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Evaluation the Efficacy of Mixed Herbal Essential Oils as a Treatment Option for Clinical Endometritis in Dairy Cattle

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Abstract

The endometritis form of uterine infection is considered as a common reproductive disorder deleterious to the reproductive performance of dairy herds. A wide variety of treatments with controversial results have been reported for endometritis, including local or systemic administration of antibiotics or disinfectants as well as hormone application. These treatments have side effects on endometrium, and antibiotic residues are found in milk following intrauterine and systemic antibiotic therapies. The aim of this study is to evaluate the possible effects of the mixed essential oil of Satureja bachtiarica Bunge, Artemisia Aucheri Boiss and Syzygium aromaticum (L.) Merr. & L.M.Perry on treatment of clinical endometritis in dairy cattle. One hundred and twenty cows with clinical endometritis were selected and randomly assigned to one of the following three groups: the HM group received mixed herbal essential oils, the OX group received 2.5 g oxytetracycline HCl, and the EX group received 1 g of ceftiofur sodium, all by intrauterine injection. The cleaning and first service conception rate was significantly higher in HM group than the EX, whereas the mean open days were lower in HM than the EX group. The number of service per conception was also significantly lower in HM group than OX and EX groups. In general, reproductive performance after herbal treatment was quite comparable to chemical antibiotic therapy and even better in some other reproductive indices. The mixed essential oils treatment represents an effective potential alternative to postpartum therapy for cows with clinical endometritis.

Key words: Dairy cattle, Endometritis, Satureja, Artemisia, Syzygium

Introduction

The endometritis is a common reproductive disorder detrimental to the reproductive performance of dairy herds. This form of uterine infection is described as a superficial inflammation of the endometrium which is combined with a

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purulent or mucopurulent uterine discharge visible in the vagina at ≥ 21 days of milking (I Martin Sheldon, Lewis, LeBlanc, & Gilbert, 2006). Clinical endometritis adversely influences reproduction and milk production. Reduced conception rate, prolonged open days, increased culling rate, and considerable financial losses are considered as common consequences for this kind of infection (Drillich & Wagener, 2018; Iain Martin Sheldon & Owens, 2018).

A wide variety of treatments have been suggested for endometritis; results of which are debated among veterinary practitioners. These therapies include local or systemic administration of antibiotics or disinfectants as well as hormone application. Intrauterine (IU) antibiotic treatment has been initiated with the aim of reducing endometritis negative effect on fertility through decreasing bacterial contamination of the uterus and its concomitant inflammation (R. Gilbert, Santos, Galvão, Brittin, & Roman, 2007). Tetracycline, penicillin, cephapirin, ceftiofur hydrochloride and other compounds have been tried in the treatment of endometritis with controversial results (Makki, Gheisari, & Ahmadi, 2016; Schlegl et al., 2020). Most studies indicate that local and systemic administration of antibiotics has a limited success in curing endometritis and that it may interfere in uterine defense mechanisms (Mollett, Elmore, Blanchard, & Berg, 1985; Oxenreider, 1982). Similarly, some field trials have demonstrated that intrauterine infusions of antibiotics in various protocols have generally failed in improving reproductive efficiency over the untreated control group (Thurmond, Jameson, & Picanso, 1993) or an alternative group treated with PGF2α (Olson, 1996; I. Sheldon & Noakes, 1998b).

Although some studies have recommended PGF2α as an effective treatment for endometritis with minimum harm to uterus and residual in milk and meat (R. O. Gilbert & Schwark, 1992; Heuwieser, Tenhagen, Tischer, Lühr, & Blum, 2000; Olson, 1996; I. Sheldon & Noakes, 1998b), there has been limited success (Hirsbrunner, Burkhardt, & Steiner, 2006) and there isn't any specific evidence for improved reproductive performance of dairy cows with clinical endometritis. Thus, uncertain efficacy of intrauterine medication, inconsistent recovery rate, possibility for suppression of the immune system, the risk for public health in term of drug residues in milk and meat as well as high cost of treatment have urged researchers to find alternative therapies.

Herbal therapies have long been used in the treatment of several human disorders (Klepser & Klepser, 1999), but information regarding the treatment of reproductive diseases in farm animals are limited. Some studies have used plant extracts in the treatment of clinical endometritis and have reported minor adverse effects on reproduction and milk production as well as low cost of the treatment, and its efficacy against a variety of disorders (Esparza-Borges & Ortiz-Marquez, 1995).

