Effect of herbal feed additives on the quality of colostrum, immunological indicators of newborn calves blood and growth energy of young animals

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Abstract. Studies have shown that the use of herbal feed additives «Fitopank» by Grey Ukrainian cows 30 days before calving had a positive effect not only on the body of cows—mothers and the quality of their colostrum, but also on the physiological state of the born calves and the growth energy of young animals. Adding of «Fitopank» to the cows diet helped to improve the quality of colostrum and its immunological value. It showed a probable increase of total protein by 10–14 % (P < 0.05) and immunoglobulin level by 23–25 % (P < 0.01). The titratable acidity of both the first and second milk yields was 16 % lower (P < 0.05) than in the control group. The use of this feed additive improved the immunological status of newborn calves. In their blood there was an increase of total protein content by 17 % (P < 0.05), the level of gamma globulins by 18% (p < 0.05), IgG concentrations by 14 % (p < 0.05), Ig M by 13 % and Ig A by 17 % (p<0.05), as well as the growth of T-lymphocytes by 46% and B-lymphocytes by 24 % (p < 0.01), compared with control group. Detected increased growth of the 8–12 months bulls (by 2.3%, p < 0.05) of the experimental group may indicate their tendency for better development of meat qualities. Experiment showed that the absolute live weight increase of young animals up to 18 months was higher than the control by 10.9 kg (p < 0.05). The study of the herbal supplements efficacy for the prevention and treatment of animals immunodeficiency, as well as their impact on the metabolic processes and productive qualities of farm animals will be the focus of our further research.

Keywords: beef cattle; feed additives; colostrum; immune status; growth energy.

Вплив рослинного препарату на якість молозива, імунологічні показники крові новонароджених телят та енергію їх росту

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Анотація. Встановлено, що застосування кормової фітодобавки «Фітопанк» коровам сірої української породи за 30 діб до отеплення мало позитивний вплив не тільки на організм корів—матерів і якість їх молозива, а й на фізіологічний стан отриманих від них телят та енергію росту молодняку. Встановлено, що додавання до раціону «Фітопанк» сприяло підвищенню якості молозива та його імунологічної повноцінності. У ньому відбувалося вірогідне зростання загального білка – на 10–14 % (P < 0.05) та рівня імуноглобулінів – на 23–25 % (P < 0.01). Титрована кислотність як першого, так і другого удою була меншою на 16 % (P < 0.01), ніж у контролі. З’ясовано, що використання кормової фітодобавки покращувало імунологічний статус новонароджених телят. В їх крові відмічалось підвищення вмісту загального білка – на 17 % (P < 0.05), рівня гамма-глобулінів – на 18 % (P < 0.01), концентрації IgG – на 14 % (P < 0.01), Ig M – на 13 % та Ig A – на 17 % (P < 0.01), а також зростання T-лімфоцитів – на 46 % і B-лімфоцитів – на 24 % (p < 0.01), порівняно з контролем. Виявлена перевага за відносною швидкістю росту в бугаїв дослідної групи, порівняно з аналогами 8–12-місячного віку (на 2,3 %, p < 0.05), може свідчити про їх схильність до кращого формування м’ясних якостей. Абсолютний приріст у відсотковій розмірці та продуктивні якості сільськогосподарських тварин будуть напрямом наших подальших досліджень.

Ключові слова: м’яча худоба; кормова фітодобавка; молозиво; імунний статус; енергія росту.
Introduction

