

VETERINARY SCIENCES

THE REPRODUCTIVE FUNCTION AND THE METABOLIC STATE OF COWS ORGANISM AFTER GIVING AN INJECTION OF A NEUROTROPIC AND METABOLIC PREPARATION IN THE LAST TEN-DAY PERIOD OF PREGNANCY

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Abstract

It has been determined that the regulatory and the biochemical processes in Holstein cow's organisms which received the neurotropic and metabolic preparation Glutam 1 M at a dose of 20 ml on the 265th-267th day of pregnancy were more favourable for the restoration of the reproductive function than in control. Thus, in the blood of cows the amount of estradiol significantly increases, followed by a false decrease in progesterone amount. The lutropin concentration significantly decreases as compared to the false decrease in control, and the amount of follitropin increases. The amount of glucose, inorganic phosphorus, albumen, cholesterol decreased by 11.8%, 11.1%, 10.6%, 13.1% respectively, and the amount of globulins increased by 5.35 ($p < 0.05$) as compared to the 265th day. These changes of the researched ingredients of cow's blood contributed to the reduction of the pregnancy period by 2 days, as well as to the increase in impregnation capacity after the first insemination procedure by 42.8%, and to the reduction of service period and the insemination index by 38.9 days ($p < 0.001$) and by 35% ($p < 0.05$).

Keywords: cows, pregnancy, Glutam 1 M preparation, estradiol, progesterone, lutropin, follitropin, impregnation capacity, service period.

One of the management directions of the female animal's reproductive functions is the use of biologically active preparations with neurotropic and metabolic actions in different periods of the reproductive cycle. The theoretical hypothesis of their application is the fact that the animals reproductive function in the vast majority of cases depends on the effects of the surrounding which stipulates the response of the female animal's reproductive system through the hypothalamo-pituitary axis. The functioning activation of the hypothalamo-pituitary-ovarian axis in a female organism in different periods of the reproductive cycle can facilitate the improvement of its reproductive function. It has been investigated that the preparations effects with metabolic and neurotropic action is revealed in morphofunctionally strained periods of the reproductive cycle. Thus, the ingredients of Glutam 1 M preparation through the intensification of the metabolic processes in nerve tissue and in the cow organism in the period of active functioning of the reproductive system promote to the ovulation as well as to the process of embryo survival in the female uterus.

In the postpartum period in cow organism there are some serious morphofunctional processes in all systems of the organism. First of all the dominant of the labor process is replaced by the lactation one to provide the life-sustaining activity of the descendant who has come to life. At the same time the reproductive system restores as well.

The processes of uterus involution as well as sensibilization of the nerves to estrogens for restoring the regulative role of hypothalamo-pituitary-ovarian system take place. The latter influences the efficiency of the next reproductive cycle. It is more than likely that the preparation to these morphofunctional changes occurs on the basis of the functional correlation relations in the prepartum period. This fact is proved by the re-

search which shows that the introduction of the biologically active preparations such as tetravit in combination with ACDf-2, as well as placenta denatured dispersive, or selenium and vitamin E into the calvers in the period of 10-20 days before the labor improves the next reproductive cycle. These facts testify to the probability of improving the cows further reproductive function through the introduction of the biologically active substances into the calvers in the period of 10-20 days before the labor. That is why looking for such substances and preparations which improve the biological indexes of cow's reproductive capacity after the labor is of great economic and selective importance.

The research goal was to enhance the cow's reproductive capacity and to study the concentration dynamics of sex and gonadotrophic hormones as well as to study some biochemical blood indexes after the introduction of Glutam 1 M preparation during the last 10-days pregnancy period.