Three plants commonly used in herbal therapies are *Artemisia Aucheri Boiss, Satureja bachtiarica Bunge and Syzygium aromaticum (L.) Merr. & L.M.Perry*. *Artemisia Aucheri* has been found to have antiseptic, analgesic, anti-inflammatory, and antioxidant properties (Loizzo et al., 2008). Also, its essential oil is enriched with high percentage of thymol and carvacrol with antibacterial and antifungal activities (Lopes-Lutz, Alviano, Alviano, & Kolodziejczyk, 2008; Ramezani, Fazli-Bazzaz, Saghafi-Khadem, & Dabaghian, 2004; Setzer, Vogler, Schmidt, Leahy, & Rives, 2004). In ethnopharmacology, A*rtemisia* species are frequently utilized for the treatment of diseases including bacterial, fungal and viral infections (Abad, Bedoya, Apaza, & Bermejo, 2012). Likewise, the antimicrobial and antioxidant activities of *Satureja bachtiarica Bunge* have been proven (Ahanjan et al., 2011), and *Syzygium aromaticum (L.) Merr. & L.M.Perry* is believed to have anti-inflammatory and antiseptic activity. The

latter is traditionally used in inducing uterine contractions (Bonjar, 2004). *Satureja bachtiarica Bunge* is traditionally used as an antiseptic and analgesic agent in Iran (Zargari, 1992) and *Syzygium aromaticum (L.) Merr. & L.M.Perry* is widely used for treatment of infectious disease in Iranian traditional medicine (Keyhanfar, Nazeri, & Bayat, 2012).

The beneficial effects of other herbs on treatment of reproductive disorders have also been investigated. For example, several studies have demonstrated the positive effects of Zataria multiflora on reproductive disorders in human (Lopes-Lutz, Alviano et al. 2008, Abdali, Jahed et al. 2015) and in dairy cattle (Hajibemani, Mirzaei et al. 2016).

In this study, we attempted to analyze the possible impact of a mixed essential oils of *Satureja bachtiarica Bunge*, *Artemisia Aucheri Boiss* and *Syzygium aromaticum (L.) Merr. & L.M.Perry* on clinical endometritis as well as reproductive performance in dairy cattle. At the same time, the efficacy of intrauterine injection of oxytetracycline and ceftiofur sodium commonly used in the treatment of clinical endometritis is evaluated and compared with the herbal group. The effectiveness of these three protocols are evaluated by calculating and comparing parameters of herd reproductive performance.

Materials and methods

Essential oils of Syzygium, Artemisia and Dianthus were purchased from Barij Essence (Iran, Kashan). A mix vial of these three essential oils was prepared for one intrauterine injection. Its total volume was increased to 50 ml by adding distilled water. The chemical composition of this herbal mixture derived by gas chromatography-mass spectrometry (GC-MS) is presented in table 1. Gas chromatography-mass spectrometry was performed by Thermoquest 2000 GC (Thermo Quest, USA) equipped with Thermo Finnigan Mass system and a DB-1 capillary column (30 m × 0.25 mm; 0.25 lm film thickness). Helium was the carrier gas. Mass range was from m/z 35-375 amu, and the mass spectra were taken at 70e V.

The field approach of this study was performed at a commercial Holstein dairy farm with 2200 lactating cow (Zagros Milk and Meat Co., Shahrekord, Iran). Cows were fed a total mixed ration (TMR), milked three times a day, and received a complete herd-health service and reproductive management. The ration composition based on the percentage of the total feed consumed included: 35% concentrate, 5% alfalfa hay, 18.5% Beet pulp, 35% corn silage, <1% wheat straw and 5.5% water. They were examined for complete reproductive examination including edendometritis at 30 days of postpartum. The examination included vaginal inspection and ultrasonographic evaluation of ovaries and uterus. The clinical endometritis was defined according to Sheldon et al, (2009) as the presence of purulent (>50% pus) or mucopurulent (approximately 50% pus, 50% mucus) uterine exudate in the vagina, 21 days or more post-partum, without any systemic signs (I. Sheldon, Price, Cronin, Gilbert, & Gadsby, 2009). In addition to the observation of the presence of abnormal uterine discharge externally on the perineum, vulva and tail, transrectal ultrasonography was used to examine the entire reproductive tract. Ultrasonographic examination included imaging of abnormal fluid at the anterior vagina, measurement of cervical size (≤7.5 cm; >7.5 cm), and imaging of abnormal fluid in the uterine lumen. Care was taken to distinguish the normal fluid accumulation during estrus. Cows with clinical endometritis had cervical size >7.5 cm or had purulent or mucopurulent discharge externally, at the anterior vagina or in the uterine lumen (Kasimanickam, Cornwell, & Nebel, 2006). Those that showed only opaque discharge, not purulent or mucopurulent (mild endometritis) were left out of this study.

