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ORIGINAL ARTICLE

Environmental justification for using of active yeast in laying hens diet

O.S. Orishchuk[1], S.V. Tsap[1], O.M. Chernenko[1], L.M. Darmograi[2], O.I. Chernenko[1], V.V. Mykytiuk[1]

[1]Dnipro State University of Agriculture and Economics, 25, S. Efremov St, Dnipro, 49600, Ukraine
[2]Lviv National University of Veterinary Medicine and Biotechnologies named after S.Z. Gzhytskyj, 50, Pekarska St, Lviv, 79010, Ukraine.

E-mail: jrcfyf8588jhsi@i.ua (or) *murolyb@ukr.net*

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The results of the studying of active yeasts *Saccharomy cescerevisiae* influence on the egg-laying capacity and morphological characteristics of the hens "NOVOgen brown" breed eggs are presented. The nutritional intervention of active yeast *Saccharomy cescerevisiae* in laying hens diet allowed to increase egg-laying capacity by 3.7-6.6% and improve the morphological composition of the eggs. However, feed conversion in experimental groups was by 5.1-7.9% lower in relation to the control group.

Keywords: Hens; combined feed; amino acid; *Saccharomy cescerevisiae*; productivity

Introduction

In the system of complete feeding of farm animals and poultry one of the promising directions for the supporting of their nutritional needs in feeding protein is the using of proteins from microorganisms. Such proteins can be equated with animal proteins of biological origin. Today the Ukrainian market is represented by yeast products mostly from foreign producers, while home ones are few. Up to the present moment, experimental data on the using of various foreign products in the feeding of farm animals have been accumulated (Tsap et al., 2016). However, the effectiveness of complex using of home-produced yeast has not been sufficiently studied, which set bounds to the possibility of outspreading the raw material base of protein sources for animals and poultry diet. In Ukraine the traditional feed yeasts are *Saccharomy cescerevisiae* and *Saccharomy cescerevisiae*, however, as it can be seen from the literature, the introduction of them into feed does not ensure its complete balance with biologically active substances (Yegorov & Chernega, 2016; Makarynska & Vorona, 2019).
The composition of yeast includes many vital amino acids, such as arginine, histidine, lysine, leucine, tyrosine, threonine, phenylalanine, methionine, valine, tryptophan. Ashes of yeast contain macro-elements - phosphorus, calcium and sodium and micro-elements - copper, zinc, manganese and cobalt (Halász, 2017). It has been established that the introduction of yeast in diet, especially for young pigs and poultry, increases the productivity of animals and the efficiency of their feeding. Protein from feed yeast at biological value is close to protein feeds of animal origin. Numerous experiments have shown that the using of feed yeast increases the gain by 15-20% and reduces the prime cost of gain by 14-17% (Sakine, 2014). Various scientists have studied the ability of microorganisms to consume nutrients, among them there are preparation of direct and indirect action. The first ones include probiotic preparations based on propionic acid, lactic acid bacteria and Bifidobacterium. Preparations of indirect action include microorganisms that do not belong to the normal microflora of the digestive tract of animals, they are dry feed supplements based on yeast cultures. Application of yeast biomass allows to solve several problems: to increase efficiency of nutrient using, productivity of poultry; to suppress the growth of opportunistic and pathogenic intestinal microflora, stimulate immunity; to contribute to the growth of economic gain of production; to ensure the environmental safety of products (Świątkiewicz et al., 2010).
Thus far in Ukraine the issue of producing quality and safe poultry products, in particular poultry slaughter products, is solved by the introduction of intensive technologies that involve the using of various feed supplements, including vitamins, probiotics, prebiotics, macro and microelements. Despite the fact that the beneficial properties of normal intestinal microflora have been known for over 100 years, the study of probiotics is only developing, the history of its formation covers no more than the 25-year period when it became known that normal intestinal microflora participates in supporting colonization resistance of intestinal mucosa and plays an important role in preventing human and animal diseases. It should be noted that yeast is a worthy alternative to other sources of protein and amino acids, but the choice of yeasts must be done very carefully about their quality, first of all it relates to the biological value of the diet. It has been proved that the using of

