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**EFFECTIVENESS OF BUSERELIN IN EARLY PUERPERIUM ON RESTORATION OF OVARIAN CYCLICITY AND SUBSEQUENT CONCEPTION IN PRIMIPAROUS DAIRY COWS**

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**ABSTRACT**

The postpartum period is an important period in the reproductive life of both dairy and beef cows due to its great influence on subsequent pregnancies. A major reproductive problem on dairy farms is the delayed restoration of postpartum ovarian activity, which is reflected in a prolonged intercalving interval, where farmers suffer large economic losses. Dairy cows have between two and five months after parturition to become pregnant again if reproductive capacity and productivity are to be kept in desirable range. Utilisation of hormones is quite common in dairy industry, but to achieve improvement in reproductive performances they have to be used with caution. The study was focused on the restoration of the ovarian cyclicity in the early postpartum period and the reduction of the service period, where a total of 50 primiparous Simmental cows were divided into experimental and control groups. The experimental group was treated with the hormone buserelin, hormonal MSD preparation - Receptal®, which is a GnRH agonist was used in dose of 5 ml on the 10th, 11th, 12th, 13th or 14th day postpartum for earlier restoration of ovarian cyclicity. It is expected that about 30-40 heifers will require some assistance during parturition. A slightly higher number of animals had earlier restoration of ovarian cyclicity and had more conception in the experimental group in contrast to the control group. Buserelin applied at day 13<sup>th</sup> or 14<sup>th</sup> looks to be relatively efficient in shortening the service period and intercalving interval in primiparous simmental cows.

**Keywords:** *primiparous cow, buserelin, ovarian cyclicity, service period.*

## INTRODUCTION

A major reproductive problem on dairy farms is the delayed restoration of postpartum ovarian activity, which is reflected in a prolonged service period and intercalving interval, where farmers suffer large economic losses. In order to improve the reproductive parameters of cows, various hormonal methods are used to establish and regulate the sexual cycle. Hormonal therapy must be accompanied by adequate nutrition. GnRH and PGF2 $\alpha$  can certainly improve the reproductive results of cows only if proper nutrition is included, especially when milk production increases. Body condition at parturition has a direct positive influence on the restoration of ovarian cyclicity after calving. One of the most common reasons for postpartum anestrus is the sudden loss of the cow's body mass as a result of a negative energy balance and due to an increase in milk production during first 2-3 months of lactation. This is result of a direct connection of a negative energy balance to the inhibition of GnRH release (Peter and Ambrose, 2009). Any deviation in the length of the estrus cycle is considered pathological. Factors affecting the length of the estrus cycle can be of different etiologies such as temperature or transport stress, but also diseases of the reproductive system. Any hormonal therapy cannot correct deficiencies in the management itself. The postpartum period is an important period in the reproductive life of both dairy and beef cows due to its great influence on subsequent pregnancies. The entire postpartum period or puerperium, is defined as the period from birth until reproductive organs return to their normal anatomical and histological condition. Any prolongation of the puerperium in cows could have a detrimental effect on reproductive performance. Essentially, the determination of this period basically depends on the continuation of normal cycles on the ovaries, the manifestation of estrus and conception (Stevenson, 2014). Dairy cows have between two and five months after parturition to become pregnant again if reproductive capacity and productivity are to be kept in desirable range. To achieve this goal in dairy cows, it is necessary to bring the ovaries and uterus back to a normal physiological state that will allow conception (Mutevelić et al., 2019). In extensive ways of keeping dairy cows, it is assumed that about 75% of cows with very high milk yield will have an extended time to restore cyclicity and between 20 and 48% of cows manifest prolonged postpartum anestrus (Peter and Ambrose, 2009). Inadequate climatic factors, high milk production, poor physical condition, poor housing conditions, inadequate nutrition qualitatively and quantitatively represent significant factors that influence the prolongation of the interval from parturition to the first ovulation. Also, as a result of these factors, postpartum disorders and diseases of infectious and non-infectious etiologies, affect the first appearance of estrus postpartum (Shresthaa, 2004). All the listed factors directly influence the prolongation of the inhibition of the release of pituitary gonadotrophins (LH and FSH), which consequently delays the normal development and ovulation of the dominant follicle and the function of the *corpus luteum*. Our research is focused on the restoration of the ovarian cyclicity in the early post-partum period and the reduction of the service period.

### **MATERIALS AND METHODS**

The research was conducted in the modern farm conditions for production of dairy cows "Spreča", Gornje Vukovije, Tuzla Canton in Bosnia and Herzegovina, where dairy cows of the Holstein-Friesian and Simental breeds are bred in free stall system.

