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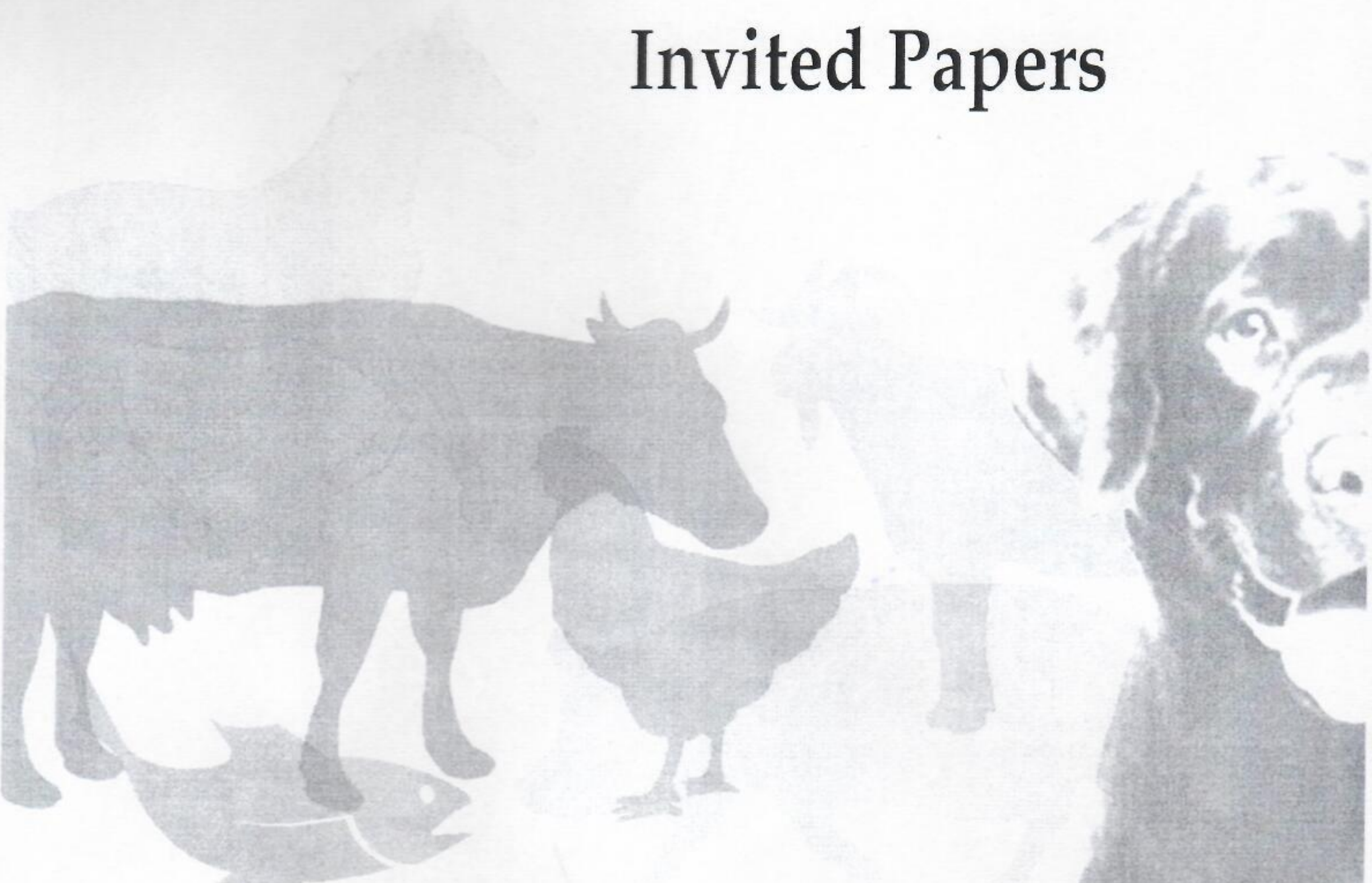

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Editors:
Asit Das
Madhu Mohini
Indranil Samanta
M. C. Pakhira
Srinibas Das
G. P. Mandal

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Tel. 9831161961
Email: indranil_roy1@yahoo.co.in

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Effects of Different Dietary Lignocellulose Levels on Growth and Litter Quality of Broilers

D. Šefer[1]*, S. Katoch[2], R. Marković[1], S. Radulović[1], D. Perić[1]* and L. Makivić[3]

[1]Department of Animal Nutrition and Botany, Faculty of Veterinary Medicine, University of Belgrade, Serbia

