



The Effect of Recipient Holstein heifers' Corpus Luteum Type, Diameter and Secretary Potential on their Pregnancy Rate during Routine ET program in Egypt

Oshba MR¹, Sosa GAM³, Nossier MB², Fadel MS¹ and El-Raey M^{3*}

¹Diagnostic Imaging and Endoscopy Unite, Animal Reproduction Research Institute, Al Haram.

²Department of Theriogenology, Faculty of Veterinary Medicine, Alexandria University

³Department of Theriogenology, Faculty of Veterinary Medicine, Benha University, P.O: 13736, Tokh, Kaliobia, Egypt.

ABSTRACT

The objective of this study was to find the correlation between the pregnancy rate of Holstein recipient heifers and their corpus luteum type, diameter and its secretary competence at the day of embryo transfer. A total of (92) recipients were selected after estrus synchronization by prostaglandin. Fresh embryos were collected from (12) donors. Corpus luteum type and diameter were determined at transfer date using transrectal ultrasound. Serum samples were collected at transfer day (7th day post heat/luteal phase) for progesterone and Estradiol-17 β assessment. Current study revealed non-significant difference between pregnant and non-pregnant recipients concerning CL diameters; regardless CL type either compact (24 \pm 0.83 and 24 \pm 0.58mm, respectively) or cavitory (26 \pm 1.1 and 27 \pm 1.4mm, respectively); however there was significant difference (P<0.05) between compact and cavitory CL diameters in non-pregnant recipient. Concerning, the relationship between CL type and pregnancy rate: 13 (39.39* %) out of 33 pregnancies had cavitory CL; whereas, 15 (25.42%) pregnancies out of 59 had compact CL. Cavitory CL secreted progesterone (5.9 \pm 1.7ng/ml) and estradiol (43 \pm 1.9 pg/ml). Compact CL recorded low level of progesterone (4.4 \pm 0.96 ng/ml), and high estradiol concentration (49 \pm 1.4* pg/ml). The percentage of cavitory CL in Holstein breed was (35.87%) versus (64.13%) for compact ones. Ratio of luteal cavity/luteal tissue was (0.48%) for Holstein recipients heifers. It was concluded that, corpus luteum type not diameter was a decisive factor in recipient selection. Luteal phase estradiol concentration was a crucial in recipient selection and should be at the lowest concentration.

Keywords: *Corpus luteum, Pregnancy, Recipient, Embryo, Progesterone, Estradiol*

(<http://www.bvmj.bu.edu.eg>) (BVMJ-36(1): 77-87, 2019)

1. INTRODUCTION

The principal use of embryo transfer in cattle is to amplify the reproductive rates of valuable animals. Ideally, embryo transfer can be used to accelerate genetic improvement and to increase marketing opportunities with purebred cows. Because

of their relatively low reproductive rate and long generation interval, In vitro embryo production (IVP) is useful to potentially increase the number of offspring from superior genetics donor cows during their reproductive life (Seidel, 1991; Palma 2001). The success of embryo transfer, establishment and maintenance of

pregnancy involve complex interaction between the embryo, uterine environment and corpus luteum (Mann et al., 1995); where bad quality recipients might result in poor embryo transfer result. Summarily, poor CL quality besides the embryo uterine asynchrony resulted in embryo under-development (Sreenan and Diskin, 1987); due to incompetence of maternal recognition and pregnancy maintenance. Moreover, rearing management and farmer's culture may gravely affect the likelihood of pregnancy in the recipient female if they badly go on after the transfer process (Camargo et al., 2006; Lestari et al., 2016).

Once transferable embryos were collected from a donor cows, the decision is to provide high quality recipients to receive these embryos to achieve the greatest number of offspring (Wright, 1981). The suitability of recipients is dependent on many factors such as the timing of estrus and the presence of an efficient corpus luteum (CL). On the point of CL efficiency, most embryo transfer technicians relies on rectal palpation to recognize and characterize the size and integrity of CL. Nowadays ultrasonography gains an increased interest to become a valuable tool to judge the genital tract potential capability especially luteal structure quality score (measurements and characteristics) (Kastelic et al., 1990 a&b; Singh et al., 1997). CL diameter has been used to categorize the recipients (Demetrio et al., 2007; Duica et al., 2007); however, the results of these previous reports were contradictory. Baruselli et al. (2001) and Duica et al. (2007) stated that when the luteal diameter increased the pregnancy rate increased. Where, recipients with large luteal structure diameter had a higher progesterone (P4) concentration (Kerbler et

