THE EFFECT OF TOASTED *ADENANTHERA PAVONINA* SEED MEAL ON HAEMATOLOGY AND BLOOD CHEMISTRY OF

FINISHER BROILER CHICKENS

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**Abstract:** The effect of graded levels of toasted *Adenanthera pavonina* seed meal (TAPSM) on haematological and biochemical indices of finisher broiler chickens was investigated in a five-week feeding trial with 84 (5-week old) broilers divided into four groups of 21 birds per group. Each group had three replicates of 7 birds. Four experimental diets were formulated to contain 0, 10, 20 and 30% TAPSM, represented as T1, T2, T3 and T4 respectively. Birds were allocated to the four diets in a completely randomized design and housed in a deep litter pen. Feed and water were offered to birds *ad libitum*. Haematological parameters showed a significant (P<0.05) decreasing trend in haemoglobin (10.20, 9.03, 8.60 and 7.63 g/dl), packed cell volume (28.33, 26.10, 25.60 and 25.20%), total red blood cell (4.23, 4.00, 3.99 and 3.99 ×106/µl), mean corpuscular volume (66.97, 65.25, 64.16 and 63.15 fl), mean corpuscular haemoglobin (24.11, 22.57, 21.55 and 19.12pg) and mean corpuscular haemoglobin concentration (35.66, 34.59, 33.59 and 30.27%) with an increase in the level of TAPSM in the diets. Total white blood cells (71.76, 73.40, 75.07 and 76.17 ×103/µl) increased significantly (P<0.05) as the TAPSM level increased. The other values were as follows: urea (4.00, 5.00, 6.67 and 10.00mg/dl), creatinine (0.33, 0.25, 0.43 and 0.46mg/dl), cholesterol (149.67, 135.67, 113.67 and 102.67 mg/dl), total protein (3.33, 3.43, 2.97 and 2.50mg/dl), glucose (146.14, 208.49, 179.66 and 135.33 mg/dl), alkaline phosphatase (105.65, 111.2, 132.67 and 145.00 iu/*l*) and aspartate transaminase (68.68, 70.00, 78.00 and 85.67 iu/*l*). In conclusion, 10% TAPSM in broiler diets most favourably influenced haematological and biochemical parameters.

**Key words:** *Adenanthera pavonina*, toasted, finisher broilers, haematology, blood chemistry.

**Introduction**

There is a need for readily available, high quality alternative plant proteins that are inexpensive and capable of reducing production cost of meat and other animal products. Intensive poultry farming in Nigeria has been greatly affected by high cost of feeds and feed ingredients, especially the protein and energy sources like soybean, groundnut cake and maize (Ani and Adiegwu, 2005). This has been blamed on the competition between man, livestock and industries on conventional feedstuff (Akinmutimi, 2004). The importance of nutrients in intensive poultry production is evidenced by the fact that 75‒80% of production cost is due to feed (Iheukwumere, 2008).

A common solution cost is by using locally available and cheap sources of feed ingredients, particularly those that do not involve competition between humans and livestock. One of the problems with the use of legumes is the presence of anti-nutritional factors (ANF). *Adenanthera pavonina* seeds have been reported to contain anti-nutrients such as taninns, phenols, cyanogenic glycosides, saponins, trypsin inhibitors and haemagglutinins (Umezuruike, 2006). Most of the processing methods employed to improve the feed value of alternative feedstuff do not completely eliminate the anti-nutrients, but only reduce their concentration to a tolerable level in the feedstuff (Akinmutimi, 2004).

*Adenanthera pavonina*,commonly called coral bean tree or red sandal wood, belongs to the family *Fabaceae*, sub-family *Mimisoideae*. The seed of *Adenantherapavonina* contains appreciable amounts of protein, crude fat and minerals compared to commonly consumed staples (Ezeagu et al., 2004). It has also been reported that 25% of the seed weight is oil, with a high digestibility in animals and humans. The roasted seeds are eaten by humans while the leaves are used as fodder for animals. Apart from that, it is also used for medicinal, timber and ornamental purposes (Orwa et al., 2009). There is an active search for alternatives to conventional feed raw materials in animal feeding. This includes evaluation of leguminous seeds (Camara et al., 2003; Kwari et al*.,* 2011; Emiola et al., 2013;Agbabiaka et al., 2013; Ukpabi et al., 2015), leaf meal (Esonu, et al., 2006; Madubuike and Ekenyem, 2006; Ukpabi et al., 2009; Obikaonu et al., 2012; Onunkwo and George, 2015;) and by-products from food and ethanol industries (Amaefule et al., 2006; Alu et al., 2013; Adeyemo and Sani, 2013). The raw seed of *Adenantherapavonina* had been reported to negatively affect the haematological and serum biochemistry of finisher broilers at levels exceeding 5% in the diet (Ukpabi et al*.,* 2015).