active yeast in laying hens feeding has a positive effect on growth, productivity, development of the digestive system, immunological status, which makes it possible to enter this product in farm poultry diets (Barrow, 1992; INATOMI, 2016).
However, researches about the effects of different amounts of active yeast on poultry productivity are insufficient. Therefore, the scientific hypothesis of our research was in fact that the feeding of laying hens with the active yeast *Saccharomy cescerevisiae* in the composition of combined feed could have a positive effect on their egg-laying capacity and morphological parameters of eggs. It was important to study the amino acid and chemical composition of the active yeast *Saccharomy cescerevisiae*.

Materials and methods

Laying hens of breed "NOVOgen brown" were the material for scientific and economic experiments.
According to the methodology (Ibatulin & Zhukors`kiy, 2017) 250 laying hens were selected for the experiment, they were analogues by age, live weight, productivity, clinical state of health. Next hens were divided into five groups of 50 birds in each by random sampling - the 1st group was control, 2nd, 3d, 4th and 5th were experimental.
In scientific and economic experiment laying hens of experimental groups were fed with combined feed containing active yeast in the amount of 0.02, 0.04, 0.06 and 0.08% instead of a similar amount of soybean meal. Laying hens from the control group received a complete combined feed which was manufactured in accordance with DSTU 4120-2002 in the feed processing building.
Scheme of scientific and economic experiment is shown in Table 1.

Table 1. Scheme of scientific experiment.

Group, n=50	**Type of feeding**
Comparative period - 5 days	
1 (control)	Complete combined feed (CF)
2 - experimental	CF+0.02% yeast *Saccharomy cescerevisiae* instead of a similar amount of soybean meal
3 - experimental	CF+0.04 % yeast *Saccharomy cescerevisiae* instead of a similar amount of soybean meal
4 - experimental	CF+0.06 % yeast *Saccharomy cescerevisiae* instead of a similar amount of soybean meal
5 - experimental	CF+0.08 % yeast *Saccharomy cescerevisiae* instead of a similar amount of soybean meal

Diets were specified for energy and nutrients content according to the recommendations (Svezhenczov et al., 2008). For the composition of diet the actual nutrient density of the feed used in the experiment was determined by chemical analysis.
The feeding of experimental hens was carried out twice a day with complete combined feed. The poultry was kept in triple-deck cages. During the study period, which continued 180 days, all groups received complete combined feed mainly from cereal. Troughs of all experimental hen groups were disconnected from the general automatic delivery line and the feed was given by hand. The age of the birds in the scientific and economic experiment was 300 days.
During the experiment the consumption of feed by poultry of each experimental group, the chemical composition and nutritional value of feed, saving livestock, live weight, and productivity were taken into account. Egg production accounting was taken into account daily.
The research of the chemical composition of combined feed and yeast samples was carried out in the Scientific Laboratory of the Zoochemical Analysis in Technology of Feeding and Breeding Animals Department of Dnipro State University of Agriculture and Economics. The determinations were carried out with methods of zootechnical and biochemical analysis (Petuhova et al., 1989): the initial moisture was determined by drying the samples in a humidity desiccator at a temperature of 60-65 °C to a constant mass; hygroscopic moisture was determined by drying samples at a temperature of 100-105 °C to constant mass; raw fibre was determined by the method of Henneberg and Stomann; raw fat was determined by S.V. Rushkovskyi method on the amount of fat-free rest in the Soxhlet extractor using ether as a solvent; raw protein was determined by Kjeldahl method; raw ash was determined by dry ashing by burning the sample in a muffle furnace at a temperature of 450-500°C.
The quality of the eggs was evaluated according to their morphological characteristics. For analysis eggs with a mass as close as possible to the average in the group were taken at the end of each month. The determination was carried out according to the generally accepted method (Fysynyn et al., 1998) according to the following indices: egg mass (weighing on weight scales ВЛКТ-500-М with balance accuracy to 0.1 g); the mass of the shell (weighing on weight scales ВЛКТ-500-М); the thickness of the shell (measured by a micrometer, since the thickness of the shell decreases in the direction from the pointed end to the rounded, thickness measurements were made in three parts of the egg: at pointed and rounded ends and in the middle part); mass of albumen and yolk (weighing on weight scales ВЛКТ-500-М); albumen index (calculated by the formula: the height of the albumen - h was divided into the sum of the large and small diameters of the albumen divided into two), index of yolk (calculated by the formula: the height of the yolk was divided into the sum of large and small yolk diameters divided into two), Haugh unit (calculated according to a special table, which determines the quality of the albumen depending on its height and weight of the egg).

