MILK PRODUCTION AND AGE IMPACT ON OVARIAN ACTIVITY DURING *POSTPARTUM* PERIOD

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Abstract: The aim of this study was to evaluate milk production and age impact on ovarian activity in *postpartum* period. The material for this study were collected from 32 clinically healthy holsteinizated cows which were selected by days average after parturition, by daily milk production and by age. Cows were divided into groups: ***by milk production*** (production average of 14–21 days *postpartum)* analogue of age: 1) < 30 l (N=14), 2) 31–40 l (N=13), 3) > 41 l (N=5); ***by age*** analogue of production: 1) I lactation (N=5), 2) II lactation (N=17), 3) III lactation (N=10). Ultrasound of reproductive system were performed with Caresono HD 9300 VET to track dynamic of ovarian size and changes of functional units. Rectal examination were performed 6 time repeated for each cow every 7±1.5 days.

It was found that dynamic of ovarian size depend on productivity in *postpartum* period (P<0.001) also dynamic of reproductive system is better expressed when productivity higher. Cows with the medium and high yield maximum size of ovary reach at 22–29 days *postpartum*. It was found that dynamic of right ovary area depend on the cow age (P<0.05). It also was found that cows with productivity >41 l per day follicles begin develop from 14–21 days when with low and medium yield from 6–13 days *postpartum*. Regardless of productivity corpus luteum begin develop from 6–13 day. During 22–29 days *postpartum* follicles begin develop for the first lactation cows and during 6–13 – for second and third lactation, and regardless of cow age corpus luteum begin growth during 6–13 days *postpartum*.

Keywords: ovarian activity, production, reproduction, *postpartum*

Introduction. The constant focus on herd reproductive performance increase has a positive impact on the development of the dairy business, the increase in the number of cows in the herd and increased milk production levels (http://www.fao.org/wairdocs/ilri/x5442e/x5442e03.htm#TopOfPage).

The *postpartum* (*pp*) period, also called the puerperium (or paripartum), is a physiological and global process of modifications occurring in the female reproductive tract after parturition that leads to recovery from the changes that took place during pregnancy (Breda et al., 2015).

Most cows in peripartum period enter a state of negative energy balance (NEB) associated with many metabolic changes which have carry over effects on the resumption and normality of estrous cyclicity. These relationships between metabolic profiles and fertility differed between first lactation cows (which are still growing but produce less milk) and mature animals (Wathes et al., 2007). Research has shown that the yield is demand of high nutrient and NEB is closely related to the lack of ovulation in *pp* period (Butler, 2003). Cow with higher milk yield has an impact for appearing of NEB which prolongs renewal period of ovarian functional activity (Sakaguchi et al., 2004).

Higher dairy milk production capacity is associated with a decrease in fertility (Butler, 2003). Cow with higher milk yield delays normal ovulation (Crowe et al., 2014) and in the early period after calving have weakly expressed estrus (Peter et al., 2009) also milk yield of early breeding first lactation heifers were significantly lower (Ettema et al., 2004).

Inactive ovaries are associated with the absence of estrus (DesCôteaux et al., 2010). Presence of developing follicles indicates activity in ovarian – gametogenesis and steroidogenesis (http://www.nadis.org.uk/bulletins/fertility-in-dairy-herds/part-4-identifying-and-treating-the-abnormally-cycling-cow.aspx). Evaluating reproductive status follicle size is an important indicator that shows that ovulatory power gain within 12 mm size (Bittar et al., 2014). It was also found that the right ovary are higher than the left when animal get mature but in three and four years old cow left ovary is higher than the right one (DesCôteaux et al., 2010).

About 95 % of dairy ovarian activity renewed up to 50 days after calving (Hafez et al., 2000) and follicular growth generally resumes within 7 to 10 days in the majority of cows (Crowe et al., 2014). I.M. Sheldon and others (2008) wrote that the first dominant follicle is selected around 10–12 days after calving. Dairy cows that do not have a NEB usually appear first ovulation about 15

day (Crowe et al., 2014). Some researchers have found that the first ovulation is observed within 2 weeks after calving (Sakaguchi et al., 2004), other that within 15-30 days (Callahan et al., 1971; Kesler et al., 1979; Lucy et al., 1991), while other assume that first ovulation after calving can occur after 3 weeks (Lucy et al., 1991). J.S. Stevenson and E.P. Call (1983) established that interval to first ovulation was 19±1 days whereas interval to first detected estrus averaged 45±3 days. For non-lactating cows first ovulation is already on the 10th day after calving and for lactating cows it delay of up to 30 days or longer (Noakes et al., 2001). With modern cows occur that average interval to first ovulation is 10 days longer (Lucy, 2001).

The first ovulation is often "silent" with a short ovulation interval (> 70 percent) (Crowe et al., 2014). Growth and persistence of the corpus luteum may be subnormal and time from the first to the second ovulation averages about 15 days as compared to about 21 days from the second to the third ovulation (Callahan et al., 1971). For the most cows in second estrus "first" estrus and ovulation are found about 35 days after calving. Older cows ovulate earlier than primiparous (Hafez et al., 2000).