The feed consists of winter and summer meals and milking is done three times daily with the DeLaVal system. Veterinary technicians are responsible for the daily inspection of cows in production and detection of estrus. In doubtful cases, veterinary doctors re-examine such cows. Insemination of cows is done on average 60 to 80 days postpartum and based on the assessment of the doctor of veterinary medicine on the farm, some inseminations can be performed even earlier depending on the general condition of the cow.

Our research included a total of 50 primiparous Simental cows, divided into experimental and control groups. The experimental group was divided into cows that were treated with the hormone buserelin (Receptal®) on the 10th, 11th, 12th, 13th or 14th day postpartum for earlier recovery of ovarian cyclicity. The period of the study was autumn/winter of 2020/2021.

For the application of buserelin, the hormonal MSD preparation - Receptal®, which is a GnRH agonist, was used (1 ml of injection solution contains 0.0042 mg of buserelin acetate - 0.004 mg of buserelin and 20.0 mg of benzyl alcohol). For the treatment, the manufacturer's recommended dose of 5ml in the early post partum period was used.

Research was conducted after dividing the experimental cows into 5 groups, i.e. on which day postpartum they received Receptal. The first group was administered on the 10th day postpartum (n=5), the second on the 11th day postpartum (n=5), the third on the 12th day postpartum (n=5), the fourth on the 13th day postpartum (n=5) and on the fifth, 14th day postpartum (n=5). All primiparous cows that had parturitions with or without the assistance of a veterinary doctor participated in the research. The control group consisted also of 25 primiparous cows. All cows in the study were monitored in the postpartum period of 20-30 days, when a transrectal ultrasonographic examination was performed to detect changes in the ovaries for cyclicity. In the period 40-100 days postpartum, artificial insemination was performed and a pregnancy examinations were performed 40- 50 days later.

Statistical data processing was carried out using the t-test to determine the significance of differences.

### **RESULTS AND DISCUSSION**

Research showed that out of 25 pregnant heifers in the experimental and control group, 11 (44%) animals of the experimental and 8 (32%) animals of the control group needed assistance during calving to correct the position of the fetus and provide more power for extraction.

Table 1. The number of primiparous cows of the experimental and control groups that had a difficult parturition.

	<b>Experimental group</b>	<b>Control group</b>
<b>Heifers with assistance during parturition</b>	11 (44%)	8 (32%)
<b>Heifers without assistance during parturition</b>	14 (56%)	17 (68%)
<b>Total number of heifers</b>	25	25

Clinical examination of the ovarian status and the established number of cyclic cows after the Receptal application on the 10th, 11th, 12th, 13th or 14th day post partum and the control group. The first three groups have mostly the same or similar values compared to the control group, while groups 4 and 5 have slightly higher values – more cyclic cows compared to the control group.

Table 2. Established number of cyclic cows in experimental groups and their controls.

<b>Confirmed cyclicity on day PP</b>	<b>Group 1.</b>	<b>Con.</b>	<b>Group 2.</b>	<b>Con.</b>	<b>Group 3.</b>	<b>Con.</b>	<b>Group 4.</b>	<b>Con.</b>	<b>Group 5.</b>	<b>Con.</b>
20-25	3	2	2	2	2	1	4	2	3	2
30	2	1	3	2	2	3	4	3	4	2

For the first three groups the results are identical as their control, while groups 4 and 5 have a slightly higher values – more pregnancies compared to the control group.

Table 3. Number of pregnant and non pregnant cows in experimental groups and control animals after artificial insemination performed 40-100 days post partum (PP).

<b>AI 40-100 days PP</b>		<b>Pregnant</b>		<b>Not pregnant</b>	
Group 1	Control	2	2	3	3
Group 2	Control	2	2	3	3
Group 3	Control	2	2	3	3
Group 4	Control	3	2	2	3
Group 5	Control	4	2	1	3

A slightly higher number of animals conceived in the groups that received Receptal, in contrast to the conception of the control group. Also, the number of cows treated with Receptal, which did not conceive in the period 40 to 100 days post partum when artificial insemination was performed, does not differ much from the number of cows that did not conceive in the control group.

The results of ovarian cyclicity after the application of Receptal in the post partum period and the success of artificial insemination have no significant differences

compared to control cows, so this result is not significant ( $p < .05$ ), which is caused by the small number of treated cows that were available at the given time for the research.

