# Effect of Oxytocin Injection on Some Biochemical Parameters and Morphological of Some Organs Trails in Holstein Crossbreed Cows

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# ABSTRACT

This study was carried out in the commercial animal farms in the Aboalkassib village during November /2018, to investigate the influence of injected of oxytocin hormone on some biochemical parameters (glucose, cholesterol, total protein, albumin, AST and ALT) and hormones (LH, FSH, estradiol and prolactin) concentrations in the carotid artery and jugular vein. A total of 30 dry adult Holstein crossbreed cows aged between 5-6 years old were divided randomly into two groups: 15 cows as a control group and 15 cows were treated with oxytocin hormone at dose 9 ml/head by injecting in intramuscular. The results showed significant (P<0.05) increase in the concentrations of glucose, cholesterol, LH, FSH, estradiol and prolactin in the treatment group compared with the control one. No significant (P<0.05) differences in the concentrations of total protein, Albumin and liver enzymes (AST and ALT) were observed among the treatment group and the control group. Also, the results showed that concentrations of glucose and most of hormones such as LH, FSH, estradiol and prolactin were higher significantly (P <0.05) in a jugular vein compared with carotid artery, while the concentration of estradiol was higher significant (P <0.05) in carotid artery compared with jugular vein.

### **INTRODUCTION**

Blood is a unique fluid containing cells that is pumped by the heart to all over the body in a system of pipes known as the circulatory system., it carries oxygen and metabolic compounds to the tissues and cells of the body and removes waste products like carbon dioxide from them (Guyton, 1981). Blood have many other functions, namely: 1) keeping conditions in the body constant. 2) Keep the acidity or pH stable and helps maintain a constant temperature in the body. 3) Has an important role in defending the body against disease. The blood vessels, including: 1) arteries, which are carrying blood away from the heart, it has thick elastic walls that struggle the flow blood and can withstand the surges of high blood pressure. 2) Veins, which are carrying blood to the heart, in those vessels the blood flow slowly and for this reason veins have thinner walls than arteries. The carotid artery and jugular vein run side-by side of the neck, one pair on the left and one on the right, the carotid artery carries oxygenated blood up to the head while the jugular vein transport deoxygenated blood down to the heart (Guyton, 1981).

Oxytocin is a peptide hormone product by the nerve cells in the hypothalamus (magnocellular neurons and paraventricularnuclei) with structure contains nine amino acids, it is travelling along axons of this gland and inter into the posterior pituitary by the axon terminals where it is stored, when the posterior pituitary actived, oxytocin is released into the bloodstream and then transport by blood vessels to the organs of the body, the half- life of oxytocin is about 3 to 5 minutes (Cimpl and Fahrenholz, 2001; Chard, 1985). The main function of oxytocin is stimulating the mammal glands to secrete the milk, furthermore, stimulate the contractions of smooth muscle cells of the reproductive system (Martinet *et al.*, 1976; Hays and Van Demark, 1953). **Keywords:** Oxytocin injection, carotid artery, jugular vein, biochemical parameters, hormones, Holstein crossbreed cows.

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There are many studies about the effect of treatment with oxytocin on various functions such as improve the production of milk (Jalal and Kawa, 2012), regulation of the reproductive performance in cows, and improve the performance of corpus luteum (Tahawy and Sharkawy, 2014; Metwelly and EL-Bwab, 1999).

The purpose of this study was to determine the effect of oxytocin injected on some biochemical parameters and some hormone concentrations and compared between the content of arteries and veins from such as those parameters in Holstein crossbreed cows.

#### **MATERIALS AND METHODS**

This study was carried out in the commercial animal farms in the Aboalkassib village (17 km south of Basrah province center) during November /2018, to investigate the effect of injection oxytocin hormone on some biochemical parameters in both the carotid artery and jugular vein, additionally, the morphology of some organs. A total of 30 dry adult Holstein crossbreed cows aged between 5-6 years old were chosen randomly and divided randomly into two groups: 15 cows as a control group and 15 cows were treated with oxytocin hormone (Manufactured by the Anova Joint Venture Company, Vietnam) at dose 9 ml/head by injecting in intramuscular. The injected of hormone was in the morning about 7:30 a.m. and for once time, after one hour, ten ml of blood was collected from each cow and from two kinds of blood vessels, carotid artery and jugular vein. Blood samples were placed into a plastic tube without anticoagulants and transported to the laboratory, which are separated by centrifugation at 3000 rpm for 15 minutes, all serum samples were frozen under -20C<sup>o</sup> until analysis time. On the next day 4 animals per group were slaughtered to

On the next day 4 animals per group were slaughtered to separating some organs such as normal oviduct, ovary, kidney, liver and uterus, after this operation, the samples were kept in tubs containing formaldehyde and then transported to the laboratory of veterinary collage/ Basrah to study the anatomic trails of the above organs according to method of Luna, (1968) by using Microtome (Jinhua Yidi Medical Appliance CO., L.T.D./ China).