Table 1. The chemical composition of the herbal mixture derived by gas chromatography-mass spectrometry (GC-MS). Compounds were identified by comparison with MS database spectra, retention time (RT), area percentage and pure reference chemicals, and are listed in order of elution from the column

No.	Compounds	RT	Area percentage
1	Alpha pinene	8.8	0.41
2	Camphene (CAS)	9.32	0.8
3	Beta Myrcene	11.25	0.21
4	Yomogi alcohol	11.94	0.2
5	Alpha terpinene	12.56	0.32
6	Cymene	13.1	6.7
7	1,8-Cineole	13.25	2.26
8	Trans-2,7-Dimethyl-4,6-octadien-2-ol	13.75	0.3
9	Gamma terpinene	14.6	1.77
10	Artemesia alcohol	15.73	0.33
11	Beta Thujone	16.42	10.1
12	Alpha Thujone	16.77	2.67
13	Bornanone	17.6	4.5
14	Verbenol	17.8	0.8
15	p-Menthone	17.9	0.2
16	Cyclopentane	18.35	0.8
17	Endo borneol	18.45	1.8
18	Menthol	18.66	0.24
19	3-Cyclohexen-1-ol	18.72	0.67
20	Beta fenchol	19.16	0.41
21	Chrysanthenyl acetate	20.74	0.4
22	Bicyclo [2.2.1] heptan-2-ol, 1,7,7-trimethyl-, acetate	21.32	0.24
23	m-Thymol	21.97	3.1
24	Carvacrol	22.2	9.57
25	Eugenol	23.24	36.75
26	Trans-caryophyllene	24.26	5.76
27	Alpha Humulene	24.9	1.1
28	Acetyleugenol	26.25	4.85
29	Caryophyllene oxide	27.3	1.35
30	10,10-Dimethyl-2,6-dimethylenebicycle [7.2.0] undecan-5.beta.-ol	28.3	0.21
31	1,2-Benzenedicarboxylic acid, bis (2-ethylhexyl) ester (CAS)	34.78	0.58
Total			99.4

Cows were bred on observed estrus after cleaning, and pregnancy diagnosis was performed by ultrasonography examination around 30 days after insemination. At the end of clinical examination, 120 cows with clinical endometritis were selected and randomly assigned to one of the following groups: (1) HM group receiving the mixed herbal essential oils, (2) OX group receiving 2.5 g oxytetracycline HCl (Oxyvet® 5%, RAZAK, Iran) (I. Sheldon & Noakes, 1998a), and (3) EX group receiving 1 g of ceftiofur sodium (Excenell® 4 g, Pfizer, Madrid, Spain) (Galvão, Greco, Vilela, Sá Filho, & Santos, 2009) diluted in distilled water. All three groups had one intrauterine injection after endometritis confirmation by ultrasonographic and visual observation. The total volume of injection for all groups was 50 ml that was injected into the uterine lumen by using the disposable catheter. After the catheter entered the vagina, the tip of the catheter was fixed in the external os. of the cervix and was passed through the cervix into the uterine lumen by gentle oscillating movements of the cervix.

Intrauterine administration of oxytetracycline and ceftiofur hydrochloride was a routine procedure for treatment of clinical endometritis at the time of our study.

After about one week, the cows were reexamined, and clean ones received hormonal treatment protocols for estrus induction based on their ovaries structure. These protocols included heat-synch, ov-synch, one PGF2α injection or two PGF2α injections 12 days apart. All cows were inseminated at observed estrus if they were detected clean. Non-clean cows retreated (if necessary) with another antibiotic dose and inseminated on observed estrous. In this case, the cows that were previously treated with oxytetracycline were treated with ceftiofur in the next round, and vice versa. Also, the cows that were not treated with the mixed essential oils, were treated with one of these two antibiotics (oxytetracycline or ceftiofur) in the next round.

The efficacy of treatment protocols was evaluated by reproductive performance parameters including cleaning rate, days open, calving to first service interval, first service pregnancy rate (FSP) and service per conception (SPC). Cleaning rate was calculated in percent by the number of detected clean cows during reexamination following treatment divided by all the treated cows in the group (Drillich, Raab, Wittke, & Heuwieser, 2005). First service conception rate was calculated separately for cleaned cows, and cleaned and non-cleaned cows together. This was for evaluation of the first treatment effect on general uterine health and the result of other treatments in progress.