al., 1997; Mann, 2009); and therefore more suitable uterine environment (Ashworth et al., 1989; Lonergan et al., 2007; Okumu et al., 2010). Progesterone plays major roles endometrial glands synthesis of the histotroph during early pregnancy (Gray et al., 2001; Wang et al., 2007; Lonergan, 2011). Histotroph is required for embryo development, migration, and implantation since it provides growth factors, amino acids, carbohydrates, and other necessary substances (Barnes, 2000; Spencer et al., 2004; Morris and Diskin, 2008). Regarding CL type; the corpora lutea were classified into compact and cavitary types (Barreiros et al., 2006). Cavitary corpus luteum was found in 40-80% of the estrous cycles of cows and heifers treated with P4 or prostaglandin; even though P4 concentrations were not influenced by the presence or absence of the CL cavity (Spell et al., 2001; Marques et al., 2002). The presence of corpus luteum with cavity CL had generated much controversy (Grygar et al., 1997; Marques et al., 2002; Looney et al., 2006, Siqueira et al., 2009). So the aim of the current study is to determine the association between CL diameter, type, serum progesterone and estrogen concentrations in recipient cattle and their pregnancy rate.

2. MATERIAL AND METHODS

This study was conducted at Elattar Dairy Farm located on Alex-Cairo desert road during the period from January to April 2017. The selected animals had regular body condition score 3, on a scale of 1-5 (Edmonson et al., 1989). Insemination was done using frozen semen from superior genetic bulls from USA companies (ABS and Semex), 0.5 ml straw of conventional semen containing (14 to 20) million sperm per insemination dose. Selected cows and

The Effect of Recipient Holstein heifers' Corpus Luteum Type, Diameter and Secretary

heifers were subjected to detailed clinicogynecological examinations prior to the experiment to confirm that they were healthy and of efficient reproductive system. Animals were fed on total mixed ration (TMR) ration and had free access to water and mineral salts.

Twelve donor cows were used as a source of fresh embryos. CIDR based superovulation protocol was used with fixed time AI for donor superovulation (table 1) according to Oshba et al. (2018). Insemination was done twice with (12h) apart. Embryos were recovered by non-surgical flushing technique on the 7th day after the insemination (day 16th) using patent flushing media (Complete flush, Agtech, USA). On the day of recovery the recovered embryos were shifted to holding media (Agtech, USA) to be classified by using stereomicroscope (Meiji) according to the International Embryo Transfer Society Manual (Wright, 1998); depending on the stage of development and quality.

For each donor enough recipients were synchronized with single prostaglandin (Cloprostenol- Estrumate®, Canada) injection - (3 days before the day of donor insemination)-. Total of (92) recipients were selected by the aid of trans-rectal ultrasound (Sonoscape A5 vet) at frequency 7MHZ for type and the diameter of CL. Embryo transfer (ET) was carried out, by nonsurgical method. Recipients were examined by ultrasonography (7MHZ) on the thirty day after ET for pregnancy diagnosis. Another confirmative examination was performed at 60-70 days post ET. Serum samples were collected from the recipients' heifer at the same day of embryo transfer (7th day post heat). Samples were submitted to Biochemistry lab of Animal Reproduction Research Institute (ARRI) to analyze them for progesterone (ng/ml) using Nova Tech progesterone Enzyme Immunoassay; while Nova Tech 17 β -Estradiol Enzyme Immunoassay was used for the quantitative determination of 17 β -Estradiol (pg/ml).

Table (1): GnRH-CIDR based superovulation program with timed insemination in dairy cow.

Days of treatment	7 AM	7 PM
0	CIDR insertion	
2	Receptal 10 μ g / IM	
4	Folltropin 80 mg I/M	Folltropin 80mg I/M
5	Folltropin 60mg I/M	Folltropin 60mg I/M
6	Folltropin 40mg I/M	Folltropin 40mg I/M
7	Folltropin 20mg I/M	Folltropin 20mg I/M +Estrumate (500 μ g I/M) + CIDR remove
8		Estrumate 500 μ g + Receptal (10 μ g I/M)
9	Timed AI	Timed AI
16	Embryo Flushing (non-surgical recovery of embryos).	