Newborn diseases are one of the most common diseases in animal husbandry. In particular, despite significant advances in growing and maintaining dairy calves in the United States, one in ten calves die before weaning (Hulbert & Moisá, 2016). For example, in the Netherlands (Sanntan-Berends et al., 2019), calf mortality before 1 year of age is approximately 16.5 %. Dividing the first year of life into three age categories, the death rate for postnatal calves (≤ 14 days) was 3.3 %; early weaned (15 to 55 days) – 4.5% and weaned > 56 days – 3.1 %. Researchers (Voljč et al., 2019) report that about 8 % of prenat al calf deaths were related to the functional condition of cows, while only 4 % to neonatal pathology. In the latter case, the season of birth and the size of the herd had a significant impact. According to other scientists, the death rate of newborn calves was as high as 40 % (Kasap et al., 2019) and it was highest from birth to six months age, as it was associated not only with born calves but also with care and feeding of cows in the dry season. During this period cows may have a high negative energy balance and fatty liver, which affects the health of both mother and calf. The authors inform (Abuelo et al., 2019) that the increased risk of diseases in the prenatal and neonatal periods is a consequence of disturbance of the redox balance in the body and the manifestation of oxidative stress, so the use of additives containing natural antioxidants can prevent animal diseases.

The most important requirements for preventing neonatal pathology are care and feeding, and in particular, keeping the time and term of the first portion of colostrum, as its maternal antibodies are effective in preventing many diseases of newborn calves (Hulbert & Moisá, 2016). Insufficient amount of immunoglobulins synthesized by the fetus is not able to protect it effectively after birth. Immunoglobulins supplied with colostrum remain the only source of passive immunity, so the constant monitoring of its quality will allow reliable transmission of passive immunity to calves (Chemenko et al., 2017). Scientists believe that controlling of colostrum feeding and improving nutrition before heifers weaning will have a positive effect on the mammary gland formation (Vailati-Riboni et al., 2018) and further cow productivity (Makau et al., 2018; Milostivty et al., 2018b), as immunobiological indicators of blood are closely linked with the productive qualities of livestock (Milostivty et al., 2018a; Elshahby et al., 2019).

It is very important to use the means for correcting the immunodeficiency states of productive and young animals. Despite the great arsenal of domestic and foreign medicines, they are not always effective and safe. Therefore, the use of biologically active natural additives of plant origin has become widespread in preventive veterinary practice, but their efficacy requires appropriate research. So, the purpose of the work was to determine the immunological value of cows colostrum, the immune status of newborn calves and the growth energy of young animals effecting by herbal feed supplement «Fitopank».

Material and methods

The study is a fragment of the research «Application of biologically active substances of plant origin for disease prevention, preservation and non-specific immunity» (state registration number 0112U08497) and «Impact of technological factors on the improving of the livestock products quality» (state registration number 0114U005590) Dnipro State Agrarian and Economic University. The experiment was performed on the Polivaniwka cattle farm in Magdalyinivka district, Dnipropetrovsk region. From the total herd of full-grown Grey Ukrainian cows in 30 days before calving, a control (n = 20) and experimental (n = 20) groups of cows-mothers and their calves (respectively n = 7 and n = 6) were formed on the principle of analogues (age, live weight, physiological state). Cows with calves were winter-stalled. All the animals involved in the experiment were clinically healthy. During the experiment, the requirements of the European Convention for the Protection of Vertebrate Animals Used for Experimental and Scientific Purposes (Strasbourg, 1986) and the Law of Ukraine “On the Protection of Animals against Cruelty” No. 3447 of 21.02.2006 were observed. The study material was serum and colostrum.

«Fitopank» feed additive is a complex composition of alcoholic infusions of seven medicinal plants combined in the author’s ratio (Filippov et al., 2002): the root of palmate rhabarb of Tungut (Rheum palmatum var. Tanguticum), dill (Anethum graveeas, L.), root of wild sunflower (Inula helenium L.), root of German iris (Iris germanis L.), poison Hemlock herb (Conium maculatum L.), the leaves of the bogbean (Menwathas Trifoliata L.) and sage (Salvia officinalis). «Fitopank» was given to cows individually at a dose of 1 ml twice a day for 30 days before calving with a rubber bottle, spreading it in 100 ml of water.

