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NEW FEED ADDITIVES BASED ON PHYTOGENICS AND ACIDIFIERS IN ANIMAL NUTRITION **

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Abstract: Low regulations for feed additives use, has been changed in EU as well as in our country, especially in use of antibiotics as growth promotors. Along years it has investigated an alternative for antibiotics as growth promotors. Essential oilsand organic acids are one of alternative feed, which acting in a few of the most important directions: as antioxidants, metabolic upgraders, growth promotors and development of pathogenic microorganizms controllers, including moulds and bacterias and as environmental protecting through control of amonia and nitrogen excretion.

The objective of this paper was to determine the composition of essential oils in a commercial product which, in combination with the blend of organic acids may be used in poultry nutrition as an alternative to antibiotic growth promoters and establish impacts on performance parameters in broilers and microbiological properties of feces. Obtained results showed that the product contained an essential oil in which over 75% of total mass account for 2-phenol- carvacrol and eugenol.

Tested product has been found to have positive effects in reduced mortality (from 4,45% to 1,5%), higher body weight by 5,55% and decreased feed conversion by 9.9%. Significant positive effect has been shown in reduction of abdominal fat content, by 70%, and it can be concluded that product in question may be successfully used in poultry nutrition.

Key words: essential oils, organic acids, broiler nutrition, performance parameters

Introduction

Regulations related to animal feed additives have undergone many changes in recent years not only in EU countries, but in our country, too, particularly regarding the application of antibiotic growth promoters. Prior to antibiotics ban as of January 1, 2006, extensive researches were carried out directed to developing alternative additives and strategies to replace antibiotic feed additives, such as: organic acids and their salts, probiotics, prebiotics, enzymes, biogenic additives (essential oils, herbal extracts, etc.) *Adams* (2005), *Dibner* (2004), *Dijk* (2004), *Mccartney* (1999).

Short chain organic acids (c1-c7,) or their salts have been used for a long time as preservatives in hay or feed raw material for the purpose of preventing mould growth and, therefore, mycotoxin production *Gedek* (1999), *Liu* (2001), *Luckstadt* (2005). Interest in blends of organic acids or their salts combined with essential oils and herbal extracts as potential replacement for antibiotic growth promoters (AGP) inhibiting or hindering growth of pathogenic bacteria in the animal gut *Luckstadt* (2005), PERIS AT ALL (2002) has increased recently.

Through dietary supplementation with acids, pH within the gut is reduced below 6 and beneficial bacteria growth is promoted at the expense of pathogenic organisms *Adams* (2005), *Bolder at all* (2000). Each acid has its specific spectrum of antimicrobial activities, while mode of action is rather similar in all of them. Owing to the combined effect of acids and low pH of water used as feed raw material, microbial growth, Salmonella in particular, is inhibited as early as in grain, and therefore, contamination of feedstuffs and animal. The amount of used acids depends, in general, on the type of raw material, that is, its buffering capacity *Luckstadt* (2005).

Biogenic additives, primarily essential oils, have been widely used in animal nutrition for many years to promote a higher immune status *Bauerman* (2006). Essential oils, the volatile aromatic extracts obtained from different parts (buds, flowers, seeds, leaves, branches, roots, etc.) of herbs (coriander, cinnamon, oregano, rosemary, sage, clove, thyme, etc.) by straining, fermentation or extraction, or by steam distillation for commercial production.

They are known for their antioxidative, antimicrobial, fungicidal and antiviral effects, are said to promote a higher immune status within the animal and are environmentally friendly in a way that they decrease ammonia concentrations and excess nitrogen excretion in the environment *Bauerman* (2006), Eilers (2006), *Luckstadt* (2005).

Adequate combination of essential oils and organic acids and their salts as an alternative to antibiotics, provides opportunity for production of "organic" meat, branded meat offered at special price. Antioxidative effects are of considerable significance at times of stress, whether on body or cell level. This effect plays an important role in poultry production, since poultry is very sensitive to stress situations *Peris at all* (2002).

The main metabolic effect of feed additives formulated in such a way is demonstrated in the liver function, reduction of fatty liver syndrome and in hormone activity. Production and application of these additives is in a continuous rise. Of particular importance is understanding of the synergistic effects of plant extracts and acids on the animal health *Wald* (2004).

