seven cases suffered from a cervical dilation problem, which did not exceed two-finger dilation. This cervical dilation was also reported by

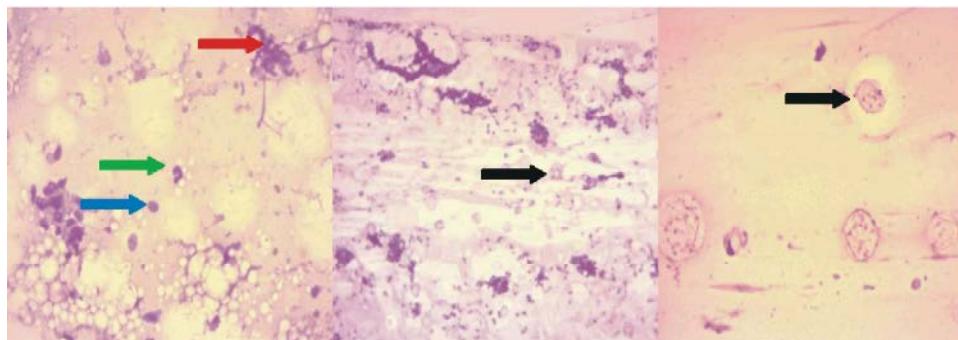


Fig. 3: Uterine smear of the mare 6. Left ($\times 40$), middle ($\times 10$), right ($\times 40$)

- : Yeasts,
- : Lymphocyte,
- : Polymorphonuclear neutrophile
- : Champignons

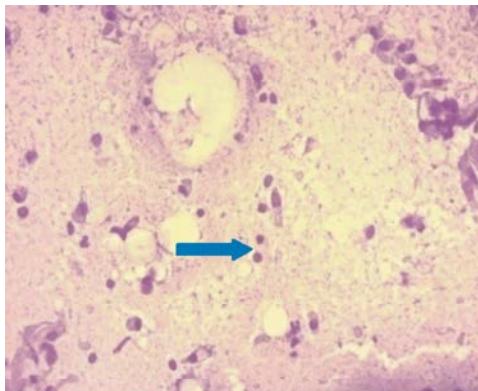


Fig. 4: Uterine smear of mare 9 ($\times 40$)

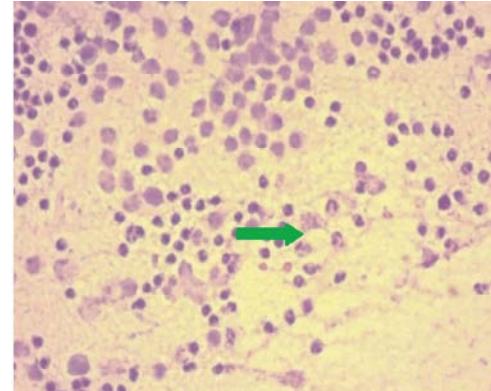


Fig. 5: Uterine smear of mare 2 ($\times 40$)

several authors Pycock, 2006 [11]. The non-dilatation of the cervix explains the retention of uterine fluids in these mares.

The presence of uterine fluids is rare at different times in the estrous cycle and its presence is common among infertile mares, 9/15. Infertile mares showed uterine fluid retention at different times of the cycle, among the 9 cases, 7 showed endometritis. Our results are similar to those reported by Pycock, 2006 [11] who suggest that a history of infertility and / or uterine fluid accumulation during the reproductive period is the most useful indicator of a sensitive mare in the practice.

Of the 7 mares that had endometritis, 2 mares had > 2 cm in diameter from the uterine fluids during estrus. Our results are consistent with those reported by LeBlanc [12].

In contrast, LeBlanc [4] reported a mare had chronic endometritis with uterine fluid retention of 5 mm, which explains why the diameter of the uterine fluids during estrus is not a reliable diagnostic criterion, so any

presence of fluid during estrus requires a cytological diagnosis.

According to the presented results, the echogenicity of the uterine fluids and the scalloped appearance is a sign of endometritis.

Mare 6 had fungal endometritis (Figure 3), which can be explained by the uncontrolled use of antibiotics and corticosteroids (Immunodepressants) at the stud level, which is also reported by Betsch [10] and Dascanio [13].

4/7 mares had lymphocyte-rich smears associated with polymorphonuclear neutrophils (Table 1), confirming the chronicity of endometritis. This is reported by Causey *et al.* [14].

The decrease of the inflammatory cells that was associated with the presence of fungi may refer to the presence of a biofilm by these fungi in the endometrium [9,15,16]. This biofilm can prevent the passage of inflammatory cells and medication to clear the infection [15].

Table 1: Ultrasound and cytological diagnosis of endometritis

Mares	Age/year	Estrous phase	Ultrasound diagnosis			
			liquid diameter/mm	Aspect	Echogenicity/ grade	Cytological findings
1.	16	Estrus	19	festoon shape	0	PNN
2.	16	Diestrus	27	festoon shape	2	PNN, lymphocytes
3.	27	Estrus	5	festoon shape	0	PNN, lymphocytes
4.	-	Estrus	15	Normal	0	No cells
5.	-	Estrus	12	Normal	0	No cells
6.	18	Proestrus	27	festoon shape	2	Yeasts, chlamydiae PNN, Lymphocytes
7.	16	Diestrus	21	festoon shape	1	PNN, lymphocytes
8.	21	Diestrus	10	festoon shape	0	No cells
9.	20	Diestrus	15	festoon shape	0	lymphocytes

Polymorph nuclear neutrophils (PNN).

CONCLUSION

This study concluded that any fluid retention in an infertile mare regardless of its diameter indicated endometritis and suspected mares must be the subject of the cytological diagnosis and other confirmatory tests for applying appropriate treatment.

ACKNOWLEDGEMENT

I would like to thank Dr. Missoum, Head of the Cytology Department and his collaborators at the Zaaroura / Tiaret Polyclinic, for the help given to the staining of the smears and their demonstration in microscopic observation; and Slimani Khaled and Professor Amara Karim for their support.

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