Influence of selected phytoadditives and probiotics on zootechnical performance, caecal parameters and meat quality of rabbits

Ľubica Chrastinová[1], Mária Chrenková[1], Andrea Lauková[2], Mária Poláčiková[1], Monika Simonová[2], Renáta Szabóová[2], Viola Strompfová[2], Ľ. Ondruška[1], I. Chlebec[1], V. Parkányi[1], J. Rafay[1], Zuzana Vasilková[3]

[1] Animal Production Research Centre, Nitra: Institute of Nutrition, Hlohovecská 2, 951 41 Lužianky, Slovak Republic; [2] Institute of Animal Physiology, Slovak Academy of Sciences, Šoltésovej 4-6, 04001 Košice, [3] Parasitological Institute of Slovak Academy of Sciences, Hlinkova 3, 04001 Košice, Slovak Republic

SUMMARY

A total of 96 unsexed, weaned NZW (New Zealand White) hybrid rabbits 5 weeks old were randomly allocated to 4 similar groups, and kept in standard cages. The animals of the experimental group (EG1) got the sage (*Salvia officinalis*) plant extract (10 µl/animal/day) in drinking water for the period of 21 days and were fed with an untreated pelleted diet B, in (EG2) they were fed with an enriched diet with supplement *Eleutherococcus senticosus* (Ginseng dry extract 30g / 100 kg feed). The rabbits of the experimental group (EG3) had *ad libitum* access to the untreated diet B and the drinking water had a culture of *Enterococcus faecium* AL 41 strain 10^9 CFU / ml; 500 µl /animal/day) in the drinking water for the period of 21 days. The control group (CG) had *ad libitum* access to the untreated diet B and the drinking water did not contain any coccidiostatic drugs during the experiment. The experiment lasted for 42 days, until the animals attained the slaughter weight of 2.5 kg. The body weight and feed consumption were registered weekly. The samples of individual feeds and complete granulated mixture were analyzed for the content of nutrients according to STN 46 7092. The samples of MLD *(Musculus longissimus dorsi)* were homogenized and analyzed for individual nutrients.

The collected caecal samples were analyzed for pH, VFA, ammonia-N and content of lactic acid. *Eimeria sp.* oocysts were enumerated in the faeces samples microscopically and expressed as counts of oocysts per 1 g of faeces (OPG). The significant differences were evaluated by a t- test. The feeding of natural substances did not influence biochemical and zootechnical parameters, and it had no negative effect on the growth performance of rabbits. It had however a positive effect on the health status and it reduced the number of *Eimeria* oocysts in the rabbit intestinal ecosystem. Among these 3 additives, compared to each other, the highest values of phagocytic activity (PA) were

determined after application of the probiotic strain *Enterococcus faecium* AL41. Phytoadditives did not influence negatively biochemical parameters in the blood, and the caecum of rabbits. They positively influenced mortality as well as they increased the body gain and the energetic value of meat.

Keywords: rabbits, phytoadditives, probiotics, meat quality

INTRODUCTION

With the advance of modern biotechnology, application of naturally occuring antimicrobial and antioxidant compounds have been preferably employed in meats because of their potential health benefits and physiology of the host, modulation of the gut flora due to their antimicrobial activity; moreover, they did not leave residua in meat and other products. The essential oil of sage *(Salvia officinalis L. aetherolum)* consisted of 25% thujon, 18% borneol and 15% cineol. *Eleutherococcus senticosus* (Siberian ginseng) contains different bioactive substances (Haviarová et al., 2006; Szaboóvá et al., 2008; Simonová et al., 2008; Chrastinová et al., 2009).

The aim of this study was to examine the effect of feed supplementation on natural substances and the effect of the bacteriocinogenic strain *Enterococcus faecium* AL41 on the zootechnical parameters, and the meat quality and biochemical parameters in the digestive tract of rabbits.

MATERIAL AND METHODS

A total number of 96 unsexed, weaned NZW (New Zealand White) hybrid rabbits, age 5 weeks were randomly allocated to 4 similar groups, and kept in standard cages.

