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Ultrasonic characteristics of the uterus and ovaries during estrus, and their relationship with pregnancy rate in dairy cows

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ABSTRACT

It was hypothesized that the accumulation of fluids in the uterine lumen reduces fertility in dairy cows. Therefore, the purpose of the present study was an evaluation of the ultrasound characteristics of the reproductive tract, including the accumulation of fluids in the uterine lumen during estrus, and the effect of these findings on pregnancy rates in dairy cows. The study was conducted on 486 lactating Holstein cows detected to be in estrous, on a large commercial dairy herd in Shiraz, Iran. Transrectal ultrasound was performed at the time of artificial insemination. Reproductive tract characteristics, comprising follicle diameters, the presence of corpus luteum in ovaries, the thickness, folding and edema of the uterus, and intrauterine fluid, were visualized and scored by ultrasonography. The cows were followed after insemination and their pregnancy rate determined. The effect of ultrasound findings were investigated in relation to pregnancy rates. The data were analyzed using logistic regression analyses. The results indicated that the pregnancy rate was significantly higher in cows with follicle size >14 mm (38.8%) compared with ≤14 mm (27.3%), after adjusting for the parity of the animals, days in milk and mean daily milk production (OR = 1.84, P = 0.005). No association between pregnancy rate and other ultrasound characteristics of the reproductive tract during estrus was observed in this study (P>0.05). In conclusion, follicle size is positively associated with the pregnancy rate of dairy cows in estrus. However, other ultrasound findings of the uterus, including intrauterine fluid, did not show any association with pregnancy rates.

Key words: follicle diameter; insemination; lumen score; corpus luteum; lactation

Introduction

Low intensity of estrous behavior and difficult prediction of ovulation time are the two most limiting factors in achieving optimum reproductive performance in dairy cows

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(HUDSON et al., 2012). The average duration of estrus in cows has traditionally been recognized to be about 15 hours, with a wide range of 2-30 hours; however, there is good evidence that estrus in 'modern' Holstein and Jersey cows, compared with heifers, is much shorter. Factors affecting estrus duration include: breed, season, the presence of a bull, nutrition, milk production, lactation and, most importantly, the number of estrous cows at one time (HURNIK et al., 1975).

The estrous cow should have suitable structures on the ovaries and favorable conditions in the uterus and cervix for establishment of pregnancy. In some cases, despite administering artificial insemination (AI) at the correct time, it does not lead to pregnancy. There are some reasons for this problem, including ovulatory follicle size at estrus (KESKIN et al., 2016). Uterine infections could occur as subclinical endometritis in dairy cows at the time of AI. Subclinical endometritis reduced reproductive performance of dairy herds in one study (SHELDON et al., 2006), but some researchers did not find any adverse effect of this uterine disease on reproductive performance (KASIMANICKAM et al., 2004). In this "silent" disease, a clear estrous mucus is observed.

The presence of a CL on one of the ovaries leads to an increase in progesterone levels in estrous cows. Measurement of milk progesterone concentrations or circulating progesterone (P4) levels have shown that between 10% and15%, or perhaps even 22% of cows are inseminated during the luteal phase (APPLEYARD and COOK 1976), during inappropriate stages of the follicular phase (BULMAN and LAMMING 1978) or in the diestrous phase or pregnancy (STURMAN et al., 2000).

Ultrasound imaging of the reproductive tract is a non-invasive method for evaluating the reproductive tract (PIERSON and GINTHER, 1987a,b). In practice, ultrasound examination has been used for evaluation of follicular dynamics and luteal development, and differential diagnosis of pathological conditions in the ovaries (EDMONDSON et al., 1986) and uterus in cattle (FISSORE et al., 1986; PIERSON and GINTHER, 1987b). Morphometric and echotexture changes of the uterus related to plasma hormone levels are detectable during the estrous cycle. Ovarian and uterus ultrasound characteristics, such as the diameter of follicle, the size and echotexture of the CL, and the shape and echotexture of the uterus, may be used to estimate the stage of the estrous cycle in each cow (PIERSON and GINTHER, 1988). Accumulation of fluids can be visualized as anechoic fluids in the uterine lumen in estrous (PIERSON and GINTHER, 1987b). Descriptions of endometrial thickness, echotexture, and shape are based primarily on a comprehensive study reported in 1987 (PIERSON and GINTHER, 1987b).

