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# Prelliminary Study on Ozone Therapy in Postpartum Endometritis of Dairy Cows

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#### Abstract

The aim of research was to evaluate the effects of intrauterine foam ozone application in parturient endometritis on first service conception rate (FSCR), the average of straws until pregnancy (ASUP), the average of days not pregnant to first service (ADOFS) and the average of days not pregnant to pregnancy (ADOP). Another objective of this study was to observe the influence of ozone against uterine bacteria. The study involved 24 Holstein and Cross Breed Holstein cows, divided into two groups (Ozone O n=12, and Control C n=12) based on the uterine findings and diagnosis (puerperal endometritis and control). In cases when endometritis was diagnosed, we used ozone foam (RIGER SPRAY®, Novagen, Italy). Ozone was applied by means of a vial containing ozone foam under pressure for 10 seconds. Bacterial and fungal samples were taken at the first uterine examination and the second probe, at the moment of artificial insemination. After the days open period, all animals were put on hormonal therapy to induce the oestrus and then were inseminated. The next step was the ultrasonographic pregnancy diagnosis after 29-32 days. Bacteriological examination of uterine aspirate samples from the animals revealed single type of bacteria in 7 cases (29.16%) and mixed bacterial isolates in 17 cases (70.84%). The most isolated bacteria from group O was Escherichia coli (91%) followed by Staphylococcus hyicus (33%), Staphylococcus bovis (25%) and Arcanobacterium pyogenes (25%) and from group C Escherichia coli (66%), then Streptococcus bovis (16%). No fungal species were found. The median days open to first service and the median days open to pregnancy were similar in both groups: group O (65.4 days) and group C (65.1 days) for ADOFS, and for ADOP 112.3 days respectively 108.9 days. The significant difference between the two groups was observed for the FSCR and for the ASUP where half of the cows from the group O were pregnant at first service.

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#### 1. Introduction

Regarding high-yielding dairy cow's management, the transition period (3 weeks before and 3 weeks after parturition) is particularly at risk because animals undergo pronounced physiological changes that can cause suppression of immunological mechanisms (Islam et al., 2014).

Relaxation of the vulva and cervical dilatation during and after the onset of parturition allows the entry of bacteria into the uterus, causing infection in 80-100% of cows by 14-21 days post-partum (Sheldon et al., 2006; Islam et al., 2013). The most common bacteria are represented by *Escherichia coli*, *Arcanobacterium pyogenes*, *Pseudomonas aeruginosa*, *Pasteurella multocida*, *Staphylococcus aureus*, *Streptococcus uberis*, *Clostridium* spp., *Prevotella* spp. and *Fusobacterium* spp. (Singh et al., 2008; Kaçar and Kaya, 2014).

These bacteria compromise animal welfare, as well as causing subfertility and infertility because of uterine inflammation and/or infection (Zobel et al., 2014) represented by metritis, clinical and subclinical endometritis.

Endometritis may prolong the days open until first service, days open until pregnancy, the intercalving period, the conception rate and the risk of cows being culled due to infertility (Đuričić et al., 2015).

The most common way to treat endometritis is either intrauterine or systemic antibiotic administration (Zobel et al., 2012; Zobel et al., 2014). Other scientific works revealed the use of non-antibiotic substances such as: garlic extract (Sarkar et al., 2006), dextrose (Brick et al., 2012), neem - *Azadirachta indica* (Kumar et al., 2013), non-steroidal anti-inflammatory drugs, PgF<sub>2a</sub>, antiseptic solutions (Troitzky, 2010) and others.

A novel antibacterial, antifungal and immunostimulant therapy in animals is represented by different pharmaceutical forms of ozone (cream, gas, injections, paillettes, foam, pearls, boluses) (Djuricic et al., 2012 a; Zobel et al., 2012; Zobel and Tkalčić, 2013; Zobel et al., 2014; Đuričić et al., 2015). Ozone (O<sub>3</sub>) is a gaseous and unstable molecule composed of three oxygen atoms, colorless, with a specific smell and a tendency for quickly transforming into the oxygen (Djuricic et al., 2012 b; Đuričić et al., 2015; Ersoz et al., 2015; Polat et al., 2015). Some of these forms can be used in postpartum endometritis because the most important ozone derivatives are lipohydroperoxides, which act directly on pathogens and activate erythrocytes and immune cells (Zobel et al., 2014; Đuričić et al., 2015; Enginler et al., 2015; Ersoz et al., 2015; Calderon et al., 2016).

The aim of this study was to evaluate the effects of intrauterine foam ozone application in parturient endometritis on first service conception rate (FSCR), the average of straws until pregnancy (ASUP), the average of days not pregnant to first service (ADOFS) and the average of days not pregnant to pregnancy (ADOP). Another objective of this study was to observe the influence of ozone against uterine bacteria.

#### 2. Materials and Methods

#### 2.1. Animals

The study involved 24 Holstein and Cross Breed Holstein cows, divided into two groups (Ozone O n=12, and Control C n=12) based on the uterine findings and diagnosis (puerperal endometritis and control), from one commercial dairy farm in the south part of Romania (Ploiești County) with temperate-continental climate over the period of 5 months (Sept.-Feb.).

The cows were housed in free-stall barns and mattress bedding. Of crucial importance was the lying/resting area. All the females spent up to 14 h per day lying with about half of the resting period ruminating (Winckler, 2014). Cows were milked three times per day (morning, noon and evening milking schedule). All cows were fed with a unique nutritional ratio twice per day. The animals from the two groups had a physiological puerperium but the exclusion criteria included birth assistance, receiving systemic antibiotic therapy within 60 days prior to calving, abnormal internal genitalia (including adhesions), BCS < 2.5, systemic diseases, retention of foetal membranes, any kind of dystocia, including caesarean section, lameness, puerperal mastitis. Only cows following the second to the fourth partus were involved in the study. The groups' average milk yield during the study was 43.95 kg per day for C group and 40.84 kg per day for O group.

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