- The animals of the experimental group (EG1) were supplied with the sage (*Salvia officinalis*) plant extract (10 µl /animal /day) in the drinking water for the period of 21 days and were fed with the untreated pellet diet B (Table 1) for growing rabbits.
- The rabbits of experimental group (EG2) were fed with the enriched diet A with supplement of *Eleutherococcus senticosus* (ginseng dry extract 30 g/ 100 kg feed).
- The animals of the experimental group (EG3) were fed with the coccidiostat-free feed mixture B and the drinking water contained a culture of *Enterococcus faecium* AL 41 strain 10^9 CFU / ml; 500 µl /animal/day)) for a period of 21 days.
- The animals of the control group (CG) were fed with the coccidiostat-free feed mixture B and the drinking water without any coccidiostat drugs during the experiment.

The experiment lasted for 42 days, until the animals attained the slaughter-weight of 2.5 kg.

The samples of individual feeds and complete granulated mixture were analyzed for the content of nutrients according to STN 46 7092. The content of digestible energy (DE) was calculated by the equation of Wiseman et al. (1992). The samples of *Musculus longissimus dorsi* (MLD) were collected immediately after death and stored at 5˚C for 24h and then the standard physicochemical analyses (according to STN 57 0185) were made. The samples of MLD were homogenized and analyzed for individual nutrients.

Table 1 Ingredients and chemical composition of feed mixtures

Feed ingredients in %	A	B	Chemical analysis in g kg^{-1} feed	A	B
Dehydrated lucerne meal	26.0	26.0	Dry matter	902	900
Dried beet pulp	17.0	17.0	Crude protein	164.3	165.8
Oats	5.0	5.0	Crude fibre	179.3	180.6
Barley	2.5	3.7	Fat	44.0	40.7
Apple pomace	7.0	7.0	Nitrogen-free extract	444.8	440.6
Wheat bran	8.5	8.5	Organic matter	832.4	827.7
Sunflower meal	17.5	17.5	Ash	69.6	72.3
Olive-seed meal	6.0	6.0	Calcium	9.15	9.37
DDGS from corn**	5.0	5.0	Phosphorus	4.9	4.1
Minerals &Vitamins	2.6	2.6	Lysine	7.5	7.4
Rape oil	1.7	1.7	Met+Cys	6.5	6.5
Carob meal	0.2	0.2	Digestible energy ($MJ.kg-^{1}$)	11.2	11.1
Wheat meal +30 g *Eleutherococcus senticosus*	1.0	0	Metabolizable energy ME ($MJ.kg-^{1}$)	10.60	10.56

*Provided per kg diet: vit. A 12000IU; vit.D_2 2500 IU; vit. E 20 mg; vit.B_1 1.5 mg; vit. B_2 7.5 mg; vit. B_6 4.5 mg; vit.B_{12} 30 µg; vit.K 3 mg; nicotic acid 45 mg; folic acid 0.8 mg; biotin 0.08 mg ; Choline chloride 450 mg; Premix minerals (per kg diet) Ca 9.2 g; P 5.2 g; Na 1.6 g; Mg 1.0 g; K 10.8 g; Fe 327.5 mg; Mn 80 mg; Zn 0.7 mg

** Distiller- dried Grains with Solubles from maize

The pH at 24 hours post mortem was measured with the portable pH-meter mod. Radelkis OP-109, a combined electrode penetrating 3 mm into the MLD. Protein and fat content were estimated using an INTRATEC 1265 spectroscope and expressed in $g.100g^{-1}$: from these values, the gross energy value of samples of meat was calculated according to:

Energy-value ($kJ.100g^{-1}$) = (16.75 × total protein content + 37.68 × total fat content).

The caecal samples from each group of three slaughtered rabbits were collected for caecal bacteria analyses. The collected samples were analyzed for pH and VFA (molar production of acetate, propionate, butyrate, valerate, capronate), ammonia-N concentrations and content of lactic acid). Body weight and feed consumption were registered weekly. *Eimeria sp*. oocysts were enumerated in the faeces samples microscopically and expressed as counts of oocysts per 1 g of faeces (OPG). The results were evaluated by a two-way ANOVA and significant differences were marked at $P<0.05$ and $P<0.01$.

