Full Length Research Paper

Research regarding the reproductive capacity and biotechnologies of cows

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The decline of fertility in the last decades is a major concern in dairy farms being caused by a series of factors analysed in the present paper. Research took place on a number of 950 dairy cows exploited in a semi-intensive system in the North-East area of Romania. On the whole analysed population, the frequency of reproduction disorders were between 12 (in January) and 27% in July, with a greater intensity starting with the end of spring up to winter season. The most frequent reproduction disorders were anoestrus, losses of heat cycles, weak heats undetected in due time, metritis, feeding and others. The batch of the cows with over 85 days of infecundity represented 88.15%, while the batch with one to 27 days infecundity was of only 1.05%, and the batches with 28 to 56 days and 57 to 84 days of infecundity represented 4.25 and 6.55%, respectively from the total of analysed population. The most days of infecundity were recorded at primiparous cows (55.90%) followed by the second-parous ones (32.10%) and multiparous cows (12.00%). The losses by infecundity and sterility over passed the rate of 25% from production. The common feature of the reproduction biotechnologies consist maximization of the genetic process for cattle, of their wealth fare and fertility state, in the conditions of reducing the price cost per product unit.

Key words: Cows, reproductive capacity, biotechnology

INTRODUCTION

The post-war period has placed the agriculture of the European Union countries in a favourable context: sustained rates of economic growth, the acceleration of the technical progress, the development of international trade and the progress of European construction. Reproduction conditions of the economic production of humpless cattle are obtained since the animals follow the breeding process meant for different productions. The female young stock for reproduction and milk production must go through the physiologic puberty in normal conditions and reach the optimal age for reproduction. The normal development of the body, the reproductive function, the level of subsequent economic productions and the health state depend on these aspects. Thus, a normal reproduction at farm level means more milk, more meat and implicitly higher profits (Maciuc et al., 2003, 2006).

The decline of fertility in the last decades has been a serious concern in the milk cow farms since this is due to a series of factors analysed in this study; thus, ensuring animals' wellbeing in the animal husbandry farms represents a complex and current issue at world level that conditions progress directly.

MATERIALS AND METHODS

Research was carried out in a three-year period (2009 to 2011) on a total livestock of 950 Friesian milk cows exploited in a semi-intensive system in the north-eastern part of Romania. In the studies on private farms, we focused on the genetic value of

the biologic material, the reproductive, improvement and feeding technologies and the management of the technological factors. In all analysed farms, the cows had the same exploitation conditions; in closed shelters with free maintenance. The shelters were provided with paddocks to ensure a better movement of animals. The interior arrangement of the shelter complied with the three functional areas such as the rest area, the movement area and the feeding area. As for litter straw, they used a thick straw layer being about 3 kg of straws per day and per animal. The litter straw was changed depending on the needs (every few days) and the discharge of dejections was made mechanically every 30 days. For the liquid dejections, each farm had a collecting ditch.

The feeding system was seasonally differentiated meaning that in the cold period of the year, they were fed conserved fodders and in the hot period, they were fed green mass in the shelter because the farms did not have a sufficiently large pasture ground to graze. The hay used was grass-clover and leguminous hay (alfalfa, clover), corn silo with 30% dry substance and 20% cellulose, green mass in summer and concentrate mixture.

The milking was done in a 'Fishbone' milking parlour having a capacity of 2 x 10 and 2 x 20 seats, respectively. The arrangement of cows was symmetric and oblique at 30° in relation with the service alley and by two rows.

Holstein-Friesian breed is considered as the world gene pool for the improvement of the milk production. It is quite a demanding breed in terms of exploitation conditions and it needs a balanced diet with high quality fodders and protein-rich concentrates. The inadequate exploitation conditions determine the immediate decrease of milk production and also reproductive problems.

The data collected by personal observations were supplemented with those from the records of farms. The analysis of the reproductive capacity supposed the following for a period of three years (2009-2011): the influence of the age at first calving on the production and reproduction indices, the insemination index, the conception rate, the gestation index, the frequency of difficult calving, the frequency of reproduction disorders and the economic efficiency of infecundity and sterility control. Since 2011, in the farms under study, they started using some biotechnologies such as the induction of estrus and ovulation and the synchronization of estrus meaning a higher focus on calving programming. The synchronization of estrus also brings about the synchronization of ovulation and, consequently, the treated females may be inseminated at a fixed time period after the end of treatment without being necessary to identify estrus. Since ovulation is variable in time from one individual to another, it is necessary to carry out a double insemination within the interval of 48 to 72 h after the end of treatment.

