



## The Effect of Intramuscular Oxytocin Injection on Milk Ejection in Dairy Cattles

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### ABSTRACT

Milking is a source of change in milk yield and milk quality determination, and one of the main events of milking is the oxytocin-mediated ejection reflex. Complete ejection of milk from the udder is due to the presence of high oxytocin during milking. The aim of this study is to investigate whether oxytocin administered intramuscularly has an effect on increasing milk ejection. In the study, 28 clinically healthy lactation Holstein cows were used, the lactation numbers and lactation periods of these cows were ignored in the evaluation. The study groups are divided into four groups, each consisting of seven cows. 50 IU oxytocin was given to group-1 five days before milking in the morning and evening, group-2 received 50 IU oxytocin once a day just before milking and the group-3 was given 50 IU oxytocin once before morning milking on the 1st, 3rd, and 5th days and the group-4 is a control group, oxytocin was not given to this group. Oxytocin injection was administered intramuscularly 30 minutes before milking. In the evaluation, the last five days of milk data before the application, five days of milk data during the application period and five days of milk data after the application period were taken into consideration. As a result of the study, we can say that the udder was not completely emptied with the intramuscular administration of oxytocin.

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## Introduction

In dairy cattles (or cows), a large portion of milk, 80-100%, is stored in the alveolar compartment of the udder. For this reason, milk extraction during breastfeeding and machine milking is very important to allow alveolar milk to be taken (Bruckmaier, 2003). The main role of the endocrine system is to synchronize udder growth and function for the animal's reproductive status and calf milk demand. Few levels of reproductive hormones, including estrogen (E), progesterone (P), placental lactogen (PL), prolactin (PRL), and oxytocin (OT), depending on the reproductive status and all of these hormones have a direct effect on the mammary gland (Neville et al., 2002). The release of milk from the alveoli into the cisternal spaces is induced by the oxytocin released from the posterior pituitary in response to tactile nipple stimulation of the calf, hand or milking machine (Al-EknaH and Homeida 1991; Belo and Bruckmaier, 2010; Bruckmaier 2005; Kabilan, 2014). Milking has an important role in determining milk yield and milk quality, and one of the main events of milking is oxytocin-mediated ejection reflex (Lollivier et al., 2002; Moberg and Prime, 2013).

Oxytocin is a peptide hormone consisting of nine amino acids released from the forebrain bundle, zona incerta, bed stria terminalis, substantia innominata and other similar nuclei, especially the supraoptic and paraventricular nuclei in the hypothalamus (Al-EknaH and Homeida 1991; Kabilan, 2014; Uzun and Sulu, 2002). Oxytocin comes from the nuclei it releases to the posterior pituitary, and from there it is transferred to the systemic circulation, it affects mainly the uterine muscle, mammary gland and seminiferous tubules, but also has receptors in the pancreas and thymus, affecting the cardiovascular system (Anonymous, 2005; Uzun and Sulu, 2002). The half-life of oxytocin in blood is 0.55 to 3.6 minutes (Neville et al., 2002).

Continuous ejection of milk depends on the presence of a high concentration of oxytocin during all milking. Disruption of milk ejection may result from peripheral inhibition of oxytocin effects on the mammary gland or inhibition of oxytocin released by the central nervous system (Bruckmaier and Blum, 1998). Not all milk can be taken from the udder, up to 90% of the milk stored in normal machine milking is ejected from the cows by

endogenous OT treatment. The remaining milk is ejected with an intravenous (i.v.) supraphysiological dose of OT (Macuhova et al., 2004).

In infections and traumas that may occur in the cow's udder, it is aimed to completely empty the udder in order to increase the effectiveness of the treatment. The aim of this study is to investigate whether oxytocin administered intramuscularly has an effect on increasing milk ejection.

## Materials and Methods

The research was conducted on 28 clinically healthy *Holstein breed cows* in a commercial livestock farm, where the same care and feeding program was performed, milking two times a day (morning and evening), milking of each animal was done at 06:00 a.m. and 04:00 p.m. The ages, lactation numbers and periods of lactating cows were not taken into account and cows were randomly grouped. The study groups were divided into four groups, each consisting of seven cows. 50 IU oxytocin was given to group-1 five days before milking in the morning and evening, group-2 received 50 IU oxytocin once a day just before milking and the group-3 was given 50 IU oxytocin once before morning milking on the 1st, 3rd, and 5th days and the group-4 was a control group, oxytocin was not given to this group. Oxytocin (Oksitosin 10 IU Injection Solution, Vetaş) (Anonymous, 2017) injection was administered intramuscularly (i.m.) 30 minutes before milking. Daily milk data of cows were collected by milk plan herd management program.

In the statistical analysis, a randomized block design plan was applied using SPSS (version 25).

Considering animal health and welfare, ethical committee permission was given by Adana Veterinary Control Institute Experimental Animals Local Board with the document dated 06.05.2019 dated 2019-6 / 1199 before the practice of studies.

## Results and Discussion

5-day average daily milk data of cows in the establishment and grouped for oxytocin application before application, application period and after application are

given in Table 1. Likewise, the data of the control group (Group 4), which is not applied, are given in the same table.

In Table 2, the difference between both groups and applications is statistically insignificant ( $P>0.05$ ).

And in Table 3, The effect of groups on daily average milk yield is statistically insignificant ( $P>0.05$ ), and the effect of applications on daily average milk yield is statistically insignificant ( $P>0.05$ ).