The energy value of eggs is calculated by the formula:

$$EV = \frac{16YM + 2AM}{EM - SM} \times 100$$

EV - energy value in 100 g egg contest, kJ;
YM - yolk mass, г;
AM - albumen mass, г;
EM - egg mass, г;
SM - shell mass, г;
16 - constant of energy in 1 g of yolk;
2 - constant of energy in 1 g of albumen.

Research results

The organization of complete feed for laying hens in the period of experiments provides an opportunity to obtain objective results and justify them logically. The basic diet of the control group was balanced with the main nutrients according to the norms and recommendations for the breed "NOVOgen brown".
Experimental groups received the same diet, but instead of soybean meal, they were fed with dry active yeast *Saccharomy cescerevisiae* . In the 2nd experimental group hens received 0.02%, in the 3d - 0.04%, in the 4th - 0.06% and in the 5th - 0.08% *Saccharomy cescerevisiae* from the mass of combined feed. In the composition of combined feed for laying hens of 1st (control) and 2nd, 3d, 4th and 5th experimental groups the set and the quantity of ingredients were the same, only the amount of soybean meal changed.
Cereals predominate in forage mixture, in particular corn grain - 35.0%, wheat grain - 25.51%, sunflower oil-cake - 15.0%, and soybean meal content varied from 12.92% to 13.00%. To balance the mineral substances and vitamins 10% of the marble chips, 0.2% of the “Miovit” premix, 0.34% of the sodium salt, 0.05% of choline chloride, 0.7% of monocalcium phosphate and 0.2% of lysine and methionine were added to the combine feed.
The chemical composition of combine feed used for feeding of laying hens of control and experimental groups was close and varied slightly in nutritional value.
Characteristics of active yeast *Saccharomy cescerevisiae* consists of dried alive yeast cells of *Saccharomy cescerevisiae* strain CNCM I-1077 (not less than 1*109 CFU/g) encapsulated by fatty acids, which does not contain genetically modified organism. The product is in powder form from beige to light brown color with a slight odor of yeast.
The chemical and amino acid analysis of the active yeast *Saccharomy cescerevisiae* (Table 2) was carried out in the Laboratory of the Scientific Research Center for Biosafety and Ecological Control of the Resources of the Agro-Industrial Complex of the Dnipro State University of Agriculture and Economics.

Table 2. The chemical composition of active yeast (*Saccharomy cescerevisiae*), %.

Index	Active yeast
Saccharomy cescerevisiae	
Total moisture	8.76
Raw fat	0.65
Raw fibre	absent
Raw protein:	
by Kjeldahl method	46.77
by Barstein method	41.64
Non-Protein Nitrogen	5.13
Raw ash	4.77
Exchange energy, MJ/kg RM	362.9

The results of the determination of the chemical composition of active yeast showed that they contain raw protein - 46.77% (by Kjeldahl method) and - 41.64% (by Barstein method), non-protein nitrogen - 5.13%, raw fat - 2.96%.
As a result of the research it was found that the level of exchange energy in 1 kg of active yeast *Saccharomy cescerevisiae* was 362.9 MJ/kg.
The amino acid composition of active yeasts (Table 3) was characterized by the advantage of nonessential amino acids such as glutamic acid (14.5%), aspartic acid (8.09%) and essential amino acid as leucine (9.2%), lysine (8.9%), phenylalanine (8.63%), isoleucine 5.6%), threonine (5.5%) and serine (5.3%).
The high level of glutamic acid in the yeast improves the taste of the feed, which resulted in increased appetite and better in taking of the feed.
The content of key amino acids in yeast *Saccharomy cescerevisiae* in 1 kg was 31.7 g of lysine and 9.5 g of methionine.