(Folltropin-V®, Bioniche, USA); (Estrumate®, MSD); (Receptal®, MSD)

Statistical analysis:

The statistical analysis was carried-out Chi²-test using SAS, 2004 and unpaired t-test using prism 5, version 5.01, GraphPad

software, Inc 2007. <http://iruler.net/> (2018) software was used to calculate ultra-

sonographic images measurements after screen calibration.

3. RESULTS

Relation between CL diameter and recipient pregnancy rate

Table (2) revealed non-significant difference between pregnant and non-

pregnant Holstein recipients, heifers concerning CL diameters and regardless CL type either compact (24±0.83 and 24±0.58, respectively) or cavitary (26±1.1 and 27±1.4, respectively); but there was significant difference ($P < 0.03$) between compact and cavitary CL diameters regarding the non-pregnant recipient.

Table (2): CL diameters between pregnant and non-pregnant Holstein heifers recipient

Item	Pregnant recipient	Non- pregnant recipient	$P < 0.05$
Compact CL diameter (mm)	24±0.83	24±0.58	Ns
	Ns	0.03	
Cavitary CL diameter (mm)	26±1.1	27±1.4*	Ns
Overall CL diameter (mm)	25±0.68	25±0.61	Ns

Statistical analysis was carried out using unpaired t-test

Corpus Luteum type effects on Holstein heifer recipient pregnancy rate post ET

Table (3) of the relationship between CL type (cavitary or compact) and pregnancy rate demonstrating that 13 pregnancies (39.39 %) out of 33 recipients had cavitary CL; whereas, 15 pregnancies (25.42%) out of 59 recipients have compact CL. This positive relationship between pregnancy rate and cavitary CL might be due to high

(not significant) progesterone level (5.9 ± 1.7ng/ml) secreted by cavitary and low level of estradiol in blood (43±1.9 pg/ml). In contrast, pregnant recipient had a compact CL recorded low level of progesterone (4.4±0.96 ng/ml) and high estradiol level in blood (49±1.4 pg/ml) table (4). Additionally, table (3) demonstrating that the percentage of cavitary CL in Holstein cow breed was (35.87%) versus (64.13%) for homogenous or compact ones.

Table (3): Cavitary and compact CL effects on the pregnancy rate of Holstein heifers' recipient after thirty days from ET

	Cavitary CL	Compact CL	Total
Total recipients	33 (35.87%)	59 (64.13%)	92
Pregnant recipients	13	15	28(30.43%)
Non-pregnant recipients	20	44	64(69.56% ^{**})
Pregnancy rate (PR)	13/33=39.39%	15/59=25.42%	
Chi ² of PR		6.55 ^{**}	

* = Significant at ($P < 0.01$).

Table (4): Luteal phase progesterone and estradiol concentrations in relation to CL Type in Holstein recipient heifers

Hormones	Cavitary CL	Compact CL	* $P < 0.05$
P4 (ng/ml)	5.9±1.7	4.4±0.96	Ns
Estradiol (pg/ml)	43±1.9	49±1.4*	0.02

Statistical analysis was carried out using unpaired t-test

Table (5) of the luteal cavity ratio in relation to total CL diameter revealed that the average of luteal tissue size was (13±0.20mm); average of CL cavity size

(12±0.18mm) and the ratio of luteal cavity diameter/total CL diameter was (0.48%) for Holstein recipients heifers.

Table (5): The luteal cavity ratio in relation to total cavitory CL diameter

Item	Mean ± S.E (mm)
Total CL diameter	25.25±0.33
Average of luteal tissue thickness	13±0.20
Average of CL cavity diameter	12±0.18
Luteal cavity diameter / Total CL diameter	0.48%

Measurement was calculated on images using <http://iruler.net/> after screen calibration.



Fig (1) Compact corpus luteum

Hypoechoic CL (darker) if compared to the ovarian stroma due to extensive vascularization. Echo-graphically mature CL acquires more echogenic echo texture (brightness) as reaches maturity (Red arrow in lower aspect).



Fig (2) Cavitory corpus luteum

Cavitory corpus luteum in which there is anechoic cavity, that surrounded by hypoechoic luteal tissues.

Conditions of Cavitory CL found in recipient Holstein heifers.



