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Research Article

Positive Responses to Intra-Uterine Route Administration of Natural Sahara Honeys in Cattle Clinical Endometritis

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ABSTRACT

This study was conducted to test the therapeutic efficacy of an intrauterine infusion of two varieties of honey in the treatment of bovine endometritis. Our choice focused on animals treated with antibiotics twice or three times without success. The minimum time between the last treatment antibiotics and our study is one month.Twenty-eight cows with clinical endometritiswere divided into two groups "group A and B" each group was treated with an infusion of a honey variety by intra uterine route. The monitoring of the cows was carried before infusion of honey and 24 hours and seven days after the infusion of honey by a clinical examination; ultrasound and cytology. Fertility monitoring was carried out in eleven cows by an ultrasound examination. Seven days after the honey infusion of honey "A", 100% of the cows with endometritis showed total reabsorption of uterine fluids with total disappearance of purulent discharge 12 of the 13 cows were in heat on the seventh day with translucent mucus. From a cytological point of view, the smears were rich in endometrial cells. Seven days after the infusion of honey "B", 12 Vs 15 cows presented a total reabsorption of uterine fluids with total disappearance of the purulent discharge and 02 were in heat on the seventh day with translucent mucus. Cytologically, the smears were very rich in endometrial cells. In 03 cows the purulent discharge was still present. 12 Animals were inseminated in the second estrus after treatment. The conception rate was estimated after two months of the last insemination. Conception rate was 5 in group A and 3 in group B. These results show that honey has proven its therapeutic power for bovine endometritis, by its drainage effect of pathological uterine fluids, the regeneration of the endometrial epithelium and the improvement of the fertility rate.

INTRODUCTION

Clinical endometritis is defined as the presence of a purulent discharge detectable in the vagina 21 days or more post partum, or mucopurulent discharge detectable in the vagina after 26 days post partum [1].

Endometritis in cattle is a common problem of infertility caused mainly by specific or non – specific potential pathogens [2]. Antibiotic resistance is the hot topic of this decade and in order to survive there is a need to search Alternative to counteract the pathogens and also to safeguard human and animal health [3].

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So, biological sources which stimulate the natural uterine defensive mechanisms have been suggested as alternative therapies [4]. Immunomodulators as an alternative to antibiotic therapy have been shown to be of great value in treatment of endometritis. They act mainly by three ways;

(1) Contains immunoglobulins mimic humoral immunity

(2) Chemo-attractants boost cellular immunity

(3) Antimicrobial activity like antibiotics [5]

Approaches utilizing stimulation of the uterine defense mechanism to combat infection have mentioned the single intrauterine infusion of E Coli lipopolysaccharides, oyster glycogen and

autologous plasma or hyper immune serum. These infusions act as potent chemotractant with resultant increased influx of polymorphonuclear leucocytes within the uterine lumen and significant decrease in uterine infection. However their routine use has not become popular due to high costs and poor availability [4]**.** It has been assumed that honey can act as an immunomodulator with both proinflammatory and anti-inflammatory properties [6].

Indeed, honey is a very complex mixture, containing a number of ingredients that are involved in oxidant/antioxidant physiological processes, such as hydrogen peroxide, nitrite, nitrate, glucose, glucose oxidase, iron, copper, chlorine, iodine, catalase, tyrosine, tryptophan, arginine, flavonoids and phenolic acids [7].

Cytokines interestingly represent an improved and alternative treatment against emerging pathogens [3]. Honey and its different components have the capacity to stimulate or inhibit the secretion of cytokines [6].

Based on clinical and observational reports [8] asserted that honey promotes tissue regeneration through stimulation of angiogenesis and the growth of fibroblasts and epithelial cells. Therefore, the present study was designed to evaluate the therapeutic efficacy of honey in controlling endometritis in cattle.

MATERIAL AND METHODS

A total of 28 cows with clinical endometrtis kept at twenty farms distributed in the wilaya of Tiaret, located in the Algerian highlands region were used in this study.