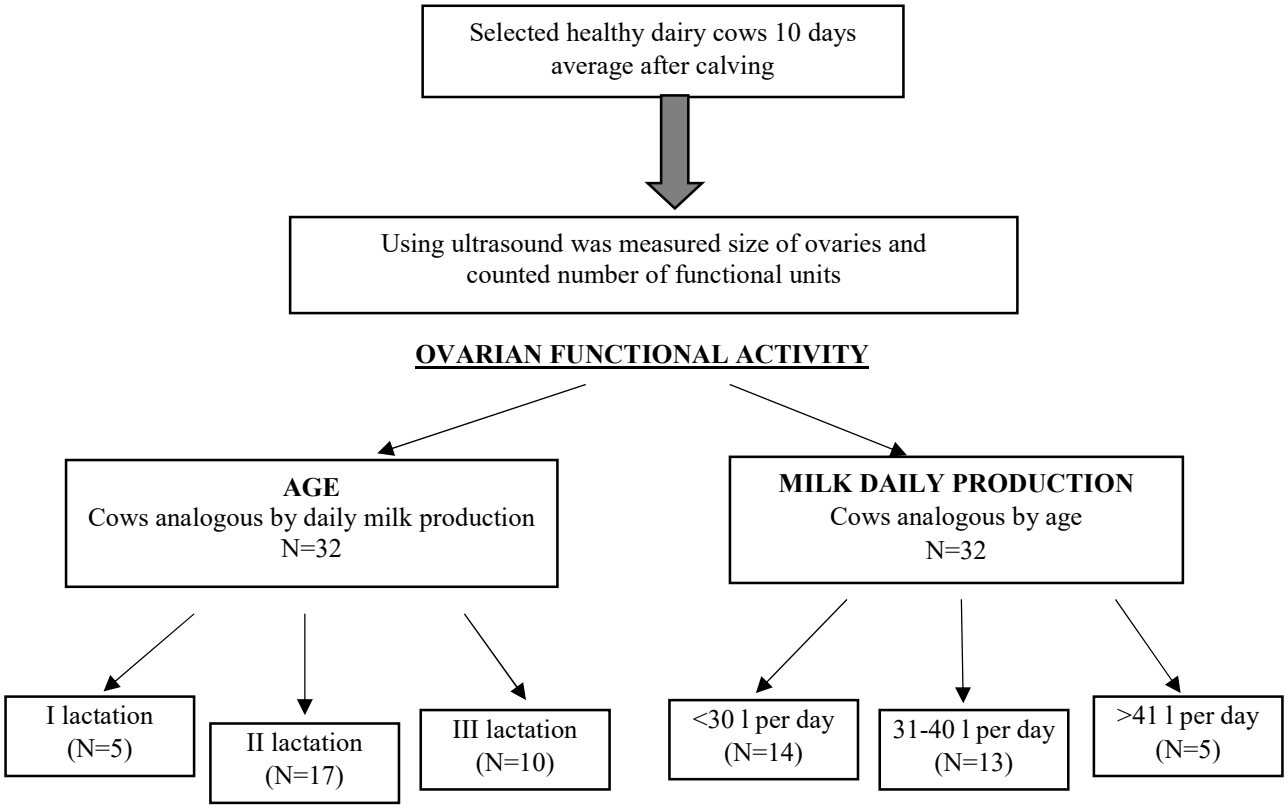
Higher productivity of the herd usually has a better reproduction. Improved reproduction of productive herds reflects better feeding, healthier cows and reproduction management systems (Lucy, 2001). But the first estrus may not reflect renewal of ovarian functional activity (Hafez et al., 2000).

Objective of this study was to evaluate milk production and age impact on ovarian activity during *pp* period; evaluate cows ovarian size depend on the productivity and age during *pp* period; evaluate cows ovarian functional units changes depend on productivity and age during *pp* period.

Material and Methods

Material for this study were collected in dairy farm in autumn-spring period. For this study were selected 32 clinically healthy holsteinized Lithuanian white & black breed cows, housing in loose method in cold barn. Cows for this study were selected about ***10 days pp*** *(average 6–13 days)*, ***by daily milk production*** < 30 l, 31–40 l, > 41 l and ***by age*** (first, second and third lactation).

Ultrasound examination were performed using Caresono HD 9300 VET (Caresono Technology CO., LTD, P.R.China) with linear 6.5 MHz frequency rectal probe. Rectal ultrasound palpation of reproductive organs were performed measuring ovarian length and width and using these parameters for counting ovarian area also counted and measured functional units (corpus luteum (CL) and follicles (F)). Information about cow age, days in *postpartum*, daily yield were taken from herd management program Delaval DelPro 4.2.



1 scheme. **Schematic view of this study**

It was decided that the duration of the study should be length of the two normal estrous cycles (as guide selected 21 days cycle) and starting at the day 10. First rectal examination were performed average 10 days *pp* and repeated 6 time for each cow every 7±1.5 days (1 examination was performed in 6–13 days *pp* N=32; 2 examination – 14–21 days *pp* N=32; 3 examination – 22–29 days *pp* N=32; 4 examination – 30–37 days *pp* N=27; 5 examination – 38–45 days *pp* N=10; 6 examination – 46–53 days *pp* N=4).

Cows were grouped by production and age. Analogous by daily milk production in 14–21 days *pp* (average of at least 30 l per day): 1) <30 l N=14, 2) 31–40 l N=13, 3) >41 l N=5. Analogous by age: 1) first lactation N=5, 2) second lactation N=17, 3) third lactation N=10.

All collected data were analyzed using qualitative data analysis statistical package IBM SPSS Statistics 19. Comparison of average were performed by ANOVA (dispersion analysis) test. Data were considered statistical significance when P<0.05 or P<0.001.

Results and Discussion

Dynamic of ovarian area.

Evaluating changes of ovarian area in different rectal examination time confirms trend of ovarian area increasing (Fig. 1) (P<0.001) during *pp* period.

Table 1. **Average comparison of LO and RO area (mm²) in 6–53 days *postpartum* (P<0.001)**

Examination time	Area of left ovaries (LO)	Area of right ovaries (RO)
I	348.75±39.30	414.13±28.32
II	407.0±45.94	446.25±33.22
III	531.84±48.15	513.19±50.49
IV	541.63±56.91	624.43±62.68
V	596.0±106.58	579.2±101.78
VI	545.0±61.97	693.0±178.22

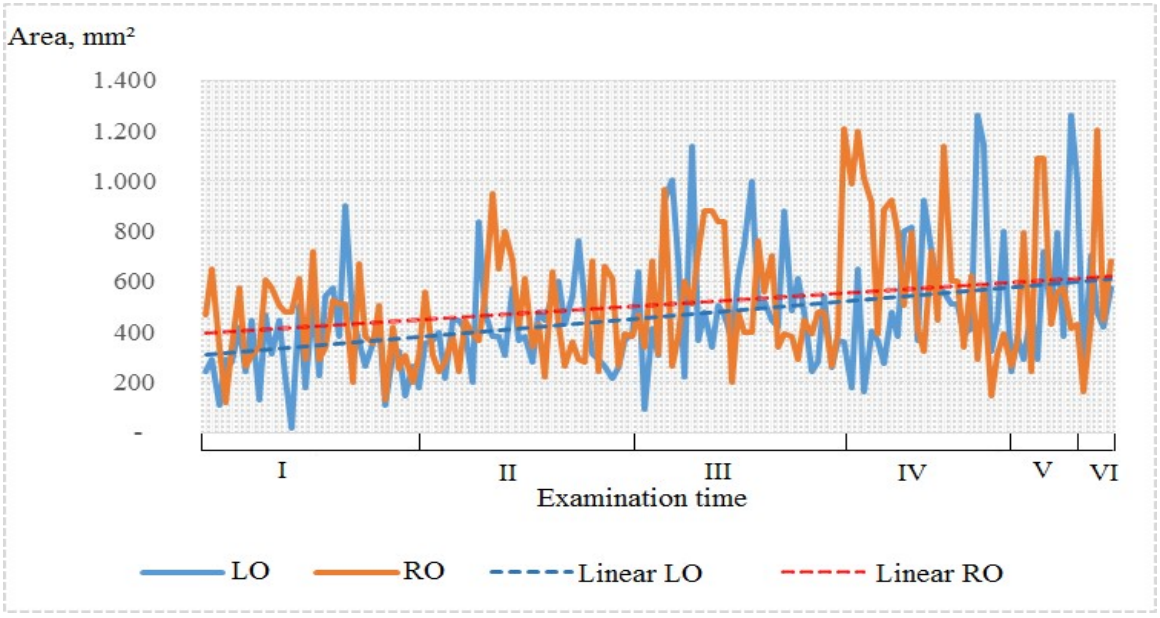


Fig. 1. **Average comparison of LO and RO area (mm²) between different examination time**