Some researchers have indicated notable ultrasound changes in the uterus and endometrium close to ovulation time in Holstein heifers (PIERSON and GINTHER, 1987a). Endometrial thickness (ET) increased at the time of normal luteolysis and reached maximum thickness on the day before ovulation (PIERSON and GINTHER, 1987b). After

ovulation, others have indicated a decrease in ET and changes in endometrial echotexture, likely due to an increase of P4 levels (BONAFOS et al., 1995; JIMENEZ-KRASSEL et al., 2009; PIERSON and GINTHER, 1987b).

LÜTTGENAU et al. (2015) concluded that ultrasound examination of cows before or during artificial insemination prevents the service of proestrous and diestrous cows, and decreases estrous detection errors (LÜTTGENAU et al., 2015). Therefore, ultrasonographic evaluation of ovarian structures and the uterus of cows at the time of artificial insemination is important. To the best knowledge of the authors, there is no report in the literature concerning the relationships between the ultrasonographic characteristics of the ovarian structures and uterus and pregnancy rates in estrous dairy cows. In particular, the authors hypothesized that the accumulation of fluids in the uterine lumen (*i.e.* enlarged uterine lumen) reduces fertility in dairy cows. The objectives of the study were an evaluation of the ultrasound characteristics of the reproductive tract, including follicle diameters, the presence of corpus luteum in the ovaries, the thickness, folding and edema of the uterus, and intrauterine fluid during estrus, and the effect of these findings on pregnancy rates in estrous dairy cows.

Materials and methods

Statement of animal rights. The experiment was performed under the approval of the state committee on animal ethics, Shiraz University, Shiraz, Iran (IACUC no: 4687/63). Also, the recommendations of the European Council Directive (86/609/EC) of November 24, 1986, regarding the protection of animals used for experimental purposes, were considered.

Animals. This prospective cohort study was performed on 486 estrous Holstein cows with various days in milk (DIM) and parity (1-4), in a large commercial dairy herd (n = 2500) in Shiraz, the capital of Fars province, southern Iran, from August 2014 to January 2015. The area has a warm climate with four distinct seasons, with peak summer temperatures of about forty-three centigrade and the coldest winter temperature below freezing. The cows were housed in a free-stall with sand for bedding. The cows were fed a total mixed ration (TMR) diet and were machine-milked three times daily. The reproductive cycles of the animals were non-seasonal, with year-round calving. The general reproductive policy of the herd was timed artificial insemination protocols (Ovsynch, HeatSynch), estrous detection and fixed time AI. Trained employees were used to recognize the signs of standing estrous, and report this information to the responsible person (4 times a day and 30 minutes every time for each paved area). Artificial insemination was performed by one trained technician, and the cows were inseminated with 20-30 million frozen-thawed sperm.

Transrectal ultrasonography. The cows in estrous were restrained in stanchions for the examination and the rectum was evacuated of feces. Ultrasound examination was conducted using a portable ultrasound machine (Easi-scan®, a versatile ultrasound machine for large animal scanning, upgrade 3), fitted with a 5-MHz linear transducer. Ovaries and cervix were localized, and all parts of reproductive tract were examined. The follicle diameters were measured and the cows grouped accordingly with some modifications (1 = small size (≤14 mm); 2 = large size (>14 mm). Also, the ovaries were checked for the presence of corpus luteum (GILBERT et al., 2005) and grouped (0 = no CL; 1 = have a CL). The diameters of their horns were measured at the greater curvature of the horn (1 = ≤3 cm; 2 = >3 cm) (BAEZ et al., 2016). Endometrial thickness (ET) was measured and grouped (1 = <2 cm; 2 = ≥2 cm). Contractility, folding and edema of the uterus were scored (1 = minimum; 2 = medium; 3 = maximum) (Figs. 1 and 2). Also, the lumen of the uterus (intrauterine fluid) was scored (0 = none; 1 = minimum; 2 = medium; 3 = maximum) (GINTHER, 1998) (Fig. 3). The cows in the first examination were followed and the occurrence of pregnancy determined at 35 days after AI, using ultrasonography.

