Département des Sciences Vétérinaires, Faculté des Sciences Agronomiques et Vétérinaires Université Ibn-Khaldoun de Tiaret, Algéria.

HISTOPATHOLOGICAL DIAGNOSIS OF ENDOMETRITIS, IN DAIRY COWS, IN TIARET AREA (ALGERIA)

(With 4 Tables and 6 Figures)

By

K. ZIDANE: A. NIAR and D. TAINTURIER*

* Département. de Pathologie de la Reproduction, Ecole Nationale Vétérinaire de Nantes, Nantes, Cedex 03, France.

(Received at 16/3/2006)

التشخيص النسيجى المرضى لحالات التهاب مخاطية الرحم عند الأبقار التشخيص النسيجى المرضى لحالات التهاب مخاطية الرحم

خالد زيدان ، عبد اللطيف نيار ، دنيال تانتوريي

التهاب الرحم يمكن أن يشمل كل هذا العضو (شامل)، ويمكن أن يقتصر على مخاطية الرحم فقط. يحدث هذا عندما تقل وسائل الدفاع في جسم الأنثى. يحدث هذا أيضا في مرحلة ولادة الأبقار، عندما تنعدم الإجزاءات الصحية في القطيع، وينجم عادة عن هذا الإستغناء المبكر عن هذه الأبقار المصابه. تم إجراء دراسة نسيجية مرضية لخمسون جهاز تناسلي لأبقار تم الإستغناء عنها مبكراً، من مجزرة مدينة تيارت (الجزائر). نتائج هذه الدراسة بينت إصابة محاطية الرحم عند ٨٠% من هذه الأبقار. الكشف الذي أجرى على هذه الأبقار قبل الذبح، لم يمكننا من تشخيص أي مرض على مستوى الجهاز التناسلي. يعتبر التشخيص النسيجي المرضى، الوسيله الوحيدة المتاحه للكشف المبكر على حالات التهاب الرحم ومخاطيته.

SUMMARY

Metritis and endometritis are inflammations of the uterus. Metritis involves the endometrium, the underlying glandular tissues and the muscular layers. Endometritis involves only the endometrium and the underlying glandular tissues. Uterine inflammation is the consequence of a rupture in balance between attack and defence factors. This is the case when hygiene rules are missing at the time of birth, which often leads to a premature reform of the female. Accordingly, an analytical and descriptive study of the genital sphere was carried out on 50 cows at the slaughter-house of Tiaret (Algeria), for various causes of infertility.

Results of this study showed that 82 % of the cases presented an attack of the endometrium. For all this animals, the ante-mortem examination did not let appear signs in favour of metritis. The uterine histopathological examination is the only way which made it possible to confirm the existence of typical uterine inflammatory lesions.

Key words: Uterine infection, endometritis, metritis, histopathology, cows.

INTRODUCTION

Metritis and endometritis are inflammations of the uterus. Metritis involves the endometrium, the underlying glandular tissues and the muscular layers. Endometritis involves only the endometrium and the underlying glandular tissues. Commonly, metritis is used to indicate both conditions. A clinical or sub clinical form of metritis may be present. Clinical metritis may be either acute, appearing quickly and generally affecting the cow's appetite and milk production, or chronic, persisting over a long period. Clinical metritis may be detected by rectal palpation as an increase in size and thickness of the uterine wall. A purulent (contains pus) vaginal discharge may or may not be present.

Sub clinical endometritis is not detectable by rectal palpation (Arthur et al. 1998).

More commonly it occurs in the chronic rather than the acute form. No vaginal discharge is evident. Sometimes examination with a speculum will reveal a purulent discharge, but not always. Cultures of the uterus may or may not verify a microbial infection. For example, many times the repeat breeder is negative on culture. Sub clinical endometritis can be positively diagnosed by microscopic examination of a uterine biopsy (Arthur et al. 1989; Dhaliwal et al. 2001). Controversy exists over the effects of metritis and endometritis on fertility in cattle. Responses of individual animals to intrauterine treatment are quite variable. Some respond well to medication and conceive while others do not. Other factors such as nutrition, hormonal imbalances and overall health of the animal must be considered. Most cases of metritis and endometritis are initiated during the period from calving to the time the uterus returns to normal size (Lewis, 1997; Griffin et al. 1974; Del Vecchio et al. 1994). The process by which the uterus returns to its normal nonpregnant size is called involution. Involution in the cow varies from 26 to 56 days after calving, with an average time of 42-47 days.

Among the world, researchers considered the annual incidence of bovine uterine infections approximately 10 to 50% (Arthur *et al.*1992; Lewis, 1997). However, the real incidence of these uterine infections cannot be known, because the means of detection and diagnosis are not always very suitable.