Average from I to VI examination time of RO area increase 167.34 % from 414.13±28.32 to 693.0±178.22 mm² (Table 1). Average from I to VI examination time of LO area increase 156.27 % from 348.75±39.30 to 545.0±61.97 mm². Average of RO area in I examination time was 118.7 % major than LO. Average of RO area in VI examination time was 127.16 % major than LO.

Most major difference of LO area established in II and III examination time were 124.84 ±2.21 mm². Average difference of LO area between I and II, IV and V, V and VI examination time varies from 58.25±6.64 mm², 54.37±57.49 mm² to 51.0±44.61 mm². Most least difference of LO area established in III and IV examination time 9.79±8.76 mm².

Most major difference of RO area established in III and IV examination time were 111.24±12.19 mm² and in V and VI examination time – 113.8±76.44 mm². Average diference of RO area between II and III also between IV and V examination time were 66.94±17.27 mm² and 45.23±39.1 mm². Most least difference of RO area established in I and II examination time were 32.13±4.9 mm².

In V examination time difference between LO and RO areas reach the minimum difference 16.8±4.8 mm² though average of LO area were larger than the RO. Meanwhile in VI examination difference between LO and RO area

reach the maximum 148.0±116.25 mm² thought average of RO area were larger than the LO.

Major difference of RO area in III and IV, and in V and VI examination observed due to intense changes on ovarian structural-functional development. It observed that average of RO area increase gradualy and area of LO increase intensily.

Dynamic of ovarian area depend on productivity

Dynamic of ovarian area depend on gradually increasing milk production (P<0.001) in 6–53 days *pp*. Figure 2 shows dynamic of ovarian area in different examination time when daily production <30 l. Average of RO area from I to V examination time increase 123.27 % (P<0.001). Average of LO area from I to V examination time decrease 66.80 % (P<0.001). Major average of RO area difference were 437.33±162.21 mm² and LO – 253.33±177.57 mm² in V examination time. Average of RO area in V examination time increase 172.6 % comparing with LO. Average of LO area when daily milk production <30 l were 381.73±38.6 mm² and RO – 424.0±36.44 mm² of all examination. Average of RO area of all examination time were 111.07 % higher then LO.

Average of RO area from I to IV examination time increase 148.95 % and LO – 178.11 %. When daily production 31–40 l per day average of LO area increase more intensive than RO (Fig. 3.).

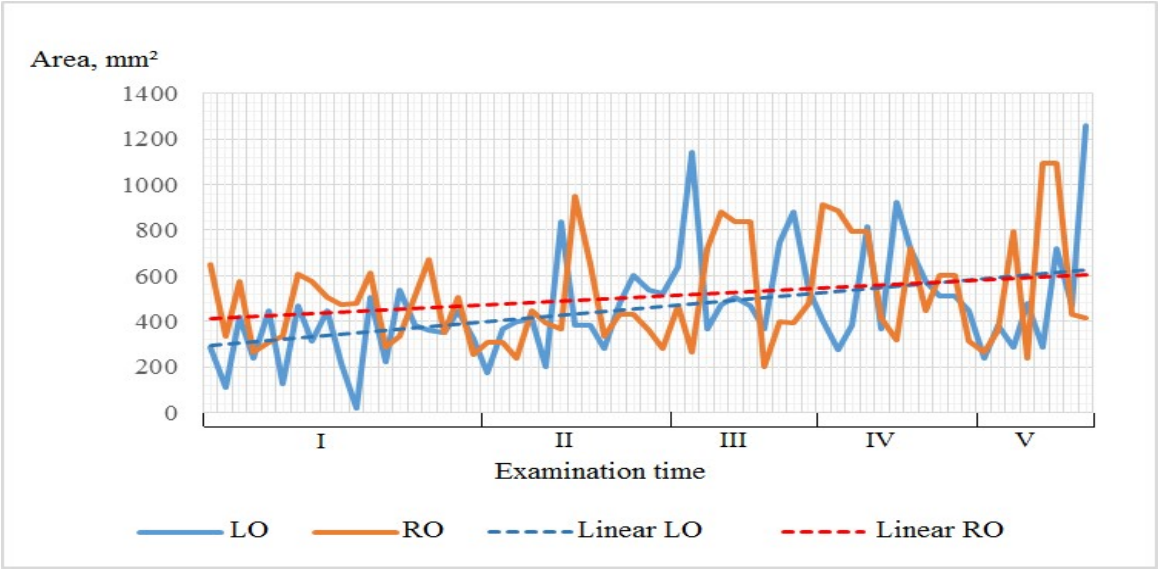


Fig. 2. **Average of LO and RO area (mm²) in different examination time when milk production <30 l per day**