Glucose concentration was measured by a chemical kit of the England Plamatec company. Cholesterol, total protein and albumin concentrations were determined by using a chemical kit of the France Biolabo company. The concentrations of liver enzymes, L-Aspartate 2-Oxaglutrate Amino Transferase (AST) and L-Alanine 2-Oxaglutrate Amino Transferase (ALT), were determined by using a chemical kit of the United Kingdom Randox laboratories limited company. Follicle stimulation hormone (FSH), luteinizing hormone (LH), estradiol and prolactin concentrations were determined by using a procedure associated with the kit of Monobind Inc – USA Company. The obtained data were analyzed statistically using the SPSS (2013). Statistically significant differences were determined at the P<0.05 level of significance.

# **RESULTS AND DISCUSSION**

Table (1) shows the effect of oxytocin injection on concentrations of some biochemical parameters in the carotid artery and jugular vein in Holstein crossbreed cow's blood. The concentration of glucose was increased significantly (P<0.05) in the treated groups in comparison with control one either on carotid artery or in the jugular vein, owing due to the fact that when oxytocin arrived at pancreas gland, it will increase both of the insulin levels and the glycogen analysis (Stock *et al.*, 1990). Also, Paolisso *et al.*, (1988) reported that treatment with

oxytocin caused a rise in blood glucose and glycogen which followed by elevate in insulin secretion. This result agrees with (Wallin *et al.*, 1989). The glucose concentration was raised significantly (P<0.05) in the jugular vein compared with carotid artery, this may be due to that, glucose is a major source of energy in the brain of rumen animals, while the source of energy in all the body of those animals is acetate, where, the jugular vein comes from the brain, so, it is normally carried much more of glucose than the carotid artery.

Cholesterol concentration was increased significantly (P<0.05) in cows treated with oxytocin compared with the untreated group. The reason for this result may be due to that oxytocin receptors become more active and more numbers when the oxytocin levels are increased in the blood, those receptors require to cholesterol to become stabilized in it is high – affinity state (Gimpl et al., 2000, 1997, 1995). According to this fact, a high number of sources of the lipids was rapidly converted to cholesterol, this will lead to an increase in concentration of cholesterol in the blood. No significant (P<0.05) differences were observed among treatment groups and between various blood vessels in concentrations of total protein and albumin, this result may be due to the dose of oxytocin was injected into cows in the present experiment, so, based on our data, we could suggest that injected Holstein crossbreed cows at dose 9 ml/head in intramuscular will does not effect on total protein and albumin levels. However, the values of those two parameters were within a normal range, which recorded by Al-Fartosi *et al.*, (2010) in female cattle.

Table 1: Effect of oxytocin injection on concentrations of some biochemical parameters in the carotid artery and jugular vein in Holstein crossbreed cow's blood. (Mean ± S.E.)

Blood vessels	Group	Biochemical parameters			
		Glucose	Cholesterol	Total protein	Albumin
		(mg/100ml)	(mg/100ml)	(g/100ml)	(g/100ml)
Carotid artery	control	30.25±1.51 d	80.65±2.73 c	5.34±0.23	3.32±0.29
	treatment	48.78±9.35 b	84.63±4.93 b	5.87±0.25	3.95±0.24
Jugular vein	control	43.98±1.36 c	71.70±6.32 c	5.35±0.31	3.35±0.21
	treatment	50.00±1.89 a	91.07±4.05 a	5.49±0.33	3.72b±0.21

Different small letters within column means significant difference (P<0.05).

No significant (P<0.05) differences in the concentrations of liver enzymes were observed among the treatment group and the blood vessels (table 2). Those values were within a normal range (Osman and AL-Busadah, 2003; AlFartosi *et al.*, 2010), that indicate to no stress state on the body tissues was presented, therefore, the enzymes of liver were in normal range.

Table 2: Effect of oxytocin injection on concentrations of AST and ALT enzymes (IU/L) in the carotid artery and jugular vein in Holstein crossbreed cow's blood. (Mean ± S.E.)