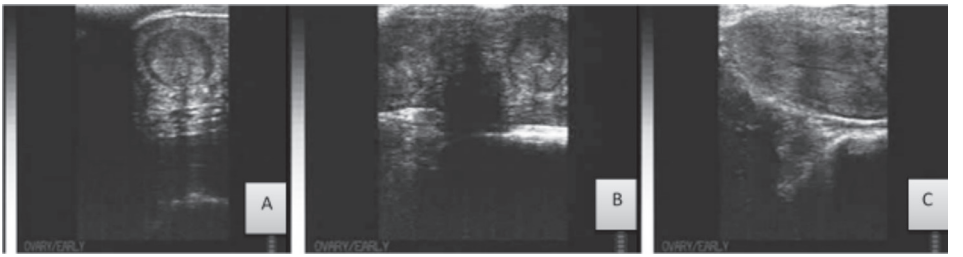

Fig. 1. Transverse section ultrasound images of the uterine horn edema in estrus. Minimum, medium and maximum edema are shown in sonograms A, B and C, respectively.

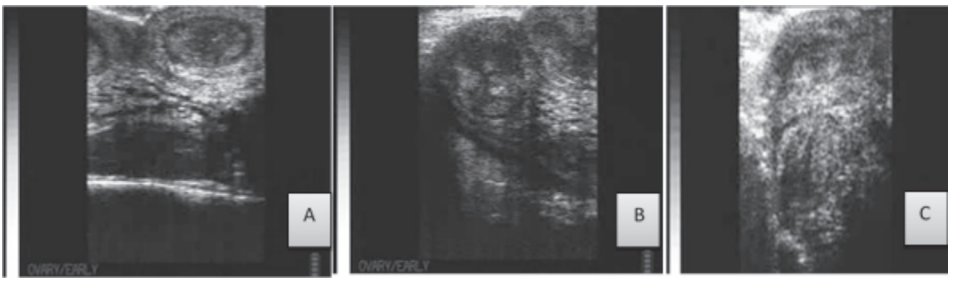

Fig. 2. Ultrasound images of the uterine horns showing fold characteristics during estrus. Increasing scores of the uterine fold are shown in sonograms from A (1), B (2) to C (3).

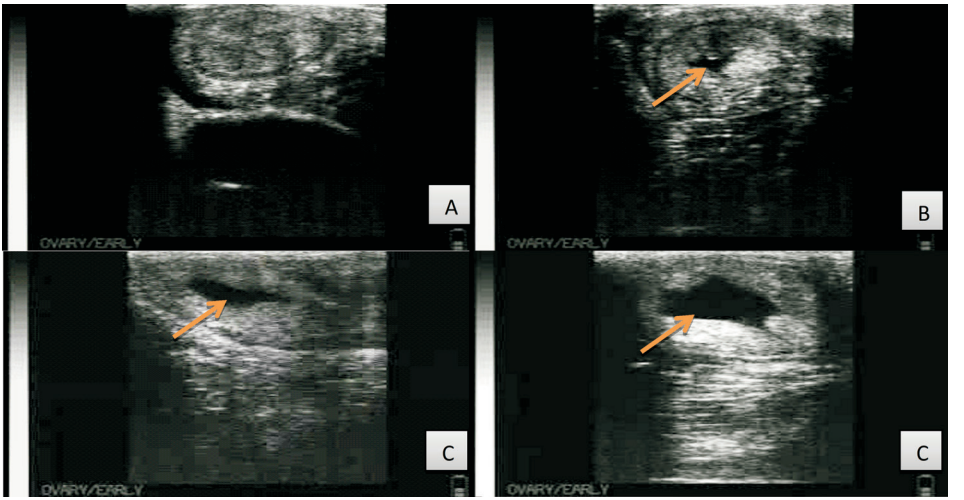

Fig. 3. Sonogram of uterine lumen scores during estrus. Maximum intraluminal fluid is shown in sonogram D (arrow), whereas no fluid was detected in the uterine lumen (sonogram A).

