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DIPEPTIDE ENTEROCIN A/P APPLIED TO GROWING RABBITS WITH ADMINISTERED METHICILLIN-RESISTANT *STAPHYLOCOCCUS EPIDERMIDIS*: EVALUATION OF GROWTH PARAMETERS AND MEAT QUALITY

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ABSTRACT

The aim of this study was to find: (1) whether bacteriocin-enterocin Ent A/P applied in rabbits drinking water can have an inhibitory effect on methicillin-resistant *S. epidermidis* P3/Tr2a; (2) whether application of the P3/Tr2a strain can negatively influence selected parameters and meat quality, and (3) if application of enterocin A/P can reduce possible negative impact of the P3/Tr2a strain. A total of 66 *post*-weaned hybrid rabbits (meat lines M91 and P91) of both sexes at the age of 35 days were divided into two experimental (EG1; EG2) and one control (CG) groups (22 rabbits in each group). Rabbits were kept in standard metal cages. They were fed with a mixture of a standard diet. At day 14, rabbits in the EG1 were administered with methicillin-resistant *Staphylococcus epidermidis* P3/Tr2a strain (in drinking water), marked with rifampicin (a dose of 500 µl/animal/day; 1.0 x 10^5 CFU/ml). Rabbits in the EG2 were administered with Ent A/P (50 µl/animal/day) supplied in drinking water (first 14 days). The rabbits had access to water *ad libitum*. Since day 22, all animals were fed only the untreated commercial granulated diet for growing rabbits. The experiment lasted for 48 days. Sampling of faecal mixtures per each group was provided at days 0/1, 14, 21 and 48 (end of experiment) to assess staphylococci and *Staphylococcus epidermidis* P3/Tr2a. At day 56 and at the end of the experiment, carcass and meat quality parameters (pH value 24 h *post-mortem*, colour lightness, water-holding capacity, protein and fat content, energetic value of the carcass yield) were measured. Methicillin-resistant SE strain did not affect negatively the selected physico-chemical parameters and nutritional quality of rabbit meat, as well as growth performance and the zootechnical parameters. In addition, in rabbits with Ent A/P, the highest daily weight gain was noted. In spite of the fact, that there was not significant reduction in the SE strain, enterocin A/P could be used as a preventive supplement in rabbit husbandry.

Key words: rabbits; meat quality; enterocin A/P; methicillin-resistance; *Staphylococcus epidermidis* P3/Tr2a

INTRODUCTION

The most critical period in rabbit breeding is weaning. Rabbits are most susceptible to various gastrointestinal infections and spoilage agents (Hermans *et al*., 2003). These problems may have a negative effect on feed consumption, growth performance and health status of animals. To reduce economic losses and to stabilise/improve the health status and gastrointestinal tract development and growth performance as well, antibiotics and growth promoters have been widely used in diet for several decades. As a consequence,

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an increase in microbiota antibiotic resistance has appeared. Serious situation has been caused due to methicillin resistant staphylococci (MRs), which are also a problem in human population. Therefore, an enormous demand is focused on reduction of MRs in rabbit breeding, especially because rabbits represent food-producing animals. Nowadays, to reduce/replace antibiotic supplements in feed diet a new approach is preferred – using nature-based supplements such as probiotics, prebiotics, synbiotics, enzymes, bacteriocins, organic acids, herbs and their extracts. They represent well-tried tools for disease prevention in various animals, including rabbits (Kellner *et al*., 1988; Falão-e-Cunha *et al.*, 2007; Lauková *et al.*, 2012; Pogány Simonová *et al*., 2019; 2020).

Previously, the beneficial effect of probiotics and/or their antibacterial products – bacteriocins in rabbits on growth performance, nutrient utilization, metabolism changes, microbial composition, health and meat quality has been described (Lauková *et al.*, 2006; 2012; Szabóová *et al.*, 2008; Kalma *et al.*, 2016; Cunha *et al.*, 2017; Chrastinová *et al.*, 2018; Pogány Simonová *et al.*, 2015; 2019). Staphylococci are a part of microbiota in the digestive tract of rabbits. Many staphylococcal strains harboured methicillin resistant genes. Therefore, they are spoilage bacteria, which may threaten rabbit health. As previously mentioned, one possibility to reduce MRs can be the use of bacteriocins. Dipeptide bacteriocin – enterocin (Ent A/P) is produced by the non-autochtonous strain *Enterococcus faecium* EK13 = CCM7419, which has been successfuly applied in different animals with a beneficial effect (Lauková *et al.*, 2006; Pogány Simonová *et al.*, 2020). *Staphylococcus epidermidis* P3/Tr2a is Gram-positive strain, which showed resistance to methicillin. Pogány Simonová *et al.* (2020) reported beneficial use of enterocins to inhibit growth of methicillin-resistant staphylococci.

