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Research Article

Comparison of Intrauterine Ozone and Rifaximine Treatment in Cows with Subclinical Endometritis

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Abstract

The aim of presented study was to compare the effect of intrauterine ozone and rifaximine treatment in cows with subclinical endometritis. The study was conducted on 53 Simmental cows with subclinical endometritis, which was diagnosed by ultrasonographic examination. According to results, interval between treatment to pregnancy (46.4±6.2 vs. 40.0±6.0), interval from calving to pregnancy (129.4±9.0 vs. 125.0±13.1), and insemination number (3.2±0.3 vs. 3.1±0.5) after treatment were similar in the groups (P>0.05). In conclusion, intrauterine ozone treatment was observed as therapeutic as rifaximine and to be an alternative treatment approach in dairy cows with subclinical endometritis.

Keywords: *Cow, Ozone, Rifaximine, Subclinical endometritis*

Subklinik Endometritisli İneklerde İntrauterin Ozon ve Rifaksimin Tedavisinin Karşılaştırılması

Özet

Sunulan çalışmanın amacı, subklinik endometritisli ineklerde uterus içi ozon ve rifaksiminin etkisini kıyaslamaktı. Çalışma, ultrasonografik muayene ile subklinik endometritis tanısı konmuş 53 adet Simental ırkı inek üzerinde yürütüldü. Sonuçlara bakıldığında; tedavi sonrası gruplar arasında tedavi gebe kalma aralığı (46.4±6.2; 40.0±6.0), buzağılama gebe kalma aralığı (129.4±9.0; 125.0±13.1) ve tohumlama sayısı (3.2±0.3; 3.1±0.5) açısından benzer sonuçlar bulundu (P>0.05). Sonuç olarak intrauterin ozon tedavisinin, rifaksimin kadar tedavi edici olduğu ve endometritisli sütçü ineklerde alternatif tedavi yöntemi olarak kullanılabileceği gözlendi.

Anahtar sözcükler: *İnek, Ozon, Rifaksimin, Subklinik endometritis*

INTRODUCTION

Reproductive efficiency is essential for sustainable dairy production and failures in reproduction cause economic losses depend on drug cost, extended interval between calving and pregnancy, decreased pregnancy per artificial insemination, reduced conception and increased culling rate [1-3]. Naturally, postpartum uterus is fully contaminated with pathogenic bacteria until 2 weeks after calving and elimination of the contamination in postpartum period has determinative effect on uterine functions and future fertility [4,5]. Most of the pathogenic bacteria (nearly 90%) that contaminate the uterus after calving are reduced until 60 d postpartum [4]. However, due to some predisposing factors such as retained placenta, subclinical hypocalcaemia, sub-optimal dry matter intake, elimination of the bacterial contamination is delayed and become a permanent infection. This permanent infection has negative and sometimes detrimental effects on ovarian cyclicity, uterine involution, endometrial regeneration and establishment of pregnancy [6,7]. Finally, the conception rate is reduced virtually 20% and interval to pregnancy is extended more 30 d in the herds with postpartum uterine infections [8].

The treatment of postpartum infections include antibacterial, antiseptic and hormone therapies [9]. Although, various systemic and intrauterine antibiotics are commonly used in postpartum infections [8,10,11], antiseptics and hormones are also suggested due to its safety for drug residue and antibacterial resistance and less inactivation in pus that can be observed in antibiotic treatment [12-15].

An alternative therapy to antibiotics, ozone is presented to use in the treatment of intrauterine infections in cows due to its safety for herd and public health [16]. Ozone is a

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gaseous and unstable molecule composed of three oxygen atoms (O_3) that shows a tendency for quickly transforming into the oxygen. In comparison with iodine and chlorine, ozone has more antibacterial activity that inactivates the bacteria, spores and viruses in a few minutes following to exposure [17,18] by its bactericidal [17], immune-stimulating [19] and anti-inflammatory [20] effects.