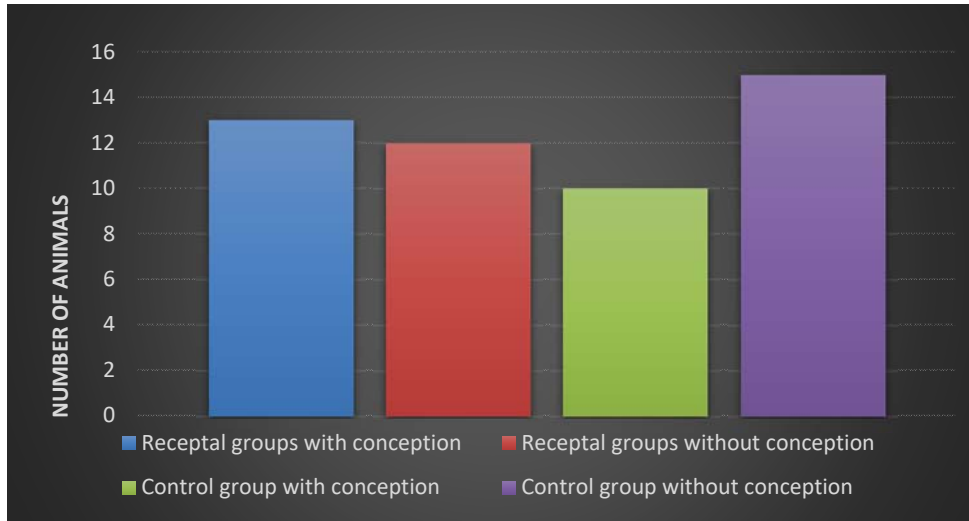


Chart 1. The chart shows the conception of primiparous cows treated with Receptal compared to the control group of primiparous cows.

The postpartum period has an important impact on reproductive performance. Despite all efforts and scientific knowledge, the fertility of dairy cows is constantly declining. Ovarian inactivity and postpartum uterine diseases, which later lead to postpartum anestrus are taken as the main contributing factor. Factors such as limited energy intake, lower body condition and postpartum diseases delay ovarian cyclicity and postpartum anestrus is common in dairy herds with high milk production. The restored cyclicity of the ovaries postpartum is influenced by the secretion of pituitary gonadotrophins (FSH and LH). In healthy cows, it is restored between the 2nd and 4th week postpartum and according to Reist et al. (2008), over 80% of cows should normally restore ovarian cyclicity within the first 35 days postpartum, which coincides with our research, while ovarian cyclicity was observed somewhat later in control cows.

After a normal parturition, according to Wattiaux (1996), the first ovulation occurs on average after the day 15th, the second on the day 32nd and the third on the day 53rd postpartum. The author points out that the first postpartum ovulation occurs as a silent one, without external signs of estrus as a normal occurrence in 76% of cows, which is consistent with our research in which we did not observe ovulation until the 15th day postpartum in both groups of cows. Our results also agree with Crowe (2008) where visible estrus is found on the day 32 (50% cows) and day 53rd (90% cows) postpartum.

In the research from Jeffrey and Benmrad (1985) on Holstein-Friesian cows that had puerperal disorders, a single application of GnRH in the period 10-14 days postpartum, reduced the time interval from parturition to the reappearance of physiological estrus and conception by 43 to 48 days. It should also be noted that cows that were administered GnRH 10-14 days postpartum had 26% to 41% fewer repeated artificial inseminations, i.e. conception was more successful. The author concludes that early postpartum treatments with GnRH improve the fertility of cows, especially in those with puerperal disorders. According to our results, the application of Receptal 10 – 14 days postpartum also led to a faster recovery of ovarian cyclicity. In our research, due to the smaller number of treated cows, the results did not show such improvements, but they indicated that the application of Receptal may restore ovarian cyclicity earlier.

In research by Leslie et al. (1984) after examining the effect of GnRH hormone administered in the period 8-14 days postpartum, in cows that had retained placenta, the authors conclude that there are no significant differences between treated and untreated cows regarding the interval from parturition to the first observed estrus, the interval from parturition to insemination, the interval from parturition to conception and the number of inseminations required to conception. However, for herds in which breeding began earlier in the postpartum period (herds having a mean less than or equal to 80 days from parturition to first service GnRH treatment resulted in a significantly shorter calving to conception interval as compared to control. Moreover, Lee et al. (1983) concluded that after GnRH application the conception rate increased by 15% to 18%, which is probably related to general and reproductive health status.

This conclusion of Lee et al (1983) agrees with our research in which we observed an increase in conception in cows that were administered Receptal 10., 11., 12., 13. or day 14th postpartum. It should be emphasized once again that due to the smaller number of cows that were included in the research, the number of cows with cyclicity and conception is not statistically significant, but it is noticeable.

### **CONCLUSIONS**

About 30 to 40% of heifers will require veterinary assistance before parturition. The application of GnRH is more effective in restoring ovarian cyclicity when administered on the 13th or 14th day postpartum. Cows receiving GnRH on these days also have a higher chance of conceiving earlier. While GnRH use in the early puerperium positively impacts the restoration of ovarian cyclicity and the timing of conception, the effects are not significant to a large extent.

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