Haematological and biochemical indices are essential indicators of health status in animals and have been indispensable in the diagnosis and treatment of many diseases (Emiola et al., 2013). Haematology and blood chemistry are also important tools for assessing the quality of feed and the health status of animals that are placed on experimental diets (Merck, 2010). Thus, the aim of this study was to examine the effect of toasted *Adenantherapavonina* seed meal (TAPSM) on the haematology and blood chemistry of finisher broilers.

**Materials and Methods**

Experimental site

The experiment was carried out at the Livestock Unit of Faculty of Agriculture Teaching and Research Farms, Abia State University, Umudike Campus, Nigeria. Umudike has coordinates of 7°31¹ East and 5°28¹ North, and lies at an altitude of 122 meters above sea level (Adiele et al., 2005).

Procurement of feed ingredients

*Adenanthera pavonina* seeds were gathered from the premises of the Abia State University, Umudike Location and the National Institute for Horticulture (NIHORT), Mbato in Okigwe, Imo State. Other feed ingredients were purchased from a commercial feed shop in Umuahia.

Processing of *Adenanthera pavonina* seeds

The raw seeds were sorted and toasted for 15 minutes at a temperature range of 60‒65°C, and the timing started immediately when the seeds were poured into the toaster. The *Adenanthera pavonina* seeds were cooled before being milled with a hammer mill. The toasted APSM was then weighed according to formulation before being incorporated in the diets.

Experimental design

A total of eighty-four, 5-week old Anak broiler chickens were used for the experiment. The birds were divided into four groups of twenty-one birds each and assigned to the four treatment diets in a completely randomized design (CRD). Each group was further sub-divided into three replicates of seven birds each. Feed and water were offered *ad libitum* and the feeding trial lasted 5 weeks.

Experimental diets

Four experimental diets were formulated by incorporating TAPSM at 0, 10, 20 and 30% dietary levels for T1, T2, T3 and T4, respectively. The gross composition of the experimental diets is shown in Table 1. The experimental diets and TAPSM were subjected to proximate analyses following the procedure of A.O.A.C. (2006) and are presented in Table 2.

Table 1.Composition of the experimental diets (%).

|  |  |
| --- | --- |
| Ingredients | TAPSM inclusion levels in diets (%) |
| T1 (0) | T2 (10) | T3 (20) | T4 (30) |
| Maize | 50.00 | 45.00 | 40.00 | 35.00 |
| Soybean meal | 25.00 | 20.00 | 15.00 | 10.00 |
| TAPSM | 0.00 | 10.00 | 20.00 | 30.00 |
| Blood meal | 2.00 | 2.00 | 2.00 | 2.00 |
| Palm kernel cake | 14.00 | 14.00 | 14.00 | 14.00 |
| Fish meal | 4.00 | 4.00 | 4.00 | 4.00 |
| Palm oil | 1.00 | 1.00 | 1.00 | 1.00 |
| Bone meal | 3.00 | 3.00 | 3.00 | 3.00 |
| Premix\* | 0.30 | 0.30 | 0.30 | 0.30 |
| Salt | 0.50 | 0.50 | 0.50 | 0.50 |
| Methionine | 0.10 | 0.10 |  0.10 | 0.10 |
| Lysine | 0.10 | 0.10 | 0.10 | 0.10 |
| Total | 100.00 | 100.00 | 100.00 | 100.00 |
| Calculated composition |
| Crude protein (%) | 23.03 | 23.37 | 21.71 | 21.05 |
| ME (MJ/kg) | 12.43 | 12.02 | 11.61 | 11.19 |