The materials and the research methods. The research was conducted at the private farm enterprise "Severtsy" Popilnia district Zhytomyr oblast in winter period under cow's separate enclosure. The materials for the research were the dapple cows of Holstein breed with live mass of 550-650 kg. The test and the experimental groups of 14 cows each were formed to study the effects of Glutam 1 M on the cow's reproductive capacity after it was introduced into the cows at the end of dry period. The cows were chosen according to their age, weight, live mass and the date of insemination. In the experiment a technical engineer artificially inseminated all the tested cows using a rectal-cervical method. The dose contained 15 mln male cells with linear and forward motion. Glutam 1 M was given to the cows of the experimental group by injection under skin near the blade bone on the 265th-267th day of their pregnancy at a dose of 20 ml. The main active substance in Glutam 1 M is glutamate monosodium and the additional one is

physiological solution. The preparation was produced by "Pharmac" enterprise (Kyiv) in accordance with standards 4881: 2007. The preparation is transparent liquid without any taste and flavor. The physiological solution at a dose of 20 ml was given to test animals (table 1). The blood for research was withdrawn from the jugular vein in the morning before feeding and before giving the preparations as well as on the next day after the procedure of giving the preparations was fully completed. The blood serum was withdrawn from the

test tube after the formation of clod. Then the blood serum was centrifuged and frozen in the test tubes of 2 ml in volume in freezers with a temperature regime -18 C. In the laboratories of the departments of obstetricians and surgery of Zhytomyr National Agroecological University blood was studied on the content of: glucose - by glucose-oxidase method; white- by refractometric method; calcium - by complexon Arsenazo; nonorganic phosphorus - with

Table 1.

The scheme of giving the preparation by injection and blood withdrawal in cows.

Indexes	Group	
	Test	Experimental
Amount of animals in which the biochemical blood content and reproductive function were researched	14	14
Amount of animals in which the blood content on hormones was researched	4	4
Preparations, ml	Physiological solution-20	Glutam 1 M 20
Pregnancy days when the preparations were given	265-267	265-267
Pregnancy days when the cows were chosen	269	265, 269

vanadium- molybdenum according to Puls in V. F. Koromyslov and L. A. Kudriavtseva modification; the amount of albumen- by bromsterol green; cholesterol - by Ilko method; triglycerides- by Sadisaem and Manning method. A semiautomatic biochemical analyzer "Rayto 1906 C" was used in the experiments. The amount of sex hormones was determined on gamma counter "Gamma 800" with a set of chemical reagents for radioimmunological determination in vitro of progesterone, estradiol, lutropin and follitropin in cattle at PE "Diagnostica Plus" Zhytomyr.

The results of the research. Glutam 1 M which was given to the cows by injection during three days

starting from the 265th day of their pregnancy promoted to the reduction of pregnancy period by two days. Herewith, the impregnation after the first insemination increased by 42.8%, the service period and the insemination index decreased by 38.9 day ($P < 0.001$) and 35% ($P < 0.05$) respectively as compared to control. The period of expulsion of afterbirth in test animals was longer than in experimental group animals by 11% (table 2).

One of the main grounds for high milk yielding is storing of a significant

Table 2

The reproductive function of test animals.

Indexes	Group, n=14			
	control		experimental	
	x ± S.X.	Cv, %	x ± S.X.	Cv, %
Dry period, days	68.2±1.40	7.98	66.1±1.31	7.66
Pregnancy, days	282.0±1.36	1.91	280.1±1.31	1.81
Expulsion of afterbirth	8.2±0.65	30.6	7.3±0.49	26.1
Calves live mass, kg	38.0±1.0	10.2	38.0±0.86	8.9
Recovering period, days	65.2±5.34	31.7	48.9±4.02*	31.9
Insemination index	2.0±0.27	54.0	1.3±0.12*	36.5
Service period, days	97.9±7.03	29.9	59.0±7.22**	47.4
Impregnation after the first insemination	28.6±12.05		71.4±12.08*	

* $p < 0.05$ ** $p < 0.001$ as compared to the control

amount of nutrients in dry period in a cow organism. They are of great importance in restoring of the reproductive function on the background of intensive lactation especially during the first three months. That is why the duration of a dry period can be a criterion which characterizes the state of nutrients storage in cows before calving. The duration of a dry period depends on the pregnancy duration. Glutam 1 M did not affect considerably the duration of a dry period. It was less by 3.1% than in control.