Statistical analyses. The data were analyzed using SPSS statistical software (version 16; SPSS Inc., Chicago, USA). The results of the ultrasonic findings of the uterus and ovaries were presented as the number of animals in each group. Associations of ultrasound findings and pregnancy rates were investigated by univariable and multivariable analysis. Univariable analysis was performed using Pearson Chi-square analysis. Pregnancy at 35 days after AI was considered as a dependent variable (1 and 0 for pregnant and non-pregnant, respectively) in the multivariable logistic regression model. Among the ultrasonic findings of the uterus and ovaries, those factors with P-value <0.2 in the univariable analysis were introduced into the multivariable model as independent variables. Also, to account for the potential confounding effect of parity (1, 2 and ≥3), mean daily milk production (kg) and DIM, they were forced to remain in the multivariable logistic model as covariates. Using a backward elimination procedure and Wald statistics, the final logistic model was constructed. In the final analysis, a P-value less than 0.05 was considered significant.

Results

Overall, 486 cows were selected for the study. A total of 98 animals were at parity 1, 156 were at parity 2 and the remaining were at parity ≥3. Mean values (± standard deviation) for daily milk production and DIM were 41 ± 9 kg and 226 ± 114 days, respectively. There was a number of missing cases for some of the variables. The missing

cases were deleted list-wise from the analysis. Therefore, 450 cases were finally analyzed for univariable and multivariable association studies.

Table 1. Associations of different ultrasound characteristics of the uterus and ovaries with pregnancy rate in 450 estrous dairy cows in a large commercial dairy herd, Shiraz, Iran

Parameter	Number of animals		P-value
	Pregnant (%), n = 155	Not-pregnant (%), n = 295	
Follicle size			
≤14mm	47 (27.3)	125 (72.7)	
>14mm	108 (38.8)	170 (61.2)	0.01
Corpus luteum existence			
No	122 (36.7)	210 (63.3)	
Yes	33 (28)	85 (72)	0.09
Endometrial thickness			
<2 cm	90 (35)	167 (65)	
≥2cm	63(33.3)	126 (66.7)	0.76
Uterine diameter			
≤3 cm	88 (36.4)	154 (63.6)	
> 3 cm	60 (31.9)	128 (68.1)	0.35
Edema score			
Minimal	69 (37.5)	115 (62.5)	
Medial	67 (34)	130 (66)	
Maximal	19 (27.5)	50 (72.5)	0.32
Folding score			
Minimal	79 (34.8)	148 (5.2)	
Medial	72 (34.1)	139 (65.9)	
Maximal	4 (33.3)	8 (66.7)	0.98
Contractility score			
Minimal	12 (30.8)	27 (69.2)	
Medial	90 (34.1)	174 (65.9)	
Maximal	53 (36.1)	94 (63.9)	0.81
Lumen score			
None	8 (37.1)	132 (62.9)	
Minimal	52 (31.3)	114 (68.7)	
Intermediate	19 (35.2)	35 (64.8)	
Maximal	6 (30)	14 (70)	0.66

The frequency distribution of the study animals according to ultrasound findings, as well as pregnancy status, is presented in Table 1. Pregnancy rates in cows with follicle diameter equal and less than 14 mm and greater than 14 mm at the time of estrus were 27.3% and 38.8%, respectively. The difference in pregnancy rates between the groups was significant (P = 0.01, Table 1). Pregnancy rates in cows without CL and with CL on their ovaries at the time of estrous was 36.7% and 28%, respectively (P = 0.09, Table 1).

No significant associations between pregnancy rate and other ultrasound characteristics, including endometrial thickness, uterine diameter and folding, edema and contractility of the uterus, were observed in this study (Table 1).

Pregnancy rates in cows without any uterine fluid *i.e.* lumen score 0 (37.1%) was slightly higher than in cows with minimum (31.3%), medium (35.2%) and maximum (30%) uterine lumen. However, the differences were not statistically significant (P = 0.66, Table 1).

Follicle size and CL existence were introduced into the multivariable logistic regression analysis while adjusting for parity of animals, DIM and mean daily milk production. Using the backward elimination procedure, follicle size remained in the final model (Table 2). The results showed that the pregnancy rate was significantly higher in cows with follicle size >14 mm compared with ≤14 mm (OR = 1.84, P = 0.005). Among the adjusting factors, only milk production showed a significant negative effect on pregnancy rate (OR = 0.97, P = 0.03).