[2]Department of Animal Nutrition, CSKHP Agricultural University, Palampur India

[3]Feed Factory, Rapić, Gradiška, Bosnia and Herzegovina

ABSTRACT

The use of raw fiber in poultry nutrition formula is often a controversial topic among many experts involved in the diet of non-ruminants.Purified lignocellulose represents a pronutritive substance that affects the viscosity of the intestinal content, increases the absorption of nutrients and reduces the number of pathogenic bacteria in small intestine. In this experiment the effects of lignocellulose in poultry nutrition was studied. Trial included 384 broilers of Cobb 500 provenance, both male and female, divided into four groups (control group: C and three experimental groups: E-I, E-II and E-III), 96 animals in each. Animals were fed with standard feed mixtures, starter (from 1st to 13th day), grower (from 14th to 28th day) and finisher (from 29th to 42th day), according to the manufacturer's recommendation. A control group (C) was fed without source of lignosellulose. Diet for experimental groups (starter and grower) contained a commercial preparation of purified lignocellulose (Arbocel® R, J. Rettenmaier & Söhne GmbH + CO. KG, Rosenberg, Germany). Preparation was added in the amount of 4 g/kg of feed for the E-I group, 6 g/kg of Arbocel® R as an expense of 0.3% soybean meal and 0.3% maize was added for the E-II group and 6 g/kg of Arbocel® R as an expense of 0.6% soybean meal was added for the E-III group. Analyzing the entire trial period (from 1st to 42th day), adding the lignocellulose in experimental E-II group resulted in the best production indicators (final body weight 2611.00 g, average daily feed intake 96.09 g, average weight gain 2569.29 g and feed to gain ratio 1.67) as well as the best litter quality (moisture content 21,98±1,67%). Based on the obtained results, it can be concluded that the use of lignocelluloses in broilers nutrition has its medical, nutritional and economical justification.

Keywords: Broilers, Lignocellulose, Addition, Production results

1. Introduction

Thanks to numerous discoveries and application of science in the last twenty years, great progress has been made in the development and improvement of livestock production world wide. There is no branch where such progress has been made as in livestock. Undoubtedly, these were accompanied by appropriate changes in the system of keeping and feeding of the poultry. Compared to other types of meat, chicken meat has a relatively low cost, as well as acceptance by all cultures and religions, which makes chicken products acceptable, desirable and appropriate in daily human consumption. Large specialized poultry farms had an important role in the rapid development of poultry production, the growing interest of producers for this type of fattening through cooperation, as well as the favorable price of broiler meat, relative to the prices of other types of meat (Jovanovic et al., 2004). Except intensive poultry keeping, broilers are also kept in less intensive fattening in different European countries, and organic broiler production has got momentum during the last ten years. The following hybrids are present in our country as well as in the world: Cobb 500, Ross 308, Hybro, Hubbard, Lohman and among them the most represented are Cobb 500 and Ross 308 (Bjedov et al., 2011).

According to the FAO and the US Department of Agriculture, production of poultry meat is rising in recent years. In 2013, 94.2 million tonnes of poultry meat was produced worldwide, with the share of broiler meat 87.4-90.3%, or 86.4 million tonnes of broiler meat. In 2009, broiler meat production increased by 1.0 million tonnes (1.4%) compared to 2008, while in 2010 more than 4.5 million tonnes (6.1%) was produced. In 2011, more than 3.0 million tons (3.8%) of broiler meat were produced, in 2012 - 1.95 million tons (2.3%), and in 2013 - 3.2 million tonnes (3.8%) of broiler meat compared to the previous year (2010, 2011 and 2012) (Sakhatskiy, 2014).

Broiler fattening depend on the genetic potential of the line hybrid, diet, fattening technology, health status and preventive measures. The application of biotechnology and genetics in the last 20 years has led to improvements in broiler production performance.. One of the tasks of the selection is to produce a broiler that has as few feathers as possible at the time of completion of fattening, as it is easier to manipulate in the slaughterhouse. Broilers have a high basal metabolism and require strictly balanced meals made up of energy-rich nutrients, which at the same time contain other nutrients necessary for the growth of their body. In broilers, the heart and lungs are barely able to provide enough oxygen

*Corresponding Author : dperic@vet.bg.ac.rs

to meet the needs of the body. With controlled management conditions and biosecurity measures, the breeders were able to produce broilers that reached a mass of 2100 g on day 35 and 2800 g on day 42, with feed conversion up to 1.47 (Bjedov et al., 2011).