The number of erythrocytes and leukocytes in blood were counted by Goryaev’s method; hemoglobin was studied by hemoglobinicyanide method; total protein, total calcium, inorganic phosphorus, reserve alkalinity in serum were determined by chemical analyzer «Stat Fox-1904a» (USA). The level of gamma globulins was determined by the turbidimetric method an concentration of immunoglobulins A, G, M by radial immunodiffusion method of G. Mamicini et al., (1965). The number of T- and B-lymphocytes in the peripheral blood was determined by spontaneous rosette formation. Colostrum composition and its quality were investigated by conventional methods (Tsetkhmistrenko & Kononskyy, 2014). Total collagen immunoglobulin content was determined by Krouse colostrometer (Denmark) using the appropriate scale on the instrument. Growth rates (average daily growth, relative live weight increase and live weight increase coefficient) for different age periods of the young animals were calculated on the basis of monthly weightings using the above methods (Pidpala, 2007).

The obtained data were statistically processed using the built-in mathematical functions of the Office Excel 2007 computer program and the Statistica for Windows 6.0 statistical suite with Student’s t test.

Results

The physiological status of the cows was examined before the application of «Fitopank» feed additive. It was established that the hematologic indices of cows in the control and experimental groups were within the reference values: the number of red blood cells were 5.6 ± 0.09 and 5.7 ± 0.07 T / l, the leukocyte number

### Table 1. Composition of cow colostrum (n = 40, $\bar{X} \pm SE$)

| Indicator                  | 1-st milking | 2-nd milking |
|----------------------------|--------------|--------------|
|                            | control      | control      | experimental | experimental |
| Total protein, g / l       | 185.5 ± 6.46 | 204.6 ± 8.51*| 165.0 ± 1.79 | 189.0 ± 2.03*|
| Immunoglobulins, g / l     | 48.2 ± 1.84  | 59.5 ± 2.25**| 32.0 ± 1.07  | 40.1 ± 2.37**|
| Titrated acidity, °Т        | 52.4 ± 1.41  | 44.0 ± 0.54**| 42.3 ± 0.98  | 35.4 ± 1.02**|

Note: * – p < 0.05, ** – p < 0.01 compared to control.
was 9.1 ± 0.0, 10 and 9.0 ± 0.18 G/l, hemoglobin content was 05.4 ± 2.65 and 103.8 ± 3.96 g/l, total protein was 72.0 ± 1.58 and 71.8 ± 2.16 g/l, total calcium concentration was 19.6 ± 0.6 and 26.1 ± 0.11 mmol/l, inorganic phosphorus was 1.93 ± 0.05 and 2.01 ± 0.09mmol/l, the reserve alkalinity was 53.4 ± 0.96 and 52.7 ± 0.83 % CO₂ respectively. The differences we found between the groups of cows in terms of blood were unreliable and testify to the satisfactory condition of the animals in the dry season.

The use of feed herbal supplements affected the immunological value of cow colostrum. It was found (Table 1) that the content of immunoglobulins in colostrum of the second milk yield decreased by 34 % in the control group and 33 % in the experimental animals compared to the first milk (p < 0.001). However, due to the effect of «Fitopank» in the colostrum of both the first and the second milk, the level of immunoglobulins was higher by 23 and 25 %, than in the control (p < 0.01). Total protein content increased by 10 and 14%, (p < 0.05). The titratable acidity of colostrum was 16 % lower (p < 0.01) than in animals of the control group.

It was found (Table 2) that the application of «Fitopank» for cows before calving had a positive effect on the immunological status of calves in the postnatal period. Live weight is one of the main economically useful traits of animal productivity in beef cattle that characterize growth, development and meat quality of young animals. The analysis of the obtained data (Table 3) shows that the bulls of the experimental groups grew intensively, developed well and in all ages according to the live weight met the requirements of the first class and elite. In the feeding period, differences in live weight of the control and experimental groups were minimal. However, with age, their growth pattern has changed. In particular, the live weight of the bulls in experimental group was by 8.3 and 6.9 kg heavier at 12 and 15 months age, and by 10.5 kg at 18 months (p < 0.05). Overall, the absolute increase in live weight of the experimental animals was higher than the control by 10.9 kg.