\*Vitamin mineral premix provides per kg diet: vit. A, 13.340 iu, vit. D3 2680 iu, vit. E iu, vit. K, 2.68 iu, Calcium panthonate, 10.68 mg, vit. B12 0.022 mg; Folic acid, 0.668 mg; Choline chloride 400 mg; Chlorotetracycline, 26‒28 mg; Manganese, 133.34 mg; Iron, 66.68 mg; Zinc, 53.34 mg Copper, 3.2 mg; Iodine, 1.86 mg; Cobalt, 0.268 mg; Selenium, 0.108 mg. ME = Metabolizable Energy (MJ/kg), calculated according to Pauzenga (1985) as ME (MJ/kg) = 37 × % CP + 81 × % EE + 35.5 × % NFE (Folorunso et al., 2016) TAPSM = Toasted *Adenantherapavonina* seed meal.

Table 2. Determined proximate composition of experimental diets and toasted *Adenanthera pavonina* (L) seed meal (TAPSM).

|  |  |  |
| --- | --- | --- |
| Parameter | TAPSM inclusion levels in diets (%) | TAPSM |
| T1 (0) | T2 (10) | T3 (20) | T4 (30) |
| Dry matter | 94.16 | 94.18 | 94.20 | 94.20 | 92.00 |
| Crude protein | 21.05 | 20.04 | 20.02 | 20.00 | 20.08 |
| Crude fibre | 11.00 | 11.16 | 13.04 | 15.00 | 14.00 |
| Ether extract | 9.12 | 8.66 | 8.65 | 8.16 | 9.04 |
| Ash | 9.52 | 9.04 | 8.56 | 8.51 | 3.10 |
| NFE | 43.47 | 45.28 | 44.16 | 42.92 | 45.78 |
| ME (MJ/kg)\* | 12.84 | 12.79 | 12.77 | 12.84 | 13.00 |

TAPSM = Toasted *Adenanthera pavonina* seed meal; NFE = Nitrogen-free extracts; \*ME = Metabolizable energy, calculated according to Pauzenga (1985) as ME (MJ/kg) = 37 × % CP + 81 × % EE + 35.5 × % NFE (Folorunso et al., 2016).

Blood sample collection and analysis

At the end of the experiment, one bird per replicate was randomly selected making a total of 12 birds. Blood samples (7.0 ml) for analysis were collected from under the wing veins of birds using 10-ml plastic disposable syringes for which 2ml were collected into a bijour bottle treated with ethylene diamine tetra acetic acid (EDTA) for the haematological assay (packed cell volume, haemoglobin, white blood cell and red blood cell) and 5mls of blood each were collected into EDTA-free bottles for serum biochemistry (total protein, blood glucose, blood urea and serum enzymes). Haematological indices were determined by the methods of Jain (1986). These are the Wintrobe’smicrohaematocrit kit for packed cell volume (PCV), the cyanmethaemoglobin method for haemoglobin (Hb), an improved Neubaurhaemacytometer for red blood cells (RBC) and white blood cells (WBC) while mean corpuscular volume (MCV), mean corpuscular haemoglobin (MCH) and mean corpuscular haemoglobin concentration were calculated from PCV, RBC and Hb. Standard methods of serum separation were used to obtain sera which were used to determine urea, creatinine, total protein, glucose, cholesterol and serum enzymes (Kohn and Allen, 1995)

Data analysis

Data obtained were subjected to statistical analysis using a one-way analysis of variance (ANOVA) as outlined in Steel and Torrie (1980). The Duncan’s multiple range test was used to separate significant treatment means where they occurred (Obi, 1990).

**Results and Discussion**

The haematological parameters of finisher broilers fed diets containing different levels of TAPSM are presented in Table 3. The values for total red blood cell, total white blood cell, Hb, PCV and MCH showed significant (P<0.05) differences among treatment means.

The PCV ranged from 28.33% in T1 (0%) to 25.20% in T4 (30%). The values of PCV obtained fall within the recommended range of 23‒55% for healthy birds (Maxwell et al., 1990; Banerjee, 2005). The results of PCV in this study were in line with the results of Alibi et al. (2011) who reported 25‒29.00% PCV in finisher broiler chickens. Low PCV in birds is an indication of iron deficiency (Iyayi et al., 2006). Since the values obtained were normal for healthy birds, it suggested that the experimental birds were not anaemic.