The second important criteria on which the reproductive capacity depends on is their milk yielding, especially in the first three months of lactation. The com-

parative analyses showed that the reduction of a dry period in experimental cows did not affect the milk yielding capacity. Thus, the milk yields in experimental cows during the first three months were 1922.1±31.01 kg, in contrast to 1860.4±65.31 in control. That is, their milk yields increased by 3.3%. But the difference in milk yields of experimental cows and control ones was within the error, that is why we can consider that the introduction of a preparation did not have negative effects on the lactation intensity.

Change in estrogens and progesterone ratio in the direction of increasing the estrogens level is one of the main moments of pregnancy development. Herewith,

the lutropin concentration decreases because of an increase in the estrogens content.

The progesterone level in cow's blood plasma in the process of a normal pregnancy development reduces. It has been determined that the estradiol and progesterone concentration in the pregnancy period, in time and after calving changes. In 24-72 hours before calving the progesterone amount in cow's blood decreases as a result of synthesis by the amniotic coverings and by turning it into cortisone and estrogen's. In a day after calving the progesterone amount sharply decreased by 6.6 times. According to the data, in two days before calving the progesterone concentration starts decreasing sharply, but it gets low indexes in a few hours after labor.

Based on the above, a certain hormone phone is necessary for the normal development of calving stages. Under the hormone disbalance between progesterone and estrogens a contractive function of plain muscles falls down that results in the offspring detention after calving, weak labor and, as a result, in postpartum complications as well as reproductive capacity worsening. That is why the research of these hormones concentration in calvers blood when using neuthropic and metabolic preparations can be a test on positive or

negative reaction of animal's organism on their introduction.

The analyses of the hormones concentration data determined before giving the preparation by injection testifies to the fact that on the approach to the labor there is a tendency to the reduction of progesterone amount and to the increase in estradiol-17b. Herewith, after giving three injections of glutamin 1 M to the cows the estradiole-17b concentration increased by 54% ($p < 0.01$) and 28.9% as compared to the 265th day of pregnancy and to control on the 269th day. In test animals in the period from the 265th to 269th days the concentrations of lutropin and follitropin decreased by 11.5% and 45.4% ($p < 0.01$) respectively. In the experimental group cows, the concentration of lutropin decreased by 23.5% ($p < 0.05$) and by 16.4% respectively, and the concentration of follitropin increased by 5.8% and by 98.5% ($p < 0.001$) respectively as compared to the 265th day and to the control. So, it can be stated that follitropin continues to stimulate the follicula growth and functional activity. As a result, the estradiol concentration increases. At the same time the lutropin concentration decreases, that is, the support of the yellow body functional activity falls down and the progesterone content decreases (table 3).

Table 3

The amount of sex and gonadotrophic hormones in test cows blood

Hormones	Group, n=4, $\bar{x} \pm S.X.$			
	control		experimental	
	Day of pregnancy			
	265	269	265	269
Progesterone	22.43±1.203	18.51±5.082	22.3±1.09	19.4±2.12
Estradiol	81.52±6.483	142.0 ±43.82	83.4±11.58	181.7± 8.60*
Lutopine, me/l	1.68±0.035	1.49±0.094	1.68±0.058	1.28±0.019*
Follitropin, me/l	1.30±0.133	0.70±0.331**	1.31±0.038	1.39±0.030***

* $p < 0.05$ ** $p < 0.01$ as compared to the 265th day of pregnancy *** $p < 0.001$ as compared to the control

So in cow's blood who received Glutam 1 M on the 265th-267th day of their pregnancy there is an increase in estradiol followed by the decrease in progesterone. Herewith, In the blood of experimental cows the amount of lutropin significantly decreases in contrast to insignificant decrease in control, and the concentration of follitropin increases.

The investigations made by N.H. Fedosova (1994) proved that the six times introduction of ACTH on the 265th-267th day results in the reduction of pregnancy period by 1.9 day. Herewith, the concentration of cortisol and estradiol increases but that of progesterone decreases. In our research in an experimental group the pregnancy duration was shorter and calving occurred earlier as compared to the control. So, we can suppose that the preparation has effects on female hypothalamus, through releasing hormone it stimulates the secretion of ACTH by pituitary that results, firstly, in cortisol synthesis and its rush by the renal gland that stimulates synthesis of estradiol in the placenta and, secondly, together with estrogen it hampers the synthesis and the secretion of lutropin.