As seen in Table 2 and Table 3, no significant effect on the daily milk ejection of cows was detected in OT applications.

Some milk made by dairy cattles before milking is stored in the milk cistern and is ready to be removed. Most milk is removed from the udder due to the tactile stimulation of the milking machine and oxytocin release. In order for milk to be completely removed from the udder cistern, there should be an ejection event during the milking process (Bruckmaier and Blum, 1998). Oxytocin plays an important role in milk milking. Even a small amount of exogenous oxytocin can be effective in milk ejection or milk ejection can also be initiated by exogenous oxytocin application. Traditionally, oxytocin has now been used to collect harvest milk (Sagi et al., 1980).

Regarding exogenous oxytocin applications, Macuhova et al. (2004) reported that OT injection caused OT elevation in the blood level for two hours, but Bruckmaier (2003) reported that chronic oxytocin administration negatively affected milk ejection.

Bruckmaier (2003) studied the effect of oxytocin on milk ejection with i.m. injection, each with seven montofon cows, three groups and two different experiments. The three groups were control, placebo and 50 IU oxytocin (chronic administration) i.m. injection group. Oxytocin administrated daily in every morning and evening between 1-22 days. In the experiment-1, they examined milk data on the days 0, 7, 14, and 21. In the experiment-2, they examined milk data on the 1st, 8th, 15th, 22nd days with i.m. 50 IU oxytocin was applied to all groups on the specified days after pre-milking. In the experiment-1, the milk ejection, which was 95% on the zero day of the oxytocin group, on the 7th, 14th, 21st days, gradually milk ejection fell to 80% levels.

Table 1. Dairy cattle groups and 5-day milk averages.

	Before application Average of 5 days D. cattle / lt / day	Application Average of 5 days D.cattle / lt / day	After application Average of 5 days D. cattle / lt / day
1.Group (n:7)	17.56	16.46	18.77
2. Group (n:7)	16.33	17.55	19.3
3. Group (n:7)	16.47	17.58	17.57
4. Group (n:7)	18.24	16.97	17.22

Table 2. Between-subjects factors

Status	Sequence	N
Group	1.00	3
	2.00	3
	3.00	3
	4.00	3
Application	1.00	4
	2.00	4
	3.00	4

\* The difference between both groups and applications is statistically insignificant ( $P>0.05$ )

Table 3. Tests of between-subjects effects

	Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Intercept	Hypothesis	3675.700	1	3675.700	5188.979	0.133
	Error	0.268	0.378	0.708 <sup>a</sup>		
Group	Hypothesis	0.442	3	0.147	0.153	0.924
	Error	5.793	6	0.966 <sup>b</sup>		
Application	Hypothesis	3.053	2	1.527	1.581	0.281
	Error	5.793	6	0.966 <sup>b</sup>		

Dependent Variable: OT; <sup>a</sup>MS(group) + MS(application) - MS(Error); <sup>b</sup>MS(Error)

In the experiment-2, there was no difference between the milk ejection percentage values of the placebo, control and oxytocin groups on the 1st, 8th, 15th, 22nd days, while the milk ejection percentage values increased significantly compared to the experiment-1 days. Again the percentage data of the oxytocin groups in experiment-2 increased significantly compared to experiment-1. After each study, they administrated i.v. 10 IU oxytocin (Bruckmaier, 2003).

Macuhova et al. (2004) investigated the blood oxytocin values and the effect of oxytocin on milk formation with different experiments as well as intra-muscular oxytocin injection and milk ejection in montofone cows. In the examination, they also considered the intramammary pressure. They performed their studies on milk ejection on a total of 13 cows, 4 of them were primiparous. In practice, they gave 50 IU oxytocin intramuscularly and performed oxytocin applications just before milking after udder stimulation. They didn't get any reaction from 6 cows in an increase in intra-mammary pressure due to oxytocin, and in 7 cows, they reacted in an increase in the internal pressure of the udder. However, they observed a similarity in the amount of milk remaining in the udder in the reacting and non-reacting cows (Macuhova et al., 2004).

Bruckmaier (2003) and Macuhova et al. (2004) were performed OT applications after udder stimulation or just before milking or after pre-milking, but we performed our OT application 30 minutes before milking. According to our study data, OT doesn't have any significant effect on milk ejection. Although, in some of Macuhova et al. (2004)'s studies, reaction related to OT injection was observed in some cow udders, but no significant effect on milk ejection was observed and they obtained results similar to our study. In Bruckmaier (2003) experiment-1, a decrease in milk ejection was observed in chronic oxytocin administration, indicating that prolonged oxytocin administration may have different effects than the desired effect, and even chronic OT administration may have an inhibitory effect on an endogenous oxytocin release. In Bruckmaier (2003) experiment-2 (OT), the increase in milk ejection with OT supplementation for the 2nd time in the OT group showed that endogenous oxytocin, which is thought to be inhibited, was removed, and also this includes differences with the study findings made by us and Macuhova et al. (2004). In addition, in Bruckmaier (2003) experiment-2, the OT group, the control group, and the placebo group achieved significant increases in milk, which can be said to be caused by the second dose of OT in the same day. In other studies, it appeared in results with different findings according to our study, together with results supporting our study findings. When we ignore the individual physiology of each animal, the effects of the application of animals immediately before or after pre-milking are seen in different results.

In conclusion, the udder was not completely emptied with the intramuscular administration of oxytocin. We need to continue intravenous oxytocin administration to remaining milk from the udder until a new and easy intervention is found.

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