Enterocin 55 produced by non rabbit-derived strain *Enterococcus faecium* EF55 in relation with microbiota and selected parameters in broiler rabbits

Andrea Lauková[1], Ľubica Chrastinová[2], Iveta Plachá[3], Renáta Szabóová[4], Anna Kandričáková[5], Monika Pogány Simonová[6], Zuzana Formelová[7], Ľubomír Ondruška[8], Mária Goldová[9], Mária Chrenková[10], Viola Strompfová[11]

[1,3,5,6,11]Institute of Animal Physiology Slovak Academy of Sciences, Košice
[2,7,8,10]National Agricultural and Food Centre, Nitra-Lužianky, Slovakia
[4,9]University of Veterinary Medicine and Pharmacy, Košice, Slovakia

***Abstract*—** Ent55 is produced by poultry strain *Enterococcus faecium* EF55. It is substance which can be allotted to Class II enterocins; thermo-stable, small peptide. Because producer strain has shown beneficial effect in poultry and broiler rabbits as well, we decided to apply Ent55 in broiler rabbit husbandry. Ent55 showed antimicrobial activity in broiler rabbits by reduction of staphylococci, *Clostridiae*, pseudomonads and coliforms. Its beneficial effect was demonstrated by stimulation of phagocytic activity as well as by reduction of *Eimeria* spp. oocysts. GPx values were lower; it means, no oxidative stress was evoked. Moreover, it has not negative influence on growth performance and biochemical parameters. Our results indicated that enterocin produced by not-autochtonous strain can also have protective and beneficial effect in broiler rabbits.

***Keywords*— Enterocin, effect, microbiota, immunity, rabbit.**

I. INTRODUCTION

Ent55 is produced by poultry strain *Enterococcus faecium* EF55 (1, 2). It is substance which can be allotted to Class II enterocins; thermo-stable, small peptide. Its maximum inhibition activity achieved after 12 h of cultivation in broth was 12 800 AU/ml. Ent55 has a broad antimicrobial spectrum and its inhibition activity *in vivo* was demonstrated in Japanese quails; but its producer strain was shown to have also benefits in chicken. That is, Levkut et al. (3) reported inhibition activity of EF55 strain against *Salmonella* Enteritidis PT4 in chicks. Administration of EF55 strain to chicks challenged with *Salmonella* Enteritidis (SE147) resulted in increased number of blood heterophils, caecal IgA+ IEL (4). This strain also demonstrated positive impact on intestinal morphometry in the jejunum (5). To have this information we decided to test Ent55 effect in broiler rabbits. The aim was to spread information concerning the Ent55 but also to find beneficial activity in rabbits by this non rabbit-derived strain producing Ent55. In last decade, bacteriocins (enterocins) have been tested as biopreservative substances in food industry to prevent contamination by spoilage bacteria (6, 7). However, at recent time, more frequently have been presented studies with application of bacteriocins to protect animals health including our studies (8, 9, 10, 11) .

II. MATERIAL AND METHODS

2.1 Experiment design

Forty-eight *post*-weaned rabbits (aged 5 weeks, Hyplus breed) both sexes were divided into the experimental group (EG) and the control group (CG); 24 animals in each group. Animals were kept in the standard cages (0.61m x 0.34 cm x 0.33 m, the type D-KV-72 supplied by Kovobel, Czech Republic), two animals per cage. The cages allowed faeces separation. A cycle of 16 h of light and 8 h of dark was used throughout the experiment. The temperature and humidity were recorded continuously with a digital thermograph positioned at the same level as the cages. The heating and forced ventilation systems allowed the building air temperature to be maintained within 16 ± 4ºC through the experiment. The relative humidity was about 70 ± 5 %. All care and experimental procedures were approved by the Slovak Veterinary and Food Administration. The experiment was performed in co-operation with our colleagues in Nitra-Lužianky (National Agricultural and Food Centre, Slovakia). The rabbits were fed the commercial feed mixture for broiler rabbits (pelets 3.5 mm in average size) characterized by the contain of 901.75 g/kg dry matter, 828.29 g/kg of organic matter, 176.02 g/kg of fibre content, 155.89 g/kg of N-substances, 34.65 g/kg of fat content, 73.46 g/kg of ash content; digested energy was 11.01 MJ/kg and metabolised energy 10.46 MJ/kg. Samples of individual feeds and complete granulated mixture were analyzed according to STN 46 7092 (Slovak Technical Norm, 2010). The rabbits had access to feed and water *ad libitum.* Water and feed consumption by the rabbits were

controlled daily. Rabbits in EG were administered enterocin 55 (semi-purified) prepared as previously reported Strompfová and Lauková (2). Its activity was checked by agar diffusion method (13) against the indicator strain *Enterococcus avium* EA5 (our strain). Ent55 was applied in the water at a dose 50 µl per animal per day for 21 days (3 weeks). The experiment duration was 42 days.