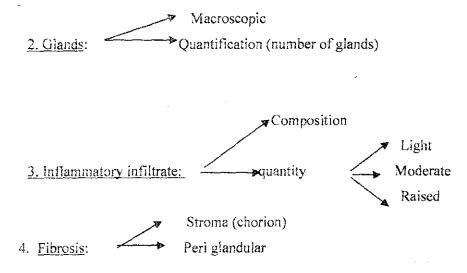
MATERIELS and METHODS

The purpose of this research task was to explore the anatomopathological incidence of uterine lesions of 50 culled cows, 2 to 6 years old (42 of Pie-noire race and 08 of Pie-rouge race, between October 2003 and January 2005, at Tiaret slaughter-house (Algeria). Uterine biopsies were collected immediately after slaughter, preserved in formol at 10%, and are analyzed at Pathological anatomy laboratory, of Parnet Hospital (Hussein Dey /Algiers). We have practised the ante-mortem and post-mortem examination on all the cows belonging to our sample. So that this study is realizable, it was necessary to quickly fix sampled tissues in a state as close as possible to the state living at the slaughterhouse of Tiaret. Thereafter, the samples are fixed at formol passed in another fixer, before carrying out the colouring of the mixtures which contain pieric acid or chloride mercuric, particularly indicated in similar studies. Thereafter, these same samples are dehydrated while successively passing them during 2 hours, in 5 alcohol bottles with 90°. This operation is followed by an impregnation of the studied samples, which consists in replacing alcohol by a soluble intermediate liquid at the same time in alcohol and paraffin. The solvent more used is xylene or toluene. This operation is followed by an inclusion and a coating of samples in a block of paraffin. Once the samples were included in homogeneous blocks, the cuts are carried out. The thickness of the cuts varies from 1 to 2, 4 µm. They will be after assembly on microscopic blades, coloured in a routine way in Hemateine and Eosin.

RESULTS

The results of the examined lesions were quantified according to criteria's retained and already recognized by many authors. These criteria are distributed as follows:

1. State of the surface coating associated with the compartment luminal



We have used these bibliographical criteria to be able to lead to a labelling of metritis and for thus classifying it in:

- 1- Acute endometritis.
- 2- Sub acute endometritis.
- 3- Chronic endometritis.

1. Gross findings:

All uteri of studied culled cows have presented at the macroscopic examination multiple inflammatory lesions, characterized by congestion, tumefaction, of hypertrophy and atrophy of uterine horns, often accompanied by clearly or flocculent exudates (Table 1).

Table 1: Revealed frequency of lesions observed with the macroscopic examination of uteri.

Frequency of observed lesions	Number of cows	Reason of slaughter	Macroscopical Exam
20,45 %	n=10	Infertility	- Uterine horns Hypoplasia; - Congestion and hypertrophy of one or the two uterine horns.
18,18 %	n=9	Sanitary	- Often, left uterine horn is pregnant; In other cases: congestion; hypertrophy of uterine horns; tumefaction.
61,36 %	n=31	Slaughter Other affections: Pulmonary, digestive, arthritis senility, and others.	Congestion; pronounced tumefaction; hypertrophy or atrophy.

2. Histopathological findings:

In 18 % of the cases, the quantification of the lesions showed only vascular congestion and oedema; it is acted in fact in these cases of lesions of the post-mortem. The remainders of the found lesions (82%) are distributed according to criteria's established as follows (Table 2).

Table 2: Lesions observed on the level of the surface layer of the uterine mucous membrane of the 50 studied uteri.

Uterine surface coating (%)		Total Animal Number (n = 41)	
Conserved Abrased	(34 %) (30 %)	n = 14 n = 12	
Ulcerated Regenerated	(20 %) (16 %)	n = 08 $n = 07$	

According to these results, 52 % of the studied uteri (N=21) presented a deterioration of uterus surface coating. Among them, 32 % presented a fibrino-leucocytic coating in the luminal compartment (N=07), which pleads in favour of a metritis diagnosis. 34 % of the uteri only preserved this surface coating (N=14). Regeneration of this area was observed in 16 % of our samples (N=7), which determines regression of the inflammatory process, therefore in cure after a preliminary state of metritis. The uterine glands belonging to 23 studied uteri presented a lesional attack, of about 56 %; among these attacks, 43.47 % of these glands were suppurated, 39.14 % were cystic, and 17.39 % showed images of dedifferentiation (a process of cure after deterioration). Only 44 % of uteri presented a normal conservation of glands without any inflammatory deterioration; this shows also the importance of the uterine affections in our sample, and our bovine breedings in general (Table 3).