Blood vessels	Group	Enzyme		
		AST	ALT	
Carotid artery	control	42.93±1.91	10.83±0.43	
	treatment	41.95±3.36	10.76±1.18	
Jugular vein	control	43.52±0.81	10.44±0.25	
	treatment	41.73±2.32	10.62±0.35	

Cows treating with oxytocin showed a significant (P<0.05) increase in the concentrations of FSH, LH, estradiol and prolactin hormones in comparison to the untreated cows (table 3). The reason of the elevation of LH hormone in the treatment group may be due to the indirect relationship between injected oxytocin and released the pulsatile LH hormone. Cimpl and Fahrenholz, (2001) reported that treatment with oxytocin could be

stimulated the production of prostaglandin hormone, which can play a role in initially hypothalamus gland to release the GnRH of the luteinizing hormone and reconvening the estrus cycle (Yu-Wen *et al.*, 2013). Additionally, when oxytocin a rise in blood caused inhibits GnRH reduction and so makes more GnRH available for stimulation of LH (Robinson *et al.*, 1976). This result was compatible with Sammelwitz and

# Rishdy *et al.* /Effect of Oxytocin Injection on Some Biochemical Parameters and Morphological of Some Organs Trails in Holstein Crossbreed Cows

Nalbandov, (1958) in their study on cows. Also, many other studies suggested that oxytocin can cause release of pituitary gonadotropins hormones (LH and FSH) in several species (Martini *et al.*, 1959; Shibusawa *et al.*, 1955). While Anant *et al.*, (1964) reported that treatment oxytocin at dose 150 units had no significant difference in pituitary hormones such as LH and FSH levels, the difference between the result of this study and our data may be due to the exchange in doses which were used in both two studies.

The reason of increase estradiol hormone concentration in treatment group can be explained by the interaction between the active of estrogen hormone and the level of oxytocin in blood, estrogen can inhibit or stimulate the expression of oxytocin receptors mRNA and increase the number of high- affinity binding sites for oxytocin in organs of female (Leung *et al.*, 1998; Wathes and Lamming, 1995; Soloff, 1975). Okuda *et al.*, (1997) reported that the receptors of oxytocin have been found in bovine granulosa cells during follicular growth, so they suggested that oxytocin probable effect on the secretion of estrogen. The concentration of prolactin was increased significantly (P<0.05) in the treatment group in comparison with control, may be due to the direct physiological role of oxytocin in the stimulation of prolactin secretion because of oxytocin initiating the positive feedback for oxytocin - prolactin excretion (Jessica and De'Nise, 2012). Furthermore, the rise in oxytocin in the peripheral plasma causes an increase in prolactin secretion (Samon et al., 1986). According to (Egli *et al.*, 2004) oxytocin initiation the lactotrophs cells to release prolactin after releasing intracellular Ca+2, therefore, they were suggested that oxytocin stimulate prolactin secretion when Ca+2 is available. The results showed that concentrations of most hormones such as LH, FSH, estradiol and prolactin were higher significantly (P < 0.05) in a jugular vein compared with carotid artery because of the glands (hypothalamus and pituitary) which are responsible on the releasing of those hormones sit in brain and the jugular vein comes from this organ, while, the concentration of estradiol was higher significant (P <0.05) in carotid artery compared with jugular vein because of the estradiol comes from ovary and transport to hypothalamus to effect on it is

Table 3: Effect of Oxytocin injection on concentrations of some hormones in the carotid artery and jugular vein in Holstein crossbreed cow's blood. (Mean ± S.E.)

secretions.

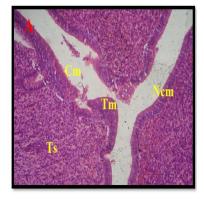
crossbreed cow's blood. (Mean ± 3.E.)									
Blood vessels	Group								
		LH (ng/ml))	FSH (ng/ml)	Estradiol (pg/ml)	Prolactin (pg/ml)				
Carotid artery	control	1.28±0.34 d	1.81±0.08 c	39.32±1.34 b	152.01±6.55 d				
	treatment	1.45±0.19 c	1.66±0.09 d	45.88±2.00 a	180.94±5.11 b				
Jugular vein	control	2.23±0.71 b	2.74±0.24 b	23.19±0.74 d	160.37±3.28 c				
	treatment	3.56±0.04 a	3.23±0.11 a	27.14±1.67 c	200.27±5.56 a				

Different small letters within column means significant difference (P<0.05).

The oviduct of animals that treated with 9 ml/head dose of oxytocin suffered from the hyperplasia of the tunica mucosa and an infiltration of inflammatory result that spread at the tunica submucosa in addition to slightly fibrosis (Figure 1). The ovary in comparison with the control group (Figure 2), the mucosa composed by a ciliated and secretory epithelial cell that lies beneath the lamina properia the muscular possessed an inner circular and outer longitudinal layer. The ovary of untreated animals composed of outer surface covered with a single layer of cuboidal cells termed the germinal layer, then the tunical albyginae, the ovary divided into an outer cortex and an inner medulla, the cortex contains a rellular connective tissue, which the follicles in different size embedded, while, the medulla consist of loose connective tissue with blood vessels and nerves, the ovary of treated animals with 9 ml/head dose showed a avacuolation of