Data obtained for all three groups, open days, service per conception and calving to first service interval were analyzed with one-way analysis of variance through general linear model, and the means were compared with Tukey test. The results are presented as least square means and standard error. The cleaning rate and first service pregnancy rate were compared with Chi-square between three treatment groups. Statistical software SAS 9.1 (SAS Institute, Inc.) was used throughout analysis, and $P < 0.05$ was considered statistically significant.

Results

A total of 120 dairy cows were included in this study, of which 39, 41 and 40 were assigned to the HM, OX and EX groups respectively. The prevalence of endometritis in the herd was around 30% of postpartum cows at the time of clean test. The results of cleaning rate, first service conception rate of cleaned cows and first service conception rate of cleaned and non-cleaned cows are presented in table 2. The treatment rate was calculated for cows of each group. In HM group, 69.2%, in OX group, 56.1% and in EX group, 40% were cleaned after one intrauterine treatment. The significant difference was seen between groups HM and EX, but no such difference was observed between groups HM and OX. Considering only the cleaned cows, the first service conception rate was significantly higher for group HM than group EX but there was no significant difference between groups HM and OX. Moreover, when all the cleaned and non-cleaned cows were considered together, the results were similar, and were significant only between groups HM and EX with higher rate for HM group (table 2).

The mean open days were significantly lower in HM group than EX group. Similarly, the service per conception was significantly lower for HM group than groups OX and EX. There was no significant difference between the groups in terms of calving to first service (table 3).

Table 2. The percent of cleaning rate and first service conception rate in cleaned cows and first conception rate in cleaned and non-cleaned cows in three treatment groups. Different letters in a column show significant difference between groups (P< 0.05)

	Cleaning rate (%)	First service conception rate in cleaned cows (%)	First service conception rate in cleaned & non-cleaned cows (%)
Mixed herbal essential oil (n=39)	69.2[a]	62.96[a]	48.72[a]
Oxytetracycline (n=41)	56.1[ab]	34.78[ab]	29.27[ab]
Excenel (n=40)	40[b]	20[b]	22.5[b]

Table 3. Mean±standard error of open days, calving to first service interval and number of service per conception in three treatment groups. Different letters in a column show significant difference between groups (P< 0.05)

	Open days	Calving to first service	Number of service per conception
mixed herbal essential oil (n=39)	106±0.2[a]	70±0.4[a]	2.02±0.22[a]
Oxytetracycline (n=41)	132±0.8[b]	70±0.3[a]	2.6±0.21[b]
Excenel (n=40)	120±0.12[ab]	64±0.37[a]	2.6±0.21[b]

Discussion

A total of 120 dairy cows were enrolled and assigned to one of three protocols for the treatment of endometritis focusing on improvement of reproductive parameters among the herd groups. The main protocol was based on the intrauterine infusion of mixed herbal essential oils, and a healthy control group was not included to avoid possible suffering of the animals involved and/or economic losses by the dairy herd. The mixed herbal medicine was a combination of essential oils of *Satureja bachtiarica Bunge, Artemisia Aucheri Boiss* and *Syzygium aromaticum (L.) Merr. & L.M.Perry*.

According to the results, intrauterine administration of mixed herbal essential oils was more effective against clinical endometritis relative to oxytetracycline and excenel. This finding is in agreement with the experiments showing antibacterial and anti-inflammatory properties of plants used in the mixed herbal medicine. According to some studies, *Artemisia Aucheri* containes analgesic, antiparasitic, antibacterial, anti-inflammatory and antiseptic agents, and is effective in treating visceral pain (Asghari, Jalali, & Sadoughi, 2012; Mahboubi & Bidgoli, 2009). Likewise, *Satureja bachtiarica* is an aromatic medicinal plant with antimicrobial, antioxidant and antiviral activities against several types of viruses (Loizzo et al., 2008; Saab et al., 2012). Moreover, *Syzygium aromaticum (L.) Merr. & L.M.Perry* possesses anti-inflammatory and antiseptic drugs active in treating uterine diseases through induction of uterine contractions (Gyrdagva, 2004).