The treatment ensuring the best synchronization of estrus and ovulation and which was used in the farms under study is the combined one of estrogens and a substance having a luteolithic effect. To induce luteolysis, they used $PGF_2\alpha$-based preparations. The current treatment based on progestins and prostaglandins is well defined and it provides an efficacious control of estrus. To better supervise the ovulation in view of blind insemination (without identifying the heat symptoms), the progestin treatment was improved with some preparations based on estrogen hormones such as Gn-RH, HCG, PMSG or $PGF_2\alpha$ (Aslan et al., 2011).

All data such as the arithmetic mean ($\overline{X}$), the average error ($\pm s\overline{x}$), standard deviation (s), the variability coefficient (V%), the regression coefficient, and the regression line (Maciuc et al., 2003) were statistically processed and synthesized in tables and figures. The statistic processing was carried out by means of SPSS 16.00 for WINDOWS programme. Our goal was to see how the cows' reproductive capacity evolved in this interval and which losses were in the conditions of a deficient reproduction (Averill et al., 2006; Boichard and Manfreddi, 1994).

RESULTS AND DISCUSSION

According to a study of Holstein Friesian Breeders Association from the USA based on a multiannual statistics, cows' fertility expressed by gestation rate diminished in the same proportion in which the milk production increased in 50 years. The study shows that the milk production of the cows owned by the breeders from the association doubled as compared to the milk production of the individuals of the same breed that had results in 1957; the average value for milk for Holstein-Friesian breed from the USA exceeds 9500 L in 305 days. Concomitantly with the steady and evolutive progress of milk production, cows' fertility decreased as the milk production increased (Hoard's, 2008).

Influence of age at first calving age on production and reproduction indexes

After analyzing the average of milk production for the entire population under study, we obtained a value of 6737 kg with limits between 3393 and 10737 kg of milk. The fat content of milk had the same high variability, with limits between 3.2 and 5.23, and 80% of the primiparous cows had the fat content between 3.83 and 4.2%. At the same time, the protein content varied between 2.7 and 3.9%, with an average of 3.35% for the first lactation and 3.20% for the second lactation.

The age at first calving was on average 809 days, and 26.5 months, respectively. The duration of gestation had limits between 277 and 287 days. The interval between calving exceeded by much the optimal value; the variation limits ranged between 303 and 937 days, and more than 50% females exceeded the 400-day threshold.

The mammary repose is the single index having an average value within the optimal limit of 60 days, but it has a very high variability between 10 and 150 days. A special situation was registered for the service-period that had the average value of 123 days and limits between 18 and 655 days. The extreme variability of this reproductive index (V% = 58%) caused significant economic losses due to the poor management of the reproduction activity.

The data from the specialized literature show the influence of the age at first calving (VF1) on the milk production from the first lactation and the entire productive life (Maciuc et al., 2003; Gonzalez-Reico et al., 2005). Thus, in the studied conducted, we noticed that heifers that gave birth at the age of 20 to 25 months registered 5734 kg of milk in their first lactation with a high fat content (4.37%) and protein content (3.42%). The primiparous heifers that gave birth at ages between 26 to 31 months registered 6497 kg of milk with a fat content of 4.09% and a protein content of 3.27%. The difference of 763 kg of milk was significant for p< 0.01 and the confidence interval (C.I) was 95%. The regression coefficient and the regression line presented in Table 1 and Figure

Table 1. Regression coefficient between L1nl (milk quantity for 305 days) and V.F.1 (age at first calving).

Specification	Standardized coefficient		Regression coefficient	t	Significance
	B	Standard error			
Constant	1107.579	188.391	0.473	5.879	0.000
V.F.1	3.087	0.207		14.935	0.000