The WBC values obtained from this study ranged from 71.76 (x103/µl) in T1 to 76.17 (x103/µl) in T4. There were significant (P<0.05) increases in the WBC as the level of TAPSM increased in the diet. A similar trend of a significant increase (P<0.05) in WBC with an increase in the level of *O. gratissium* in broiler chickens has been reported (Odoemelam et al., 2014). The increase in WBC as inclusion of TAPSM increased in the diet shows that the principal function of phagocytes which is to defend against invading microbes will be enhanced (Adedapo et al., 2012) or that the birds were reacting to inflammatory conditions resulting from the diets (Agbalaya et al., 2017).

Table 3. Haematological parameters of finisher broilers fed graded levels of toasted *Adenanthera pavonina* seed meal.

|  |  |  |
| --- | --- | --- |
| Parameter | TAPSM inclusion levels in diets (%) | SEM |
| T1 (0) | T2(10) | T3 (20) | T4(30) |
| PCV (%) | 28.33a | 26.10b | 25.60c | 25.20c | 3.52 |
| WBC (x103/µl) | 71.76b | 73.4b | 75.07a | 76.17a | 4.25 |
| RBC (x106/µl) | 4.23a | 4.00a | 3.99b | 3.99b | 0.85 |
| Hb (g/dl) | 10.20a | 9.03b | 8.60c | 7.63d | 0.44 |
| MCV (fl) | 66.97a | 65.25a | 64.16b | 63.15b | 0.86 |
| MCH (pg) | 24.11a | 22.57b | 21.55c | 19.12d | 0.70 |
| MCHC (%) | 35.66a | 34.59b | 33.59c | 30.27d | 1.55 |

a, b, c, d Means in the same row with different superscripts differed significantly (P<0.05). TAPSM – Toasted *Adenanthera pavonina* seed meal, SEM – The standard error of the means, PCV = Packed cell volume, WBC = White blood cells, RBC = Red blood cells, Hb = Haemoglobin, MCV = Mean corpuscular volume, MCH = Mean corpuscular haemoglobin, MCHC = Mean corpuscular haemoglobin concentration.

Red blood cell values for birds on T1 were significantly (P<0.05) different and higher than those on diets T2, T3 and T4. The red blood cells (RBCs) are responsible for the transportation of oxygen and carbon dioxide in the blood, hence higher values indicate a greater potential for this function and a better state of health (Olugbemi et al., 2010). A low value of RBC observed in birds fed diets containing 30% TAPSM may be attributed to a high level of TAPSM in the diet or due to low quality of feed and protein deficiency (Awoniyi et al., 2000). The values for RBC obtained ranged from 4.23 (×106/µl) in T1 to 3.99 (×106/µl) in T4. The values obtained in this study were within the normal reference values of 2.0–4.0 reported for chickens (Banerjee, 2005).

The haemoglobin values obtained ranged from 7.63g/dl in T4 to 10.20g/dl in T1. Treatment T1 had the highest value which was significantly (P<0.05) different from T2, T3 and T4. Haemoglobin values decreased as the level of TAPSM in the diet increased. The results fall within the recommended haemoglobin concentration of 7‒18.6g/dl for healthy birds (Pellet and Young, 1980). A reduction in the haemoglobin values obtained in birds fed diets 3 (20% TAPSM) and 4 (30%TAPSM) indicated a possibility of poorer transportation of oxygen from the respiratory organs to peripheral tissues and carbon dioxide for excretion (Murray, 2009).

Mean corpuscular volume (MCV) values obtained were: 6.97 fl for T1, 65.25 fl for T2, 64.16 fl for T3 and 63.15 fl for T4. There was no significant (P>0.05) difference between T1 and T2 but they differed significantly (P<0.05) from T3 and T4 which were similar (P>0.05). The values obtained were higher than the values reported in literature (Oko et al., 2011).

Mean corpuscular haemoglobin (MCH) values showed significant (P<0.05) differences among the treatments. The values obtained decreased as the level of TAPSM in the diets increased from 0 to 10%. Values of MCH in this study were below the range of 53‒97 pg reported in literature (Orwa et al., 2009). Since MCH is an indicator of the oxygen carrying ability of the RBC (Ugwuene, 2011), the blood of the birds fed diets 1 (0%) and 2 (10%) may be more efficient than the blood of others in performing respiratory functions.