2.2 Sampling and microbial analyses

Faecal sampling was carried out on day 0-1 (the start of experiment; 10 mixture samples from 48 rabbits – initial microbial background), on day 21 (3 weeks of enterocin 55-Ent55) application; five mixture samples from each group) and on day 42; the end of experiment, 3 weeks after Ent 55 cessation; five mixture samples from each group). The microbial counts were checked to follow antimicrobial effect of Ent55 using the standard microbiological method (ISO, International Organization for Standardization): the appropriate dilutions of samples in Ringer solution (pH 7.0; Oxoid Ltd., Basingstoke, Hampshire, United Kingdom) were plated onto media according to ISO: M-Enterococcus agar for enterococci (ISO-7889, Difco, Detroit, USA), Baird-Parker agar supplemented with egg yolk tellurite solution (ISO 21527-1, Becton and Dickinson, Cockeysville, USA) for isolation of coagulase-positive staphylococci (CoPs), Mannitol Salt agar, *Clostridium difficile* agar with the supplement SR0096E and 7% (v/v) defibrinated horse blood (SR0050, ISO 15883, Oxoid Ltd., Basingstoke, Hampshire, United Kingdom) were used to enumerate coagulase-negative staphylococci (CoNS) and *Clostridium* spp. Mac Conkey agar (Oxoid) was used to detect coliform bacteria. *Pseudomonads* were isolated on Cetrimide agar (ISO 16266, Becton and Dickinson). The plates were incubated at 30 ºC and/or 37 ºC for 24-48 h depending on the bacterial genera. The bacterial counts were expressed in colony-forming units (log10) CFU per gram ± SD. On day 21 and 42, three animals (n=3) from each group were slaughtered. They were electrically stunned and killed by cutting the carotidis and jugular veins (14). One g of caecal content and appendix was treated according to the standard microbiological dilution method mentioned above. Caecal and appendix samples were plated on the media as already indicated.

2.3 Blood analysis including phagocytic activity and enzyme gluthation-peroxidase

Blood (n=6) was sampled from the marginal ear vein (*vena auricularis*) on day 0-1, mixed blood samples, and on day 21, 42 from each group. Phagocytic activity (PA) was measured using the direct counting procedure with microspheric hydrophilic particles (MSHP). Ingestion of MSH particles by polymorphonuclear cells (PMNs) was determined with a modified test (15): 50 ml of MSH particle suspension (ARTIM, Prague, Czech Republic) was mixed with 100 ml of blood in an Eppendorf-type test tube and incubated at 37°C for 1 h. Blood smears were then prepared and stained by May-Grünwald and Giemsa-Romanowski (Merck, Germany). In each smear, 100 cells were observed to determine the relative number of white cells containing at least three engulfed particles (phagocytic activity), and the index of phagocytic activity (IPA, number of engulfed particles/total number of neutrophils and monocytes observed). The percentage of phagocytic cells was evaluated using an optical microscope (Motic BA 400 Biological Microscope- Motic China Group Co., Ltd.) by counting PMN up to 100.

The activity of glutathione-peroxidase (GPx; U/gHb) in blood (n=6) was determined using a specific commercial kit (RANSEL, Randox, United Kingdom) according to the spetrophotometric assay procedure of Paglia and Valentine (16).

Total proteins (g/l), albumine (g/l), triglycerides, cholesterol, calcium (mmol/l), glucose (g/l), alanine-aminotranspherase (µkat/l) were performed using commercial kits from Randox (United Kingdom). Blood (n=6, from each group) was sampled into dry non-heparinized Ependorf tubes; blood serum was separated by centrifugation at 3000 x *g* for 10 min, then stored frozen in plastic vials until analysis.

2.4 *Eimeria* oocysts counting, acids in caecal content, growth performance

To detect *Eimeria* sp. oocysts the quantitative MacMaster method was used (1968); oocysts counts were expressed in OPG/g (detected oocysts per gram of faecal sample). Lactic acid and organic acids (acetic, propionic and butyric) were evaluated using gas chromatography (Perkin Elmer, USA). Weight mass, feed conversion amd mortality were checked, measured and mathematically calculated.