2. Digestive tract of poultry

Digestive tract of poultry is a tubular system extending from the oral cavity to the cloaca, and consists of specialized parts with associated glands. The digestive system, and in particular the intestine, is the environment in which the most important physiological and biochemical digestion processes take place, which are influenced by appropriate morphological, microbiological and physico-chemical factors (MC Lelland, 1975).Besides macro conditions, within the digestive system we can also talk about micro conditions, that is, different ecological niches in certain parts of the gut (Clarke and Bauchop, 1977). The surface of epithelial cells, the mucus lining the fringes or the interior of the crypts, as well as the intraluminal contents provide different conditions for the development of certain types of bacteria. On this basis, it can be concluded that the conditions for the development of bacteria in the digestive tract of animals differ vertically from the esophagus to the rectum and horizontally from the lumen to the depth of the intestinal crypts. The microflora in each habitat is a typical, stable, highest order community composed of a large number of different bacterial species. These communities are also characteristic of the type of animal, but also depend on the diet, the type of meal and the holding conditions of the animal.

Trypsin, protease and amylase activity increased rapidly during the first three weeks of broilers life, which is not the case with lipase. However, lipase activity did not increase until day 21, and is thought to be a limiting factor in digestion. Administration of high fat feed did not significantly increase lipase activity until day 21. Several significant changes were observed in the development of nutrient transport systems during ontogeny in broilers. During the first week of broiler life, the absorption of proline in the small intestine is high compared to glucose absorption. Since the relative growth rate of broilers is greatest during the first week, then amino acid intake may be parallel to this growth pattern. An increase in glucose absorption was seen during the second week. Due to allometric bowel growth, a decrease in the bowel relative to body weight was seen during the second week of broiler life. This may be another reason for the increase in glucose absorption that occurs during this period. There is a temporary increase in proline absorption during the sixth week. This increase is in parallel with the first post-juvenile myth and the increase in absolute growth rate. It is important that the absorption capacity of the gut closely matches the nutritional needs of the broiler (Kuenzel, 1994).

3. Fiber in poultry nutrition

Carbohydrates make up the largest component in the feed of domestic animals. Carbohydrates, as the most common ingredient in animal feed, are structural and supporting material in the body of plants, and have a significant role as a reserve of energy produced by photosynthesis. Structural carbohydrates, together with lignin, give the shape and firmness of the plant, while the nonstructural ones represent energy reserves. Of the total amount of dry matter in the tissues of plants, carbohydrates account for about 70%, while in the grain there are up to 85% of carbohydrates. On the other hand, carbohydrates are essential sources of energy in the body of animals, and they have a significant role in ensuring normal digestive tract peristalsis. Polysaccharides (glucans) are polymers of monosaccharides and are divided into homoglucans composed of the same monosaccharide units and heteroglucans composed of different monosaccharide units and their derivatives (Binkley, 1988). The modern approach defines carbohydrates as polyhydroxy aldehydes, ketones, alcohols or acids, and their simple derivatives, which create these during hydrolysis. Insoluble carbohydrates in plants, and especially cellulose, are responsible for the stability and their mechanical strength, while carbohydrates of higher solubility, such as starch, serve as an energy depot. According to Jovanovic et al. (2004), poultry has a natural ability to digest starch, glycogen, sucrose, maltose and simple sugars glucose and fructose, while lactose or milk sugar is very poorly digested due to limited lactase activity in the gut.

Fibers are defined as skeletal remains of plant cells that are not susceptible to digestion by the digestive enzymes present in the animal body and represent a quantitatively respectable fraction of feed of plant origin. The variation in the amount and structure of the fibers is large between different plant nutrients. They consist of polymeric carbohydrates originating from the plant cell wall, including non-carbohydrate components such as lignin, which are not or are minimally digestible in the small intestine.The U.S. Food and Nutrition Board defines "total dietary fiber" as a set of "dietary fiber" consisting of indigestible carbohydrates and lignins originating from plants, and consisting of isolated, indigestible carbohydrate components with proven positive physiological effects on people.