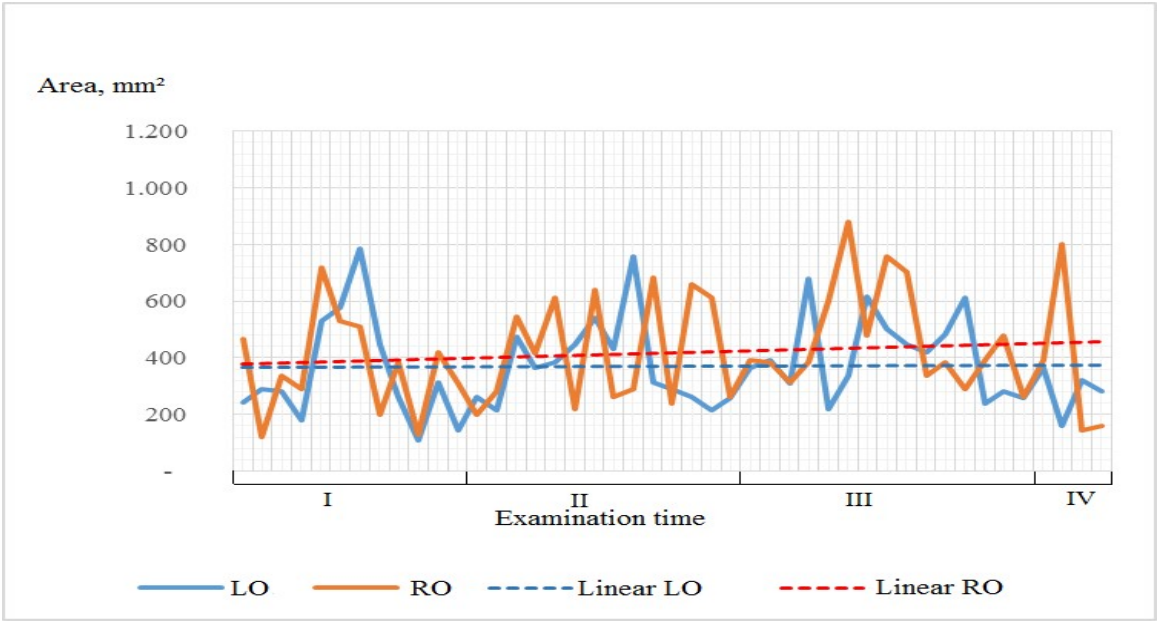


Fig. 3. **Average of LO and RO area (mm²) in different examination time when milk production 31–40 l per day**

Figure 4 shows dynamic of ovarian area in different examination time when daily production >41 l. Observed three ovarian areas increasing peak in this group: two of LO in II and III examination and one of RO in III examination. Average of RO area in I examination time was 532.8±97.59 mm² 125.19 % higher than LO 425.6±69.87 mm². Average of RO area in V examination time was 693.0±178.22 mm² 127.16 % higher than LO 545.0±61.98 mm². Average of RO area from I to V examination time increase 130.07 % and LO – 128.05 %. When daily production >41 l per day both ovaries area increase similarly.

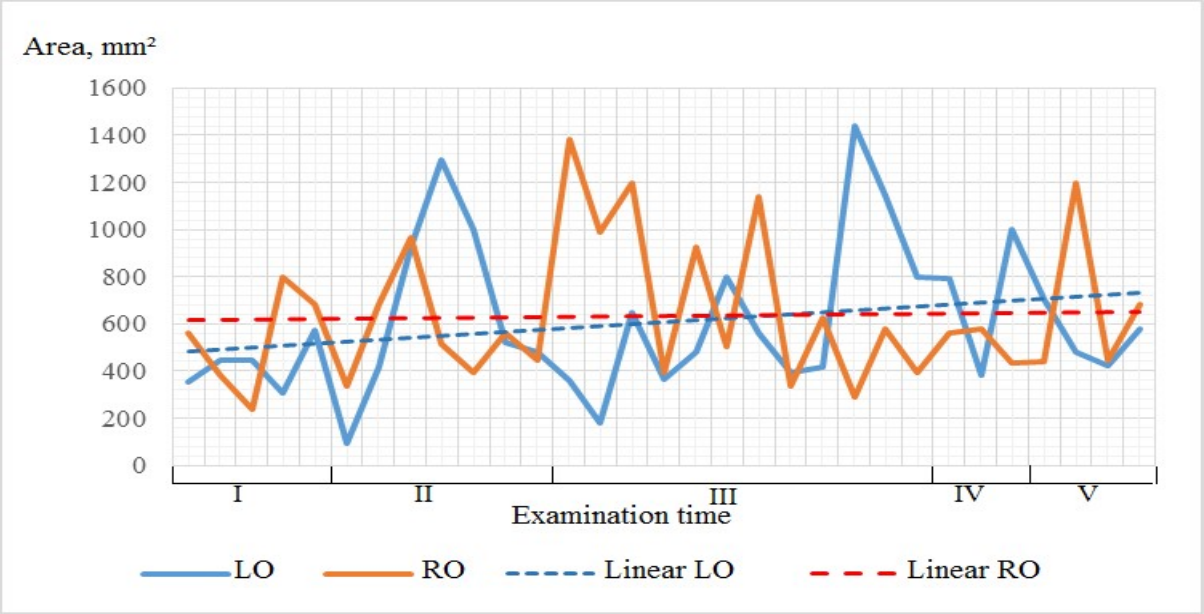


Fig. 4. **Average of LO and RO area (mm²) in diferent examination time when milk production >41 l per day**

Dynamic of ovarian area depend on the age

Observed that average of RO area of first lactation cows (Fig. 5) in I examination time was 278.4±69.13 mm² 125.63 % higher than the LO 221.6±103.56 mm². Observed that average of RO area in V examination time were 57.14 % lower than LO. Average of RO area from I to V examination time decrease 57.47 % while average of LO area increase 126.35 %. It was observed that dynamic of RO areas depend on cow age (P<0.05) in 6–53 days *pp*.