Statistical evaluation of the results was performed using one-way ANOVA test with Tukeys post hoc test (the level of significance set at $p<0.05$ ± standard deviation - SD).

III. RESULTS

Antimicrobial activity of Ent55 in faeces was demonstrated by significant reduction of CoNS in EG on day 21 compared to EG on day 42 (2. sampling to 3. sampling, a:b, $p<0.001$, Table 1). *Pseudomonas* spp. in EG were reduced in 2. sampling-day 21 compared to day 0-1 (1. sampling, a:b, a:c, $p<0.05$). Coliforms were reduced in EG on day 42 (sampling 3.) compared to day 0-1 (1. sampling, a:c, $p<0.05$; d:c $p<0.05$). On day 21 coliforms were reduced significantly in EG compared to CG (d:e, $p<0.05$) with reductive tendency on day 42 (c:f, $p=0.054$).

TABLE 1
PROFILE OF BACTERIA IN FACES OF BROILER RABBITS DURING THE EXPERIMENT (LOG10 CFU/G) ± SD

Sampling	EG	CG
(1) Day 0-1 (n=8)		
Enterococci	3.20 (0.36)	3.20 (0.36)
LAB	3.02 (0.46)	3.02 (0.46)
CoNS	3.50 (0.10)[a]	3.50 (0.10)
CoPS	2.20 (0.23)	2.20 (0.23)
Cl. spp.	2.71 (0.40)	2.71 (0.40)
Ps.spp.	5.43 (0.90)[a]	5.43 (0.90)
Coliforms	4.50 (1.63)[a]	4.50 (1.63)
(2) Day 21 (n=5)		
Enterococci	3.47 (0.23)	3.56 (0.83)
LAB	3.21 (0.70)	2.83 (0.77)
CoNS	3.30 (0.15)[b]	3.06 (0.58)
CoPS	2.43 (0.24)	2.24 (0.09)
Cl. spp.	4.54 (0.82)	4.63 (0.90)
Ps.spp.	4.18(0.30)[b]	4.44 (0.62)
Coliforms	2.95 (0.65)[d]	3.95 (0.37)[e]
(3) Day 42 (n=5)		
Enterococci	3.85 (0.69)	2.68 (1.11)
LAB	3.28 (0.89)	2.23 (1.32)
CoNS	4.04 (0.08)	3.96 (0.21)
CoPS	2.19 (0.92)	2.14 (0.53)
Cl. spp.	4.81 (1.44)	5.80 (0.90)
Ps.spp.	4.32 (0.88)	4.89 (0.12)
Coliforms	1.53 (0.84)[c]	3.23 (1.46)[f]

EG-experimental group, CG-control group, Day 0-1, start of experiment, Day 21, after 3 weeks of application, Day 42, end of experiment-3 weeks after cessation, LAB-lactic acid bacteria, CoNS-coagulase-negative staphylococci, CoPS, coagulase-positive staphylococci, *Cl.* Spp.-*Clostridium* spp.; Ps spp.-*Pseudomonas* spp., Coliform bacteria; Significant reduction in CoNS of 1. sampling compared to 2. sampling (a:b, $p<0.05$); *Pseudomonas* spp. were lower in 1. sampling compared to 2. sampling (a:b, $p<0.05$), Coliforms were lower in 3. sampling compared to 1. sampling (a:c, $p<0.01$); On day 21, coliforms were significantly reduced in EG compared to CG (d:e, $p<0.05$) with reduction tendency on day 42 (c:f, $p=0.054$);

Microbial counts in caecum and appendix were lower than in faeces of broiler rabbits. Similarly as in faeces, the counts of CoNS were reduced in EG on day 21 (2. sampling) compared to day 42 (3. sampling, a:b, $p<0.01$, Table 2). On day 21, *Pseudomonas* spp. were lower in EG compared to CG (b:b, difference 0.64 log cycle). Coliforms were also lower in EG compared to CG on day 21 (c:c, difference 1.64 log cycle). On day 42, *Clostridium* spp. in EG and coliforms were lower compared to CG with difference 0.46 log cycle (d:d) and 1.66 log cycle (e:e).