The mean corpuscular haemoglobin concentration (MCHC) values were 35.66% for T1, 34.59% for T2, 33.59% for T3 and 30.27% for T4. The values decreased significantly (P<0.05) as the level of TAPSM increased in the diet, which is an indication of the presence of anti-nutrients in the experimental diets, which invariably had an adverse effect on blood formation.

Biochemical parameters of finisher broilers fed graded levels of TAPSM are presented in Table 4. Serum biochemistry is a generalized medium of assessing the health status of animals (Frandson, 1981). There were significant differences (P<0.05) in the biochemical parameters measured in this study.

Table 4. Biochemical parameters of finisher broilers fed diets containing graded levels of TAPSM.

|  |  |
| --- | --- |
| Parameter | Level of inclusion of TAPSM (%) |
| T1 (0) | T2 (10) | T3 (20) | T4 (30%) | SEM |
| Urea (mg/dl) | 4.00b | 5.00b | 6.67b | 10.00a | 1.86 |
| Creatinine (mg/dl) | 0.33b | 0.25c | 0.43a | 0.46a | 0.04 |
| Cholesterol (mg/dl) | 149.67a | 135.67b | 113.67c | 102.67d | 9.87 |
| Total protein (mg/dl) | 3.33a | 3.43a | 2.97b | 2.50c | 0.15 |
| Glucose (mg/dl) | 146.14b | 208.49a | 179.66a | 135.33b | 8.53 |
| ALP (iu/*l*) | 105.65d | 111.20c | 132.67b | 145.00a | 2.85 |
| AST (iu/*l*) | 68.67c | 70.00c | 78.00b | 85.67a | 3.99 |

a, b, c, d Means in the same row with different superscripts differed significantly (P<0.05). TAPSM – Toasted *Adenanthera pavonina* seed meal, SEM – The standard error of the means, ALP – Alkaline phosphatase, AST – Aspartate transaminase.

The values obtained for urea in T1, T2 and T3 were similar (P>0.05), but differed significantly (P<0.05) from T4. High blood urea concentrations recorded in the birds as the level of TAPSM in the diets increased and as in T4 may be due to the protein quality of TAPSM (Kecceci et al., 1998).

Creatinine is a waste product of muscle metabolism and a good measure of kidney function (Ologhobo et al., 1993). Creatinine values ranged from 0.46mg/dl in T4 to 0.25mg/dl in T2 with T4 having the highest value. The values of creatinine obtained in the present studies were lower than 1‒2mg/dl reported for chickens (Reece and Swenson, 2004), which therefore suggested that there was no muscle wastage and birds did not survive at the expense of body reserves (Akinmutimi, 2004).

Cholesterol values ranged from 149.67mg/dl in T1 to 102.67mg/dl in T4. The treatments differed significantly (P<0.05), the values obtained in birds fed diets T1 (0%TAPSM) and T2(10% TAPSM) were within the normal range of 125‒200mg/dl reported for healthy birds (Bolu and Adelakun, 2013) while the values obtained in birds fed diets T3 and T4 were below the normal range of 125‒200mg/dl reported for healthy birds. *Adenantherapavonina* seed meal contains high crude fibre and this factor may have accounted for the depressed cholesterol level obtained in birds fed diets T3 and T4. Intakes of 9‒16.5g/day of varieties of soluble fibres have been shown to produce net reductions in serum cholesterol levels in humans (Reece and Swenson 2004), and may also be applicable to birds.

Total protein values ranged from 3.43mg/dl in T2 to 2.50mg/dl in T4. There were significant (P<0.05) differences among the treatment means. The values obtained in T1 and T2 were within the normal range (4.0–5.2mg/dl) reported for chickens (Reece and Swenson, 2004). Since total protein is usually a reflection of the protein quality fed, it would seem that the significant decrease with an increase in the level of TAPSM is an indication of the presence of a poor quality protein and poor protein utilization of test diets possibly caused by anti-nutritional factors in TAPSM.