The second hypothesis on the increasing of estrogen level in the in the cow's blood after giving Glutam 1 M is based on a probable effect of the preparation on a fetus. The main mechanism provoking labor is known to be an increase in secretion level of fetus corticosteroids which stipulate for an intensive secretion of estrogens and prostaglandins F2a by the tissues of placenta

and uterus that results in the decrease in progesterone level. That is, the increase in estrogen content in the cow's blood can be stipulated by the preparation effect on fetus hypothalamus, that will be followed by the increase in ACTH and glucocorticosteroids content due to releasing hormone.

The results of L.I. Drozdovas (2010) research showed that before calving the placenta synthesizes ACTH. That is why the third hypothesis on the increase in estradiol concentration in the cow's blood after giving them Glutam 1M can be an increase in ACTH production by the placenta before calving, in its turn it promoted to corticosteroids synthesis in renal glands as well as it stimulated the labor.

To determine the effects of Glutam 1M preparation on metabolites changeability it is necessary to consider the fact that blood was taken from each cow on a specific day of pregnancy before giving the preparations and on the next day after the last injection. That is why it is necessary to analyze the changes in the concentration of the indexes among the animals in each group.

In the cow's blood in the experimental group after giving the preparation by the injection the concentration of glucose, phosphorus nonorganic, albumen, cholesterol significantly decreased by 11.8%, 11.1%, 10.6%, 13.15 respectively, and the globulin content increased by 5.35 ($p < 0.05$) as compared to the one on the 265th day (table 4).

Table 4

Blood metabolites of the test cows, $\bar{x} \pm S.X.$, (n=14)

Metabolite	Group		
	control	experimental	
	Day of pregnancy		
	268	265	268
Protein g/l	75.59±0.928	77.13±0.517	76.59±0.612
Albumen g/l	32.77±0/397	38.75±1.514	34.29±1.061 ¹
Albumen %	43.48±0.796	50.08±1.853	44.81±1.431 ¹
Globulin %	56.52±0.796	49.92±1.853	55.19±1.431 ¹
Glucose mm/l	3.16±0.092	3.35±0.065	3.01±0.083 ²
Cholesterol mmol/l	3.23±0.143	3.76±0.152	3.31±0.146 ¹
Triglyceride mmol/l	0.50±0.042	0.45±0.037	0.43±0.026
Phosphorus nonorganic mm/l	1.80±0.052	1.77±0.032	1.58±0.028 ^{3*}
Calcium mmol/l	2.70±0.046	2.52±0.065	2.85±0.064 ²

¹p<0.05; ²p<0.01; ³p<0.001 real between pregnancy days in the middle of a group; * p<0.001 real in the periods between control.

The increase in estradiol concentration in experimental cow's blood also proves to the decrease in cholesterol content. The follitropin increase probably activates the estrogens which are connected with albumen, it resulted in the decrease in its content in blood. The growth in the amount of estrogens intensified the glycolysis. It is proved by the decrease in the concentration of glucose, phosphorus nonorganic and by the increase in the amount of calcium.

Metabolites of glycolysis mostly were included in Krebs cycle for storing the energy-rich compounds, as the amount of glycerides did not increase. The increase in calcium content can be explained by the fact that when the above mentioned processes were stimulated by Glutamin 1 M, its content in blood was at the lowest level 2.5-3 mmol/l. It is well known that calcium content is "one of the perfect constant in the organism". That is why compensation reactions occurred and provided the use of this metabolite without reducing its concentration lower than at a permissible level, and later they prolonged the increase in its reserve in blood.

In such a way the above described regulative and biochemical processes in cow's organism which received the preparation on the 265th-267th day were more favorable for the recovering of the reproductive function than in control due to storing a greater amount of energy-rich compounds.

Conclusions. Glutam 1 M given to cows by injection under skin at a dose of 20ml once a day, during three days starting from the 265th day of pregnancy results in the reduction of pregnancy duration by two days and improves their reproductive capacity: Increases the cow's impregnation after the first insemination by 42.8%; reduces service period as well as insemination index by 38.9 day (p<0.001) and 35% (p<0.05) respectively as compared to control.

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