Therefore, the aim of this *in vivo* study was to find: (1) whether bacteriocin-enterocin Ent A/P applied in rabbits drinking water can have an inhibitory effect on methicillin-resistant *S. epidermidis* P3/Tr2a; (2) whether application of the P3/Tr2a strain can negatively influence selected parameters and meat quality, and (3) if application of enterocin A/P can reduce possible negative impact of the P3/Tr2a strain.

MATERIALS AND METHODS

Experimental design

A total of 66 *post*-weaned hybrid rabbits (meat lines M91 and P91) of both sexes at the age of 35 days, reared in an intensive rabbitry in the Slovak Republic, were used in the experiment. The rabbits were randomly divided into two experimental (EG1; EG2) and one control (CG) groups (22 rabbits in each group). They were kept in standard metal cages (two animals per cage). The rabbits were fed *ad libitum* with a commercial diet (KV; TEKRO Nitra, Ltd. Slovak Republic), granulated to 3.5 mm in diameter. Water was provided *ad libitum* using nipple drinkers. The ingredients and chemical composition of the diet are presented in Table 1.

The diet was prepared according to procedures of the AOAC (2005) and Van Soest *et al.* (1991). Rabbit in CG had access to untreated diet and drinking water *ad libitum*. Feed diet did not contain any coccidiostatic drugs during the experiment. Rabbits in the EG1 group received *Staphylococcus epidermidis* (SE) P3/Tr2a strain (rifampicin marked variant, to differ it from other staphylococci) at dose of 500 µl, 10^5 CFU per animal per day at day 14 since the start of the experiment. SE was applied into rabbits drinking water for 7 days. The rabbits in EG2 group received 50 µl of Ent A/P/animal/day, applied into their drinking water, during first 14 days of the treatment period to control preventive effect of this Ent dipeptide. A cycle of 16 h of light and 8 h of dark was used throughout the experiment. The temperature and humidity in the housing area were continuously monitored using a hydrothermograph, which was situated on the same level as the cages (the average humidity and temperature during the year were maintained at values of 60 ± 5 % and 17 ± 3 °C). The experiment was performed in cooperation with the Institute of Animal Physiology, Centre of Biosciences of the Slovak Academy of Sciences in Košice. Animal care and the experimental procedure were approved by the Slovak Veterinary and Food Administration and by the Ethic Commission of both institutes. The experiment was approved by the State Veterinary and Food Administration of the Slovak Republic, no. SK CH 17016, SK U 18016.

Table 1. Nutrient content of commercial granulated diet for growing rabbits

Nutrient content	g.kg^{-1} in original feed	g.kg^{-1} in dry matter
Dry matter	886.65	1000.00
Crude protein	155.35	174.94
Crude fibre	132.37	149.29
Crude fat	20.30	22.89
Ash	90.08	101.60
Starch	238.71	269.22
Acid detergent fibre	151.69	171.08
Neutral detergent fibre	295.10	332.83
Calcium	15.90	17.94
Phosphorus	4.89	5.51
Magnesium	2.57	2.90
Sodium potasium	1.21	1.36
Iron	564.70*	636.88*
Zinc	97.77*	110.27*
Copper	20.50*	23.12*
Metabolizable energy (MJ.kg^{-1})	11.16	11.02

*mg.kg^{-1} feed

Evaluation of zootechnical parameters

The average initial and final live weights (g) were monitored during the experiment. Body weight of each experimental animal was recorded every week during the whole study; the average daily weight gain (ADWG) was calculated mathematically. Weight feed mixture was checked daily and the average daily weight gain and feed conversion were calculated mathematically.

Mortality and morbidity of animals were also recorded daily over the whole period of the experiment. Five animals (at the age of 56 days and at the end of the experiment) from each group were slaughtered and sampled. Rabbits were slaughtered in an experimental slaughterhouse after electro-stunning (90 V for 5 sec) by cutting the *carotid* and *jugular veins* and bleeding out. *Musculus longissimus thoracis and lumborum* (MLTL) were separated by removing the skin and connective tissue, chilled and stored at 4 °C for 24 h until physico-chemical analysis.