Table 3. Amino acid composition of active yeast.

Name of amino acid	Consentration	
	g/kg	**%**
Alanine	23.2	6.5

Arginine	18	5.09
Aspartic acid	28.6	8.09
Glutamic acid	51.4	14.5
Glycine	18.6	5.26
Histidine	6.4	1.8
Isoleucine	19.8	5.6
Leucine	32.5	9.2
Lysine	31.7	8.9
Methionine	9.5	2.7
Phenylalanine	30.5	8.63
Proline	16.6	4.7
Serine	18.7	5.3
Threonine	19.7	5.5
Tyrosine	11.7	3.3
Valine	16.2	4.6

The content of the same essential amino acids in soybean meal, which was replaced from the diet by the above-mentioned supplement, was only 22 g of lysine and 4.5 g of methionine, respectively.
The productivity of laying hens depends not only on complete feeding, but also on the availability and balancing of diet in all nutrients. Feed complete in nutrition improves the quality of eggs and increases the productivity.
From the given data it is seen that the poultry productivity during the experiment in the control group was 5120 eggs, in the 2nd experimental group was 5309; in the 3d experimental group was 5355; in the 4th experimental group was 5468 and in the 5th experimental group was 5381.
Thus, during the entire period of scientific and economic experiment the egg-laying capacity of hens of experimental groups in relation to the control group increased: in the 2nd group by 3.69%; in the 3d group by 4.26%, in the 4th group by 6.63% and in the 5th group by 5.21%, and more eggs were received in the experimental groups from both the primary and the average laying hen.
The live weight of productive poultry is an indicator of the its general state and depends on both the value of the diet and the amount of feed and productivity.
Analysis of the live weight of the hens from experimental groups showed that the live weight of the poultry at the beginning of the experiment in all groups was about 1780-1830 g, i.e., it was practically the same. But at the end of the experiment the live weight of the poultry from the 4th experimental group decreased, indicating a more intense course of metabolic processes in the organisms of experimental hens.
The introduction into the diets of the 2nd, 3d, 4th, 5th experimental groups of the above-mentioned supplement showed that the input of feed for 10 eggs in the control group was to 2.14 kg of feed, while in the experimental groups it was by 5.1-7.9% less compared to the control.
The analysis of egg quality indices of hens from experimental groups (Table 4) showed that the eggs mass of the laying hens from the 3d and 5th experimental groups was 63.4-63.5 g and it was by 0.95-1.1% higher compared to the control group. At the same time, the lowest index of egg mass among experimental groups was observed in the 4th group, which in our opinion occurred as a result of an increase in their egg production.

Table 4. Morphological indices of eggs of laying hens $(n=10, \bar{X} \pm SD)$.

Index	**Group**				
	Control	**Experimental**			
	I	**II**	**III**	**IV**	**V**
Egg mass, g	62.8 ± 1.2	63.1 ± 0.1,5	63.4 ± 0.9	63.0 ± 1,1	63,5 ± 0,8
Albumen mass, g	35.6 ± 0.4	35.5 ± 0.8	35.4 ± 1,1	34.8 ± 0.5**	33,8 ± 0,7***
Yolk mass, g	18.1 ± 0.6	18.2 ± 1,2	18.6 ± 0.4**	18.4 ± 1,1	20,2 ± 0,9***
Shell mass, g	9.1 ± 0.5	9.4 ± 0.7	9.4 ± 0.2*	9.9 ± 0.4***	9,4 ± 0,3*
Albumen index, %	0.08 ± 0.01	0.07 ± 0.02	0.09 ± 0.01	0.10 ± 0.03*	0.09 ± 0.04
Yolk index,%	0.46 ± 0.03	0.47 ± 0.02	0.48 ± 0.05	0.47 ± 0.04	0.45 ± 0.06
Shell thickness, mm	0.30 ± 0.01	0.32 ± 0.07	0.31 ± 0.05	0.33 ± 0.08	0.32 ± 0.04
Haugh unit	80.60 ± 0.64	82.8 ± 0.77***	75.4 ± 0.44***	80.9 ± 0.91	82.1 ± 0.76***
Energy value, kJ	678.8 ± 2.58	673.4 ± 1.85	682.8 ± 5.93***	684.2 ± 2.34***	723.7 ± 1.89***

Note.* - P <0.05; ** - P <0.01; *** - P <0.001 compared with control.