In addition, some studies have reported that uterine bacterial contamination is low during the first weeks after calving and endometrial inflammation is not always concurrent with bacterial infection (R. Gilbert et al., 2007). Hence, it is likely that our mixed herbal essential oils act as an anti-inflammatory agent, or reduces the bacterial load to a point where inflammatory stimulus diminishes and uterine defenses gain dominance.

As mentioned, some of the compounds used in this study, such as Artemisia Aucheri, have anti-inflammatory properties in addition to antiseptic properties. However, the aim of the present study was to evaluate the effect of herbal essential oils in the treatment of clinical endometritis as

an alternative treatment. Therefore, the therapeutic effect of intrauterine injection of these essential oils was compared with the common treatment of this condition, which is intrauterine injection of some antibiotics. Although flunixin meglumine is used in some treatment protocols for endometritis in dairy cows, it is not a common treatment and is usually used in combination with other antibiotics such as oxytetracycline (Königsson, Gustafsson, Gunnarsson, & Kindahl, 2001; Tek et al., 2010) and intrauterine injection of antibiotics is usually the treatment of choice. Therefore, in this study, intrauterine injection of herbal essential oils was compared with intrauterine injection of antibiotics as an alternative treatment.

Animals with clinical endometritis show reduced fertility. Cows with endometritis usually experience delayed resumption of ovarian cycle after calving, prolonged postpartum luteal phases, low conception and submission rates, long calving to conception interval and high culling rate (Drillich & Wagener, 2018; Mohammed, Mann, & Robinson, 2019). All these abnormalities eventually lead to longer mean open days, which determine herd's reproductive management efficiency. Based on our results, calving to first service interval showed no significant difference among groups, but open days for HM group was significantly lower than OX group. In addition, number of service per conception for HM group was significantly lower than groups OX and EX. Conversely, herbal treatment was more effective in treatment of endometritis, and led to higher first service conception (FSC) rate for HM group. The total group (cleaned and non-cleaned) had similar results too, representing the effectiveness of our mixed essential oils, even in the cases which needed retreatment with another antibiotic, and probably left less negative effects on the endometrium.

The findings of studies on the effects of intrauterine administration of antibiotics on the reproductive parameters of dairy cattle are controversial. Some have reported that intrauterine infusion of either oxytetracycline or penicillin has no influence on time interval to pregnancy relative to untreated cases (Thurmond et al., 1993). Kutti et al (2000) have observed no significant effect on conception rate and calving to conception interval in the cases of severe endometritis in comparison with untreated cases (Knutti, Kupfer, & Busato, 2000). In contrast, some studies have reported that infusion of cephapirin benzathine and ceftiofur hydrochloride to cows with subclinical and clinical endometritis improves reproductive performance (Reppert, 2015). In another study, intrauterine cephapirine had no significant effect on resolution of clinical signs compared to untreated animals, but resulted in shorter time to pregnancy (LeBlanc et al., 2002). On the other hand, antibiotic treatment is presumed to interfere in normal uterine defense mechanisms via lowering bacterial antigens, leading to disruption of neutrophil migration, the release of inflammatory mediators and chemotactic factors into the lumen and endometrium (Bondurant, 1999). Irritation and coagulation necrosis of endometrium have been reported as the side effects of oxytetracycline therapy, which negatively affect uterine defense mechanisms and self-healing ability (R. O. Gilbert & Schwark, 1992; Hussain & Daniel, 1991).

Furthermore, residues of some antibiotics (such as oxytetracycline) administered during peak milk production appear in milk causing deleterious effects on the milk quality. Of course, presence of drug residues in food products is unpopular with the public health and thus, the risk of antibiotic resistance and the economic losses due to milk withdrawal make the intrauterine antibiotic therapy for endometritis unprofitable (Makki et al., 2016; Tan, Huang, Jiang, & Hu, 2007). However, endometritis is a localized inflammation of the uterine lining, which interferes in embryo survival and

implantation so the treatment is necessary to reduce the load of pathogenic bacteria, halt and reverse inflammatory changes as well as enhance regeneration.