These cows were the subject of four successive diagnoses:

1) Anamnesis:

We mainly focused on

- The duration of infertility
- Placental retentions
- Presence of purulent discharge
- Coupling numbers or artificial inseminations
- Treatments carried out
- The health status of the cow

The presence of a purulent discharge was highlighted by visual examination of traces of pus on the internal face of the tail, or by means of transrectal exploration after perineal cleaning and vaginally introducing clea-gloved-hand to control the presence of uterine discharge and its color.

Only cows with clinical endometritis and treated 2 to 3 times with classical antibiotics without success were selected. So that there is no interference between the honey and the drugs used, the minimum time between the last treatment and our study is one month.

2) Ultrasound Examination of the Uterus:

The ultrasound examination was carried out using two ultrasound scanners "DRAMINSKY ISCAN" and "DRAMINSKY 4 vet mini" connected to linear probes to measure the diameter of uterine fluids and control their echogenicity.

3) Cytological Examination of the Endometrium:

The cytological examination was carried out using a cytobrush attached to an artificial bovine insemination gun, covering the whole with a plastic sheath.

The smears were stained by the Papanicolaou technique, at the cytology department of Tiaret polyclinic.

4) Treatment with Honey Infusion:

Two samples of Algerian honey: jujube honey (honey A) and euphorbia honey (honey B) originating from Algerian Sahara were obtained directly from beekeepers. Raw honeys used in this study were not submitted to thermal treatments or pasteurization, or any other operation able to alter natural composition. Honey samples were stored at 4°C in the dark until use.

The cows were divided into two groups. Each group was treated with a variety of honey: (Group A treated with honey A: 13 cows) and (Group B: treated with honey B: 15 cows). The two groups were treated with an intrauterine infusion of honey for each cow. The honey has been diluted to 70%, and infused using a sterile bovine artificial insemination sheath (70 cc).

The follow-up of the cases was made using an ultrasound and cytological examination twice after the treatment, 24 hours and the seventh day post-treatment respectively.

Nine cows were artificially inseminated and two by a natural projection in the second estrus after treatment. The diagnosis of pregnancy was made by an ultrasound examination.

RESULTS AND DISCUSSION

Variety of treatments have been used on cows following periparturient disease. Antibiotic treatment has been advocated for endometritis and metritis [9].

Each use of an antimicrobial drug is inherently associated with selective pressure for the emergence of resistant bacteria, which stresses the importance of their prudent use [10]. It is obvious that increasing antibiotic resistance is associated with decreasing clinical efficacy and could exacerbate animal welfare and economic consequences [11].

Recently, besides traditional procedures of treatment of endometritis in cows, some new preparations have been used, which stimulate different mechanisms of uterine immune system. These preparations are designated as biologically active immunomodulators. These substances stimulate uterine defense mechanism in endometritic cows and thereby help in removal of bacteria from the uterus [4].

It has been assumed that honey can act as an immunomodulator with both proinflammatory and anti-inflammatory properties [6]. The treatment of endometritis with honey in this study is justified by its efficacity in the treatment of endometritis and its harmlessness on the endometrium in the mare [12-13]. The jujube honey (honey A) used in this study is the most expensive and is starting to be the most used in Algerian traditional medicine. The euphorbia honey (honey B) has been used with great success in the treatment of wounds and in surgical operations [14-17].

Group "A":

24 hours after the infusion of honey A 10 cows still had a purulent discharge; 06 of them had a strong increase in uterine fluids and their echogenicity as well as an increase in the thickness of the uterine wall. In all cows; the smears were very rich in PMNs.

When honey “A” was first administered, the sharp increase in uterine fluids can be explained by the fact that, the outflow was created by its osmotic effect due to its high sugar content.

According to [12] intra-uterine infusion of a solution of 70% of honey caused after 6 hours a fluid accumulation within the uterine lumen of the mare probably due to its osmotic effect.