The pH value was determined after 24 h (*post--mortem*) using a Radelkis OP-109 (Jenway, England) with a combined electrode penetrating 3 mm into the samples. The electrical conductivity (µS.cm^{-1}), defined as the location of muscles, was evaluated using PMV 51 (TecPro GmbH, Germany). Colour characteristic was expressed by a CIE *L*a*b* system (lightness-L*, 0: black and 100: white; redness and greenness-a*; yellowness and blueness-b*) using a Lab. Miniscan. Lightness at room temperature was also measured.

The physico-chemical characteristic and chemical composition of meat were determined by the standard method (STN 570185). The content of water, protein and fat was estimated using a FoodScane TM – Meat Analyser (FOSS, Denmark) by the FT IR method (Fourier Transform infrared Spectroscopy); expressed in g.100g^{-1}. From these values, the energy value was calculated according to the equation of Strmiska *et al.* (1988):

Energy value (kJ.100g^{-1}) = 16.75 x protein content + 37.65 x fat content.

Water-holding capacity was determined by the compression method at constant pressure (Hašek and Palanská, 1976; Rafay *et al.*, 2008). The analysed samples MLTL (0.3 g in weight) were placed on previously weigthed filter paper (Schleicher and Shuell No. 2040B, Dassel, Germany) with tweezers. Together with the papers, the samples were sandwiched between Plexiglass plates and then subjected to pressure of 5 kg for 5 min. The results were calculated from the difference in weight between the slips with aspirating spots and the pure filter paper.

Microbiota enumeration

Faeces were sampled at day 0/1 (at the start of the experiment; 10 mixture samples from all rabbits – initial microbial background), at day 14 (2 weeks after the start of the experiment; 6 mixture samples from each group), at day 21 (56 days of age) and at day 49 (84 days of age), 6 mixture samples in each group. To test microbiota, the faecal samples (1 g) were treated using a standard microbiological method; dilutions in a Ringer solution (1:9, pH 7.0; Oxoid Ltd., Basingstoke, Hampshire, England) were prepared according to the International Standards Organization (ISO, 2001). The appropriate dilutions were plated onto Mannitol Salt Agar (ISO 6888, Difco) for coagulase-negative staphylococci (CoNS) and Baird-Parker agar supplemented with egg yolk tellurite solution (ISO 6888, Becton and Dickinson, Cockeysville, USA) to enumerate coagulase-positive staphylococci (CoPS). Brain-heart agar enriched with rifampicin was used to determine *S. epidermidis* P3/Tr2a. The plates were incubated at 37 °C for 48 h. Bacterial counts were expressed in colony-forming units per gram ± standard deviation (log_{10} CFU).

Statistical analysis

The results are represented as the mean ± standard deviation (SD). Mean values within the same row having different superscripts indicate significant difference using Tukey test ($p \leq 0.05$).

RESULTS AND DISCUSSION

The diet was composed of dehydrated lucerne meal, extracted rapeseed meal, wheat bran, oats, malt sprouts, DDGS (dried distillers grains with soluble), barley, sodium chloride, mineral and vitamin mixture and limestone. The chemical composition of the diet is presented in Table 1. The animals were found in good health conditions throughout the experiment. A significant increase in live weight was found in EG2 (Ent A/P) ($p \leq 0.01$) compared to CG. In the EG1 group (SE P3/Tr2a-methicillin resistant strain marked with reifampicin) a decreasing tendency in live weight was detected before SE P3/Tr2a strain cessation at the end of the experiment compared with the control animals.