Although ozone has been prepared in various forms such as cream, gas, injections, paillettes, foam, pearls, boluses, the lipohydroperoxides foam and ovuli, ozone products are mostly preferred in veterinary medicine especially in the treatment of uterine infections [21].

The objective of this study was to compare the effect of intrauterine ozone and an intrauterine antibiotic, rifaximine, treatment on pregnancy rate in the cows that were not pregnant until about 90 day after calving although inseminated several times.

MATERIAL and METHODS

Animals and Experimental Groups

Fifty-three Simmental cows, which were housed in a commercial dairy farm in Erzurum, were in the 3rd lactation with average 5025±320 kg milk yield per year and were not pregnant in the 90th day after calving although artificially inseminated at least 2 times, were taken to the study from October 2013 to March 2014. The cows were fed with corn and grass silage, and concentrate from the same food producer on each farm and milked twice daily. The cows in the study had body condition score between 2.5-3.5 according the described classification by Edmonson et al.[22]. Production parameters such as fertility, milk yield, health status and vaccination in the farm, followed by a herd management software program (Nedap® Livestock Management, Netherlands)

The cows were examined by clinical examinations (vaginoscopy, rectal palpation, rectal temperature) and ultrasonography (Agroscan AL®, Noveko International Inc., Angoulême, France) to diagnose intrauterine infections. In these examinations, status of cervical orifice (erythematous and fibrotic changes), character of cervical discharge (normal, purulent, mucopurulent or pus), changes in ovarian structures (follicular development, luteal structures, or cysts) and uterine health (normal, increased heterogeneity and uterine fluid) were evaluated. Cows with subclinical endometritis was identified by detection of fluid accumulation in ultrasonographic examination as described by Kasimanickam et al.[1]. However, cytological and microbiological examinations were not performed due to inappropriate farm conditions. Clinically positive cows were not taken to the study and treated with the conventional antibiotic and hormone treatment. The remaining 53 cows, which had no clinical signs of metritis, were divided into ozone (Group 1; n=30) and rifaximine (Group 2; n=23) treatment groups.

The cows in Group 1 were treated by intrauterine ozone foam (Sanofoam Spray®, Agriprom, Netherland), whereas the cows in Group 2 were treated by intrauterine rifaximine foam spray; Fatroximin®, Vetaş, Istanbul, Turkey). All treated cows were artificially inseminated starting from the first natural oestrus following the treatment. Pregnancy was diagnosed in the 35th day after artificially insemination by ultrasonography. The cows, which were not pregnant until 200th days after calving (G1 n=4; G2 n=5), were excluded from the study and culled because of infertility.

To assess the reproductive performance for the treated cows, interval from treatment to pregnancy, interval from calving to pregnancy, and insemination number were measured. The study was performed by the permission of Animal Use and Ethics Committee of Atatürk University (Permission number: 8/151-2014).

Statistical Analysis:

The fertility parameters were analysed using Independent Samples Test t (IBM SPSS Statistics 20, 2012). Results were considered as significant when P value was <0.05.

RESULTS

At the beginning of the study, 53 cows, which were not pregnant despite consecutive inseminations until about 90 day after calving, were clinically healthy for uterine disorders. However, pregnancy was detected average 46 and 39 days after intrauterine treatment in Group 1 and Group 2, respectively. Additionally, every pregnancy could be provided by 3 consecutive inseminations, averagely *(Table 1)* in both groups. According to the result, interval between the treatment and pregnancy, interval between calving to pregnancy, and insemination number were not statistically significant between the groups *(Table 1)*. Forty-four of 53 cows were pregnant until 200th days.