The objective of this paper was to determine the composition of essential oils in a commercial product which, in combination with the blend of organic acids may be used in poultry nutrition as an alternative to antibiotic growth promoters and establish impacts on performance parameters in broilers and microbiological properties of feces.

Material and Methods

Active principles and quantitative determination of phytobiotic feed additive were identified by gas chromatography coupled with mass spectrometry. Usual analytical procedures of separating the constituent ions on separation column and mass spectrum recording in «SIM» and«SCAN mode». *Packard (1999)* were employed.

Dietary Treatmetns and Animals

Throughout the trial, chickens were fed two balanced treatment diets: group I – trial diet I control and group II – trial diet II with mixture acids and botanicals, 3kg/t. U obe smeše je dodat enzim fitaza. Nutrients and energy content of trial diets is given in Table 1.

Composition of mixtures for chicken feeding was calculated according to *TablesAEC (1987)* and chemical analyses results of used raw materials. Chemical composition of raw materials and complete feeds was determined in accordance with *AOAC methods (2000).*

The trial involved 900 day-old chicks. Chickens were randomly assigned in 2groups (450 birds in each group) and housed in battery pens within the house system under controlled ambient conditions. Chickens had *ad libitum* access to feed and water. Trial lasted 42 days. Body weight was recorded at

the start and end of the trial, and feed consumption at the end of the trial and following production parameters were determined: average daily gain, average daily feed intake and feed utilization (kg gain/kg consumed feed).

Table 1. Nutrients and energy content in trial diets

CONTENTS	TRIAL DIETS I		TRIAL DIETS II	
	1-28days	28-42days	1-28days	28-42days
Crude protein [%]	21.87	18.87	21.87	18.87
Crude fibre [%]	3.47	4.15	3.47	4.15
Calcium [%]	0.99	0.89	0.99	0.89
Available phosphorous [%]	0.27	0.25	0.27	0.25
Lysine [%]	1.24	0.98	1.24	0.98
Methionine + Cystine [%]	0.86	0.73	0.86	0.73
ME Poultry [MJ/kg%]	13.00	12.80	13.00	12.80

After being denied access to feed for 12 hours, broiler chickens were slaughtered.Edible organ meats (liver, heart, gizzard) were weighed per groups, and chilled carcasses individually. Total 20 representative carcasses - 10 from each treatment group – were selected for dissection. Carcasses were cut apart in the following parts : breast, thighs, back, wings. Meat, skin and bones were manually separated from breasts and thighs and recorded for each individual bird. Mass of edible parts and breast and thigh meat was expressed in grams per a bird, and share of abdominal fat as a percentage of live body mass.

Data for body weight, feed conversation, edible part yield, and fat share in meat were statistically processed by analysis of variance and the differences between groups were tested for significance.

Swabs taken from broilers from trial and control groups were incubated in broth at 37°C for 24 hours and inoculated on nutrient culture media,

a) Blood agar
b) Mac Conkey agar
c) Sabouraud agar.

Feces from trial and control groups was incubated at 37°C for 24 hours in Selenite broth with enriched Rappaport-Vassiliadis broth. After incubation, direct inoculation on blood agar, Mac Conkey agar and Sabouraud agar was performed.

Results and Discusion

Utilized modern analytical method for confirmation of quantitative and qualitative composition of essential oil, being main component for phytobiotic feed additives production, it has been determined that 2-phenol-carvacrol and eugenol account for over 75% of the essential oil total mass. These two compounds provide phytobiotic potential to essential oil, that is, to patented final product. Out of more important compounds, ranging from 1 – 7%, α and γ-terpinen, o-cimen and thymol should be mentioned. The presence of other terpene compounds and cyclic phenols, accounting for about 20 components , is of no therapeutic importance. Essential oils content ranged in the level of 2%. In Table 2 are given results of examined essential oil composition.

Table 2: Sample 1- GC-MS analyses

NO.	RT	COMPONENT	%
1.	10.11	α-terpinen	1.10
2.	10.29	o-cimen (p-cimen)	7.64
3.	15.90	timol	1.07
4.	16.39	karvakrol	28.65
5.	17.92	eugenol	46.77

Besides aromatic oils, the tested product contained 46% of organic acids (lactic, fumaric, citric and formic). Tested product was added in concentration of 3 kg in the compound feed.

Tested product has been found to have positive effects in reduced mortality, higher body weight and decreased feed conversion. Significant positive effect has been shown in reduction of abdominal fat content, by 70%.