Table 2. Effect of treatment on zootechnical parameters of rabbits (means ± SD)

Tested of parameters n = 22	Control	EG1 *S. epidermidis* P3/Tr2a	EG2 Ent EK13
Initial live weight (Day 35 of age; 1 day of experiment), g	1013 ± 119	1140 ± 140	1061 ± 151
Intermediate live weight (Day 21 of experiment), g	1760 ± 218	2015 ± 186[a]	1999 ± 212
Final weight (Day 84 of age), g	2648 ± 272	2871 ± 288	3044 ± 339
Daily gain, ($g.d^{-1}$)	34.10	36.10	41.30[A]
Feed conversion ratio between 1 and 21 days of the experiment, ($g.g^{-1}$)	2.64	2.79	2. 37
Feed conversion ratio between 22 and 48 days of the experiment, ($g.g^{-1}$)	3.90	4.26	4.17
Feed conversion ratio per kg gain	3.30	3.50	3.20
Carcass weight (Day 21), g (*n* = 4/group)	2006 ± 45	2203 ± 89	2305 ± 58
Carcass yield, % (Age at slaughter 56 d)	52.4 ± 0.85	53.84 ± 1.22	52.30 ± 0.65
Carcass weight (Day 47), g (*n* = 4/group)	2643 ± 165	2678 ± 78	2758 ± 109
Carcass yield, (%) (Age at slaughter 82 d)	55.83 ± 1.80[b]	52.85 ± 0.84	53.72 ± 1.96
Mortality (n)	3	1	1

[a, b] – $p \leq 0.05$; [A] – $p \leq 0.01$ significant differences from control

Average daily gain was higher in EG1 (6 %) and EG2 (21 %) compared to the CG: CG – 34.1 g, EG1 (*S. epidermidis* P3/Tr2a rif^{R}) – 36.1 g, EG2 (Ent A/P) – 41.3 g. Beneficial effect of Ent A/P on the growth of rabbits was observed. No significant differences were found between the experimental groups in carcass value in the fattening experiment (Table 2).

Regarding the processing technology para–meters and characteristics of *MLTL* muscles, only slight difference was found between the individual components, which corresponds to the results of other authors (Bianospino *et al.*, 2006; Dalla Zotte and Szendrő, 2011; Chrastinová *et al.*, 2016; Pogány Simonová *et al.*, 2015; Lauková *et al.*, 2016; 2017; Kalafová *et al.*, 2014; 2015).

The pH value of rabbit meat in the experimental group EG1 was lower compared to EG 2 but differences were not significant ($p > 0.05$). The pH value depends on the balance of muscle energy metabolism and represents a key role in the maintenance of meat quality during storage. It determines the environmental microbial balance because of the bacteriostatic effect of low pH on meat (Pogany Simonová *et al.*, 2010). In our study, the pH at 24 h *post-mortem* showing lower values in groups EG1 and EG2 compared to the control. However, pH at 48 h values were in agreement with the literature and no statistical differences among groups are reported.

The pH values (6.00 – log molc) 48 h *post-mortem* were obtained, which could be explained by depletion of glycogen reserve in muscles during refrigeration and by longer storage time. Meat pH affects many meat properties including water-holding capacity, muscle fat content and colour. Losses of water in meat, leading to increased pH and a decrease of the muscle protein, which is closer to the isoelectric point, result in a lower hydratation level.

Electric conductivity was increased in the CG group ($p \leq 0.05$) compared to EG1 and EG2. Colour lightness parameter was slightly decreased in the EG2 experimental group in comparison to CG and EG1.

Table 3. The selected processing technology parameters and chemical characteristic of *MLTL* muscles 24 h *post mortem* (mean ± SD; n = 4)

Parameter	Control	EG1 *S. epidermidis* P3/Tr2a	EG2
	at 56 days of age		
Content of water (g.100 g^{-1})	74.56 ± 0.22	74.97 ± 0.15	74.96 ± 0.37
Total proteins (g.100 g^{-1})	22.97 ± 0.38	22.84 ± 0.37	23.05 ± 0.19
Content of fat (g.100 g^{-1})	1.32 ± 0.12^{b}	1.16 ± 0.19	1.43 ± 0.20^{b}
Collagen (g.100 g^{-1})	0.72 ± 0.11	0.86 ± 0.12	0.78 ± 0.09
Energetic value (kJ.100 g^{-1})	434.30 ± 10.69	426.15 ± 11.13	439.84 ± 8.75
pH_{24} values	6.00 ± 0.02	5.99 ± 0.05	5.93 ± 0.04
Electric conductivity (µS.cm^{-1})	1.35 ± 0.27	1.52 ± 0.64	1.46 ± 0.68
Colour Lightness	46.21 ± 3.04	50.93 ± 2.06	49.76 ± 3.34
Water holding capacity (g.100 g^{-1})	26.15 ± 1.69^{c}	22.49 ± 4.03	21.0 ± 4.34
	at 82 days of age		
Content of water (g.100 g^{-1})	74.49 ± 0.64	74.19 ± 0.79	73.82 ± 0.53
Total proteins (g.100 g^{-1})	23.67 ± 0.48	23.70 ± 0.24	23.62 ± 0.21
Content of fat (g.100 g^{-1})	0.95 ± 0.15	0.84 ± 0.19	1.12 ± 0.22
Collagen (g.100 g^{-1})	0.72 ± 0.11	0.86 ± 0.12	0.78 ± 0.09
Energetic value (kJ.100 g^{-1})	431.97 ± 13.31	428.71 ± 6.26	437.84 ± 10.06
pH_{48} values	5.94 ± 0.03	5.99 ± 0.05	5.93 ± 0.04
Electric conductivity (µS.cm^{-1})	3.38 ± 0.68^{bc}	1.52 ± 0.64	1.46 ± 0.68
Colour Lightness	48.00 ± 3.99	49.20 ± 3.68	51.20 ± 5.47
Water holding capacity (g.100 g^{-1})	22.61 ± 4.52	26.83 ± 3.09	24.31 ± 3.16