Table 3: Glandular lesions observed with histological examinations of the 50 studied uteri:

Nature of the glandular lesion	Percentage of animals	Number of animals	
Conserved Glands	44 %	n = 18	
Attacked Glands:	56 %	n = 23	
1/ cystic	39.14 %	n = 09	
2/ Suppurated	43.47 %	n = 10	
3/ Dedifferentiated	17.39 %	n = 04	

Always in our results, the inflammatory infiltrate generally was of lymphoplasmocytic nature, sometimes histiocytic, and one (01) only case presented lymphoid nodule. We have noted that in 76 % of the 41

attacked uteruses (N = 31), the inflammatory infiltrate was present; in 16 % of the cases (N = 5), this infiltrate was light, but in approximately 84 % of the cases (N = 26), it was very significant, which confirms our endometritis or metritis diagnosis. Fibrosis was often moderate in our samples, and generally has interested the glandular circumference; in the stroma, fibrosis was very spindly (Table 4).

Table 4: Inflammatory infiltrate observed with the anatomo-pathological examination of the 31 studied uteruses.

Nature of Inflammatory infiltrate	Percentage	Number of animals
Light	16%	n = 5
Moderated	18 %	n = 6
Raised	66 %	n = 20

As we already mentioned before, 82 % of our samples (N = 21), generally presented inflammatory lesions classified as follows:

- 1- Acute endometritis: It is characterized by the prevalence of a vasculo-exudative (oedema or congestion): The "oedematous" type is characterized by an imbibitions of stroma cells, and by a séro-albumin exudate; this type of lesion is very common, momentary, and generally pain-killer (Photo 1). The "congestive" type follows on the other hand upon a brutal and intense vasodilatation of the blood capillaries (Photograph 2).
- 2- Sub Acute endometritis: It is characterized also by exaggeration and prevalence of intra tissue cellular phenomena, compared to the vasculo-exudatifs phenomena which appear minors or fugacious, and clinically blurred or unapparent. It prevails especially on the level of external coating and the level of the cytogenous chorion (Photograph 3). In this type of lesions, there is a destruction of the external coating, which is completely abrased. The cells are then in this case hyper chromatic (of dark colour). Always in the sub acute form, we can see the same external deteriorations as the preceding one, with in more, invasion of the cytogenous stroma or chorion by inflammatory cells, polynuclear and histiocytes. In other cases, we can note a fibrinogen extravasation, and fibrin formation (Photo 4).
- 3- Chronic endometritis: At the time of a chronic endometritis, we can note the same lesions as those already quoted in a sub acute case, but these lesions can be sometimes irreversible and lead to fibrosis and sclerosis of touched tissues. Purulent endometritis were often met, which are characterized by a massive exudation of polymorphnuclear cells; the latter arrive by infiltration at the level of the inflammatory hearth,

release their lysosomales enzymes which cause external destruction of glands; this involves the complete disappearance of the glandular structure. This type of inflammation is intermediate between a sub acute and chronic nature. The endometrium can in certain cases be regenerated with new repaired tissues and newly formed vessels, new glands, and a young and cellular conjunctive tissue; it is about the process of cure. The sub acute inflammation can then pass to chronicity, with however the appearance of irreversible lesions. In this case, there will be a sclerosing chronic endometritis, which results in an excessive dilation of glands diameter. In addition to these lesions, we can observe a significant peri-glandular fibrosis, with many layers of fibroblasts visible around the gland (Photo 5). Chronic endometritis can also take a purulent form, characterized by a massive destruction of the coating surface and a significant inflammatory infiltrate, clinically diagnosed by the formation of pus (Photo 6).

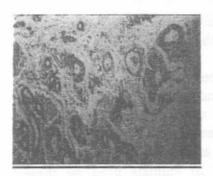


Photo 1: Acute Endometritis.

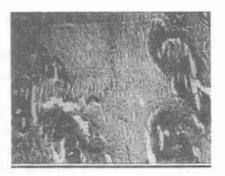


Photo 2: Congestive Acute Endometritis.



Photo 3: Sub Acute Endometritis (Polynuclear Invasion).



Photo 4: Sub Acute Endometritis (Degenerative type).

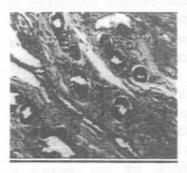


Photo 5 : Sclerosant chronic Endometritis.

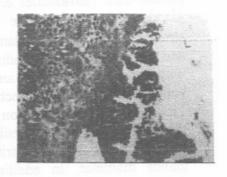


Photo 6: Purulent chronic Endometritis chronique

DISCUSSION

Criteria and models of our results were based on several researches made previously by many authors (Chaffaux *et al.* 1987; Manspeaker and Haaland, 1983).