RESULTS AND DISCUSSION

The study was carried out in the Animal Production Research Centre Nitra, Slovak Republic. 96 rabbits were used in the experiment. They were weaned at 35 days and divided into 4 groups (2 rabbits per cage). We did not find large differences among the experimental groups according to feed intake, body weight and carcass-value in the fattening experiment (Table 2). Feed consumption per kg gain of live weight was lowest in the group with the supplement of *Eleutherococcus senticosus* (ginseng dry extract 30 g.100 kg^{-1} feed). The rabbits were slaughtered before the morning feeding to observe the fermentation processes in the caecum. Concentration of the observed VFA shows, that the highest intensive processes were observed in the caecum of rabbits in the experimental groups (Table 4; Table 4a).

The results of the antimicrobial activity of the essential oils, described by Delamare et al. (2007) reveal, that the oils of *Salvia officinalis* inhibited the growth of *Bacillus cereus, B. megatherium and B. subtilis*; a partial inhibitory effect was observed against *Escherichia coli* and *Staphylococcus aureus* (Szabóová et al., 2008). The reduced counts of st. aureus were obtained in samples from rabbits supplemented by AL41 (Pogány Simonová et al., 2009).

Table 2 Effect of treatment on the performance of rabbits

Parameter	EG1	EG2	EG3	Control
	Salvia	Ginseng	EF AL41	
Number of animals in groups	24	24	24	24
Initial live weight (35 d), g	970	950	830	845
Intermediate live weight (56 d), g	1824	1832	1692	1734
Final weight (77 d), g	2530	2625	2524	2504
Feed conversion ratio between 35th and 56th day ($g.g^{-1}$)	2.93	2.89	2.87	3.45^{**abc}
Feed conversion ratio per kg gain	3.50	3.55	3.43	3.59
Mortality (n)	2	1	3	2
Daily weight gain, (g/d)	37.14	39.88	40.33^{**A}	39.5
Carcass value (%)	52.25	52.44	52.31	52.26

**P<0.01 Significant difference

Table 3 Physicochemical characteristics of rabbit meat (MLD) 24 h *post mortem*

Parameter ($g.100g^{-1}$)	EG1	EG2	EG3	Control
Content of water	76.07±0.36	75.23±0.15	76.57±0.15	76.5±0.4
Total proteins	21.37±0.5	22.27±0.15	21.93±0.15	22±0.26
Content of fat	$3.53±0.8^{*bcd}$	1.5±0.1	1.73±0.2	1.5±0.2
Energetic value ($kJ.100g^{-1}$)	$491.3±8.3^{** bcd}$	429.49±3.8	$432.69±11.8^{*d}$	425.0±11.3
pH_{24}	5.71±0.03	5.72±0.04	5.54±0.03	5.65±0.04
Water holding capacity	$32.8±2.9$	$35.3±1.4^{*acd}$	31.1±5.2	32.2±2.1

*P<0.05; **P<0.01 Significant difference

Table 4 Qualitative parameters in the caecum on the 56th day of age

Parameter (n=3)	EG1	EG2	EG3	Control
pH	6.14±0.01	6.12 ±0.01	6.12 ±0.32	5.24±0.01
N-NH_3 (mmol.1^{-1})	9.31±4.95	10.91±1.47	10.91±1.47	18.8±0.76**abc
Lactic acid (g.100g^{-1})	0.059±0.01	0.057±0.01	0.02±0.01	0.016±0.01
VFA in the caecum (mmol.100g^{-1})				
Acetic acid	4.58±0.49	4.20±0.23	4.25±0.21	5.69±0.31
Propionic acid	0.23±0.04	0.33±0.09	0.28±0.05	0.42±0.17
Butyric acid	1.39±0.05	1.40±0.04	1.21±0.09	1.36±0.28
Other VFA	0.12±0.02	0.16±0.02	0.11±0.02	0.17±0.06