$^{b, c}$ Means in the same row followed by different letters differ significantly $P < 0.05$; EG – experimental group.

Increases in the lightness (*L**) and yellowness (*b**) at day 82 of age were found in both experimental groups (EG1 and EG2) compared to CG. Higher values of yellowness could be connected to free radicals produced by lipid oxidation during storage and/or manipulation, which can oxidise hem pigments causing discolouration of meat and meat products (Münch, 2004). There is also a positive correlation between water-holding capacity and intramuscular fat content (Hernández *et al.*, 2000) as well as the ultimate pH (Lambertini *et al.*, 1996). Ent A/P treatment led to the increased concentration of total protein and fat in rabbit meat; energetic value of meat was also increased significantly in EG2. The leanest part of the body is the loin (MLD) with average lipid content of 1.8 g.100 g^{-1} (Dalle Zotte and Szendrő, 2011); these observations were also confirmed in this study, measured in *MLTL* samples.

The feed intake after *S. epidermidis* P3/Tr2a application can be explained by the increase in feed consumption by the animals. The application of *S. epidermidis* P3/Tr2a did not cause higher mortality and in the Ent A/P group of rabbits the same mortality was found. Young animals are usually susceptible to gastrointestinal diseases caused by feed-borne pathogens, particularly colibacillosis and clostridiosis (Takáčová *et al.*, 2012). Staphylococci are supposed to be the second most frequent infection in rabbits (Hermans *et al.*, 2003). However, they are also a part of microbiota in the digestive tract of rabbits. At day 14 (two weeks of Ent A/P application in EG2), staphylococcal count in faeces was well-balanced in all groups; their count was even the same compared to samples from day 0/1 (Table 4). In the faeces of EG1, *S. epidermidis* P3/Tr2a strain reached amount up to 10^2CFU.g^{-1} at day 21 (day 7 of its administration). However, count of *S. epidermidis* strain decreased until the end of experiment (up to 1.0 CFU.g^{-1}, log 10). The inhibitory activity of bacteriocins on methicilin-resistant staphylococci isolated from various sources is described in several works (Růžička *et al.*, 2004; Goller, Romeo, 2008; Freeman *et al.*, 1989; Chaieb *et al.*, 2007; Christensen *et al.*, 1982; Lauková *et al.*, 2017; 2020; Pogány Simonová *et al.*, 2020). No significant anti-staphylococcal effect of applied Ent A/P was noted in the presented experiment, contradictory to previously observed *in vitro* and *in vivo* results during enterocins administration (Lauková *et al.*, 2012, 2020; Pogány Simonová *et al.*, 2009, 2020).

CONCLUSION

It is obvious, that methicillin-resistant *S. epidermidis* P3/Tr2a did not have any negative effect on rabbit's health. Rabbits in both experimental groups even showed higher weight gain and lower mortality compared to control group. However, in rabbits with Ent A/P the highest daily weight gain was noted. In spite of the fact, that there was

Table 4. Counts of staphylococci and *S. epidermidis* P3/Tr2a strain (log10 CFU/g ± SD) in the faeces of rabbits and treatment with Ent EK13 = Ent A/P