Fig (3)

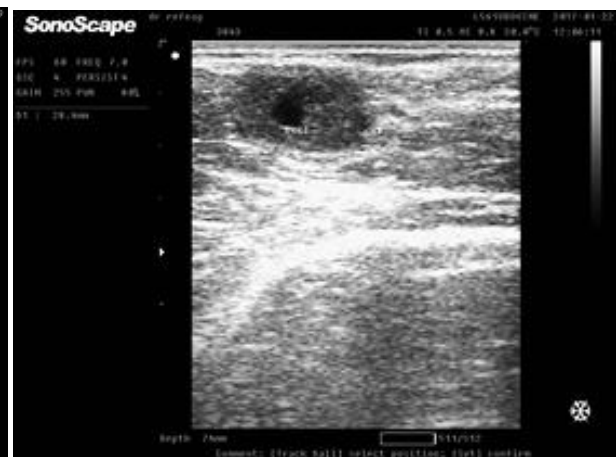


Fig (4)

Fig (3and 4): The anechoic cavity in the CL could be central or eccentric, respectively.



Fig (5)



Fig (6)

Fig (5 and 6) The anechoic cavity in the CL could be irregular or round circular in shape, respectively



Fig (7)

Fig (7) Echogenic fibrin strands are occasionally observed within the fluid- filled cavity of the CL



Fig (8)

Fig (8,9 and 10) In some cases, uniform hyperechogenic tissue completely fills the cavity. There are specular reflexions of new luteal tissue intermixed with fibrin strands, which are known to be very echogenic



Fig (9)



Fig (10)

4. DISCUSSION

Recipient cows selection is crucial for embryo transfer program success and stand on the same important level of donor cows selection; as the recipient will have a profound effects on several aspects including conception rate, calving success, calf performance and cost. So, upon initial selection of a certain recipient group one usually must choose the best prospect by eyeballing them besides developing an efficient methods and criteria that aid the selection process. In between such criteria is CL type and diameter. Rectal palpation alone may not yield an accurate luteal size determination; so it was very important to perform a thorough reproductive examination ideally by ultrasound (Looney et al., 2006). The current results indicated that the ultrasonographic luteal diameter wasn't a significant point that affects the recipient pregnancy rate, where the pregnant recipient had an average CL (25 ± 0.68 mm) in diameter (Rang: 20-33.8mm). Whereas, non-pregnant recipients had an average (25 ± 0.61 mm) luteal diameter (Rang: 14.3 - 35.3mm). The current findings run in a harmony with many studies that didn't find any positive correlation between pregnancy rates and luteal diameter (Spell et al., 2001; Bényei et al., 2006; Rodríguez et al., 2007). On the other hand, these findings were not comparable to those previously reported by Baruselli et al. (2001) and Marques et al. (2002) who reported that the pregnancy probability was affected only by CL diameter, but not by P4 plasma concentration.

Regarding CL type, the current study findings revealed that the cavitory CL exerts a positive significant effects ($\text{Chi}^2=6.55^{**}$) on recipient cows pregnancy rate (39.39%) versus compact CL type (25.42%). Cavitory CL was first reported by Pierson and Ginther (1984), but cavities were not fully described until 3 years later (Pierson and Ginther, 1987). Kastelic et al. (1990a&b) and Spell et al. (2001) demonstrated that the ability to produce P4 and maintaining pregnancy was equivalent in both CL types, so they differing only in morphology. Barreiros et al. (2006); Spell et al. (2001) and Marques et al.

(2002) showed non-significant difference in (P4) concentration with cavitory or compact corpora lutea at the time of embryo transfer. The current results came in harmony with these previous reports; although the current results revealed that the recipient cows the have cavitory CL recorded the highest PR and their CLs secret slightly higher level of progesterone (5.9 ± 1.7 ng/ml) than compact or homogeneous ones (4.4 ± 0.96 ng/ml). Progesterone prepared the uterine environment to receive and stimulate the embryo's development by acting on the uterine nutrients and growth factors that affecting the implantation success (Gray et al., 2001; Spencer et al., 2004; Gonella et al., 2010). Thus, higher progesterone concentrations will in turn promote endometrial adaptation to become receptive (Kayacik et al., 2006; Looney et al., 2006; Siqueira et al., 2009). The current finding came in agreement with Grygar et al., (1997) who reported that cows with luteal cavities displayed higher plasma progesterone levels, also cavitory CLs in pregnant cows contain greater luteal tissue volume and secretory activity than homogeneous CLs. On contrary, Garcia and Salaheddine (2000), Marques et al. (2002) and Barreiros et al. (2006) suggested that neither presence nor the size of luteal cavity affect the success and maintenance of cow's gestation in embryo-transfer program regardless the treatment protocol or cattle breed.