The sharp increase in uterine fluids and their echogenicity, the infiltration of inflammatory cells and even the thickness of the uterine wall 24 hours after the infusion of honey "A", was probably due to an inflammation of the endometrium caused by honey, which is confirmed by the massive influx of PMNs (Fig. 2B). It is clear that after 24 hours the honey will be very diluted by uterine fluids (Fig. 1B).

In accordance with [18] honey at low concentration induces the chemotactic activity of isolated neutrophils. Honey at the concentration of 1% (w/v) incubated with MM6 cells (and human monocytes) for 24 h increased the TNF-a, IL-1b and IL-6 release [19-20]. It’s well known that TNF-α stimulates the production of IL-8 a member of the CXC family of chemokines and a potent chemoattractant of neutrophils [21-22]. The inflammatory phase, of wound healing is characterized by immune cell migration into the wound, especially neutrophils and macrophages.

24 hours after the infusion of honey "A" 3 cows presented a thinning of the discharge with a decrease in the amount of uterine fluids and the thickness of the uterine wall. The process could be the same as previously but with a more spontaneous drainage of secretions, in fact the presence of PNN was observed for the 13 cases. A spontaneous drainage of secretions was also observed by [12] in a mare only six hours after the infusion of honey.

Seven days after the honey infusion, 100% of cows with endometritis showed total reabsorption of uterine fluids with total disappearance of purulent discharge 12 of the 13 cows were in heat on the seventh day with translucent mucus. From a cytological point of view, the smears were rich in endometrial cells (Fig. 1 and 2).

According to Chaudhary et al. [23] honey at very low dilution facilitates cell proliferation and attainment of confluence towards faster wound healing and modulation of cardinal epithelial genes. Both TNF-α and IL-1β stimulate the release of various growth factors, including platelet derived growth factor and transforming growth factor-beta [24].

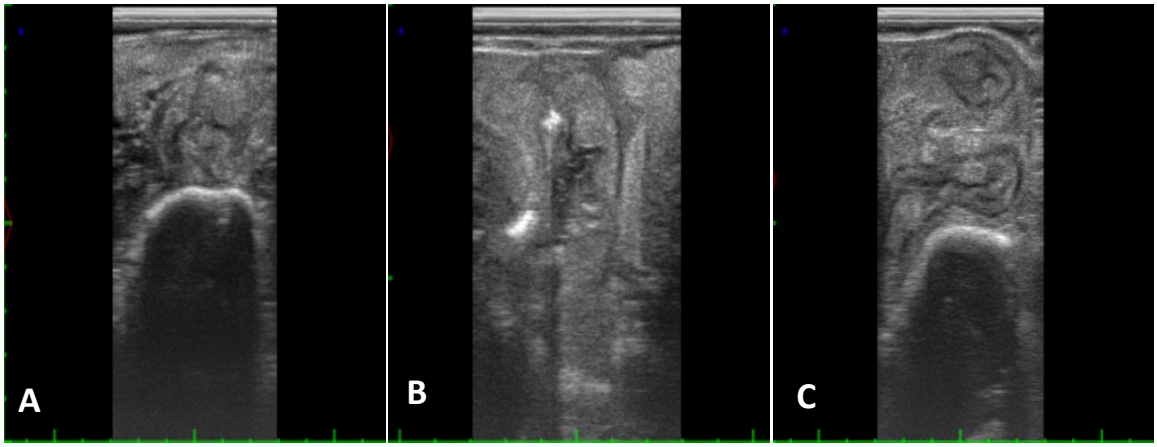

Figure 1: Ultrasound images of cow no 02: Ultrasound image of the uterine horn before infusion of honey "A". (A) horn with hyperechogen liquid 14 mm, uterine wall 7 mm , increased thickness of the uterine wall;: Ultrasound image of the uterine horn 24 hours after the infusion of honey "A". (B) Increased amount of uterine fluid and echogenicity 24 mm, uterine wall 14 mm; (Ultrasound image of the uterine horn seven days after the infusion of honey "A". (C) Disappearance uterine fluid and reduction of the thickness of the wall.