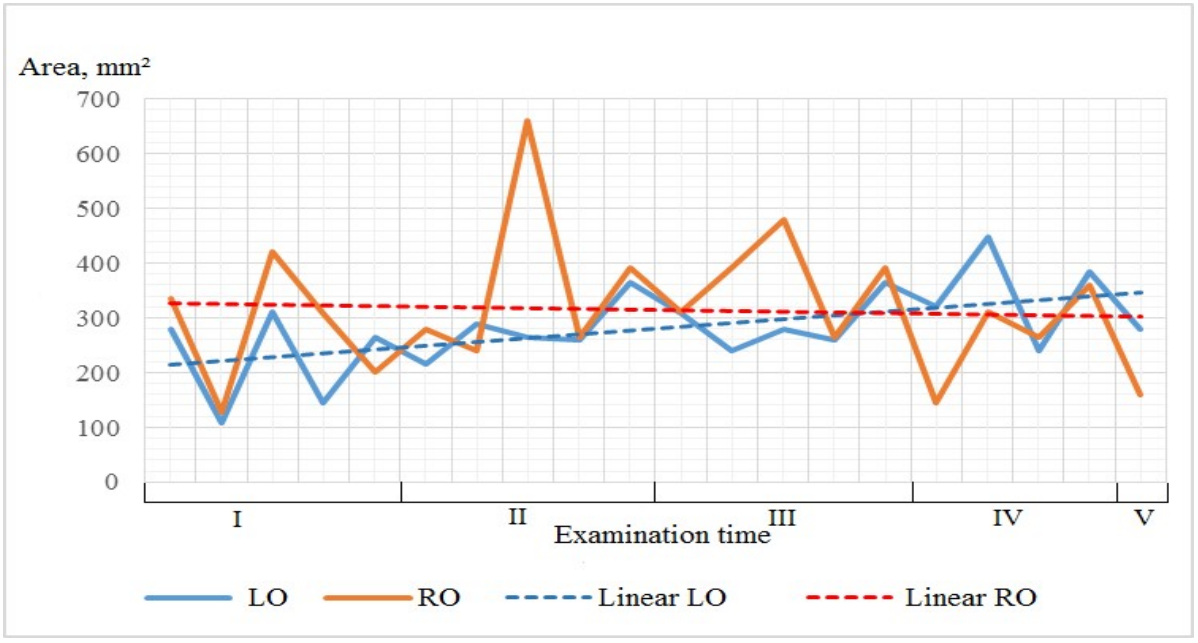


Fig. 5. **Dynamic of ovarian area (mm²) depend on examination time for the first lactation cow**

Observed that average of RO area of second lactation cows (Fig. 6) in I examination time was 455.29±37.83 mm² 118.89 % higher than LO 382.94±57.26 mm². Observed that average of RO area in VI examination time was 693.0±178.22 mm² 127.16 % higher than LO 545.0±61.98 mm². Average of RO area from I to VI examination time increase 152.21 % than LO – 142.32 %.

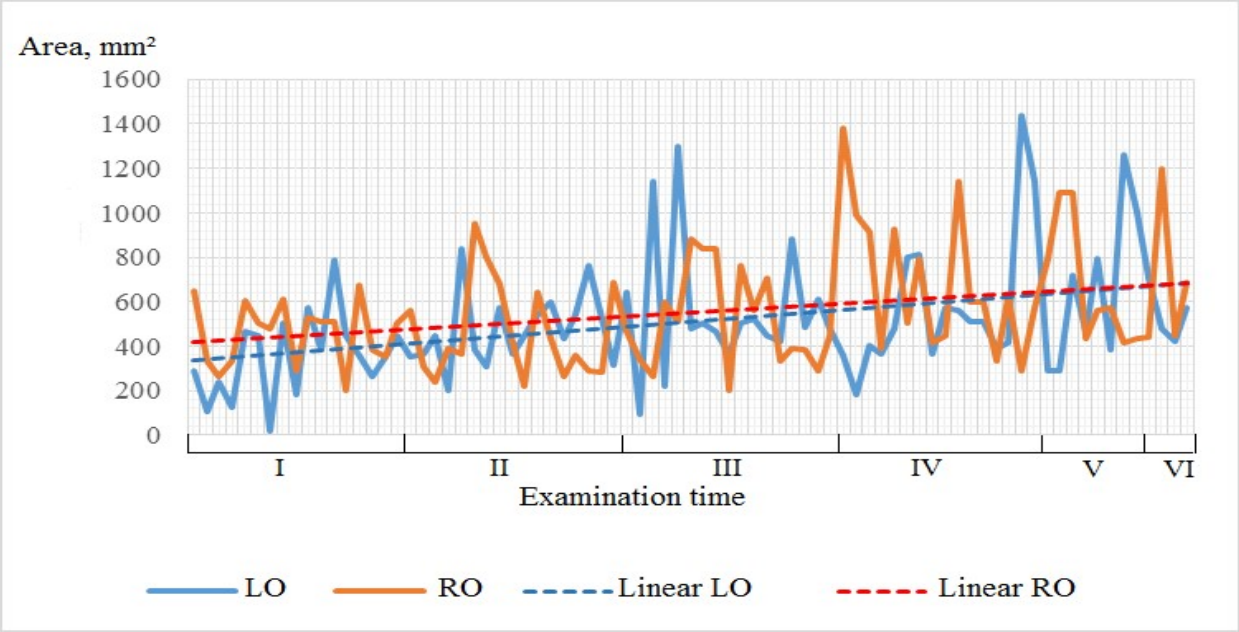


Fig. 6. **Dynamic of ovarian area (mm²) depend on examination time for the second lactation cow**

Observed that average of RO area of third lactation cows (Fig. 7) in I examination time was 412.0±50.35 mm² 116.32 % higher than LO 354.2±71.45 mm². Observed that average of RO area in V examination time was 50.0 % lower than LO. Average of RO area from I to V examination time decrease 58.25 % than average of LO area increase 135.52 %.

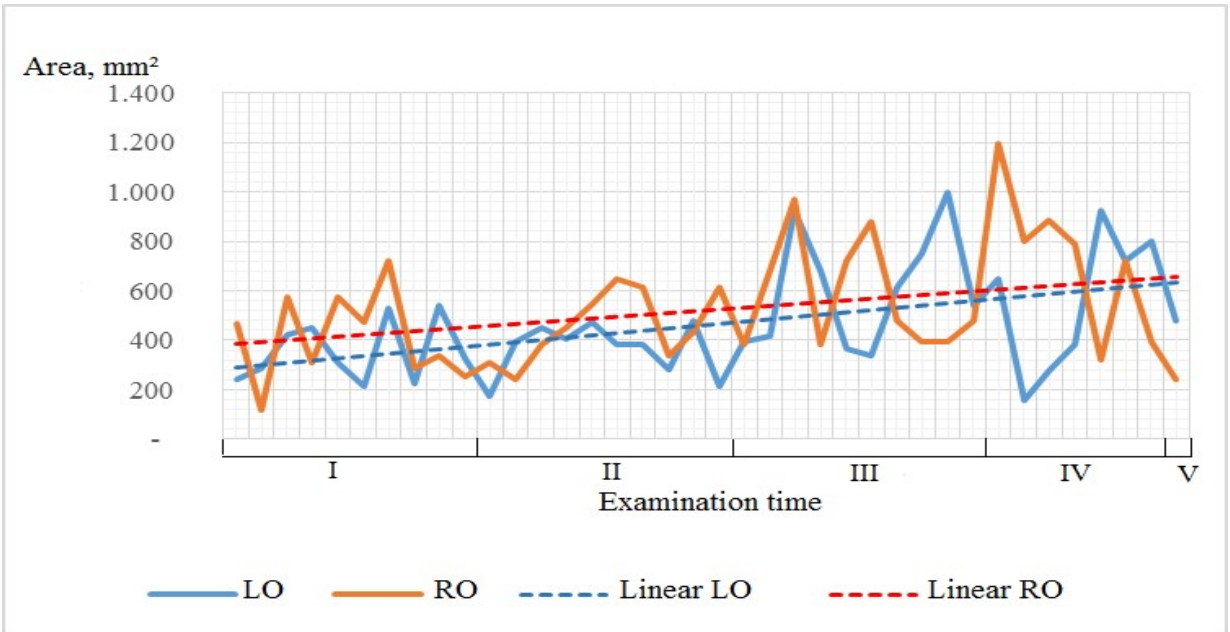


Fig. 7. **Dynamic of ovarian area (mm²) depend on examination time for the third lactation cow**

Dynamic of ovarian functional units depend on production

In ovaries with oestrous cycle can be found developing functional units – follicles and corpus luteum. In different examination time in ovaries were calculated functional units per cow.