A well-known advantage of traditional medicine is its ability to provide holistic therapy for interrelated diseases. Indeed, natural materials such as plant extracts as stimulators of uterus defense mechanisms have been considered as an alternative therapy for the treatment of reproductive disorders like infections. Schnellbach (1990) reported that an intrauterine infusion of Eucalyptus compositum solution improves first service and overall conception rate in cows with mild signs of endometritis compared with an untreated control group (Schnellbach, 1990). An alternative therapy with Zataria multiflora has been introduced for treatment of clinical endometritis in dairy cattle, which is as equally effective as penicillin-streptomycine therapy (Hajibemani, Mirzaei, Ghasrodashti, & Memarzadeh, 2016). Similarly, Esparza *et al*., (1995) found that a combination of plant extracts provides the necessary agents for the treatment of clinical endometritis without any deleterious effect on the milk quality (Esparza-Borges & Ortiz-Marquez, 1995). Also, Kumar et al., (2006) successfully applied garlic extract for the treatment of endometritis (Kumar et al., 2006). Intrauterine infusion of neem oil and extract of neem bark was examined for treatment of sub-clinical endometritis in repeat breeding cows. Infusion of neem oil fraction was found effective in this study and could significantly improve the conception rate (Brahmanand, Shukla, Kumar, & Kumar, 2019). In this regard, our mixed herbal essential oils significantly reduced the open days, and can be considered as a new therapeutic approach. Unfortunately, no reliable scientific information on clinical trials of treating cattle endometritis by essential oils of Satureja bachtiarica, Artemisia Aucheri and *Syzygium aromaticum (L.) Merr. & L.M.Perry* is available to compare our findings.

The results of this study indicate that intrauterine administration of mixed herbal essential oils of *satureja bachtiarica Bunge, Artemisia Aucheri Boiss* and *Syzygium aromaticum* can effectively treat dairy cows diagnosed with clinical endometritis, and improve their reproductive performance. The results were completely comparable to those of chemical antibiotics and even better in some reproductive indices. Thus, this herbal treatment represents an effective potential alternative to postpartum therapy for cows with clinical endometritis.

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Conflict of interest

The authors declare that they have no conflict of interest.

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ارزیابی کارایی مخلوطی از عصاره‌های روغنی گیاهی به عنوان یک روش درمانی برای اندومتریت بالینی در گاوهای شیری

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چکیده

بیماری اندومتریت یک از انواع شایع عفونت‌های رحمی است که سبب افت بازده‌ی تولیدمثلی گله‌های گاو شیری می‌شود. درمان‌های متعددی برای رفع این مشکل مورد استفاده قرار می‌گیرند که نتایج متفاوتی نیز از آن‌ها گزارش شده است. از آن جمله می‌توان به تزریق داخل رحمی یا سیستمیک آنتی‌بیوتیک‌ها و درمان‌های مختلف هورمونی اشاره کرد. درمان‌های آنتی‌بیوتیکی سیستمیک و داخل رحمی علاوه بر ایجاد عوارض جانبی مانند تأثیر سوء بر بافت اندومتریوم رحم، سبب ایجاد بقایای آنتی‌بیوتیکی در شیر نیز می‌شوند. لذا هدف از پژوهش حاضر ارزیابی استفاده از مخلوط عصاره‌ی روغنی گیاهان مرزه بختیاری، درمنه کوهی و میخک به صورت تزریق داخل رحمی در درمان اندومتریت بالینی در گاو شیری می‌باشد. در این مطالعه تعداد ۱۲۰ رأس گاو شیری مبتلا به اندومتریت بالینی انتخاب شده و به صورت تصادفی در ۳ گروه درمانی قرار گرفتند. گروه HM: دریافت مخلوط عصاره‌ی روغنی، گروه OX: دریافت ۲/۵ گرم oxytetracycline HCl و گروه EX: دریافت ۱ گرم ceftiofur sodium. تمام درمان‌ها به صورت تزریق داخل رحمی انجام شد. میزان پاسخ به درمان و آبستنی در اولین تلقیح در گروه HM به طور معنی‌دار بیش از گروه EX بود. همچنین تعداد روزهای باز در گروه HM به طور معنی‌دار کمتر از گروه EX بود. تعداد تلقیح به ازای آبستنی نیز در گروه HM کمتر از گروه‌های EX و OX بود. در مجموع نتایج مطالعه‌ی حاضر نشان داد که درمان داخل رحمی اندومتریت بالینی با مخلوط عصاره‌ی روغنی کاملا قابل مقایسه با آنتی‌بیوتیک‌های رایج بوده و حتی در ارزیابی برخی از شاخص‌های تولیدمثلی به طور معنی‌دار بهتر بود. لذا این ترکیب گیاهی را می‌توان به عنوان یک جایگزین آنتی‌بیوتیک‌های رایج در درمان عارضه اندومتریت در گاو شیری به کار گرفت.

کلمات کلیدی: گاو شیری، اندومتریت، مرزه، درمنه، میخک

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