TABLE 2

PROFILE OF BACTERIA IN CAECUM OF BROILER RABBITS DURING THE EXPERIMENT (log10 CFU/g) ± SD

Sampling	EG	CG
(2) Day 21 (n=3)		
Enterococci	1.09 (0.33)	1.36 (0.63)
LAB	1.37 (0.72)	1.23 (0.41)
CoNS	3.49 (0.17)[a]	3.77 (0.08)
CoPS	2.27 (0.29)	1.94 (0.42)
Cl. spp.	5.10 (0.00)	5.10 (0.00)
Ps.spp.	3.71(0.23)[b]	4.35 (0.43)[b]
Coliforms	1.94 (1.50)	3.58 (1.34) [c]
(3) Day 42 (n=3)		
Enterococci	3.85 (0.69)	1.54 (0.60)
LAB	3.28 (0.89)	1.27 (0.35)
CoNS	4.04 (0.08)[b]	3.87 (0.29)
CoPS	2.19 (0.92)	1.95 (0.42)
Cl. spp.	4.81 (1.44)[d]	5.27 (0.29[d]
Ps.spp.	4.32 (0.88)	4.44 (0.51)
Coliforms	1.53 (0.84)[e]	3.19 (1.98[e]

EG-experimental group, CG-control group, Day 21, after 3 weeks of application, Day 42, end of experiment-3 weeks after cessation, LAB-lactic acid bacteria, CoNS-coagulase-negative staphylococci, CoPS, coagulase-positive staphylococci, *Cl.* Spp.-*Clostridium* spp.; Ps spp.-*Pseudomonas* spp., Coliform bacteria; On day 21 (2. sampling) CoNS in EG were lower than at day 42 (3. sampling (a:b, $p<0.01$); at day 21, *Pseudomonas sp*p. were lower in EG compared to CG (b:b, difference 0.64 log cycle); Coliforms in EG were lower than in CG (c:c, differemce 1.64 log cycle); On day 42, in EG *Clostridium* spp., and coliforms were lower than in CG with differences 0.46 log cycle (d:d), 1.66 log cycle (e:e);

Coliforms were reduced also significantly in appendix of EG on day 21 compared to day 42 (a:b, $p<0.05$, Table 3). On day 42, *Clostridium* spp., P*seudomonas* spp. and coliforms were lower in EG compared to CG with differences 0.87 log cycle (c:c), 1.07 log cycle (d:d) and 1.11 log cycle (b:e). CoPS, enterococci and LAB were neither influenced by Ent55 in faeces, nor in caecum and appendix.

TABLE 3

PROFILE OF BACTERIA IN APPENDIX OF BROILER RABBITS DURING THE EXPERIMENT (log10 CFU/g) ± SD

Sampling	EG	CG
(2) Day 21 (n=3)		
Enterococci	3.00 (0.12)	1.47 (0.53)
LAB	1.90 (1.07)	2.33 (1.24)
CoNS	2.78 (0.48)	2.34 (0.25)
CoPS	2.60 (0.71)	1.57 (0.58)
Cl. spp.	5.06 (0.03)	5.04 (0.00)
Ps.spp.	4.60(0.87)	5.10 (0.00)
Coliforms	2.28 (1.21)[a]	5.10 (0.00)
(3) Day 42 (n=3)		
Enterococci	1.74 (1.00)	1.63 (0.50)
LAB	1.10 (0.20)	0.90 (0.00)
CoNS	3.50 (0.17)	3.33 (0.25)
CoPS	1.62 (0.13)	1.83 (0.33)
Cl. spp.	4.23 (1.03)[c]	5.10 (0.00)[c]
Ps.spp.	4.11 (0.73)[d]	5.18 (1.48)[d]
Coliforms	3.68 (2.60)[b]	4.79 (2.16)

EG-experimental group, CG-control group, Day 21, after 3 weeks of application, Day 42, end of experiment-3 weeks after cessation, LAB-lactic acid bacteria, CoNS-coagulase-negative staphylococci, CoPS, coagulase-positive staphylococci, *Cl.* Spp.-*Clostridium* spp.; *Ps*. spp.-*Pseudomonas* spp., Coliform bacteria; reduction of coliforms in EG on day 21 compared to EG on day 42 (a :b, $p<0.05$), On day 42, reduced counts of *Clostridium* spp., *Pseudomonas* spp. and coliforms in EG compared to CG with differences 0.87 log cycle (c:c), 1.07 log cycle (d:d), 1.11 log cycle (b:e).

On day 21, phagocytic activity was stimulated in EG compared to CG by significant increase (a:a, $p<0.01$) as well as on day 42 (b:b, $p<0.001$, Table 4). The values of GPx were as follows: on day 21 in EG-146.70 (22.93) compared to CG-144.14 (20.73 U/g Hb). On day 42 they were decreased in both, EG-128.08 (34.94) U/g Hb; 124.76 (30.78) IN CG.