Observed that in group with <30 l milk production per day (Fig. 8) number of CL increase and reach peak in III examination time and after peak number of CL decrease. Number of F increase and reach peak in IV and V examination. In group with milk production <30 l per day established intense increase of CL and gradual increase of F.

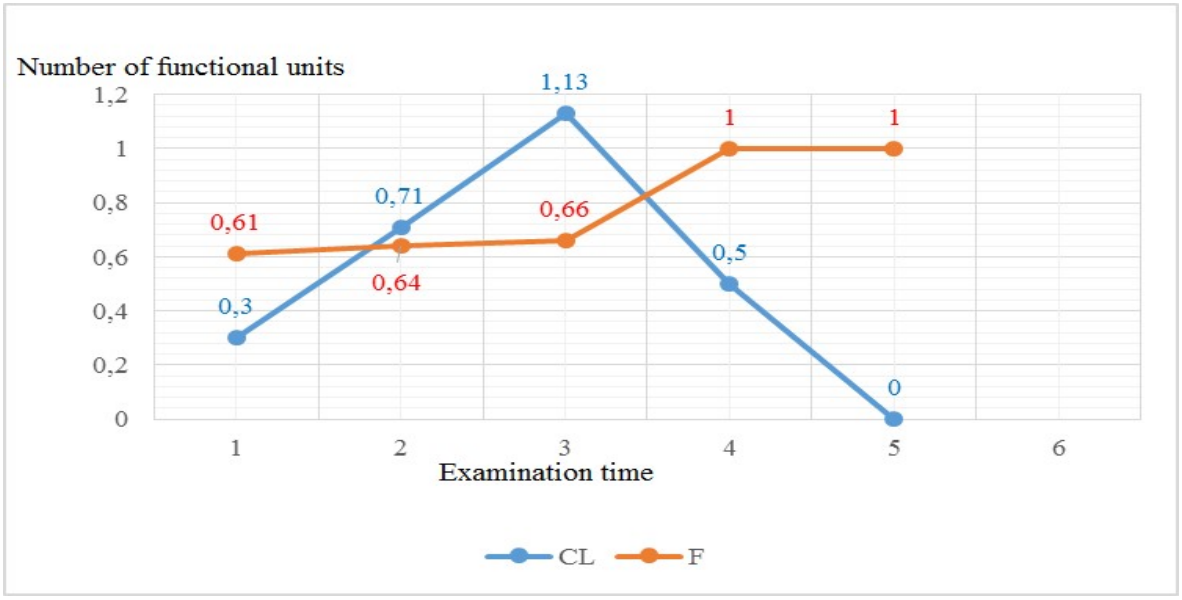


Fig. 8. **Distribution of functional units per group when milk production <30 l per day**

Observed that in group with 31–40 l milk production per day (Fig. 9) number of CL reach peak twice in III and V examination and decrease in IV examination. Number of F increase and reach peak in III examination time. In group with milk production 31–40 l per day established gradual increase and two peaks of CL number, intense increase of F till peak and consistent decrease after peak. Both number of CL and F reach peak in III examination.

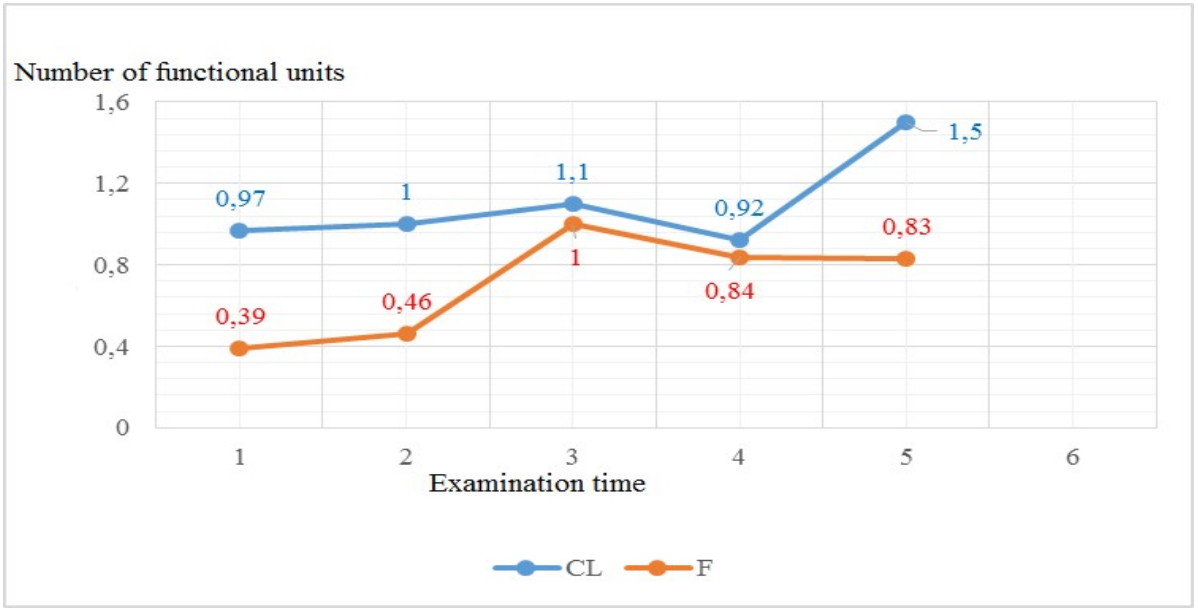


Fig. 9. **Distribution of functional units per group when milk production 31–40 l per day**

Observed that in group with > 41 l milk production per day (Fig. 10) number of CL reach peak twice in II and IV examination time. Number of F increase and reach peak in III examination time. In group with milk

production >41 l per day established intense increase and intense decrease of CL number and intense increase and gradual decrease of F number.