By weight of albumen the advantage was in the 2nd group of laying hens, which received 0.02% *Saccharomy cescerevisiae* in combined feed, and it was 35.5 g, which is higher than in other experimental groups. Despite the fact that in the 5th experimental group where hens received *Saccharomy cescerevisiae* in the amount of 0.08% in the combined feed, the productivity of the poultry was slightly lower, but the egg mass, the yolk mass, the Haugh unit and the energy value were higher by 1.1; 11.6 ($P<0.001$); 1.9 ($P<0.001$) and 6.6% ($P<0.001$) respectively. This, in our opinion, is related to the best using of amino acids from this feed supplement.
The albumen and yolk indices in all experimental groups were practically the same, and if they were slightly lower, it was due to the lower height of both egg albumen and egg yolk.
From the quality parameters of the albumen, Haugh units have the highest connection with its indices, because these indices are determined on the basis of measuring the dense protein. The optimum value of Haugh units for nutritious eggs is 65-87. The research found that the Haugh units in both the control and experimental groups were within the normal range and amounted about 75.4-82.1.
It should be noted that the introduction of dry yeast into combined feed for laying hens also led to an increase in the energy value of eggs. It is subject to poultry from the experimental 5th group, which received 0.08% feed supplement as a part of the diet; the energy value of eggs was 723.7 KJ ($P<0.001$) versus 678.8 KJ, which is more by 6.6% of the control group. And only in experimental 2nd group the energy value of eggs was lower by 0.8% for the control group, in our opinion which is connected with a decrease in the productivity of the poultry in this group.

Discussion
It should be noted that the using of microbial biomass for the enrichment of feed with protein and essential amino acids in the conditions of intensive poultry farming is one of the most important problems in the future. Humanity develops in such a way that it is unlikely to be able to provide people with traditional methods of food producing. Growing microorganisms does not depend on climatic and weather conditions, does not require cropping areas, is responded to automatization. Yeast is one of the advantageous groups of microorganisms to produce protein supplements. The protein content in cells of some yeast strains is from half to two thirds of dry substance. The share of essential amino acids is up to 10% (in soybeans rich in lysine, it contains no more than 6%) (Artemieva & Logvinova, 2018).
The leading egg poultry farms in Ukraine have known the secret of high organoleptic qualities of eggs - it is the adding to the diet 3% carotenoid or selenium-containing yeast of the combined feed during and the peak of ovipositioning. Studies show that a poultry of intensive breeds, that have been consuming combined feed with yeast for a long time, has stable habitus and continued period of ovipositioning (Utterback et al., 2005, Lu et al., 2019).
The vitamin background, created in the body due to the yeast supplement, contributes to the enrichment of egg yolk with a complex of vitamins B and carotene in an optimal ratio. This ensures a convincing increase in the mass and fertility of eggs, an increase in the percentage of hatchability and aligning brood (Rodríguez-Navarro et al., 2002).
Thus, probiotic preparations in the form of feed supplements are becoming more usable in foddering animals and poultry for both with therapeutic and prophylactic purposes. The market of this group preparations develops actively daily and is filled with samples of home and foreign production.

Conclusion
Introduction of active yeast *Saccharomy cescerevisiae* to diet of laying hens allowed to increase egg-laying capacity by 3.7-6.6%. Adding a probiotic supplement to the laying hens diet influenced positively over the morphological composition of the eggs. Thus, in experimental groups of laying hens the mass of eggs increased by 0.3-1.1%, yolk mass by 0.5-11.6% and shell mass by 3.3-8.8% compared with the control group.
The increase of amount of active yeast in the combined feed up to 0.08% in the 5th experimental group was accompanied by an increase in the energy content of eggs to 723.7 versus 678.8 kJ in the control group, which is higher by 6.6%.

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