TABLE 4
PHAGOCYTIC ACTIVITY IN EXPERIMENTAL AND CONTROL RABBITS AND INDEX OF PHAGOCYTIC ACTIVITY (%) ± SD

	EG	CG
PA21	49.00 (2.37)[a]	43.83 (2.40)[a]
PA42	50.50 (1.05)[b]	43.33 (2.50)[b]
IPA21	2.35 (0.15)	2.33 (0.14)
IPA42	2.20 (0.18)	2.13 (0.23)

On day 0-1, PA was 43.00 (1.67)% and IPA 2.30 (0.17); On day 21, EG:CG, a:a, $p<0.01$; On day 42, EG:CG, b:b, $p<0.001$; EG-experimental group-Ent55 application, CG-control group; PA21- phagocytic activity on day 21 (3 weeks application); PA42-phagocytic activity on day 42 (3 weeks cessation); IPA-index of phagocytic activity;

Biochemical parameters, such as total proteins, albumine, triglycerides, cholesterol, glucose, ALT and Ca were influenced in the range of reference values; wether higher, they were decreased or increased wether low (Table 5). On day 21 and 42, decrease of *Eimeria* oocysts was noted in EG rabbits compared to CG (Table 6). EG samples compared to CG were oocysts absent; in EG oocysts were not counted. On day 21, acids values in caecal contents were slightly increased (Table 7).

TABLE 5
BIOCHEMICAL PARAMETERS IN BLOOD SERUM

n=6	**EG**	**CG**
TP (g/l)		
Day 0-1	48.5 (2.9)	48.5 (2.9)
Day 21	52.9 (5.0)	53.2 (1.9)
Day 42	57.3 (2.5)	56.3 (3.3)
ALB (g/l)		
Day 0-1	41.8 (5.2)	41.8 (5.2)
Day 21	45.0 (3.7)	42.6 (1.4)
Day 42	44.7 (2.9)	46.9 (2.5)
TRIG (mmol/l)		
Day 0-1	2.13 (0.33)	2.13 (0.33)
Day 21	2.00 (0.50)	1.74 (0.19)
Day 42	1.83 (0.13)	1.90 (0.31)
CHOL (mmol/l)		
Day 0-1	3.57 (0.54)	3.57 (0.54)
Day 21	3.26 (0.41)	3.02 (0.21)
Day 42	2.75 (0.26)	2.83 (0.27)
GLU (g/l)		
Day 0-1	7.69 (0.37)	7.69 (0.37)
Day 21	8.52 (1.08)	8.60 (1.16)
Day 42	7.69 (0.39)	7.44 (0.56)
ALT (µkat/l)		
Day 0-1	0.05 (0.02)	0.05 (0.01)
Day 21	0.11 (0.04)	0.10 (0.02)
Day 42	0.09 (0.03)	0.12 (0.04)
Ca (mmol/l)		
Day 0-1	2.89 (0.10)	2.89 (0.10)
Day 21	3.07 (0.09)	3.15 (0.19)
Day 42	3.15 (0.15)	3.12 (0.09)

TABLE 6
EIMERIA OOCYSTS IN OOCYST PER GRAM OF FAECES (OPG/g)

Sampling	EG	CG
Day 21 (n=5)	neg	1760 (41.95)
Day 42 (n=5)	neg	600 (28.63)

EG-experimental group, CG-control group, Day 21, after 3 weeks of application, Day 42, end of experiment-3 weeks after cessation

TABLE 7
LACTIC ACID AND OTHER ACIDS IN CAECAL CONTENT OF BROILER RABBITS

	EG	CG
Day 21		
LA (g/100g)	0.084	0.097
AA (mmol/l)	10.908	7.637
PA (mmol/l)	0.615	0.345
BA (mmol/l)	1.921	1.488
Day 42		
LA (g/100g)	0.102	0.106
AA (mmol/l)	7.628	7.201
PA (mmol/l)	0.377	0.399
BA (mmol/l)	1.634	1.451

EG-experimental group, CG-control group, Day 21, after 3 weeks of application, Day 42, end of experiment-3 weeks after cessation; LA-lactic acid, AA-acetic acid , PA-propionic acid, BA-butyric acid

An average live weight mass at the end of experiment in rabbits of EG reached 2796.33 (152.47) g. In CG it was slightly less-2624.76 (68.24) g. It was reached from the initial values of 985.00 (105.82)g for CG and 984.17 (113.75) g for EG. No mortality was observed. Regarding the feed conversion in the period of 3 weeks, it was 2.91 g per g live weight mass in CG and in EG it was 2.45 g/g of live weight mass.