The corpus luteum is a transient endocrine organ that plays a dynamic role in estrous cycle, fertility regulation as well as pregnancy maintenance (Okuda et al., 2001). The primary function of the CL is to produce progesterone (Okuda et al., 2001). Besides progesterone; CL also produces a variety of other hormones in between estradiol (Shutt et al., 1975; Elbaum and Keyes 1976; Einspanier et al., 1991 and Gregoraszczyk, 1991). Luteal estradiol acts as a potent autocrine and/or paracrine regulator within the porcine (Pitzel et al., 1990) and human (Maas et al., 1992) CLs. Whereas, in cattle, luteal estradiol enhances PGF_2 production through estrogen receptors that demonstrated on bovine CL on days 8, 14, and

18 of the estrous cycle (Kimball and Hansel 1974;Grazul et al., 1989); so the low concentration of luteal estradiol affects the bovine luteal cells functions (Grazul et al., 1989), and enhances its micro-dialysis (Liebermann and Schams, 1995). Moreover, very low concentration of luteal estradiol (10-15 M) stimulated oxytocin release from micro-dialyzed bovine CL (Liebermann and Schams, 1995). Consequently, high estradiol concentration was negatively correlated with conception rate (Kajaysri, 2006). The current results showed a significant difference ($P<0.02$) between cavitory (43±1.9 pg/ml) and compact (49±1.4*pg/ml) CLs concerning estradiol level during the luteal phase of the recipient cows; this finding came in harmony with these previous reports which might explain why recipient cows that have cavitory CL recorded the highest PR. In contrast, some reports indicated that increased preovulatory concentrations of estradiol resulted in increased fertilization success by influencing the sperm transport (Hawk, 1983), improved embryo survival (Miller and Moore, 1976), as it enhanced its quality and viability (Atkins et al., 2013 and Jinks et al., 2013). Furthermore, it improved the pregnancy success (Perry et al., 2005); through enhancing the uterine environment (Miller et al., 1977; Perry and Perry, 2008 a&b). These positive effects of estradiol in these earlier studies were recorded in the normal cyclic cow's outcomes not recipient ones that were subjected to synchronization protocols to prepare them hormonally. Moreover, the source of estradiol in these previous experiments was the follicular growth not CL origin. So, the current study provides a crucial factor on which we can select and enhance the recipient cow's PR; this factor is the recipient cow luteal estradiol concentration.

The percentage of cavitory CLs recorded in the present experiment and in Holstein cow breed was lower (35.87%) than that found by other researchers. Marques et al. (2002) observed ratio ranged between 42.9 to 45.4 % for *Bos indicus* × *Bos Taurus* recipients' heifers,

respectively. This difference might be due to breed difference or due to the difference in synchronization treatment regimen. Conclusions: Corpus luteum types not diameter are decisive factor in recipient selection where cavitory CL was good for pregnancy maintaining. Luteal phase estradiol concentration is crucial in recipient selection and should be at the lowest concentration. The percentage of cavitory CL for Holstein recipients was (35.87%) and the ratio of luteal cavity to the luteal tissue was (0.48%).

5. REFERENCES

- Ashworth, C.J., Sales, D.I., Wilmut, I., 1989. Evidence of an association between the survival of embryos and the periovulatory plasma progesterone concentration in the ewe. *J. Reprod. Fertil.*, 87 (1):23-32.
- Atkins, J.A., Smith, M.F., MacNeil, M.D., Jinks, E.M., Abreu, F.M., Alexander, L.J., Geary, T.W., 2013. Pregnancy establishment and maintenance in cattle. *J. Anim. Sci.*, 91:722-733.
- Barnes, F.L. 2000. The effects of the early uterine environment on the subsequent development of embryo and fetus. *Theriogenology*, 53:649-658.
- Barreiros, T.R.R., Blaschi, W., Borsato, E.A., Ludwig, H.Ê., Meira, da Silva, D.R., Seneda, M.M., 2006. Comparison the pregnancy rates between bovine recipients with cavitory or compact corpus luteum after cloprostenol or fixed time embryo transfer. *Semina: Ciências Agrárias, Londrina*, 27(4): 657-664.
- Baruselli, P.S., Marques, M.O., Madureira, E.H., Costa Neto, E.P., Grandinetti, R.R., Bo, G.A., 2001. Increased pregnancy rates in embryo recipients treated with CIDR-B devices and eCG. *Theriogenology*, 55:157. (Abstract).
- Bényei, B., Komlósi, I., Pécsi, A., Pollott, G., Marcos, C.H., de Oliveira Campos, A., Lemes, M.P., 2006. The effect of internal and external factors on bovine embryo transfer results in a tropical environment. *Anim. Reprod. Sci.*, 93(3-4):268-279.
- Camargo, L.S.A., Viana, J.H.M., Sá, W.F., Ferreira, A.M., Ramos, A.A., Vale Filho,