granulosa cells and a necrosis in cumulus oophorus of follicular tissues in addition to edematous fluids (Figure 2). The wall of the uterus of untreated cows consisting of mucosal layer, the endometrium and fibro-muscular layer and myomerrium muscular tissue, the endomertrium contain a simple columnar epithelium with stromal connective tissue, while, in the treated group, there are avtecine glands hyperplasia with aggregation of an inflammatory cells (Figure 3). The kidney in the untreated group divided into cortex that contain of the convoluted tubules and the renal corpuscles, while the medulla represents the loops of henle and collecting ducts (Figure 4). On the other hand, the kidney of the treated group observed the dilation in some convoluted tubules and infiltration of inflammatory cells at the interstitium with the congestion of blood vessels of medullary zone (Figure 4). The liver of the untreated

group encapsulated by a thin connective tissue capsule that send a septum into the parpanchyma and divided to from hepatic lobules, each lobule observes of hexagonal hepato cycles plates which radiating toward the central vein, the sinusoid travelled in between the hepatocytes



and drained into the central vein, whereas the liver treated animals observed of the dilation of some sinusoids and the congestions of the central veins (Figure 5).

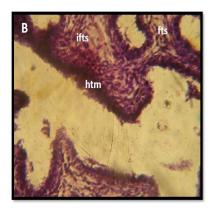
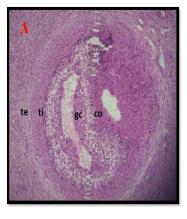


Figure 1: Oviduct of control group (A) showing, Tunica mucosa (Tm), Tunica submucosa (Ts), normal ciliated mucosa (Cm) and normal non ciliated mucosa (Ncm). Oviduct of treated group (B) showing, hyperplasia of tunica mucosa (htm), infiltration of inflammatory cells in tunica submucosa (ifts), early fibrosis of tunica submucosa (fts). H&E stains. 10x.



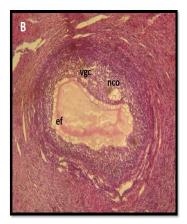


Figure 2: Ovary of control group (A) showing, granulosa cell (gc), theca interna (ti), theca externa (te) and normal cumulus ophorus (co). Ovary of the treated group (B) showing, vacuolation of granulosa cell (vgc), necrosis of cumulus ophorus (nco) and edematous fluid (ef). H&E stains. 10x.

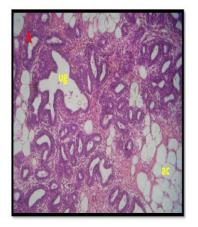




Figure 3: Uterus of control group (A) showing, normal uterine gland (ug), adipocyte (ac) of endometrium. Uterus of the treated group (B) showing, hyperplasia of uterine gland (hug), with present of some inflammatory cells around uterine glands (icug) H&E stains. 10X.

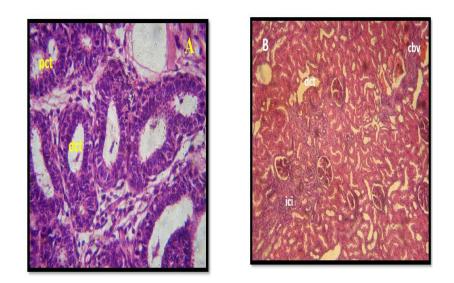
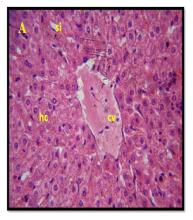


Figure 4: kidney of control group (A) showing, normal proximal convoluted tubules (pct), distal convoluted tubules (dct). Kidney of the treated group (B) showing, dilation of some convoluted tubules (dct), inflammatory cells infiltration in the interstitium (ici), with congested some blood vessels (cbv) H&E stains. (40X and 10X respectively).



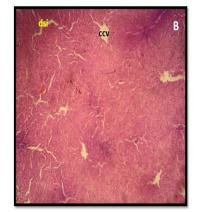


Figure 5: Liver of control group (A) showing, central vein (cv), hepatocytes (hc), and sinosuides (si). Liver of the treated group (B) showing, dilation of some sinusoids (dsi), with marked congestion of some central veins (ccv) H&E stains. 10X.

# CONCLUSION

From our results, we concluded that injected oxytocin hormone at dose 9 ml/head in intramuscular, almost, increase the concentrations of glucose, cholesterol, LH, FSH, estradiol and prolactin in the blood of Holstein crossbreed cows. Also, the carotid artery content more level of estradiol hormone, while, the jugular vein has more levels of glucose, LH, FSH, and prolactin compared with carotid artery.

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