According to Jovanovic et al. (2004), crude fibers are divided according to their type and origin into cellulose, hemicellulose and lignin (highly soluble fibers found in the weed of the cereal grain and the woody part of trees and shrubs). Fibers affect the development of the gastrointestinal tract, intestinal morphology and enzyme secretion, as well as nutrient absorption in animals, and these effects depend primarily on the physico-chemical properties of the fibers, but also on the age of the animals (Mateos et al., 2012). The physicochemical properties of the fibers primarily depend on the type of polysaccharides that make up the cell wall, i.e. their intermolecular bonds determine the solubility of the fibers, and they relate to water solubility, gel formation ability, crystallization, as well as the ability to aggregate into complex cell wall structures of plants.The fiber is predominantly found in plant cell walls and consists of NSPs and some other non-carbohydrate compounds including lignin, proteins, fatty acids and waxes with which the fiber is inextricably bound (Bach Knudsen, 2001). The main problems associated with the use of NSPs in poultry feed are the viscosity and water binding capacity. Studies have shown that viscosity is due to the presence of soluble pectins or -glucans that, even in small amounts, significantly increase intestinal viscosity. On the other hand, undissolved polysaccharides such as cellulose and xylan can bind water in limited quantities, without too much affecting the viscosity of the digestive tract.The addition of certain NSPs in the poultry diet adversely affects the digestion of starch, proteins and lipids. This effect is attributed to the presence of viscous sugars that interfere with the diffusion and transport of lipases, oils and mycelium of bile salts within the gastrointestinal chyme.

Increased viscosity of the intestinal contents may interfere with the interaction between the substrate and lipases or bile salts in the small intestine and thus impair the absorption of certain nutrients. The β -glucans found in barley and oats are thought to form complex bonds with digestive enzymes and reduce their activity. However, in contrast, Ikegami et al. (1990) have shown that the activity of GIT enzymes in rats increases after eating nutrients that increase the viscosity in the digestive tract, so it can be inferred that although some NSP fractions have an antinutritional role in poultry metabolism, it is possible that some beneficial properties may still be related with NSP fermentation end products.

The fiber content of plant nutrients depends on the type and part of the plant, as well as the vegetation stage. Young green or canned plants and root tubers contain the least amount of fiber that increases with the vegetation phase. Hay and especially straw are very rich sources of fiber. Grains contain a relatively small amount of fiber, and the differences are related to the presence of weeds. The content of BEM in the by-products varies relatively, which is conditioned by the type and method of processing the feedstock. Feed of animal origin do not contain fibers, except those left behind in the digestive tract after the processing of whole animals into meat meal.

The use of raw fiber in poultry nutrition formulas is often a controversial topic among many non-ruminant nutritionists. On the one hand, international organizations (Isa, Lohman) consider crude fiber an essential component, since their presence in feed causes, among other things, an increase in gastric size, improves digestibility of starch, and limits or reduces feathering. On the other hand, many nutritionists still avoid the significant use of fiber in poultry nutrition on the grounds that fiber is not a significant source of energy, that is, the presence of fiber in feed inevitably leads to its energy dilution. In addition, traditional fiber sources are associated with some negative properties, such as the possibility of contamination with mycotoxins. One of the reasons that fibers in the poultry diet are still under-accepted may be the fact that different fibers have different effects primarily on the digestive tract, but also on other non-ruminant organ systems.

Insoluble fibers present in feed exert a dominant influence on the function of the intestine and thus modulate nutrient utilization. Definitely, the digestibility of starch as well as the passage of intestinal contents are directly influenced by the insoluble fiber present in feed, thereby reducing the risk of colonization of harmful bacteria. Insoluble fibers affect the health of the intestine through two different mechanisms of action, which are related to the faster transit of the intestinal contents, but also to the increased number of goblet cells. Goblet cells are a special type of epithelial cells whose primary function is the formation of mucin, an integral component of intestinal mucus. It is a well-known fact that harmful bacterial species cannot be so easily attached to colon intestinal mucosa and colonized, so that increased goblet cells have a positive effect on the health of the digestive tract and the maintenance of eubiosis. Also, insoluble fibers have a very high water binding capacity, so binding water in the upper parts of the hose releases the same water by osmotic pressure in the lower parts, which makes it available for resorption and does not appear in the outside, or directly affect the humidity of the mat.