Table 2. Results of multivariable logistic regression analysis for associations of the ultrasound characteristics of the uterus and ovaries with pregnancy rate in 450 estrous dairy cows in a large commercial dairy herd, Shiraz, Iran

Parameter	B	SE	Wald	P-value	Odds ratio (OR)	95% CI for OR
Constant	0.100	0.714	0.020	0.888	-	-
Follicle size >14 mm*	0.612	0.218	7.895	0.005	1.844	1.203, 2.825
Lactation 1**			3.194	0.202		
Lactation 2	0.218	0.274	0.638	0.425	1.244	0.728, 2.227
Lactation ≥3	-0.195	0.271	0.516	0.473	0.823	0.483, 1.401
DIM	0.001	0.001	0.071	0.790	1.0	0.998, 1.002
Mean daily milk (kg)	-0.030	0.014	4.714	0.030	0.971	0.945, 0.997

* Compared with ≤14 mm; ** Reference category

Discussion

Associations of the ultrasound characteristics of the ovaries and uterus with pregnancy in estrous dairy cows were investigated in the present study. The follicle size during estrus was significantly associated with pregnancy rate. Other variables, including accumulation

of fluid in the uterus, did not show any significant relationship with pregnancy in estrus dairy cows.

The pregnancy rate in estrous cows with follicle size ≤14mm was significantly lower than cows with follicle size >14 mm. Previous studies have indicated that pregnancy rate is affected by the ovulatory follicle, although the results are conflicting (LOPES et al., 2007; PERRY et al., 2005; VASCONCELOS et al., 2013). Ovulation in cows with a larger ovulatory follicle resulted in greater pregnancy rates than in cows with smaller ovulatory follicles (KESKIN et al., 2016; LAMB et al., 2001; SÁ FILHO et al., 2010). Also, ovulation of follicles ≤ 11 mm had no sequel on the pregnancy rate or embryonic/fetal viability (PERRY et al., 2005). It has been indicated that larger ovulatory follicles while the cow is in heat are related with higher estradiol (E2) and P4 levels, and a larger CL. Higher estradiol levels facilitate sperm / oocyte transportation in the reproductive tract, and improve the uterine environment for early embryonic development (HAWK 1983). Furthermore, the CL formed following ovulation from a larger follicle, is large and produces high progesterone levels. One of the important factors in early embryonic development in dairy cows is a high P4 concentration (LOPES et al., 2007; PERRY et al., 2005; WILTBANK et al., 2011). However, in some researches larger follicles were associated with lower fertility, as a result of oocyte aging (REVAH and BUTLER 1996). Also, some studies indicated that pregnancy rates were higher in cows with small and young ovulatory follicles (LYNCH et al., 2010). It was also found that follicles that were small in size were associated with high embryo survival (LYNCH et al., 2010). Other studies showed no effect of follicle size on pregnancy rate in dairy cows (BRUSVEEN et al., 2009). Taken together, it seems that most researches, including our present study, supports a positive association of follicle size with pregnancy rates. A systematic review and meta-analysis of the literature in the future could be helpful to obtain a general consensus in this regard.

In our study, although pregnancy rates in cows without CL at the time of heat was greater than in cows with CL, the difference was not significant. Incomplete spontaneous luteal regression during proestrus has been frequently reported in dairy cows (BRUSVEEN et al., 2009; GIORDANO et al., 2012; MARTINS et al., 2011). Cows with incomplete luteolysis and low plasma P4 levels showed prominent subfertility. Parity can affect CL regression. Thus a higher percentage of primiparous had complete lueolysis than multiparous cows, after treatment with cloprostenol (GIORDANO et al., 2012; MARTINS et al., 2011). In contrast, in studies using dinoprost, researchers did not find any effect of parity on the complete CL regression rate (BRUSVEEN et al., 2009; WILTBANK and PURSLEY 2014). The effect of parity on luteolysis may be related to the different luteolytic products used in the studies.