IV. DISCUSSION

Bacteriocins were identified almost a century ago; it is believed that 99% of bacteria produce at least one bacteriocin (18). The kingdom of bacteriocins is rich and contains a diversity of small proteins from Gram-positive and Gram-negative bacteria. Several bacteria produced by lactic acid producing Firmicutes offer potential application (18). Bacteriocins are even anticipated to be good candidates for therapeutic application in order to fight against pathogenic bacteria (19). The genus Enterococcus belongs to Firmicutes and a large number of bacteriocins produced by enterococci (mostly enterocins) have been described and characterized at the biochemical and genetic level (20, 21, 22, 23, 24, 25). Enterocins are known to have frequently a broad antimicrobial spectrum. Many *in vitro* studies were related to this statement (20, 24, 22, 23). However, concerning the application abilities in animals to protect husbandry has been reported only limitedly. Our working group has published several beneficial effects reached by different enterocins produced by different strains in especially food-producing animals such as broiler rabbits, poultry but also in horses (8, 9). Previously, bacteriocins were characterized that they act against closely related bacteria (26) but many studies have spread last decade this statment because e.g. enterocins can inhibit more or less related bacteria, spoilage bacteria including (27). Ent55 also showed antimicrobial inhibition effect against Gram-positive CoNS and *Clostridium* spp.; however, coliform bacteria and *Pseudomonads* were also inhibited by significant reduction or by bacteriostatic effect on their growth. Similarly as Ent55, our other enterocins confirmed this inhibition; it is e.g. Ent M (produced by probiotic strain *E. faecium* AL41-CCM8558) reduced staphylococci, coliforms, *Pseudomonas* spp. and Clostridiae (28) or firstly described Ent4231 produced by *E. faecium* CCM4231- animal/ruminal origin strain also showed antimicrobial activity (8). In spite of the fact that amino sequence analysis of Ent55 is not tested, their properties are more similar to Class II enterocins. Moreover, producer strain of Ent55 (*E. faecium* EF55) was very active in chicken (1). Interesting is inhibition of bacteria in appendix. Appendix has importance from the aspect of differentiation and forming of lymphoid cells which after migration in the secondary lymphatic organs (spleen, lymphatic nodes and lymphoid tissue of the digestive tract-GALT) are multiplied (29). It shows that appendix can play role in eliminating and killing of undesirable microbiota performing by this protection of organism.

Phagocytic activity is important parameter; there polymorphonuclear leucocytes are responsible for non-specific immune response and in first line share of phagocytosis introdefence of the host to infectious and inflammatory actions (30). This non-specific immunity has been positively-increasingly influenced in our previous studies by using the other enterocins such as Ent4231 or Ent2019 or EntM (8, 9, 10). We could suppose that rabbits adopt to substance because prolonged stimulative effect on PA was measured. This stimulative and protective effect of enterocins could be associated with increase of PA. Moreover, oxidative stress was not evoked by additive. In the case of EntM-producing strain AL41 in poultry even lower counts of GPx were measured in EG compared to CG (31). Very important and original is finding that *Eimeria* oocysts were reduced. We dont know exact mode of action; however, it could be explained probably through the immunity stimulation; it means higher immunity, less sensitivity to agens as *Eimeria* also are. Pogány Simonová et al. (32) Szabóová et al. (8) also reported reduction of *Eimeria* oocysts in case of Ent2019 or/and Ent4231; it was studied at first in association with *Eimeria* oocyst reduction in rabbits. Caecal acids were increased only in case of acetic acid. Biochemical parameters reported were in accordance with those reported by Lauková et al. (9) after EntM application with any significant impact. Effect of enterocins on growth performance is varied from Ent to Ent used. However, it seems that this parameter is more influenced by Ent-producing strains than by enterocins themselves.

V. CONCLUSION

Ent55 showed antimicrobial activity in broiler rabbits by reduction of staphylococci, *Clostridiae*, pseudomonads and coliforms. Its beneficial effect was demonstrated by stimulation of phagocytic activity as well as by reduction of *Eimeria* spp. oocysts. GPx values were lower; so no oxidative stress was evoked. Moreover, it has not negative influence on growth performance and biochemical parameters. Our results indicated that also enterocins produced by not-autochtonous strain can have protective and beneficial effect in broiler rabbits.

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