Glucose values ranged from 135.33mg/dl in T4 to 208.49mg/dl in T2. The values recorded fall within the recommended range of 130‒270mg/dl for chickens (Reece and Swenson, 2004; Bolu and Adelakun, 2013). The values were also in line with the recommended range of 230‒250mg/100ml for birds (Rubin, 2011). The values obtained differed significantly (P<0.05) between treatments. Generally, birds can maintain a high and relatively constant blood sugar level even in low feed intake (Olomu, 1995)

Serum enzyme activities have been used as indices of toxicity as well as for monitoring protein quality. Alkaline phosphatase (ALP) values ranged from 145.00 iu/l in T1 to 105.65 iu/l in T4. There were significant (P<0.05) differences among the treatment means. The values increased as the level of TAPSM increased in the diets. ALP is predominantly found in the liver, kidney, intestine and placenta (Shipman et al., 2013). Aspartate transaminase (AST) values ranged from 68.67 iu/l in T1 to 85.676 iu/l in T4. The values increased significantly (P<0.05) among treatments, which is an indication that TAPSM had a negative effect on the functioning of the liver and the kidney (Tennant, 1997).

**Conclusion**

The results obtained from this study indicated that finisher broilers could tolerate the 10% TAPSM inclusion level in the diet without any negative effect on the haematology and serum biochemistry. Feeding of TAPSM to broiler chickens is safe as no mortality was recorded throughout the period of the experiment. *Adenanthera pavonina* seed toasted for fifteen minutes at 60‒65oC did not satisfactorily eliminate the anti-nutrients in the seeds. Increased values of alkaline phosphatase in the blood across treatments suggested an increased activity of the liver due to the presence of toxic factors.

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UTICAJ SAČME TOSTIRANOG SEMENA BILJKE *ADENANTHERA PAVONINA* NA HEMATOLOŠKE I BIOHEMIJSKE PARAMETRE BROJLERA U ZAVRŠNOM TOVU

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R e z i m e

Ispitivan je uticaj različitih količina tostiranog semena biljke *Adenanthera pavonina* (engl. *toasted Adenanthera pavonina seed meal*‒ TAPSM) na hematološke i biohemijske indekse brojlera, u petonedeljnom ogledu sa 84 pilića (starih pet nedelja) raspoređenih u četiri grupe po 21 jedinke. Svaka grupa je imala tri ponavljanja sa 7 brojlera. Četiri ogledna obroka bila su formulisana tako da sadrže 0, 10, 20 odnosno 30% TAPSM, koji su predstavljeni kao T1, T2, T3 odnosno T4. Po modelu potpuno slučajnog plana, predviđena su četiri obroka za ptice, koje su bile smeštene u objektu sa dubokom prostirkom. Hrana i voda su pilićima bili ponuđeni *ad libitum*. Hematološki parametri su ukazali na značajan (P<0,05) pad hemoglobina (10,20, 9,03, 8,60 i 7,63 g/dl), hematokrita (28,33, 26,10, 25,60 i 25,20%), ukupnih eritrocita (4,23, 4,00, 3,99 i 3,99 ×106/µl), prosečne zapremine eritrocita (66,97, 65,25, 64,16 i 63,15 fl), prosečne mase hemoglobina po eritrocitu (24,11, 22,57, 21,55 i 19,12pg) i srednje koncentracije hemoglobina u eritrocitu (35,66, 34,59, 33,59 i 30,27%) sa porastom nivoa TAPSM u obrocima. Ukupni leukociti (71,76, 73,40, 75,07 i 76,17 ×103/µl) povećavali su se značajno (P<0,05) kako se nivo TAPSM povećavao. Druge vrednosti su bile kao što sledi: urea (4,00, 5,00, 6,67 i 10,00 mg/dl), kreatinin (0,33, 0,25, 0,43 i 0,46mg/dl), holesterol (149,67, 135,67, 113,67 i 102,67 mg/dl), ukupni proteini (3,33, 3,43, 2,97 i 2,50mg/dl), glukoza (146,14, 208,49, 179,66 i 135,33 mg/dl), alkalna fosfataza (105,65, 111,2, 132,67 i 145,00 iu/*l*) i aspartat transaminaza (68,68, 70,00, 78,00 i 85,67 iu/*l*). Da zaključimo, 10% TAPSM u obrocima za brojlere najpovoljnije je uticao na hematološke i biohemijske parametre.

**Ključne reči:** *Adenanthera pavonina*, tostiran, brojleri u završnom tovu, hematološki parametri, biohemijski parametri.

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