In the current study, the pregnancy rate in estrous cows with uterine diameter ≤ 3 cm was higher than in the other group, but there was no significant difference between the groups. It was suggested that many of the cows with uterine diameter >3 cm were multiparous cows, and parity may affect uterine diameter, as reported previously (BAEZ et al., 2016). A relationship was found between the total size of the uterus close to the time of AI, and fertility in dairy cattle (YOUNG et al., 2011). These researchers found that multiparous and primiparous cows had a greater and lower percentage of large uteruses, respectively. Additionally, cows with the largest uterine size had lower conception rates compared to cows with a medium and small uterus. The results of their study were disturbed due to the mixing of parity variables in the analysis of the effect of total uterine size on fertility (YOUNG et al., 2011). In another study, there was a negative regression between uterine size and fertility in dairy cows, particularly for multiparous cows (BAEZ et al., 2016).

A positive relationship was reported between endometrial thickness (ET) near the time of AI and pregnancy per AI (YUVAL et al., 1999). More estrous cows with endometrial thickness less than 2 cm were pregnant; although the pregnancy rate in cows with endometrial thickness ≥2 cm was near to that of cows with endometrial thickness less than 2 cm. The mean maximum thickness of the dorsal surface of uterine body was reached on the day before ovulation (GINTHER, 1998). The uterus was thicker during estrus than during diestrus. Souza et al (2011) measured the endometrial thickness of both uterine horns during hormonal protocol treatment. Ovulation rates in cows with ET ≤ 8mm were lower than in cows with ET >8 mm. Also the percentage of pregnant cows per AI was lower in cows with ET ≤ 8mm (SOUZA et al., 2011). Ultrasound measurement of ET 48 h after PGF2α treatment in an Ovsynch regime was a good index for prediction of ovulation failure and success in pregnancy. Likely, subfertility in Holstein cows with decreased ET was due to low circulating estradiol levels near the time of AI, poor progesterone priming, or incomplete luteal regression during timed AI protocols (SOUZA et al., 2011).

We modified the scores for uterine folding into three groups (minimal, medial and maximal folding). Differences in pregnancy rates between the groups were not statistically significant. Apparently, during the luteal phase the cranial portions of the horns tend to fold into layers. This is compatible with reduced edema at this time. As fluids enter the tissue during estrus, the resulting internal pressure may straighten or unfold the cranial portion and reduce the tortuosity of the remaining portion of the horns (GINTHER, 1998). We studied the estrous cows at the time at which it was expected they would have minimal progesterone concentration, maximal concentration of estrogen and, therefore, minimum folding of the uterus. Most of the cows in this study had minimum and medium uterine folding.

The results of the evaluation of uterine edema indicated that there was no statistically significant difference in pregnancy rates between the groups. Researchers indicated that endometrial edema has a role in assessing the phase of the estrous cycle of mares and, in turn, assisting in the decision on the optimal time of mating (PYCOCK and NEWCOMBE 1996). Improper cervical dilation was indicated in estrous mares with extreme uterine edema or a lower volume of uterine fluid on the first or 2nd day of estrous (McKINNON et al., 1988). Retention of uterine edema after second ovulation or an increase in edema after breeding is indicative of persistent mating-induced endometritis, angiogenesis, or lymphatic lacunae (LEBLANC and McKINNON et al., 2011).

The uterine contraction scoring had no significant effect on pregnancy rates in estrous dairy cows. It is suggested that fertilization take places when the lowest number of sperm reach the ampulla. Researchers have indicated that clitoral massage had no effect on the non-return rate of Holstein cows. The skill of the individual clinician and dairy herd manager likely have a greater effect on fertility than the individual components of the insemination process (COOPER et al., 1985). Pregnancy rates in cows were not affected by oxytocin administration during AI (HAMALI et al., 2011). There is a temporal association between increased E2 concentration, increased endometrial thickness, and the unfolding of the uterine horns (GINTHER, 1998). Non-ultrasonic studies have indicated that under estradiol influence, contractions before ovulation originate near the uterine body and move toward the oviducts. After ovulation, peristaltic contractions move toward the cervix. In the ultrasonic observations, uterine contractility was observed during the preovulatory period. This may reflect movements in the cranial direction, aiding in the transport of sperm to the oviduct. The changes in the ultrasonic uterine appearance of heifers were comparable with changes in a mare's uterus during the estrous cycle (GINTHER and PIERSON, 1984). Others indicated that uterine ultrasonic specifics can be used as biological indicators of the uterus exposed to P4 and E2 (GINTHER, 1998).