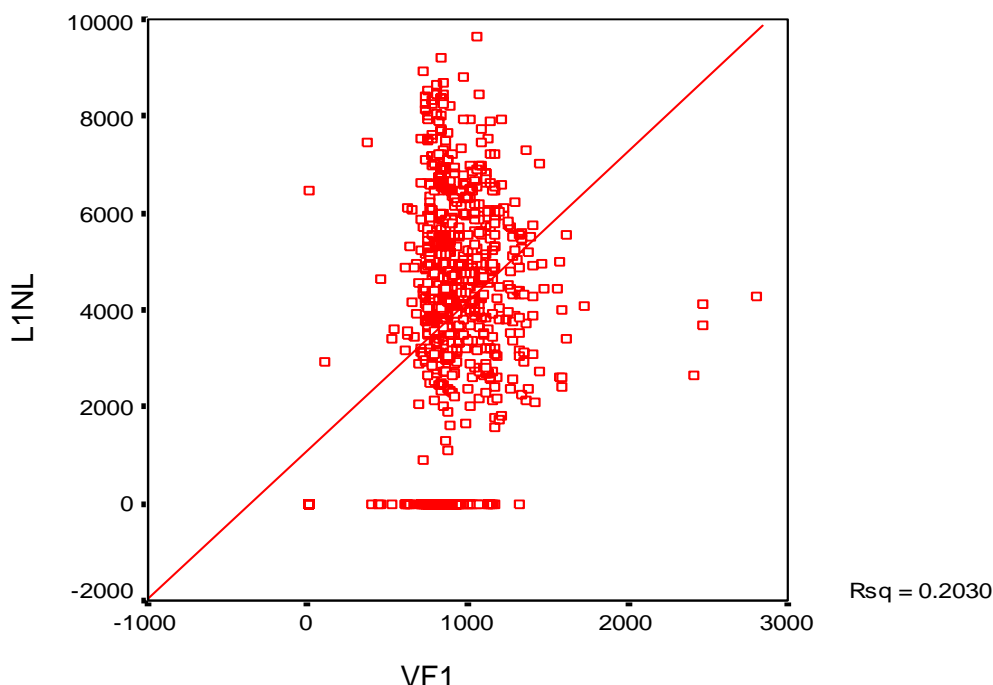a Dependent on variable: milk 305 days (L1NL); V.F.1, age at first calving.

Figure 1. Regression line between the milk quantity for 305 days (L1NL) and the age at first calving (V.F.1).

Table 2. Annual evolution of gestation index in cows and heifers.

Specification (%)	Year		
	2009	2010	2011
Pregnant cows	49.00	51.50	57.50
Pregnant heifers	31.50	35.50	39.00
Total %	80.50	87.00	96.50
% NR	70.50	75.30	83.70

1 illustrate the influence of the age at first calving on the milk production indices.

Analysis of breeding activity

By following the frequency of artificial inseminations during 2009 to 2011, we noticed a relatively uniform distribution situated at a 20 to 25% level, with some particular situations. Thus, in 2009, April registered the highest number of inseminations (35%) whereas February registered 11% artificial inseminations. For 2010, the evolution was similar and for year 2011, the highest number of inseminations was reached in May and June. It must be mentioned that the in 2011 they resorted to the application of some biotechnologies as well as the induction of estrus and ovulation, estrus synchronization and a higher focus on calving programming.

The situation of cows and heifers in the three years for the study (Table 2) shows that the reproductive activity took place within optimal parameters in the last two years (2010 and 2011) when pregnant cows exceeded 50% of livestock and they reached the maximum possible in the last year. At the same time, the non return index (% NR) or «non return» registered the best values in the last year of analysis. This reproduction index characteristic both for cows and bulls was expressed in percentages by registering the cows which do not come into heat after
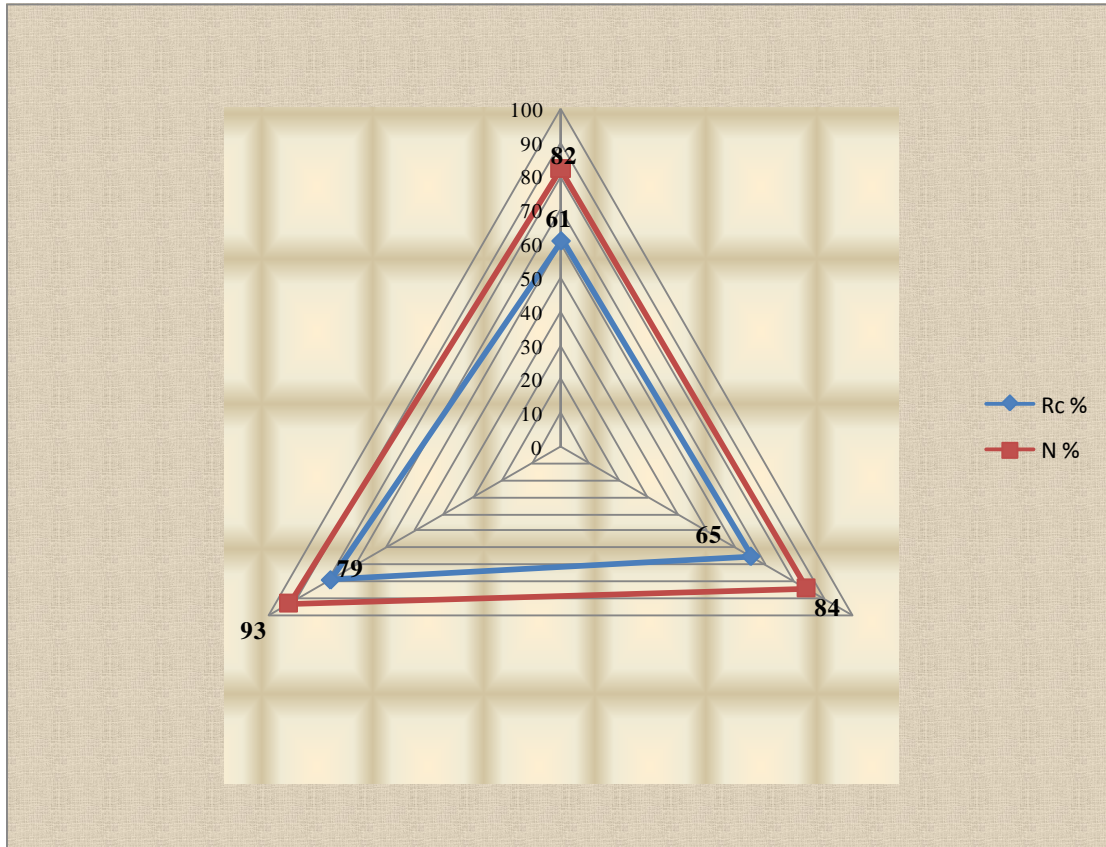
Figure 2. Conception rate after the first insemination (Rc %) and the birth rate index (N%).