*P<0.05 Significant difference

Table 4a Qualitative parameters in the caecum on the 77th day of age

Parameter (n=3)	EG1	EG2	EG3	Control
pH	6.11±0.01	6.12 ±0.01	6.12 ±0.32	6.08±0.01
N-NH_3 (mmol.1^{-1})	10.3±6.6	10.91±1.47	11.63±1.87	9.1±6.6*abc
Lactic acid (g.100g^{-1})	0.029±0.01	0.052±0.07	0.02±0.01	0.044±0.01
VFA in the caecum (mmol.100g^{-1})				
Acetic acid	5.00±0.09	4.40±0.75	5.43±0.06	5.34±0.52
Propionic acid	0.40±0.09	0.29±0.03	0.37±0.11	0.45±0.03
Butyric acid	1.57±0.24	1.14±0.27	1.49±0.08	1.42±0.28
Other VFA	0.15±0.02	0.12±0.02	0.14±0.02	0.13±0.03

*P<0.05 Significant difference

The slaughter parameters and the quality of meat were practically similar in each experimental group (Table 3).The application of natural herb additives and the probiotic strain *Enterococcus faecium* AL 41 increased the concentration of the total protein and fat in the rabbit meat and the energetic value of meat also increased significantly (P<0.05) in the experimental groups. All animals were found in good health conditions during the trial. The application of *Eleutherococcus senticosus* reduced the mortality and increased the feed conversion ratio between the 35th and 56th day of age in the rabbits (P<0.05) and the average daily weight gain (P<0.01).

CONCLUSIONS

Feeding with natural substances and the application of the probiotic strain *Enterococcus faecium* AL 41 in the drinking water did not influence the biochemical and zootechnical parameters; also it had no negative effect on the growth performance of rabbits. It had a positive effect on the health status and it reduced the number of *Eimeria* oocysts in the rabbit intestinal ecosystem.

REFERENCES

Chrastinová, Ľ., Chrenková, M., Lauková, A., Poláčiková, M., Simonová, M., Szabóová, R., Strompfová, V., Ondruška, Ľ., Chlebec, I., Parkányi, V., Rafay, J., Vasilková,Z. 2009. Utilization of an extract product from ginseng supplementation in diets and different energy levels of granulated feed in the nutrition of rabbits. In: Archiva Zootechnica 12:1, p.72-79

Delamare, A.P.L., Moschen-Pistorello I.T., Atii-Serafini L.A.L., Echeverrigaray, S. 2007. Antibacterial activity of the essential oils of *Salvia officinalis* L. and *Salvia triloba* L. cultivated in South Brazil. Food Chem., 100, p.603-608

Haviarová, M., Chrastinová, Ľ., Szabóová, R., Simonová, M., Strompfová, V., Faix, Š.,Vasilková, Z., Plachá, I., Lauková, A., Rafay, J., Poráčová, J. 2006. *Eleutherococcus senticosus* and rabbits breeding. In: Proc.VII Days of Nutrition and Veterinary Dietetics, Sept.2006, Košice, Slovakia, p.101.

Simonová, M., Szabóová, R., Chrastinová, Ľ., Lauková, A., Haviarová, M., Strompfová, V.,Plachá, I., Faix, Š., Vasilková, Z., Mojto, J., Rafay, J. 2008b: The use of a ginseng extract in rabbits. Proceedings of the 9th World Rabbit Congress, Verona, Italy, June 2008, 809-813.

Szabóová, R., Chrastinová, Ľ., Strompfová, V., Simonová, M., Vasilková, Z., Lauková, A., Čobanová, K., Plachá, I., Chrenková, M., Mojto, J., Ondruška, Ľ. 2008a: Combined effect of enterocin CCM4231 and sage in rabbits. Proceedings of the 9th World Rabbit Congress, Verona, Italy, June 2008, 815-819.

Szabóová, R., Chrastinová, Ľ., Strompfová, V., Simonová, M., Vasilková, Z., Lauková, A., Plachá, I., Čobanová, K., Chrenková, M., Mojto, J., Jurčík, R. 2008b: Combined effect of bacteriocin-producing *Enterococcus faecium* CCM4231 and sage in rabbits. Proceedings of the 9th World Rabbit Congress, Verona, Italy, June 2008, 821-825.

STN 46 7092 Metódy skúšania krmív (Methods of feeds analyses), Bratislava, 1985

Wiseman,J., Villamide, M.J., De Blas, C., Carabaňo, M.J., Carabaňo, R.M. 1992: Prediction of the digestible energy and digestibiblity of gross energy of feed for rabbits. 1. Individual classes of feeds. In: Anim. Feeds. Sci. Technol., 39, p. 27-38.