An important indicator characterizing the level of life productivity of young animals is the value of the live weight increase over certain periods of time. It should be noted that the average daily increase of experimental bulls during the whole period of rearing was high enough and the changes in keeping and feeding after weaning did not affect the health of the experimental young animals (Table 4).

It is significant that from 8 to 12 months, the average daily increase of experimental bulls was 767–830 gr, so, they effectively used herbal feed additives in this age period. However, the increase in live weight of the experimental group animals was higher than the control by 63.1 g or 8.2 % (p < 0.05). In the future (15–18 months) differences were less significant, but the predominance (7.3 %) of experimental bulls remained.

A more objective and complete dynamics of growth rate is the immunological status of calves in the postnatal period.

Table 2. Immunological indices of ten-day-old calves due to the effects of «Fitopank», $\overline{X} \pm SE$

| Indicator                      | Control group, n = 7 | Experimental group, n = 6 |
|--------------------------------|----------------------|---------------------------|
| Total protein, g / l           | 57.6 ± 2.54          | 67.4 ± 2.28*              |
| Gamma globulins, g / l         | 17.0 ± 0.51          | 20.1 ± 0.75*              |
| Immunoglobulins G, g / l       | 14.0 ± 0.37          | 16.0 ± 0.24*              |
| Immunoglobulins M, g / l       | 1.4 ± 0.07           | 1.8 ± 0.11*               |
| Immunoglobulins A, g / l       | 0.9 ± 0.12           | 1.5 ± 0.13*               |
| T-lymphocytes,%                | 27.5 ± 1.41          | 40.2 ± 2.53**             |
| B-lymphocytes, %               | 7.6 ± 0.26           | 9.4 ± 0.42**              |

Note: * – p < 0.05, ** – p < 0.01 compared to control.

Table 3. Dynamics of live weight of small bulls, kg $\overline{X} \pm SE$

| Age, month | Group                      |
|------------|----------------------------|
|            | control, n = 7 | experimental, n = 6 |
| Newborns   | 28.7 ± 0.45 | 28.3 ± 0.44          |
| 6          | 183.6 ± 0.86 | 184.8 ± 1.02         |
| 8          | 230.5 ± 1.35 | 231.1 ± 1.69         |
| 12         | 324.1 ± 2.04 | 323.4 ± 3.54*        |
| 15         | 385.2 ± 2.18 | 392.1 ± 1.61*        |
| 18         | 436.2 ± 1.42 | 446.7 ± 2.04*        |
| Absolute live weight increase | 407.5 ± 1.98 | 418.4 ± 1.63*        |

Note: * – p < 0.05 compared to control.

Table 4. Average daily increase of bulls live weight, by periods of growth, g $\overline{X} \pm SE$

| Age, month | Group                      |
|------------|----------------------------|
|            | control, n = 7 | experimental, n = 6 |
| 0–8        | 827.1 ± 5.99 | 831.1 ± 7.31        |
| 0–15       | 779.2 ± 2.97 | 795.2 ± 4.01        |
| 0–18       | 742.3 ± 2.93 | 762.1 ± 3.28        |
| 8–12       | 767.2 ± 13.62| 830.3 ± 10.48*      |
| 8–15       | 724.6 ± 9.38 | 754.1 ± 9.91        |
| 8–18       | 674.4 ± 6.51 | 706.9 ± 6.67        |
| 12–18      | 612.6 ± 9.01 | 624.6 ± 9.96        |
| 15–18      | 557.4 ± 12.59| 596.7 ± 15.13       |
given by the rate of relative growth rate and the rate of body weight increasing with age. It was found (Table 5) that the relative growth rate decreased with age and was almost at the same level. Youngsters of all groups grew and developed normally. The obtained data indicate a sufficiently high level of their productive qualities.

At the same time, better growth rate of bulls in the experimental group compared to control group (by 2.3%, p < 0.05) at 8–12 months of age, may indicate better meat quality.