Table 1. *Effects of G1 (ozone foam) and G2 (rifaximin foam) on fertility parameters in cows*
Tablo 1. *G1 (ozon köpük) ve G2' nin (rifaximin köpük) ineklerde fertilite üzerine etkisi*

Parameters	Group	n	Mean±SE
Interval from calving to treatment	G1	26	83.1±9
	G2	18	85.0±11.1
Interval between treatment to pregnancy (d)	G1	26	46.4±6.2
	G2	18	40.0±6.0
Interval from calving to pregnancy (d)	G1	26	129.4±9.0
	G2	18	125.0±13.1
Insemination number after treatment	G1	26	3.2±0.3
	G2	18	3.1±0.5

P>0.05

DISCUSSION

The presence of pathogenic bacteria in uterus causes inflammation and histological lesions in endometrium, which delays uterine involution as well as perturbs embryo survival [23]. In addition to repressing the release of pituitary LH, bacterial products or inflammations threaten postpartum follicular development and ovulation mechanism in cattle [23,24]. Thus, infections extend the period of days open and days to first service conception and decrease the conception rate [3,25,26]. According to Gautam et al.[27] metritis and endometritis are the most significant disorders for subfertility in dairy cows. As described in woman with in-vitro fertilization failure, subclinical endometrial infection has a role in implantation failure, spontaneous abortion, and preterm birth. Bacterial endotoxins and inflammatory mediators produced by the host: cytokines and chemokines [28] were accused in these failures.

The objectives of the present study were to evaluate the influence of intrauterine ozone administration on fertility in Simmental cows. The number of days open and number of artificial inseminations until pregnancy were the evaluation criteria in the study.

Intrauterine ozone administration, which was provide a potent antimicrobial activity for a wide range of micro-organisms and high oxidation potential that lead to fast transformation into free oxygen, as an antiseptic treatment approach in case of intrauterine infections was reported in previous studies [21,29-32]. Additionally, ozone also increases host immunity by activating erythrocyte metabolism and local tissue immune response. Thus, it leads to micro-environmental healing [20] and provide a cure in undiagnosed metritis cases.

Some of the disinfectant solutions, which were used in treatment of uterine infections, might cause permanent inflammations and damages and subfertility in endometrium [13,16]. On the contrary of this findings, Zobel et al.[16] suggested ozone as a non-irritant disinfectant in uterus. After contact of ozone and endometrial tissue, immunomodulative capacity and following disinfectant effect of ozone increased. Thus, days open period could be shortened in cows with endometritis [33]. Also Zobel [31] stated decreased insemination number and increased fertilization rate after ozone treatment by reducing possible spermicidal effect of endometrial inflammation.

In the current study, a difference in fertility parameter was not observed in rifaximine and ozone treatment groups. Similarly, Zobel [31] was also stated compatible cure rates between ozone and antibiotic treatment in case of intrauterine infections. Moreover, previous reports were also suggested intrauterine ozone treatment alone or combined with parenteral antibiotics was a more efficacious treatment for retained placenta in cows when compared to hormonal and parenteral antibiotic treatment modalities [15,32]. Additionally, combined treatment with prostaglandin $F_{2\alpha}$ with ozone was stated as slightly more effective than cephapirin in postpartum endometritis cases with the advantage of no drug residue in milk and meat [31]. In the presented study, interval between treatments to pregnancy were also similar in rifaximine and ozone treatment groups. Additionally, similar results in insemination number were detected in the groups.

In the current study, similar open days results with Duricic [21,34] were achieved in cows with endometritis following to ozone treatment. In these studies days open period were varied between 118 and 133. Likewise, the period was about 129 d in this study.

In recent years, the dairy herds are trying to establish new strategies such as homeotherapy, immunotherapy, and disinfectant use to minimize antibiotic consumption. These approaches become a trend especially in intra-mammary and intrauterine disease. According to results, ozone treatment was as therapeutic as rifaximine treatment during endometritis in cows. Due to ozone treatment has some advantages such as its non-irritant structure, safety for drug residue in milk, prevention against possible bacterial resistance, and inexpensiveness, the ozone treatment may be an alternative approaches to intrauterine antibiotics in dairy herds.

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