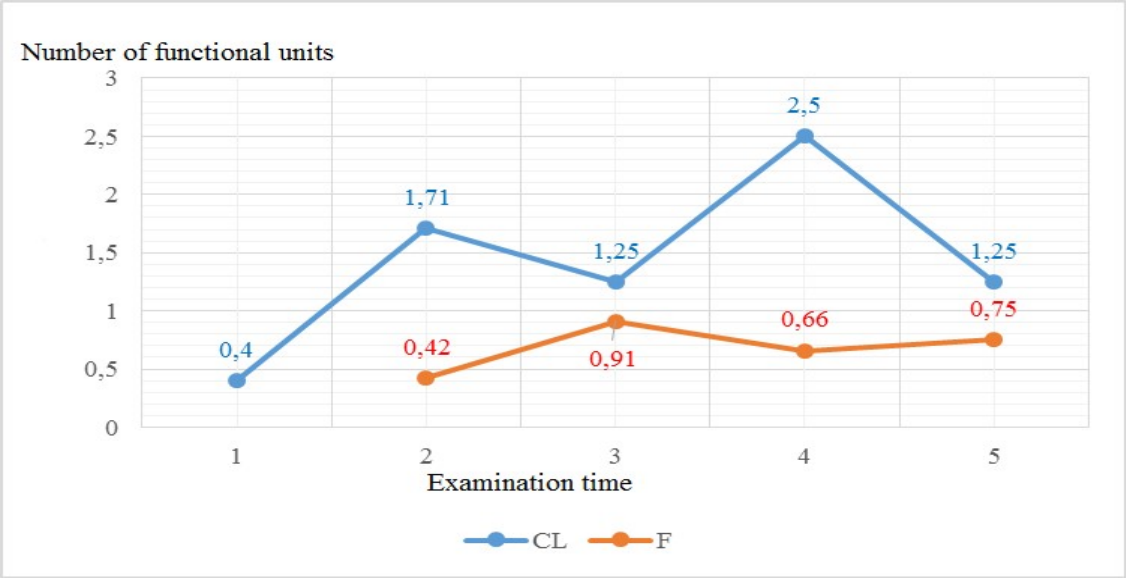


Fig. 10. **Distribution of functional units per group when milk production >41 l per day**

Dynamic of ovarian functional units depend on the age

Number of CL per group for first lactation cows (Fig. 11) increase and reach peak in II examination time. Number of F start increase from III examination time and reach peak in V examination. Observed intense increase of CL number until peak and intense decrease after peak. Observed intense increase of F number.

Number of CL per group for second lactation cows (Fig. 12) decrease in II examination time and reach maximum number in III and V examination times. Number of F decrease in II examination time and reach maximum number in III examination time. To the first peak of CL number observed intense increase, and gradualy increase and decrease until second peak of CL number. Observed intense increase number of F until peak and gradual decrease after peak.

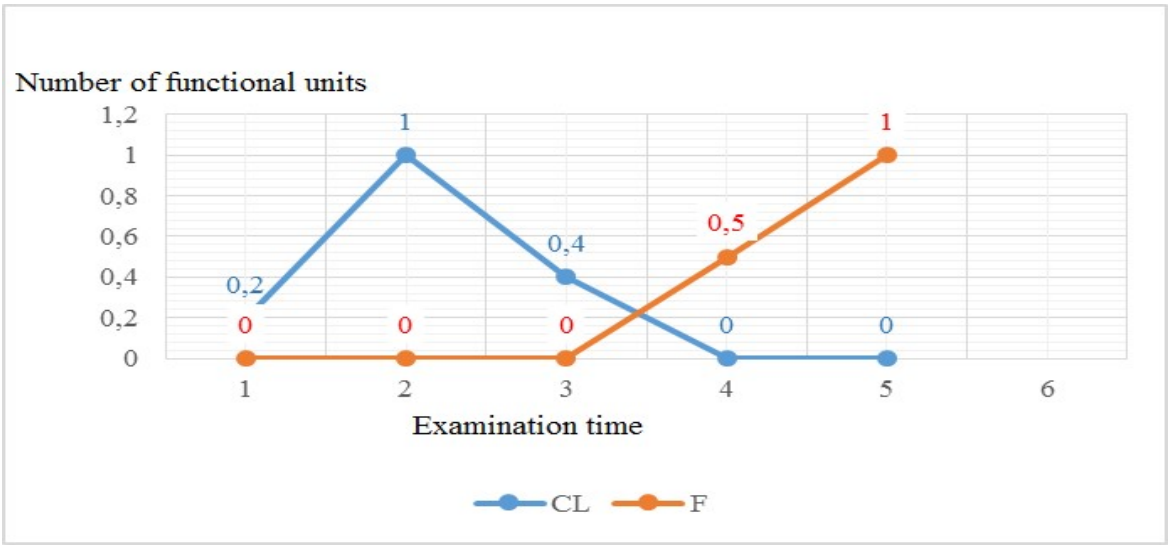


Fig. 11. **Number of ovarian functional units per group for first lactation cows in *postpartum* period**

Number of CL per group for third lactation cows (Fig. 13) decrease in II examination time and reach maximum number in V examination. Number of F reach peak in IV examination time. Observed gradual increase of CL number until peak and sudden decrease after peak. Observed intense increase of F number until peak and gradual decrease after.

Discussion

Was observed that during 6–53 days *postpartum* period RO ($P<0.001$) and LO ($P<0.05$) areas has tendency

to increase. In this study area of RO increase more evenly (P<0.001) than LO. Average of RO area from I to VI examination time increase 167.34 % while LO 156.27 %. This match our observation that ovaries area increase until 53 day *pp*.