Based on laboratory tests, the results of which are shown in Table 4, it can be concluded that the trial group fed with dietary supplemented acidifiers and aromatic extracts had significantly lower number of bacteria in gastrointestinal tract, E.Coli was detected in one sample only, while in control group presence of bacteria was identified in 7 samples -Entero bacteria.

Swabs taken from the surface of the skin of slaughtered birds from trial and control groups were examined and showed no growth in tested culture

Table 3: Production parameters of broiler chicken trial

PARAMETERS	TRIAL GROUPS	
	I	II
Number Of Chickens	450	450
Broj Uginulih Index (%)	20 4.45[a]	7 1.55[b]
Number Of Feeding Days	42	42
Starting Body Weight [G]	38.48	38.48
Finishing Body Weight [G] Index [%]	2160[a] 100	2280[b] 105.55
Feed Conversion [G/G] Index [%]	2.14 [a] 100	1.93 [b] 90.20
Edible Part Yield [G] Index [%]	1650 [a] 100	1751 [b] 106.12
Abdominal Fat Share [%] Index [%]	1.91[a] 100	0.57[c] 29.84

a,b, means within the same row followed by different letters are significantly different ($p<0.01$)
a,c, means within the same row followed by different letters are significantly different ($p<0.001$)

media, as was expected because of employed poultry slaughter technology. Pathological examination of the intestinal tracts of sacrificed birds showed no changes. Production parameters realized in broiler chicken feeding are shown in table 3.

Table 4. Results of microbiological examination of feces

	K	O
E. coli Hemolitica	+++ 7*	+ 1*
E.Coli 0157	+++ In all	+ 3*
Micrococus sp.		
Campilobacter uni		
Bacilus spp.		
Diplococus spp.		
Streptococus fecalis	++ 6*	+ 2*
Enterococus fecalis	+++ 9*	+ 2*

K -control group
O - trial group
* number of infected samples

Based on the obtained results it can be concluded that tested product gave better-than expected results and that it may be successfully used in the poultry meat production.

Conclusion

Based on the obtained results, the following may be concluded:

1. Tested essential oil is a blend in which over 75% of total mass account for 2-phenol- carvacrol and eugenol.
2. Tested product has been found to have positive effects in reduced mortality, higher body weight and decreased feed conversion. Significant positive effect has been shown in reduction of abdominal fat content, by 70%.
3. Tested product has positive effect on microbiological properties of feces
4. Tested product is recommended for applications in animal nutrition.

NOVI DODACI NA BAZI FITOBIOTIKA I ZAKIŠELJIVAČA U ISHRANI ŽIVOTINJA

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Rezime

Zakonska regulativa u oblasti dodataka hrani za životinje u zemljama EU a i kod nas se zadnjih godina značajno promenila, naročito kod upotrebe antibiotika kao promotora rasta. Dugi niz godina radjena su opsežna istraživanja u pronalsku alternative antibioticima, promotorima rasta. Eterična ulja su jedna od alternativnih hraniva koja deluju u nekoliko najvažnijih pravaca: kao antioksidanti, poboljšivači metabolizma, kontrolori rasta i razvoja patogenih mikroorganizama, uključujući dejstvo na plesni i bakterije, u zaštiti životne sredine kroz kontrolu količine amonijaka i izlučenog azota

U ovom radu ispitivan je sastav eteričnih ulja u komercijalnom preparatu koji se u kombinaciji sa smešom organskih kiselina može primenjivati u ishrani brojlera kao alternativa promotorima rasta i utvrđivanje uticaja na proizvodnje performanse brojlera kao i na mikrobiološku sliku fecesa.

Rezultati ispitivanja su ukazali da ispitivani preparat sadrži etarsko ulje u

kome preko 75% ukupne mase čine dva fenola karvakrol i eugenol.

Ispitivani preparat je imao pozitivno dejstvo na smanjenje mortaliteta (sa 4,45% na 1,5%) , na povećanje težine za 5,55%, smanjenje feed conversion za 9,8%. Naročito izražen pozitivan efekat je na smanjenje udela abdominalne masnoće, za 70%.i sa sigurnošću se može preporučiti za primenu u ishrani živine

Ključne reči: eterična ulja, organske kiseline, ishrana brojlera, proizvodne performanse

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