The traditional source of fiber in our conditions is mainly wheat bran, which still contains insufficient fiber (about 10%) and carries with it the risk of the presence of mycotoxins. The solution could be to use oat bran containing a high percentage of insoluble fibers, but they are rarely found on the market, and mycotoxin contamination is possible. For this reason, the use of insoluble crude fiber concentrates (CFCs) whose chemical composition and purity would differ significantly from standard sources of fiber in the diet of non-ruminants is imposed. According to Hetland et al. (2004), insoluble fibers have traditionally been viewed as an inert nutrient diluent with little or no nutritional value in the diet of monogastric animals. However, more recent studies indicate a positive role of insoluble fiber in the development and health of the digestive tract of poultry, thereby increasing the absorption of essential nutrients and controlling animal behavior (Mateos et al., 2012).

Although earlier studies (Sklan et al., 2003) indicated a negative effect of fiber on feed consumption, digestibility of basic nutrients and generally production performance, more recent data (Gonzalez-Alvarado et al., 2010) indicate that fiber materials in moderation improve performance of broilers. The type and amount of fiber present in food affects the development of the gastrointestinal tract and the production results of broilers in fattening (Jim'enez-Moreno et al., 2009). A specific feature of the various fibers is their solubility, which is crucial for the health and welfare of the poultry. Plant roots, sugar beets, as well as individual fruits (apples, oranges, etc.) are sources of primarily soluble fiber, unlike all types of cereals that contain a high percentage of insoluble fiber. Although soluble fibers may show a prebiotic effect, their presence in feed increases the viscosity of the intestinal contents, which reduces the digestibility of starch, fat and protein, or binds certain nutrients, thereby adversely affecting their digestibility (Iji et al., 2001). Unlike soluble, insoluble fibers increase the passage rate of the intestinal contents thereby reducing the possibility of accumulating toxic substances in the digestive tract. They also stimulate the development of intestinal fibrils, have a positive effect on the digestibility of starch and prevent cannibalism (Farran and Akilian, 2014).

Insoluble and swollen fibers like the cellulose complex fill the digestive system since they have good water-binding capacity. The swollen fiber has an effect on the stimulation of the gut edge receptors, thus facilitating the passage of feed through the gut. On the other hand, soluble and fermentable fibers form the nutrient base for lactic acid bacteria in the back bowel. Among fermentable fibers, pectin present primarily in the sugar cane and apple pulp has a very important role as an energy source for animals. Unlike young animals whose diet should be formulated without the use of large amounts of soluble and fermentable sources of fiber because digestion in the posterior bowel is not well developed, older categories of animals are able to use these feed sources effectively. Increased intake of soluble fibers has a positive effect on the digestibility of energy, while the presence of insoluble fibers decreases the digestibility of energy, indicating that the ratio of soluble and insoluble fibers affects the digestibility of food.

4. Lignocellulose, role, significance and mechanism of action

Lignocellulose is a wood-derived product and is used as a high-quality source of fiber in animal nutrition. It consists of carbohydrates (cellulose, hemicellulose) and lignin, which is not a well-defined substance, but an aromatic polymer derived from three phenyl-propane derivatives, namely coumaryl, coniferyl and sinapyl alcohol. Pectin is also present, and certain minerals and salt are also present in the traces (Singh et al., 2014). All plants contain lignocellulose, with lignin being almost insoluble among them and providing physical strength and strength to the cell wall of the plant. On the other hand, the presence of lignin in lignocellulose prevents its efficient enzymatic degradation and subsequent microbial fermentation of the resulting sugars (Zeng et al., 2014).Due to its complex structure, lignocellulose is highly insoluble and cannot be directly used in microbial processes. However, exposing lignocellulose to enzymatic hydrolysis releases fermentative sugars, which can be used by microorganisms to produce enzymes for their growth (Meng and Ragauskas, 2014).

Shivus and Denstadli (2010) found that the presence of lignocellulose in a meal prolongs the residence time of feed in the muscular part of the stomach, which increases the efficiency of exogenous (added) enzymes. First of all, the effect described should be expressed in the digestion of proteins, since the first step in their digestion is hydrolysis in the stomach by the action of gastric acid. Similar results related to proteolytic enzyme activity and increased amino acid digestibility were also found by Yokhana et al., (2014). On average, the addition of 0.8% lignocellulose to the broiler meal increased the digestibility of essential amino acids by 5.8%, on the basis of which the matrix value for insoluble CFC was calculated allowing for the reduction of soybean meal during meal optimization.

Farran et al. (2013) reported that lignocellulose added to feed in the amount of 0.8% increased protein digestibility and meat yield, that is, significantly reduced the amount of abdominal fat in broilers in fattening. The results obtained are also confirmed by Bogusławska-Tryk et al. (2015). who proved that lignocellulose in the amount of 0.5-1.0% had a positive effect on the composition of the gut microbiota and the degree of microbial fermentation in the gut.