The Effect of Recipient Holstein heifers' Corpus Luteum Type, Diameter and Secretary

- V.R., 2006. Factors influencing in vitro embryo production. *Anim. Reprod.*, 3:19-28.
- Demetrio, D.G., Santos, R.M., Demetrio, C.G., Vasconcelos, J.L., 2007. Factors affecting conception rates following artificial insemination or embryo transfer in lactating Holstein cows. *J. Dairy. Sci.*, 90(11):5073-5082.
- Duica, A., Tovio, N., Grajales, H.A., 2007. Factors that affect the reproductive efficiency of the recipient within a bovine embryo transfer program. *Revista de Medicina Veterinaria*, 14:107-124.
- Edmonson, A.J., Lean, I.J., Weaver, L.D., Farver, T. and Webster, G., 1989. A body condition scoring chart for Holstein dairy cows. *J. dairy sci.*, 72(1): 68-78.
- Einspanier, A., Jarry, H., Pitzel, L., Holtz, W., Wuttke, W., 1991. Determination of secretion rates of estradiol, progesterone, oxytocin, and angiotensin II from tertiary follicles and freshly formed corpora lutea in freely moving sows. *Endocrinology*, 129: 3403-3409.
- Elbaum, D.J., Keyes, P.L., 1976. Synthesis of 17 β -estradiol by isolated ovarian tissues of the pregnant rat: aromatization in the corpus luteum. *Endocrinology*, 99:573-579.
- Garcia, A., Salaheddine, M., 2000. Ultrasonic morphology of the corpus lutea and central luteal cavities during selection of recipients for embryo transfer. *Reprod. Dom. Anim.* 35: 113-118.
- Gonella, A.D., Grajales, H.A., Hernández, A.V., 2010. Receptive uterine environment: maternal control, control embryo, embryonic death. *Rev. MVZ Córdoba*, 15 (1):1976-1984.
- Gray, C.A., Taylor, K.M, Ramsey, W.S., Hill, J.R., Bazer, F.W., Bartol, F.F., Spencer, T.E., 2001. Endometrial glands are required for preimplantation conceptus elongation and survival. *Biol. Reprod.* 64(6):1608-1613.
- Grazul, A.T., Kirsch, J.D., Slanger, W.D., Marchello, M.J., Redmer, D.A., 1989. Prostaglandin F_{2a}, oxytocin and progesterone secretion by bovine luteal cells at several stages of luteal development: effects of oxytocin, luteinizing hormone, prostaglandin F_{2a} and estradiol-17 β . *Prostaglandins*, 38:307-318.
- Gregoraszczuk, E., 1991 The interaction of testosterone and gonadotropins in stimulating estradiol and progesterone secretion by cultures of corpus luteum cells isolated from pigs in early and mid-luteal phase. *Endocrinol. Jpn.*, 38:229-237.
- Grygar, I., Kudlác, E., Dolezel, R., Nedbálková, J., 1997. Volume of luteal tissue and concentration of serum progesterone in cows bearing homogeneous corpus luteum or corpus luteum with cavity. *Anim. Reprod. Sci.*, 49(2-3):77-82.
- Hawk, H.W., 1983. Sperm survival and transport in the female reproductive tract. *J Dairy Sci* 66:2645-60.
- Jinks, E.M., Smith, M.F., Atkins, J.A., Pohler, K.G., Perry, G.A., Macneil, M.D., Roberts, A.J., Waterman, R.C., Alexander, L.J., Geary, T.W., 2013. Preovulatory estradiol and the establishment and maintenance of pregnancy in suckled beef cows. *J. Anim. Sci.*, 91:1176-1185.
- Kajaysri, J., 2006. Comparison of estradiol concentrations and conception rate between cows with different estrus behaviors. *KKU Vet J.*, 16 (3):15-22.
- Kastelic, J.P., Bergfelt, D.R., Ginther, O.J., 1990a. Relationship between ultrasonic assessment of the corpus luteum and plasma progesterone concentration in heifers. *Theriogenology*, 33(6): 1269-1278.
- Kastelic, J.R., Pierson, R.A., Ginther, O.J., 1990b. Ultrasonic morphology of corpora lutea and central cavities during the estrous cycle and early pregnancy in heifers. *Theriogenology*, 34 (3): 487-498.
- Kayacik, V., Salmano lu, R., Polat, B., Özlü, A., 2006. Evaluation of the corpus luteum size throughout the cycle by ultrasonography and progesterone assay in cows. *Turk. J. Vet. Anim. Sci.*, 29 (6):1311-1316.
- Kerbler, T.L., Buhr, M.M., Jordan, L.T., Leslie, K.E., Walton, J.S., 1997. Relationship