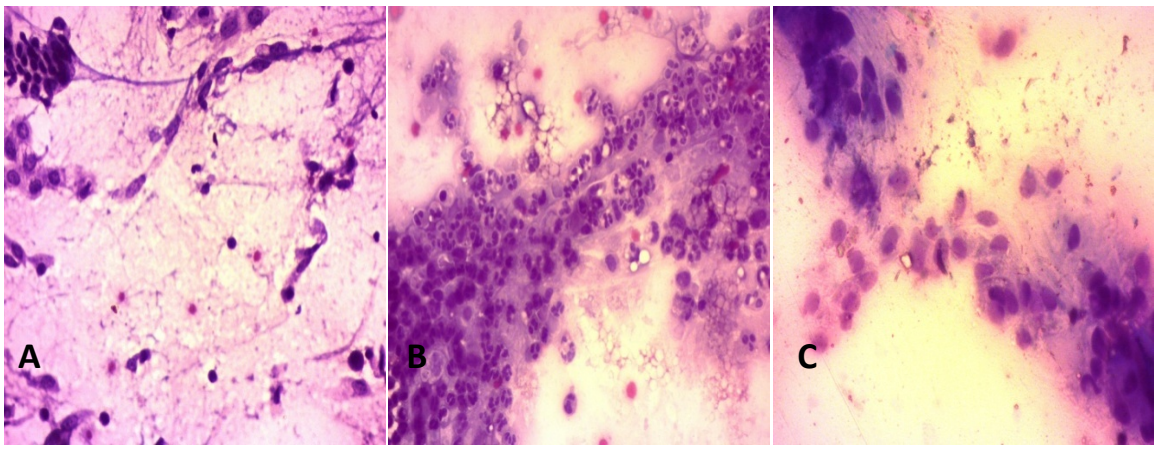

Figure 2: Endometrial smear from cow no 02: Endometrial smear before infusion of honey "A" (A) some lymphocytes, PMNs and endometrial cells. Endometrial smear 24 hours after infusion of honey "A"(B): very rich in PMNs. Endometrial smear seven days after infusion of honey "A" (C); marked decrease in PMNs and richness in endometrial cells.

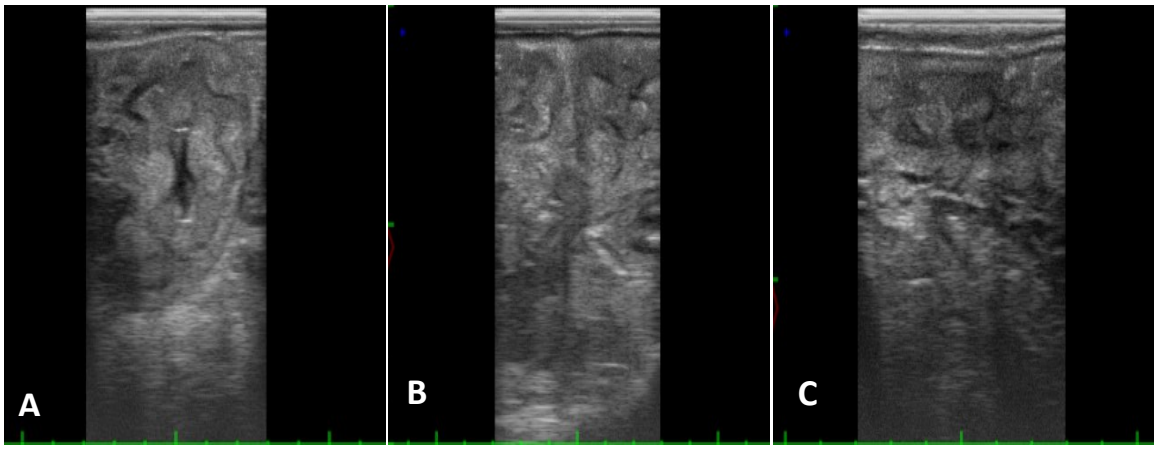

Figure 3: Ultrasound images of cow no 04: Ultrasound image of the uterine horn before infusion of honey "B" (A) horn with hyperechogen liquid; Ultrasound image of the uterine horn 48 hours after the infusion of honey "B" (B) decreased uterine fluid; Ultrasound image of the uterine horn seven days after the infusion of honey (C)Disappearance of liquids.