the artificial insemination or mated compared to the total number of inseminated cows.

As for the conception rate after the first artificial insemination (Rc-%), we noticed that the highest level was registered in 2011 (79%) whereas in the previous years, it was 61 and 65% (Figure 2). For this indicator followed for a three-year period, differences were significant with a threshold where p < 0.01 and the confidence interval was 95 % (Gonzalez - Reico et al., 2005).

Taking into account the number of cows and heifers annually inseminated, we calculated the birth rate index (N%) based on the ratio between the number of individuals obtained (alive and viable) and the total number of cows and heifers inseminated for the entire population under study (Schneider et al., 2005). The results obtained indicate that the best birth rate was registered in 2011, the values of this index being 93% whereas in the other years (2009 to 2010), the value of this index was approximately at the same level; 82 and 84%. The differences identified were significant for p < 0.01 and C.I. = 95%. The obtaining of this synthetic index of the reproductive function at a level below the optimal one is tightly connected to the frequency of reproduction disorders and the loss of calves by abortions during the

gestation period or within the first days after calving.

By following the dynamics of calving in the three consecutive years (Tables 3 and 4), we noticed that primiparous individuals represented 35.01% meaning a substantial refreshing of livestock. Thus, in the period under study, we introduced exploitation percentage of more than 30% pregnant heifers concomitantly with the removal from livestock of old individuals having poor performances, udder defects, genital or podal disorders or flawed animals (with physical and genital defects). In 2009, we registered a lower value and deficient results due to the fact that ever since that year we have monitored farms and we have had to intervene in the feeding rations (balanced from the energetic-protean and vitamin-mineral viewpoint) and the organization of the insemination and calving schedule and the application of reproductive biotechnologies. Primiparous individuals represented 35.01% of the total number of calving in the three years of the study.

Frequency of reproductive disorders and economic impact

As for the frequency of normal and difficult calving in the

Table 3. Calving frequency in the period under study (2009 to 2011).

Year	Calving		Total			
			Primiparous		Adult	
	n	%	n	%	n	%
2009	950	100.0	293	30.84	657	69.15
2010	950	100.0	330	34.73	620	65.26
2011	950	100.0	375	39.47	575	60.52
Total of 2009 - 2011	2850	100.0	998	35.01	1852	64.98

Table 4. Frequency of normal and difficult calving in the period under study (2009 to 2011).