These authors especially noted chronic lesions of metritis, but they also noted lesions of sub acute endometritis characterized by cellular infiltration, and an attack of the uterine surface coating; we raised this same type of lesions in our sample of cows, although our study was made on a sample not selected, and not presenting good and apparent metritis clinical signs, contrary to animals used by these authors. In our results we bring back an incidence of sub acute and chronic metritis of about 82 %; this percentage appears enormous, but it once again reflects the gravity of the situation of bovine breeding in Algeria, and the obvious lack of hygienic conditions, which make that our cows are prone to an intense bacterial pressure during postpartum period.

Stevenson and Call (1988) carried out an assessment of eight (08) investigations in the U.S.A; this assessment reveals that the only genital infections reach an average of 36, 9 % of milking cows with a variation of 19, 9 % to 81,6 %. According to these authors, genital infections compete with mastitis, lameness and metabolic diseases. The incidence of sub acute and chronic metritis was 32,9 % in France, in a study carried out by Meissonnier and Enriquez (1998) on 2024 milking cows belonging to 84 herds; according to these authors, an incidence of metritis higher than 20 % in any herd is regarded as abnormal. Arthur et al. (1998) report that endometritis in Great Britain and in North

European are primarily due to non-specific pathogenic germs, and that the latter have a very significant effect on fertility; these endometritis influences short-term fertility by considerable extension of birth-conception interval with increase in the number mating or I.A., and by generating sterility when irreversible changes occurs at uterine level. Among all results published to date, we manage to understand that metritis and endometritis constitute the major plague of contemporary breeding even in the most developed countries such USA, Great Britain and France. If authors brought back prevalence's which could exceed 80 % in certain cases, we can imagine the Algerian situation, where hygienic problems are permanent in our bovine breeding, and disease prevention against the most great reproductive diseases is almost non-existent.

CONCLUSION

This study was made on a not selected livestock; seldom presenting at the clinical examination, signs of genital attack; on the other hand, macroscopic examination of parts of hysterectomy could detect significant uterine inflammatory lesions. The microscopic examination was more convincing, since it enabled us to find three varieties of endometritis anatomo-pathological lesions, with a high percentage of sub acute endometritis, associating a polynuclear infiltration, and an epithelial attack; Chronic endometritis, with a less significant frequency than the preceding ones, but with generally irreversible lesions, concerning the underlying structure of the endometrium; Acute endometritis, less significant on the reproductive level, because they are of an alarming diagnosis, generally causing curiosity and speed intervention of the stockbreeders, since they are sometimes accompanied by fugacious general symptoms.

REFERENCES

- Arthur, G.H.; Noakes, D.E. and Pearson, H. (1992): Veterinary Reproduction and Obstetrics; 6, Edition Philadelphia: Baillière Tindall.
- Arthur, G.H.; Noakes, D.E.; Pearson, II. and Parkinson, T.J. (1998): Veterinary reproduction and obstetric. 7th Edition, W.B. Saunders Company, Limited, 703 pages.

- Chaffaux, S.T.; Recorbet, Y.; Bhat, P.; Crespeau, F. and Thibier, R. (1987): Biopsies de l'endomètre au cours du post-partum pathologique chez la vache, Rec. Méd. Vét., 163 (2) 199-209.
- Dhaliwal, G.S.; Murray, R.D. and Woldehiwet, Z. (2001): Some aspects of immunology of the bovine uterus related to treatments for endometritis. Anim. Reprod. Sci., 67:135-152.
- Griffin, J.F.T.; Hartigan, P.J. and Nunn, W.R. (1974): Non-specific uterine infection and bovine fertility. I. Infection patterns and endometritis during the first seven weeks post-partum. Theriogenology, 1:91-106.
- Griffin, J.F.T.; Hartigan, P.J. and Nunn, W.R. (1974): Non-specific uterine infection and bovine fertility. II. Infection patterns and endometritis before and after service. Theriogenology, 1: 107-114.
- Del Vecchio, R.P.; Matsas, D.J.; Fortín, S.; Sponenberg, D.P. and Lewis, G.S. (1994): Spontaneous uterine infections are associated with elevated prostaglandin $F_{2\alpha}$ metablite concentrations in post-partum dairy cows. Theriogenology, 41:413-421.
- Lewis, G.S. (1997): Uterine health and disorders. J. Dairy Sci., 80: 984-994.
- Manspeaker, J.E. and Haaland, M.A. (1983): Incidence and degree of endometrial periglandular fibrosis in parity I dairy cows. Veterinary Medicine, Small animal clinician, 6, 943-946.
- Meissonnier, E. and Enriquez, B. (1998): Infection utérines du postpartum: Epidémiologie, bactériologie et thérapeutique antiinfectieuse, CD des journées des GTV.
- Stevenson, J.S. and Call, E.P. (1988): Reproductive disorders in the periparturient dairy cow. J. Dairy Sci., 71, 2572-2583.