Lignocellulose affects the health of the small intestine by preventing the colonization of harmful bacteria through two different mechanisms of action, which are related to increasing the number of goblet cells and accelerating the passage of intestinal contents. Rezaei et al (2011) demonstrated a significant increase in goblet cell numbers, and Hetland et al. (2004) reported acceleration of the passage of intestinal contents when a moderate amount of chlorofluorocarbons was present in the broiler meal.

5. Material and methods

In order to study the justification for the use of purified lignocellulose in broiler diets, tests were conducted to provide detailed insight into the production performance (growth, consumption and conversion of feed) and the humidity of the broiler mat in fattened meals with different amounts of added purified lignocellulose.

The group control system was organized at the farm Pileprom doo, Srbac, Republic of Srpska (BiH) for 42 days on 400 broilers. Cobb provenance broilers originating from the commercial incubator station Insta d.o.o., Srbac, Republic of Srpska, (BiH) were used for the experiment. Tests were performed on one-day old broilers of both sexes with an average body weight of 41.84 g. During the experiment, all experimental groups were fed mixtures of standard feedstock that fully met the needs of broilers at all stages of fattening. The diet program included three fattening periods of 1-42 days, during which three nutritionally different concentrated mixtures were applied in pelleted form: starter (from 0-13 days, crushed pellets), grower (14-28 days, pellets 3 , 5 cm) and a finisher (29-42 days, 3,5 cm pellets) manufactured by Rapic d.o.o., Farmofit, Gradiška, Republic of Srpska (BiH). The basal diet was formulated to meet the maintenance and growth requirements of the animals used in the experiment.

The main task of the study was to determine the effect of broiler diet with different amounts of purified lignocellulose on health and production performance. Therefore, minimal corrections were made in the feed of the experimental groups to achieve the set target (Tables 1 and 2). The mixtures for the experimental groups differed only in the fact that in the first two mixtures (starter and grower) a preparation of purified lignocellulose was added in quantities of 4 g/kg of feed and 6 g/ g of feed for the first and second experimental groups, respectively 6 g / kg for the third experimental group with a 0.6% reduction in soybean meal. A commercial preparation of Arbocel® (R, J. Rettenmaier & Söhne GmbH + CO. KG, Rosenberg, Germany) containing about 70% acid detergent fibers (ADF) and 24% acid detegent lignin detergent (ADL) was added to the meal for all three experimental groups. (Table 1, 2)

Control measurements of specimens were performed individually during the migration of one-day broilers, as well as at the end of each broiler fattening phase. The measurements were made on an electronic scale with an accuracy of 1 g. On the basis of the measurement results, the average body weight of the broilers at the end of each phase was calculated, as well as at the beginning and end of the experiment, summarized. From the differences in body mass at the beginning and end of each phase, the total gain was calculated.

During the experiment, at the end of each phase, the amount of feed consumed for each group was accurately measured. From the obtained data on feed consumption and growth, feed conversion was calculated separately for each phase as well as for the whole trial. On day 28 of the experiment, five samples were taken from each box from all layers of litter (4 near the walls and one from the middle). Samples from each group were pooled and thoroughly mixed, and the moisture content was determined by drying at 103 ± 2 ° C to constant weight (ISO 1442: 1997).

6. Production performance

6.1. Body weight of broilers

The broiler body weight in the experiment is shown in Table 3 from which it is observed that the broilers had a uniform body weight at the beginning of the experiment (41.71 ± 1.41-42.16 ± 1.31 g) and no statistically significant differences were found ($p < 0.05$) between different groups. After three weeks of fattening, the highest body weight (826.20 ± 118.94 g) was achieved by the group of broilers (E-II), which received a higher amount of lignocellulose preparations through feed and was statistically significantly higher ($p < 0.05$) in relative to the broiler body weight of the control (772.10 ± 117.47 g) and the first experimental group (782.10 ± 96.93 g). At the end of the experiment, only the broiler group fed with a smaller amount of lignocellulose added achieved a slightly lower body weight (0.02%) than the control group. The largest body mass was achieved by the experimental group of broilers (E-II), which were reared on the feed with a higher amount of lignocellulose. A statistically significant difference ($p < 0.05$) was found between the average broiler mass of the O-II experimental group and the other observed groups (Table 3).