- between maternal plasma progesterone concentration and interferon-tau synthesis by the conceptus in cattle. *Theriogenology*, 47(3):703-714.
- Kimball, F.A., Hansel, W., 1974. Estrogen cytosol binding proteins in bovine endometrium and corpus luteum. *Biol. Reprod.*, 11:566-577.
- Lestari, T.D., Ismudiono, I., Sardjito, T., Srianto, P., 2016. The success of embryo transfer in dairy cattle recipient using beef cattle embryos. *Scientific Papers-Animal Science Series: Lucriri tiin ifice - Seria Zootehnie*, 65:203-208.
- Liebermann, J., Schams, D., 1995. Effects of estradiol-17b on oxytocin and progesterone release of the bovine corpus luteum during the estrous cycle. *J. Reprod. Dev.* 41:187-196.
- Lonergan, P., 2011. Influence of progesterone on oocyte quality and embryo development in cows. *Theriogenology*, 76:1594-1601.
- Lonergan, P., Woods, A., Fair, T., Carter, F., Rizos, D., Ward, F., Quinn, K., Evans, A., 2007. Effect of embryo source and recipient progesterone environment on embryo development in cattle. *Reprod. Fertil. Dev.*, 19(7):861-868.
- Looney, C.R., Nelson, J.S., Schneider, H.J., Forrest, D.W., 2006. Improving fertility in beef cow recipients. *Theriogenology*, 65 (1):201-209.
- Maas, S., Jarry, H., Teichmann, A., Rath, W., Kuhn, W., Wuttke, W., 1992. Paracrine actions of oxytocin, prostaglandin F_{2a}, and estradiol within the human corpus luteum. *J. Clin. Endocrinol. Metab.*, 74:306-312.
- Mann, G.E., 2009. Corpus luteum size and plasma progesterone concentration in cows. *Anim. Reprod. Sci.*, 115 (1-4):296-299.
- Mann, G.E., Lamming, G.E., Fray, M.D., 1995. Plasma oestradiol and progesterone during early pregnancy in the cow and the effects of treatment with buserelin. *Anim. Reprod. Sci.*, 37(2):121-131.
- Marques, M.O., Arruda, R.P., Madureira, E.H., Oliveira, C.A., Baruselli, P.S., 2002. Effect of the corpus luteum cavity on plasma progesterone concentration in *Bos Taurus* x *Bos indicus* embryo recipient heifers. *Revista Brasileira de Reprodução Animal*, Belo Horizonte, 26:238-240.
- Miller, B.G., Moore, N.W., 1976. Effect of progesterone and oestradiol on endometrial metabolism and embryo survival in the ovariectomized ewe. *Theriogenology*, 6:636.
- Miller, B.G., Moore, N.W., Murphy, L., Stone, G.M., 1977. Early pregnancy in the ewe: effects of oestradiol and progesterone on uterine metabolism and on embryo survival. *Aust. J. Biol. Sci.*, 30: 279-88.
- Morris, D., Diskin, M. 2008. Effect of progesterone on embryo survival. *Animal*, 2:1112-1119.
- Okuda, K., Uenoyama, Y., Berisha, B., Lange, I. G., Taniguchi, H., Kobayashi, S., Kobayashi, S., Miyamoto, A., Schams, D., 2001. Estradiol-17b is produced in bovine corpus luteum. *Biol. Reprod.*, 65: 1634-1639.
- Okumu, L.A., Forde, N., Fahey, A.G., Fitzpatrick, E., Roche, J.F., Crowe, M.A., Lonergan, P., 2010. The effect of elevated progesterone and pregnancy status on mRNA expression and localization of progesterone and oestrogen receptors in the bovine uterus. *Reprod.*, 140:143-153.
- Oshba, M.R., Sosa, G.A.M., Nossier, M.B., Fadel, M.S. and El-Raey, M., 2018. Comparison between estradiol and GnRH-CIDR based programs on super ovulation in Holstein cows/heifers. *Int. J. Adv. Res.*, 6(10), 615-620
- Palma, G.A., 2001. Producción in vitro de embriones bovinos. In: Palma GA, editor. *Biotechnology de la Reproducción*. Buenos Aires: INTA. Pp. 225-294.
- Perry, G.A., Perry, B.L., 2008a. Effect of preovulatory concentrations of estradiol and initiation of standing estrus on uterine pH in beef cows. *Domest. Anim. Endocrinol.*, 34:333-338.
- Perry, G.A., Perry, B.L., 2008b. Effects of standing estrus and supplemental estradiol on changes in uterine pH during a fixed-time AI protocol. *J. Anim. Sci.*, 86:2928-2935.
- Perry, G.A., Smith, M.F., Lucy, M.C., Green, J.A., Parks, T.E., Macneil, M.D., Roberts, A.J., Geary, T.W., 2005. Relationship