Year	Normal calving out of 950						Difficult calving out of the 950					
	Total		Primiparous		Adult		Total		Primiparous		Adults	
	n	%	n	%	n	%	n	%	n	%	n	%
2009	917	96.52	273	93.17	629	95.73	33	3.47	20	6.83	28	4.27
2010	925	97.36	316	95.75	602	97.10	25	2.63	14	4.25	18	2.90
2011	937	98.63	364	97.07	568	98.78	13	1.36	11	2.93	7	1.22
Total of 2009 - 2011	2779	97.50	953	95.49	1799	97.13	71	2.49	45	4.5	53	2.86

dynamics of the three years, we noticed that normal calving represented 97.50% of all, whereas difficult calving represented 2.49%. The primiparous individuals registered a higher frequency of difficult calving than the multiparous animals (4.5%) as compared to 2.86%.

For the entire period under study, the frequency of reproductive disorders was between 12% (in January) and 27% in July and it was higher at the beginning of spring until the cold season. The cows with 85 days of infecundity represented 88.15%, those with 1 to 27 days of infecundity represented 1.05%, and those with 28 to 56 days and 57 to 84 days of infecundity represented 4.25 and 6.55% of the entire population analysed. Primiparous cows had a high infecundity (55.90%) followed by second parous cows (32.10%) and multiparous ones (12.00%).

By reproductive disorders, they registered both calf losses and the reduction of milk production. The explanation regarding this evolution throughout the period under analysis resides in the poor results obtained in the first two years of analysis when the farms had problems in terms of animal feeding, the poor programming of inseminations and liters and an unbalanced structure of livestock.

Among the most frequent reproductive disorders are anoestrus, losses of heat cycles, weak estrus unidentified in due time, insemination of cows having vaginal discharge caused by metritis and other disorders, deficient nutrition a.o. (Senosy et al., 2011). Thus, in 2009, the cost of sanitary-veterinary treatments amounted to 48.5 EURO/cow, in 2010 they amounted to 43.70 EURO/cow, and in 2011 they amounted to 32.39 EURO/cow. In the three years (2009 to 2011), expenses amounted to 43,379.23 EURO; results that reflected into the price of the milk produced, and the highest share was noticed in 2009 when losses exceeded 25% of production.

DISCUSSION

The reproductive activity is one of the factors having a strong impact in rendering the milk cow breeder's activity profitable. As highlighted in the literature cited in the results section, breed and age at first calving affected the level of production and once with the increasing production, cow's fertility decreases. As highlighted in the literature cited, we demonstrate that between the age at first calving and milk production was a medium to strong correlation of 47%; regression coefficient was 0.473 with $p < 0.001$ and CI of 95%. Also, we showed that breeding technology and weather affect cow's fertility, and in the year in which the application of biotechnology was used for induction of estrus and ovulation, and estrus synchronization, an improved schedule for calving, and breeding indicators were higher. Thus, by judicious organization of reproduction a higher number of veal's and a higher milk production were achieved, while decreasing treatment necessary costs for genital diseases of cattle or newborn veal's. Also, the application of reproduction biotechnology for this breed, maximize the genetic progress, health and fertility, while reducing the cost price per unit of product.

Reproductive biotechnologies represent the assembly of methods, processes or operations that are used to

obtain, in optimal conditions, some new generations of more and more successful animals. Some bio-technologies include certain bio-techniques such as the off-season induction of estrus and ovulation, induction of polyovulation, estrus synchronization, calving programming, etc. In terms of chronology and the complexity of techniques, they suppose these biotechnologies fall into four generations: artificial insemination, embryo transfer, "*in vitro*" fecundation, embryo cloning and sexing, transgenesis which is in full process of study and the sporadic application in diverse breeds.

Thus, the evaluation and elaboration of some methods and techniques that may help in using the valuable females more efficiently represent a major necessity in the growth and improvement of milk cow livestock.

Conclusions

The reproductive capacity is influenced by a series of genetic factors (breed, individual, close inbreeding, sexual maturity, so on), environmental factors (temperature, humidity, air currents, so on) and technological factors (food: insufficient protein produces disturbances in endocrine activity, sexual anovulatory cycles occur without release of ovules, stabulation, movement, body care and hygiene, monitoring of breeding and so on). The primiparous cows registered the highest losses caused by their physiological state as compared to the adult animals where the reproductive disorders were more reduced.

The economic calculations related to the losses caused by the infecundity-sterility state in the cow farms demonstrate with concrete data that these losses are important and differ from one year to another and from one farm to another depending on the exploitation technological factors and the management of the reproductive function.

The use of artificial insemination and state-of-art biotechnologies in the milk cow farms have transformed animals' reproduction from the organizational viewpoint into a process guided and controlled by people, with the possibility to maximize the genetic progress, the health and fertility state while reducing the cost price per product unit.

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