Trial days	Parameter/group	Control	EG1 *S.epidermidis P3/Tr2a*	EG2 *Ent EK13*
Day 0/1	Staphylococci MSA	3.87 ± 0.17		
Day 14	Staphylococci MSA	3.27 ± 0.15	3.16 ± 0.54	3.41 ± 0.13
Day 21	Staphylococci MSA	3.60 ± 0.44	3.02 ± 0.36	3.30 ± 0.47
	BHI+R	ND	1.66 ± 1.20	ND
Day 48	Staphylococci MSA	4.12 ± 0.46	3.93 ± 0.34	4.13 ± 0.48
	BHI+R	ND	0.90 ± 0.00	ND

Day 0/1 = Sampling was performed at time 0 – at the beginning of the experiment MSA Staphylococci; BHI+R-Brain-heart infusion /agar enriched 100 µg rifampicin (rif^R) was used to detect the strain *S. epidermidis* SE P3/Tr2a; ND = not determined; $P > 0.05$; not significant differences from control; EG – experimental group

not significant reduction in SE strain, enterocin A/P could be used as a preventive supplement in rabbit husbandry.

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REFERENCES

AOAC. (2005). Official Methods of Analysis. 18th Edition. Association of Official Analytical Chemists, Gaithersburg, USA.

Bianospino, E., Wechsler, F. S., Fernandes, S., Roça, R. D. O., & Moura, A. S. A. (2006). Growth, carcass and meat quality traits of straightbred and crossbred Botucatu rabbits. *World Rabbit Science*, 14, 237–246. https://doi.org/10.4995/wrs.2006.563.

Cunha, S., Mendes, Ă., Rego, D., Meireles, D., Fernandes, R., Carvalho, A. & Martins da Costa, P. (2017). Effect of competitive exclusion in rabbits using an autochtochtonous probiotic. *World Rabbit Science*, 25, 123–134.

Dalle Zotte, A. & Szendro, S. (2011). The role of rabbit meat as functional food. *Meat Science*, 88, 319–331. DOI: 10.1016/j.meatsci.2011.02.017.

Falão-e-Cunha, L., Castro-Solla, L., Maertens, L., Marounek, M., Pinheiro, V., Freire, J. & Mourão, J. L. (2007). Alternatives to antibiotic growth promoters in rabbit feeding: A review. *World Rabbit Science*, 15, 127–140.

Freeman, D. J., Falkiner, F. R. & Keane, C. T. (1989). New method for detecting slime production by coagulase negative staphylococci. *Journal of Clinical Pathology*, 42(8), 872–874.

Goller, C. C. & Romeo, T. (2008). Environmental influences on biofilm development. *Current Topics in Microbiology and Immunology*, 322, 37–66.

Hašek, A. & Palanská, O. (1976). Determination of water holding capacity in meat by instruments at constant pressure. *Poultry Industry*, 18, 228–233. [in Slovak]

Hernández, P., Pla, M.,Oliver, M. A. & Blasco, A. (2000). Relationships between meat quality measurements in rabbits fed with three diets of different fat type and content. *Meat Science*, 55, 379–384. DOI: 10.1016/S0309-1740(99)00163-1.

Hermans, K., Devriese, L. A. & Haesebrouck, F. (2003). Rabbit staphylococcosis: difficult solutions for serious problems. *Veterinary Microbiology*, 91(1), 57–64.

Chaieb, K., Chehab, O. & Zmantar, T. (2007). *In vitro* effect of pH and ethanol on biofilm formation by clinical ica-positive *Staphylococcus epidermidis* strains. *Annals in Microbiology*, 57, 431–437.

Chrastinová, Ľ., Lauková, A., Chrenková, M., Polačiková, M., Formelová, Z., Kandričáková, A., Bino, E., Ščerbová, J., Bučko, O., Ondruška, Ľ., Szabóová, R. & Jurčík, R. (2018). Effects of Enterocin M and Durancin ED26E/7 substances applied in drinking water on the selected carcass characteristics and meat quality of broiler rabbits. *Archiva Zootechnica*, 21(1), 5–17.

Christensen, G. D., Simpson, W. A. & Bisno, A. L. (1982). Adherence of slime-producing strains of *Staphylococcus epidermidis* to smooth surfaces. *Infections and Immunology*, 37(1), 318–326.

Kalafová,A., Bulla, J., Bučko, O., Emrichová, J., Zbýňovská, K., Petruška, P., Schneidgenová, M., Chrastinová, Ľ., Ondruška, Ľ., Jurčik, R., Čupka, P., Mellen M. & Capcarová, M. (2014). Meat quality of female rabbits following administration of epicatechin. *Maso International*, 4(1), 71–75.