The Effect of Recipient Holstein heifers' Corpus Luteum Type, Diameter and Secretary

- between follicle size at insemination and pregnancy success. *Proc. Natl. Acad. Sci., USA* 102: 5268-5273.
- Pierson, R.A., Ginther, O.J., 1984. Ultrasonography of the bovine ovary. *Theriogenology*, 21(3): 495-504.
- Pierson, R.A., Ginther, O.J., 1987. Reliability of diagnostic ultrasonography for identification and measurement of follicles and detecting the corpus luteum in heifers. *Theriogenology*, 28(6): 929-936.
- Pitzel, L., Jarry, H., Wuttke, W., 1990. Effects of oxytocin on in vitro steroid release of midstage small and large porcine luteal cells. *Endocrinology* 126:2343-2349.
- Rodríguez, J.M., Giraldo, C.E., Castañeda, S.P., Ruiz, T.C., Olivera, M.A., 2007. Multifactorial analysis of pregnancy rates in embryos transfer programs in Colombia. *Rev MVZ Córdoba*, 12(2): 978-984.
- Seidel, G.E., 1991. Applications of embryo transfer, In: *Training manual for embryo transfer in cattle*. Pp. 3 - 13.
- Shutt, D.A., Shearman, R.P., Lyneham, R.C., Clarke, A.H., McMahon, G.R., Goh, P., 1975. Radioimmunoassay of progesterone, 17-hydroxyprogesterone, estradiol-17b and prostaglandin F in human corpus luteum. *Steroids*, 26:299-310.
- Singh, J., Pierson, R.A., Adams, G.P., 1997. Ultrasound image attributes of the bovine corpus luteum: structural and functional correlates. *J. Reprod. Fertil.*, 109:35-44.
- Siqueira, L.G., Torres, C.A., Souza, E.D., Monteiro, P.L. Jr, Arashiro, E.K., Camargo, L.S., Fernandes, C.A., Viana, J.H., 2009. Pregnancy rates and corpus luteum-related factors affecting pregnancy establishment in bovine recipients synchronized for fixed-time embryo transfer. *Theriogenology*, 72(7):949-958.
- Spell, A.R., Beal, W.E., Corah, L.R., Lamb, G.C., 2001. Evaluating recipient and embryo factors that affect pregnancy rates of embryo transfer in beef cattle. *Theriogenology* 56 (2): 287-297.
- Spencer, T.E., Burghardt, R.C., Johnson, G.A., Bazer, F.W., 2004. Conceptus signals for establishment and maintenance of pregnancy. *Anim. Reprod. Sci.*, 82-83:537-550.
- Sreenan, J.M., Diskin, M.G., 1987. Factors affecting pregnancy rate following embryo transfer in the cow. *Theriogenology* 27(1): 99-113.
- Wang, C.K., Robinson, R.S., Flint, A.P., Mann, G.E., 2007. Quantitative analysis of changes in endometrial gland morphology during bovine oestrus cycle and their association with progesterone levels. *Reprod.* 134:365-371.
- Wright, J.M., 1981. Non-surgical embryo transfer in cattle, embryo-recipient interaction. *Theriogenology* 15:43-56.
- Wright, J.M., 1998. Photographic illustrations of embryo developmental stage and quality codes. In: Stringfellow DA, Siedel SM, editors. *Manual of the International Embryo Transfer Society* (3rd ed). IETS, Savoy, Illinois; Pp. 167-170.