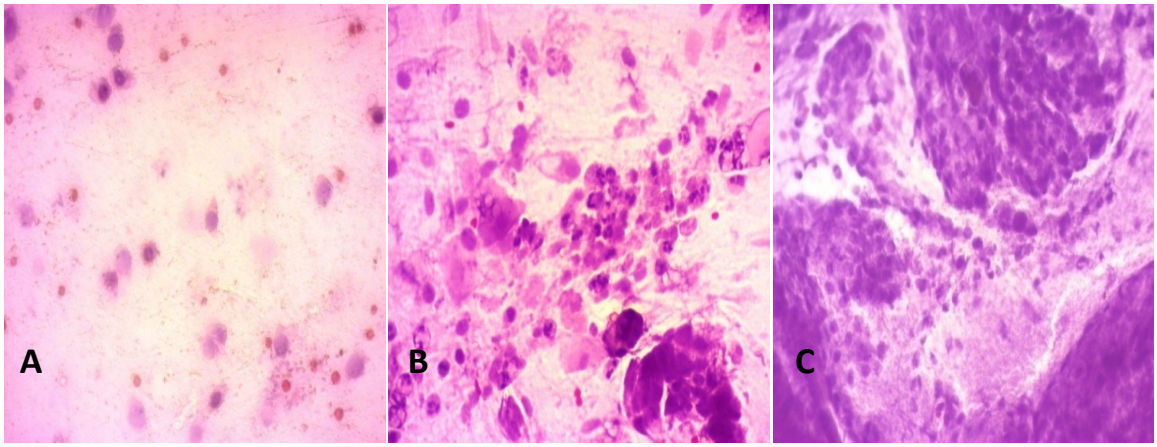

Figure 4: Endometrial smear from cow no 09: (A) some lymphocytes, PMNsand Endometrial smear before infusion of honey "B"; (B) rich en PMNs Endometrial smear 24 hours after infusion of honey "B"; (c) Endometrial smear seven days after infusion of honey "B" rich in endometrial cells.

Group "B":

48 hours (due to an independent delay of our will we could not do the exam after 24 hours of 3 cows for this group) after the infusion of honey "B", these 3 cows with clinical endometritis presented total reabsorption of uterine fluids, and presence of incompletely dry pus on the inner side of the tail.

24h after the infusion of honey "B", 02 cows showed an increase in uterine fluids and echogenicity, 06 cows had purulent discharge with a strong decrease in uterine fluids and 04 presented a total disappearance of uterine fluids. The smears were also very rich in PMNs.

The process would probably be the same as with honey A given the presence of PMNs, however the speed of action of honey B is certainly faster. Seven days after the infusion of honey "B", 12vs 15 of the cows suffering, presented a total reabsorption of uterine fluids with total disappearance of the purulent discharge and 02 were in heat on the seventh day with translucent mucus. Cytologically, the smears were very rich in endometrial cells.

In 03 cows the purulent discharge was still present; one of these cows was suffering from chronic acidosis.

12 Animals were inseminated in the second estrus after treatment 6 in group A (4 artificial insemination and 2 natural projection) 6 in group B (5 artificial insemination and 1 natural projection). The conception rate was estimated after two months of the last insemination. conception rate was 5 in group A and 3 in group B (2 natural projection and IA failed).

CONCLUSION

Results obtained herein with use of honey in bovine endometritise specially those antibiotic none-responding, has shown promising results in terms of therapeutic efficacy and conception rate following recovery of cows. This therapy is very economical under field conditions.

CONFLICT OF INTEREST

The authors declare that they have no conflict of interest.

INFORMED CONSENT

Informed consent not applicable in the current study.

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