Kalafová, A., Bulla, J., Bučko, O., Emrichová, J., Zbýňovská, K., Petruška, P., Schneidgenová, M., Chrastinová, Ľ., Ondruška, Ľ., Jurčik, R., Mellen, M. & Capcarová, M. (2015). Quality of meat rabbits after application of epicatechin and patulin. *Potravinarstvo – Slovak Journal of Food Sciences*, 9(1), 124–132.

Kalma, R. P., Patel, V. K., Joshi, A., Umatiya, R. V., Parmar, K. N., Damor, S. V., Chauhan, H. D., Srivastava, A. K. & Sharma, H. A. (2016). Probiotic supplementation in rabbit: A review. *International Journal of Agriculture Sciences*, 8, 2811–2815.

Kellner, R., Jung, G., Hőrner, T., Zähner, H., Shnell, N., Entian, K. D. & Gőtz, F. (1988). Gallidermin: a new lanthionin-containig polypeptide antibiotic. *European Journal of Biochemistry*, 177(1), 53–59.

Lambertini, L., Bergoglio, G., Masoero, G. & Gramenzi, A. (1996). Comparison between Provisal and Hyla rabbit strains. I. Slaughtering performances and muscle composition.

In: *Proc. 6th WRSA Congres*, Toulouse, vol. 3, 195−199.

Lauková, A., Strompfová, V., Skřivanová, V., Volek, Z., Jindřichová, E. & Marounek, M. (2006). Bacteriocin-producing strain of *Enterococcus faecium* EK 13 with probiotic character and its application in the digestive tract of rabbits. *Biologia (Bratislava)*, 61(6), 779−782. DOI: 10.2478/s11756-006-0191-9.

Lauková, A., Pogány Simonová, M., Chrastinová, Ľ., Kandričáková, A., Ščerbová, J., Plachá, I., Čobanová, K., Formelová, Z., Ondruška, Ľ., Štrkolcová, G. & Strompfová, V. (2017). Beneficial effect of bacteriocin-producing strain *Enterococcus durans* ED 26E/7 in model experiment using broiler rabbits. *Czech Journal of Animal Science*, 62, 168−177.

Lauková, A., Chrastinová, Ľ., Pogány Simonová, M., Strompfová, V., Plachá, I., Čobanová, K., Formelová, Z., Chrenková, M. & Ondruška, Ľ. (2012). *Enterococcus faecium* AL 41: Its enterocin M and their beneficial use in rabbits husbandry. *Probiotics and Antimicrobial Proteins*, 2012, 4, 243−249.

Lauková, A., Pogány Simonová, M., Chrastinová, Ľ., Plachá, I., Čobanová, K., Formelová, Z., Chrenková, M., Ondruška, Ľ. & Strompfová, V. (2016). Benefits of combinative application of probiotic, enterocin M producing strain *Enterococcus faecium* AL41 and *Eleutherococcus senticosus* in rabbits. *Folia Microbiologica*, 61, 169−177.

Lauková, A., Bino, E., Kubašová, I., Strompfová, V., Miltko, R., Belzecki, G. & Pogány Simonová, M. (2020). Characterisation of Faecal Staphylococci from Roe Deer (*Capreolus capreolus*) and Red Deer (*Cervus elaphus*) and Their Susceptibility to Gallidermin. *Probiotics and Antimicrobial Proteins*, 12(1), 302−310.

Münch, S. (2004). Chemistry of fat and accompanying substances. *Fleichswirtschaft*, 4, 163−167. [in German]

Pogány Simonová, M., Chrastinová, Ľ. & Lauková, A. (2020). Autochtonous Strain *Enterococcus faecium* EF2019(CCM7420), Its Bacteriocin and Their Beneficial Effects in Broiler Rabbits: A Review. *Animals*, 10(7), art. no. 1188.

Pogány Simonová, M., Lauková, A., Žitňan, R. & Chrastinová, Ľ. (2015). Effect of rabbit-origin enterocin-producing probiotic strain *Enterococcus faecium* CCM7420 application on growth performance and gut morphometry in rabbits. *Czech Journal of Animal Science*, 60, 509−512.