### Discussion

Serum protein studies have been widely used to evaluate the functional status of newborn calves (Kalaeva et al., 2019). Usually determination of the concentration of total protein (STP) and immunoglobulin G (IgG) is a reliable method of determining the effectiveness of passive transfer of calves immunosuppression (Wilm et al., 2018; Gavrilin et al., 2018). However, data on their normal serum content of newborn calves are quite limited, especially in the early days of the postnatal period. Scientists report (Staněk et al., 2019) that the concentration of IgG determined in the serum of newborn calves by radial immunodiffusion averaged 13.7 g/l (with fluctuations in the range of 1.5 to 46.6 g/l) and significantly depended on the breed, herd size and period of year. Guided by the criterion of IgG < 10 g/l, scientists have concluded that 34.6% of calves had passive transfer immunity syndrome (FPT).

The concentration of IgG in the serum of newborns is an important factor in preventing the morbidity and death of calves (Elshohaby et al., 2019) and depends on the quality of colostrum. Citing data from foreign researchers, it is reported (Chernenko et al., 2017) that mortality among calves with serum Ig less than 12 mg/ml 24 hours after birth was 6.8%, and with a blood Ig concentration above 12 mg/ml only 3.3%. It was noted (Wilm et al., 2018) that immediately after birth, before feeding the colostrum, the content of STP and IgG were respectively at the levels of 46.1 and 6.0 g/l. After 24 h after feeding the first portions of colostrum, the concentration was at the level of 58.3 and 22.2 g/l, and on the tenth day it was 57.8 and 16.1 g/l, respectively. Therefore, IgG concentration, compared to its maximum initial serum level (24 h after the first colostrum intake), «decreased» at a rate of 0.69 g/l every day. Using a total protein of < 55 and 60 g/l as a passive immunity failure assessment, researchers (Pempek et al., 2017) found that 7.6–26.8% of all auctioned calves in Ohio (USA) had it. 18.8% of calves had clinical signs of diarrhea, 19.3% had dyspepsia and 30.7% had navel inflammation. But only 0.5% (0 to 1.3%) of calves had respiratory diseases.

It was noted (Ellis et al., 2018) that feeding colostrum with 150 g of IgG provided sufficient production of antibodies to respiratory syncytial virus (PC virus) in the nasal mucosa of calves, and therefore the study of colostrum in a practical environment using a special hydrometer (colostrometer) can be considered as one of the simple and reliable methods of quality control (Chernenko et al., 2017). The existence of a reliable relationship between the content of immunoglobulins of classes G and M in the serum of three-month-old calves with signs of their milk productivity (r = 0.31-0.63; P <0.05), allows to confirm the feasibility of immunological studies of blood for early prognosis of dairy cow productivity (Milostiviy et al., 2018a).

It has been reported (Abuelo et al., 2019) that the colostrum content of immunoglobulins of class G and total protein depended on stress resistance of cows, and indirectly affected the viability of their calves. So, increase of total protein and immunoglobulins in the colostrum of cows by application of the biologically active substances of plant components of «Fitopank», will positively affect the immunological parameters of newborn calves, which is proven by our studies.

It is known (Hulbert & Moisá, 2016) that behavioral responses and stress of calves after weaning may affect their immunity when transferred from individual to group retention. Various stressors at an early age (decoration, castration, etc.) lead to a decrease in passive transmission of immunoglobulins and the development of immunodeficiency. Despite the availability of anesthetics and analgesics, the evidence suggests that they also cause leukocyte function inhibition when used during these painful procedures. Therefore, the use of natural herbal additives for the prevention and correction of immunodeficiency of animals is a promising research area that deserves attention.

Unfortunately, there is limited data in the scientific literature regarding the mechanism of action of many herbal immunotropic drugs. Their immunomodulatory effects are usually associated with the production of cytokines and mediators (Markova, 2004). This is quite justified, as the immune system works by the interaction.