6.2. Average broiler gain during fattening

The average increase of body mass of broilers during the experiment is shown in Table 4, from which it is observed that the addition of larger quantities of lignocellulose in the meal resulted in the highest daily gain in the second (784.00 ± 118.81 g) and third (743.97 ± 117.98) g) experimental group of broilers during the first half of the experiment. The same trend was maintained in the second part of the experiment (21-42 days) where the highest average daily increase was achieved by the E-II group (1781,80 ± 278,99 g). In the whole trial (1-42 days), the lowest average daily gain was achieved by control broilers (2378.28 ± 332.14 g), and th ehighest ADG was in E-II broilers (2569.29 ± 266.50 g). (Table 4)

6.3. Feed consumption and conversion of broiler

Daily consumption of feed is shown in Table 5 from which it can be seen that the control group of broilers consumed the usual amounts of feed during the experiment. In the first phase of the experiment (day 1-21), food consumption did not differ significantly among the experimental groups of broilers fed with mixtures to which different amounts of lignocellulose preparations were added. Similar trend was observed in the second phase of the experiment (21-42 days), where the best appetite (3,254 g) was found in the group of broilers (E-II) to which a large amount of lignocellulose preparations was added to the food without reducing the participation of soybean meal. (Table 5)

Considered for the whole trial summarized, the addition of lignocellulose did not affect food consumption, so the control group achieved better consumption by 6.3 and 2.5% compared to E-I and E-III, respectively, and lower consumption by 0.80 % relative to E-II group. (Table 6)

The average conversion of food during the experiment is shown in Table 7. The data show a positive effect of the addition of lignocellulose in the food, except that in the first phase of the experiment the worst conversion was achieved by control group broilers (1.394), which was 2.5; 4.2 and 1.8% lower than the realized conversion of E-I, E-II and E-III broilers. In the second phase of the experiment (days 21-42), the same trend was maintained so that the best conversion was achieved by E-I broilers (1,822) and the worst broilers (1,973) by the control group. In the whole fattening period, from the first to forty-second days, the control group broilers achieved the highest conversion. (Table 7)

6.4. Determination of carpet humidity

Based on the results of the statistical analysis, it was found that adding more lignocellulose to the broiler feed resulted in lower average rug humidity in E-II (21.98 ± 1.67%) follwed by E-III (24.10 ± 1.81%)) groups compared to control group broilers (29.32 ± 1.73%).The average humidity of the E-II and E-III broiler rugs was statistically significantly ($p < 0.05$) lower than the average broiler rug humidity of the control group. (Table 8)

7. Conclusion

Insoluble fiber also has a positive effect on nutrient utilization, so the presence of insoluble fiber in the meal prolongs the residence time of the feed in the muscular part of the stomach, which increases the efficiency of exogenous (added) enzymes. First of all, the effect described should be expressed in the digestion of proteins, since the first step in their digestion is hydrolysis in the stomach by the action of gastric acid. On an average, the addition of 0.8% lignocellulose to the broiler meal increased the digestibility of essential amino acids by 5.8% allowing for the reduction of soybean meal during meal optimization. With the addition of a larger amount of lignocellulose preparations to the feed resulted in the best economic return.

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Table 1. Raw material composition of broiler feed mixtures in fattening, (%)

Nutrients	Mixtures, (%)		
	I - starter	II - grover	III - finisher
Corn	40,86	46,40	44,83
Wheat	15,00	15,00	20,00
Soybean meal	31,10	21,86	17,75
Fullfat soya	7,00	12,00	12,00
Soybean oil	1,71	0,78	1,68
Stock chalk	1,44	1,26	1,24
Monocalcium phosphate	1,02	0,86	0,66
Salt	0,20	0,19	0,19
Sodium bikarbonate	0,17	0,15	0,15
Vitamin-mineral supplement	1,50	1,50	1,50
Total	100,00	100,00	100,00

Table 2. Chemical composition of broiler feed mixtures in fattening (%)