Pogány Simonová, M., Chrastinová, Ľ., Kandričáková, A., Kubašová, I., Formelová, Z., Chrenková, M., Miltko, R., Belzecki, G., Strompfová, V. & Lauková, A. (2019). Enterocin M and sage supplementation in post-weaning rabbits: Effects on growth performance, caecal microbiota, fermentation and enzymatic activity. *Probiotics and Antimicrobial Proteins*, 2020, 12(2), 732−739. DOI: 10.1007/s12602-019-09584-z.

Pogány Simonová, M., Chrastinová, Ľ., Lauková, A., Strompfová, V., Faix, Š., Vasilková, Z., Ondruška, R., Jurčík, R. & Rafay, J. (2009). *Enterococcus faecium* CCM 7420, bacteriocin PPB CCM 7420 and their effect in the digestive tract of rabbits. *Czech Journal of Animal Science*, 54, 376−386.

Pogány Simonová, M., Chrastinová, L'., Mojto, J., Lauková, A., Szabóová, R. & Rafay, J. (2010). Quality of rabbit meat and phyto-additives. *Czech Journal of Food Science*, 28, 161−167.

Pogány Simonová, M., Lauková, A., Žitňan, R. & Chrastinová, Ľ. (2015). Effect of rabbit-origin enterocin-producing strain *Enterococcus faecium* CCM7420 application on growth performance and gut morphometry in rabbits. *Czech Journal of Animal Science*, 60(11), 509−512.

Pogány Simonová, M., Galová, N., Bino, E. & Lauková, A. (2020). Využitie enterocínov na inhibovanie rastu meticilín rezistentných stafylokokov (Enterocin use to inhibit methicillin resistant staphylococci). *Slovenský veterinársky časopis (Slovak Veterinary Journal)*, 45(1), 40−41.

Rafay, J., Novotná, K., Mojto, J., Bozic, A. & Chrenek, P. (2008). Some meat utility and quality traits of transgenic rabbit. *Slovak Journal of Animal Science*, 41(3), 121−125.

Růžička, F., Holá, V. & Votava, M. (2004). Biofilm detection and the clinical significance of *Staphylococcus epidermidis* isolates. *Folia Microbiologica*, 49(5), art. no. 596.

Strmiska, F., Holčíkova, K., Simonová, E., Mrázova, E., Hodeková, J., Votašaková, A., Pristašová, M., Strmiska, J., Strmisková, G., Kruparová, M., Gola, J., Papajová, H., Mareš, J., Kostkanová, E. & Ehrenhaft, B. (1988). Požívatinové tabuľky, I. − Potravinové suroviny, [Food Tables, I. − Food components]. *Journal of Food and Nutrition Research*, 189.

STN 570185. (1962). Slovak Technical Standards. Examination of meat, meat products and canned meat and prepared meats in cans. *Chemical and physical methods*, Bratislava, Slovakia.

Szabóová, R., Lauková, A., Chrastinová, Ľ., Pogány Simonová, M., Strompfová, V., Plachá, I., Čobanová, K., Vasilková Z. & Chrenková, M. (2011). Enterocin 4231 produced by *Enterococcus faecium* CCM 4231 and its use in rabbits. *Acta Veterinaria (Beograd)*, 61(5-6), 523–529. DOI: 10.2298/AVB 1106523S.

Szabóová, R., Chrastinová, Ľ., Lauková, A., Haviarová, M., Simonová, M., Strompfová, V., Faix. Š., Vasilková, Z., Chrenková, M., Plachá, I., Mojto, J. & Rafay, J. (2008). Bacteriocin – prodicing strain *Enterococcus faecium* CCM4231 and its use in rabbits. *International Journal of Probiotics & Prebiotics*, 3(2), 77−82.

Takáčová, J., Ryzner, M., Plachá, I., Čobanová, K., Faixová, Z. & Faix, Š. (2012). *Thymus vulgaris* essential oil in poultry nutrition. In: *Book of Abstracts from 25th Days of Animal Physiology*, 17-19 October; Košice, Slovakia, p.72, ISBN: 978-80-968618-8-0.

Van Soest, P. J., Robertson, J. B. & Lewis, B. A. (1991). Methods for dietary fiber, neutral detergent fiber, and nonstarch polysaccharides in relation to animal nutrition. *Journal of Dairy Science*, 74, 3583−3597. https://doi.org/10.3168/jds.S0022-0302(91)78551-2.