### Table 5. The relative growth rate and the coefficient of live weight increase of bulls, \( \bar{X} \pm SE \)

| Indicator        | control, n = 7 | Group experimental, n = 6 |
|------------------|----------------|---------------------------|
|                  | Relative speed, % |                             |
| 0–8 months       | 155.7 ± 0.66     | 156.4 ± 0.68               |
| 8–12 months      | 33.8 ± 0.63      | 36.1 ± 0.51*               |
| 8–15 months      | 50.3 ± 0.67      | 51.6 ± 0.71                |
| 8–18 months      | 61.7 ± 0.61      | 63.6 ± 0.69                |
| 0–15 months      | 172.3 ± 0.42     | 173.1 ± 0.44               |
| 0–18 months      | 175.3 ± 0.38     | 176.2 ± 0.37               |

| The coefficient of live weight increase |
|----------------------------------------|
| 6                                     | 6.41 ± 0.11      | 6.54 ± 0.11               |
| 8                                     | 8.05 ± 0.16      | 8.18 ± 0.14               |
| 12                                    | 11.31 ± 0.18     | 11.77 ± 0.21              |
| 15                                    | 13.45 ± 0.22     | 13.88 ± 0.24              |
| 18                                    | 15.22 ± 0.25     | 15.82 ± 0.26              |

Note: * – p <0.05 compared to control.
of T-, B-cells, natural killer cells (NK cells), macrophages and mediators (interferons, interleukins, etc.). We are inclined to believe that the improvement of colostrum quality and, as a result of the immunological status of newborn calves due to the effects of «Fitopank», are directly related to the composition of the plants that are part of this feed additive. In particular, about 200 compounds are isolated from rhubarb, including anthraquinones, anthrons, stilbenes, flavonoids, acylglycosides and naps. These components have great pharmacological activity, including antitumor, hepatoprotective, anti-inflammatory, antimicrobial, and analgesic action (Cao et al., 2017). Dill essential oils have antibacterial, antifungal, antioxidant, insecticidal, anti-inflammatory, antispasmodic, and other properties (Chahal et al., 2017). Numerous types of wild sunflower are used alone or as an important ingredient in various drug for the treatment of cardiovascular, respiratory, urinary, central nervous and digestive disorders, for the treatment of skin, liver, fungal and bacterial infections. A number of phytochemicals, including alkaloids, essential oils, flavonoids, terpenes and lactones isolated from herbs of the Inula sorts, explain the traditional use of these plants as medicines (Das et al., 2019). Types of iris are well known as sources rich in isoflavonoids, which have antioxidant and antiinflammatory activity (Ibrahim et al., 2017), capable of exhibiting anti-ulcer and anti-inflammatory properties, having a positive effect in chronic neurodegenerative diseases (Kowalczyk et al., 2017). Bogbean is rich in chemicals (betulinic acids and triterpene saponins) with anti-inflammatory and immunomodulatory effects, and also has antineoplastic properties associated with the induction of apoptosis in neoplastic cells (Kowalczyk et al., 2019). Polyphenols and biologically active compounds in the composition of sage have a therapeutic effect in neurodegenerative diseases and various cancers (Jakovićević et al. 2019). However, some components of «Fitopank» have strong toxic properties. In particular, hemlock causes an intoxication with the hypersalivation, anorexia, hyperthermia, tachycardia, hyperpnea, muscle spasms, unsteady gait, ataxia and hyperreflexia, and even death (Cortinovis & Caloni, 2015). Therefore, the use of herbal feed additives to correct the immune deficiency of the body is a promising research direction. However, the ratio and content of the active components of the plants should be a subject of careful study, as some of them may have strong toxic properties.

**Conclusions**

The effectiveness of herbal feed additives for the prevention and treatment of animals immunodeficiency is very promising scientific research in the field of veterinary medicine. Application of the herbal feed additives «Fitopank» for dry cows helped to increase the level of immunoglobulins in colostrum by 23–25 % (P < 0.01), increase the level of immunoglobulins by 13–17 % (P < 0.05) in serum of newborn calves and T- and B-lymphocytes by 24–46 % (P < 0.01), as well as an increase of relative growth rate of bulls and their absolute growth to 18 months age.

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