Our result indicated no significant differences in uterine lumen scoring between the groups. Increasing uterine edema and luminal fluids are both closely related to increasing E2 levels, but the edema decreases slowly after the estradiol peak, compared to the loss of luminal fluids (GINTHER, 1998). This seems reasonable, because the loss of luminal fluids can occur rapidly through the cervix, whereas the loss of the tissue fluids requires time-consuming resorption or passage into the lumen (PIERSON and GINTHER, 1987b). The passage of endometrial fluid through the labia occurs on about day 1 of the estrus cycle in most cattle, as indicated by the blood-tinged mucus (HAFEZ and HAFEZ 1987). This phenomena may be related to the decreased volume of uterine luminal fluid observed in the ultrasound studies on day 1 (PIERSON and GINTHER, 1987b). Lower conception rates were associated with a luminal diameter greater than 2 mm and echoic content (ABDALLA et al., 1994). No significant usefulness of protective administration

of oxytocin in combination with dinoprost or ergometrin was observed for reproductive performance after parturition in lactating dairy cows (HOLICKOVA et al., 2015). The oxytocin released by clitoral massage facilitates sperm movement into the reproductive tract (BOZKURT et al., 2007).

Conclusions

In conclusion, current results indicate that although follicle size affects pregnancy rates in estrous cows, other uterus ultrasound findings of during estrus did not have any significant effect on the pregnancy rate of Holstein dairy cows. It seems that it may be related to some individual variations in the ultrasound characteristics of the uterus of estrous cows. Pregnancy will occur in estrous cows with ovulatory follicles with a diameter larger than 14 mm, and ultrasound echogenicity and morphometry in normal ranges. So, in practice, careful rectal ultrasonography of the reproductive tract of estrous cows will be helpful.

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AHMADI, M. R., A. MOGHEISEH, B. MIHANDOOST, M. ANSARI LARI: Ultrazvučne značajke maternice i jajnika za vrijeme estrusa i njihov odnos sa stopom gravidnosti u mliječnih krava. Vet. arhiv 89, 279-294, 2019.

SAŽETAK

Pretpostavlja se da nakupljanje tekućine u materničnoj šupljini smanjuje plodnost u mliječnih krava. Cilj je ovoga istraživanja stoga bio procijeniti ultrazvučne značajke reproduktivnog sustava, uključujući nakupljanje tekućine u maternici za vrijeme estrusa, te njihov utjecaj na stopu gravidnost u mliječnih krava. Istraživanje je

provedeno na 486 holštajnskih krava uzgajanih u velikom komercijalnom stadu, u Shirazu, Iran. Sve krave su bile u laktaciji i s otkrivenim estrusom. Transrektalni ultrazvuk učinjen je u vrijeme umjetnog osjemenjivanja. Značajke reproduktivnog sustava, koje su obuhvatile promjer folikula, prisutnost žutog tijela u jajnicima, debljinu, nabor i edem maternice te intrauterinu tekućinu, promatrane su i procijenjene ultrazvučno. Krave su nakon osjemenjivanja praćene te im je određena stopa gravidnosti. Analiziran je utjecaj pokazatelja određenih ultrazvukom na stopu gravidnosti. Podaci su obrađeni logističkom regresijskom analizom. Rezultati su pokazali da je stopa gravidnosti, nakon prilagođavanja pariteta životinja te dnevne količine mlijeka i srednje dnevne količine proizvodnje (OR = 1,84, P = 0,005), bila znakovito veća u krava s folikulima većima od 14 mm (38,8 %) u usporedbi s onima od 14 mm i manjima (27,3 %). Nije uočena povezanost između stope gravidnosti i drugih ultrazvučnih značajki reproduktivnog sustava za vrijeme estrusa praćenog u ovom istraživanju (P>0,05). Zaključeno je da je veličina folikula pozitivno povezana s stopom gravidnosti mliječnih krava u estrusu. Kakogod, drugi ultrazvučni nalazi maternice, uključujući intrauterinu tekućinu, nisu pokazali povezanost sa stopom gravidnosti.

Ključne riječi: promjer folikula; osjemenjivanje; lumen score; žuto tijelo; laktacija