Nutrient	Starter 0-13 days				Grover 14-28 days				Finisher 29-42 days
	K	O-1	O-2	O-3	K	O-1	O-2	O-3	
Metabolic energy Kcal/kg	2990	2984	2979	2985	3045	3037	3033	3040	3150
Humidity	10,93	10,93	10,87	10,88	11,09	11,1	11,03	11,04	11,11
Total ash	5,92	5,92	5,89	5,88	5,56	5,56	5,54	5,53	5,16
Crude protein	22	21,95	21,87	21,84	20	19,26	19,89	19,87	18,5
Raw fiber	3,4	3,82	3,97	3,95	3,25	3,67	3,85	3,82	3,15
Total fat	5,21	5,21	5,19	5,2	5,31	5,31	5,29	5,3	6,19
Calcium	0,95	0,95	0,95	0,95	0,9	0,9	0,9	0,9	0,8
Phosphorus total	0,67	0,67	0,66	0,66	0,62	0,62	0,61	0,61	0,56
Phosphorus usable	0,44	0,44	0,44	0,44	0,42	0,42	0,42	0,42	0,40
Sodium	0,16	0,16	0,16	0,16	0,16	0,16	0,16	0,16	0,16
Chlorine	0,19	0,19	0,19	0,19	0,19	0,19	0,19	0,19	0,19
NFE	52,44	52,21	52,21	52,25	54,79	55,1	54,4	54,44	55,89
Lysine (total)	1,30	1,30	1,30	1,30	1,18	1,18	1,18	1,18	1,06
M + C (total)	0,98	0,98	0,98	0,98	0,88	0,88	0,88	0,88	0,80
Threonine (total)	0,87	0,87	0,87	0,87	0,80	0,80	0,80	0,80	0,72

Vitamin A 13500 IU, Vitamin D3 5000 IU, Vitamin E 80 IU,Vitamin K3 4 mg, Vitamin B1 4 mg,Vitamin B2 6 mg, Vitamin B6 5 mg, Vitamin B12 0.025 mg, Vitamin C 25 mg, Biotin 0.15 mg, Niacin 60 mg, Calcium pantothenate 16.5 mg, Holin hlorid 750 mg, Folic acid 2 mg, Iodine 1 mg, Selenium 0.3 mg, Iron 40 mg, Copper 20 mg, Manganese 100 mg, Zinc 80 mg, Antioxidant 125 mg, Endo-1,3(4)-beta-glucanase (4a15) EC3.2.1.6 - 152 U, Endo-1,4-beta-xylanase (4a15) EC 3.2.1.8 – 1220 U, 6-phytase (4a1640) EC3.1.3.26 – 500 FTU

Table 3. Broiler body weights during the experiment, g

Fattening period	Groups			
	K	E-I	E-II	E-III
X̄± SD				
Days 1-21	730,38±109,44[a]	739,94±98,06[a]	784,49±118,81[b]	743,97±117,98[a]
Days 22-42	1647,90±330,58[a]	1649,90±269,32[a]	1784,80±278,99[b]	1709,10±305,97[c]
Days 1-42	2378,28±332,14[a]	2389,84±263,01[a]	2569,29±266,50[b]	2453,07±307,11[c]

Legend: same lettersa,b,c,ab p<0,05;

Table 4. Average broiler gain during the experiment, g

Fattening period	Groups			
	K	E-I	E-II	E-III
X̄± SD				
Days 1-21	730,38±109,44[a]	739,94±98,06[a]	784,49±118,81[b]	743,97±117,98[a]
Days 22-42	1647,90±330,58[a]	1649,90±269,32[a]	1784,80±278,99[b]	1709,10±305,97[c]
Days 1-42	2378,28±332,14[a]	2389,84±263,01[a]	2569,29±266,50[b]	2453,07±307,11[c]

Legend: same letters a,b,c p<0,05

Table 5. Average daily consumption during broiler fattening, g

Fattening period	K	E-I	E-II	E-III
Days 1-21	51,27	50,64	52,63	51,24
Days 22-42	154,47	143,21	154,99	149,72
Days 1-42	103,02	96,93	103,81	100,48

Table 6. Total feed consumption during broiler fattening, kg

Fattening period	K	E-I	E-II	E-III
Days 1-21	1,076	1,063	1,105	1,076
Days 22-42	3,250	3,007	3,254	3,144
Days 1-42	4,327	4,071	4,360	4,220

Table 7. Feed conversion during broiler fattening, kg

Fattening period	K	E-I	E-II	E-III
Days 1-21	1,394	1,359	1,337	1,369
Days 22-42	1,973	1,822	1,823	1,839
Days 1-42	1,788	1,673	1,669	1,691

Table 8. Rug humidity

Parameter	Groups			
	K	E-I	E-II	E-III
$\overline{X}\pm$ SD				
Humidity, %	29,32±1,73[a]	26,56±2,42[ab]	21,98±1,67[c]	24,10±1,81[bc]

Legend: same letters a,c,ab,bc p<0,05;