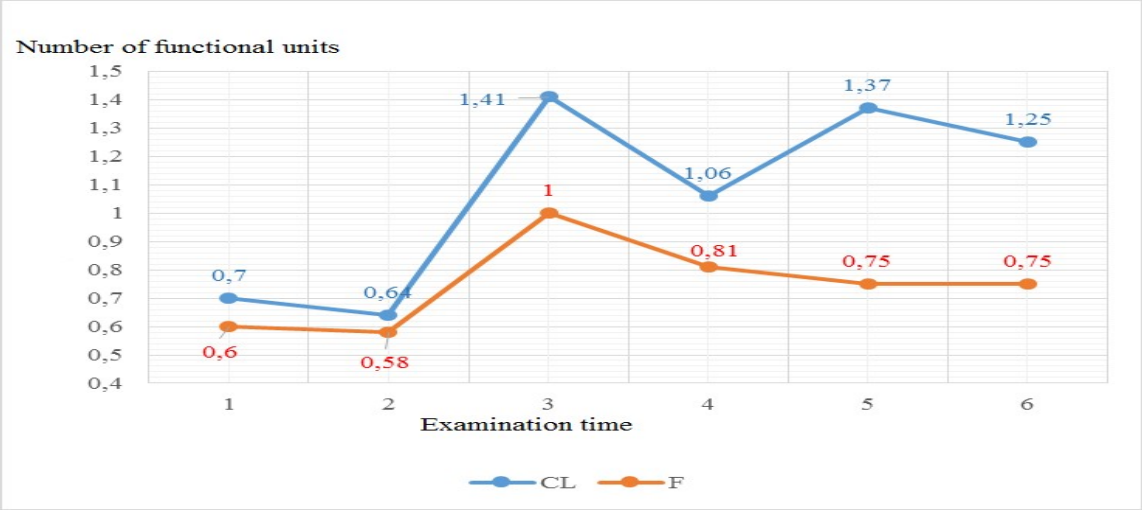


Fig. 12. **Number of ovarian functional units per froup for second lactation cows in *postpartum* period**

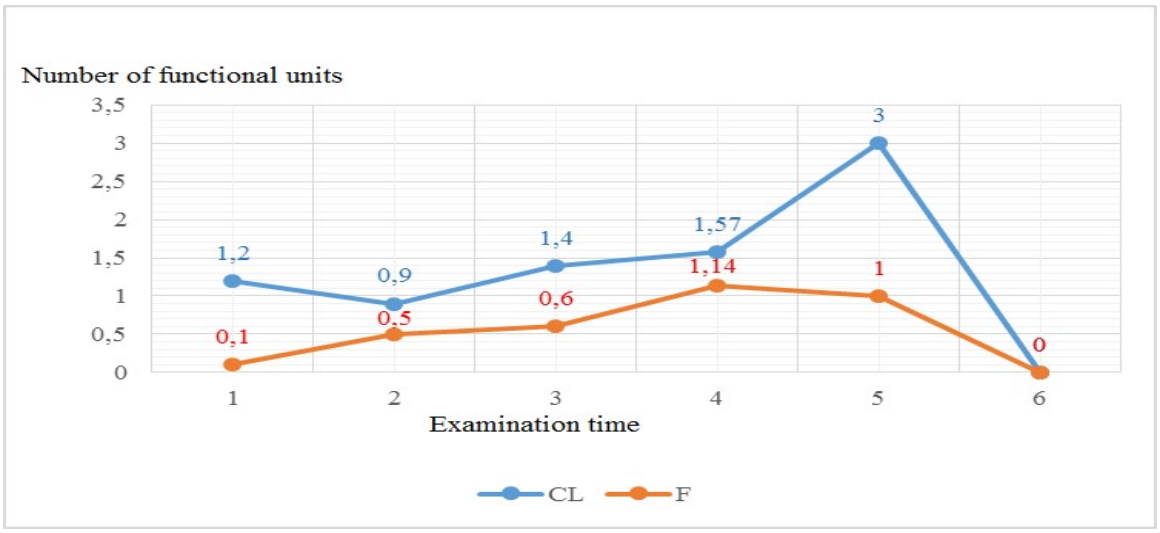


Fig. 13. **Number of ovarian functional units per group for third lactation cows in *postpartum* period**

It was observed that biggest difference of RO area were between III and IV examination time 111.24±12.19 mm², V and VI examination time 113.8±76.44 mm². While biggest difference of LO area were in II and III examination time 124.84 ±2.21 mm². Changes in RO area betake in 22–37 and 38–53 days *pp* while in LO 14–29 days. RO had two area increasing peak and LO one but increase of LO area were better expressed. L. DesCôteaux and others (2010) found that ovarian size varies because of emerge of functional structure and depend on the stage of estrous cycle. D.W. Salisbury and others (1961) admit that precense of functional derivatives increases overall size of the ovary.

Most varies of ovarian area observed in cows with low and medium yield. With productivity <30 l and 31–40 l per day RO area increase respectively 111.07 % and 111.09 % comparing of all examination times data while with productivity >41 l per day RO area increase 104.54 % than LO. W. R. Butler (2003) believe that increased capacity for milk production in dairy cows has been associated with a decline in fertility this is with less ovarian activity. In S.W. Beam and W.R.Butler (1999) opinion reduced reproductive performance has been associated with high milk yields.

M. Masiulis (2003) predicate that dynamic of ovarian size depend on cow age. It was found that with increasing animal age right ovary remains higher than the left. Areas of RO was higher than LO performed examination in 6–13 days *pp* for first, second and third lactations cows. Was found that performed examination of ovarian area in 46–53 days *pp* LO area were higher than RO for first lactation cows and ovarian areas become almost identical for second and third lactation cows.

Independent of productivity CL begin develop in 6–13 days *pp*. Observed that in cows with productivity <30 l and 31–40 l of milk per day growth of F begin from 6–13 days *pp* and with productivity >41 l per day in 14–21 days. Independent of cows age CL begin develop in 6–13 days *pp.* Observed that F begin develop in 6–13 days *pp* for second and third lactation cows while for first lactation between 22–29 days. J.C. dos S. Breda and others (2014) established that about 10–40 days *pp* is observed significant cows ovaries cyclical renewal. E.S.E. Hafez and B. Hafez (2000) wrote that the older cows ovulate before than primiparous. The first estrus may not reflect the ovarian functional activity of renewal.

J.C. dos S. Breda and other (2014) also state that cows with high-yield productivity have low incidence of ovarian activity and lower dominant follicle. Our data contrary to the author‘s assertion that high yield cows ovaries are less active, we observed that cows with >41 l milk per day have two increasing peaks of CL number and two increasing peaks of F number.

W.C. Wagner and W. Hansel in 1968 carry out the studies found that ovaries at the 30 day *postpartum* reaches its maximum weight. In the study we also found that the second and third lactation cows ovaries reaches maximum size in 22–29 days *pp*.

Conclusions

Dynamic of ovarian areas depend on productivity during *pp* period (RO r=0.296**, LO r=0.314**, P<0.001). Productivity does not have a negative impact on the cow ability to recover after parturition. Herd can be completing with high production cows without negative effect on the duration of *pp* period. Corpus luteum in the cow ovaries established during 6–13 days independently of productivity and the follicles in high yield cows recorded in 14–21 days. Dynamic of ovarian area depend on age during *postpartum* period (RO r=0.209*, P<0.05). No statistical significance was found between ovarian functional unit dependence on the productivity or on the age during *pp* period.

** Correlation is significant at the 0.01 level

* Correlation is significant at the 0.05 level

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