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EFFECT OF SIMULTANEOUSLY ADMINISTERED SALINOMYCIN AND FLAVOPHOSPHOLIPOL UPON THE PERFORMANCE OF BROILER CHICKENS

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Summary

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The combination of flavophospholipol (FPL) and salinomycin (SLM) – the formulation Flavosalin-premix (FLS), administered with fodder at a level of 65 mg/kg forage for 56 days in broiler chickens, resulted in increased live weight gain by 2.9% for both genders (2.6% in male birds and 4.8% in females). The administration of the drug decreased the forage consumption and improved feed conversion ratio by 1.1% and 4% respectively. There was a tendency towards increase in the yield and decrease in the weight of viscera and bones (including fasciae) on the account of the muscle mass.

The treatment with Flavosalin-premix caused favourable changes in the chemical composition of meat – increased protein content by 3.48% in white musculature and by 1.32% in red musculature as well as decreased fat content (by 1.83%) and decreased moisture content in white musculature (4.62%). It did not result in deposition of flavophospholipol residues in the musculature and viscera, and salinomycin traces disappeared at the 6th day.

Key words: anticoccidial drugs, drug interactions, flavophospholipol, flavosalin, salinomycin

INTRODUCTION

It is known that the control of coccidiosis as one of the most problematic diseases in broiler chickens, is related to the necessity of continuous application of coccidiostatics. In some cases, this requires a simultaneous use of other medications, including ergotropic drugs; for instance, the application of the coccidiostatic drug salinomycin (SLM) and the ergotropic drug flavophospholipol (FPL).

Salinomycin is an ionophore antibiotic with a broad spectrum of activity against all important avian coccidial species, including strains, resistant to other coccidiostatics (Morrison et al., 1979; Yvore et al., 1980; Hamman et al., 1993). The risk of appearance of resistance towards SLM is small (Migaki et al., 1978; Chappel & Babcock, 1979).

Flavophospholipol is a glycopeptide antibiotic, active mainly against Gram-positive organisms (von Wasielewski et al., 1965; von Wasielewski & Lembke, 1969). It enhances the live weight gain and the productivity contributing to the better utilization of forage (Turek et al., 1967; Krüger & Vollrath, 1969; Vogt, 1969; Vogt & Gürocak, 1970). No cross resistance with other antibiotics is observed (Lembke & von Wasielewski,

1969). It also helps to restore the sensitivity of some microorganisms that acquired resistance to other antibacterial drugs through elimination of R-plasmid resistance (Spring, 1978; George et al., 1982; Pilloud, 1983). The drug interaction and especially the rate of the ergotropic effect, observed during the simultaneous application of FPL and SLM, is a topic of both theoretical and practical importance.

Thus we performed studies upon the formulation "Flavosalin-premix" (FSL) containing FPL and SLM intended for prophylaxis of coccidiosis in chickens and for elimination of R-plasmid resistance in infections caused by *Escherichia coli*, salmonellae etc.

MATERIALS AND METHODS

Animals

The experiments were performed on 140 broiler chickens, 1 day–old, 4-line Cornish and Plimouthrock hybrids from both genders and with initial weight of 38.9–40.3 g.

The birds were housed in battery cages on a grided flour under hygienic conditions corresponding to the age cathegories. The feeding was in a group, with a standard compound feed according to Bulgarian State Standard. Depending on the age, the feeding of the chickens included 2 periods: from day 1 to day 35[1] and from day 36 to day 56[2].

The chickens had free access to both forage and water.

Drugs

The used drugs were as followed:

- Halofuginone (HLF) (Stenorol, STR containing 0.6% or 12% active substance, Scientific Institute of Chemistry and Pharmaceutics, Sofia, Bulgaria);
- FPL (Pharmastim-premix, containing 0.25% FPL, Biovet Co., Peshtera, Bulgaria);
- SLM (Salinopharm premix, containinig 3% SLM, Biovet Co., Peshtera, Bulgaria);
- Combination of FPL and SLM (Flavosalin-premix, FLS, containing 0.25% FPL and 3% SLM, Biovet Co., Peshtera, Bulgaria)

Experimental design

The chickens were divided into 7 groups with 20 birds (both genders) each with equal body weight: 1 group negative control, 5 groups positive controls and 1 experimental group:

- Group I: negative control, not treated;
- Group II: positive control, treated with HLF (0.6% or 12% STR)[3] in dosage rate of 3 mg/kg forage[4];
- Group III: positive control, treated with FPL at 5 mg/kg forage[4];
- Group IV: positive control, treated with FPL at 5 mg/kg forage and HLF (0.6% or 12% STR) in dosage rate of 3 mg/kg forage[4];
- Group V: positive control, treated with SLM at 60 mg/kg forage[4];

[1] Composition of the forage, used within the first period: 67% corn, 6% sunflower meal, 17.6% soya meal, 2.5% meat and bone meal, 1.5% yeasts, 0.11% lysine, 1.56% dicalcium phosphate, 0.65% lime stone, 0.3% salt, 0.2% trace elements mixture for birds, 0.5% premix No. 5, 2% fat and 0.018% santoquine.

[2] Composition of the forage used within the second period: 30% "Konnie" corn, 31.75% wheat, 6% sunflower meal, 8% soya meal, 17% "Tila" meal, 4% meat and bone meal, 0.6% premix No. 5-91, 0.2% trace elements mixture for birds, 0.6% lime stone, 1.5% dicalcium phosphate, 0.3% salt.

[3] Chickens received 0.6% HLF up to the age of 3 weeks and thereafter – 12% HLF.

[4] The concentration refers to the active substance(s).

- Group VI: experimental, treated with the combination FPL+SLM as FSL at 65 mg/kg forage[5] (SLM at 60 mg/kg forage and FPL at 5 mg/kg forage);
- Group VII: positive control treated with the combination FPL+SLM as FSL at 65 mg/kg forage[5] (SLM at 60 mg/kg forage and FPL at 5 mg/kg forage) and HLF (0.6% or 12% STR)[6] at 3 mg/kg forage[5].

All groups of chicken were treated with the forementioned drugs through their daily addition to forage for one fattening period.

The general condition and behaviour of birds were monitored daily. At the age of 14, 28, 35 and 56 days the birds were weighed (after 16 h fasting). The body weight, the total and average daily live weight gain (averaged for both genders and separately for males and females), the forage consumption of one chicken and the feed conversion ratio per 1 kg live weight gain were recorded.

Slaughtering data were analysed. For this purpose, at day 56, 6 birds from each of groups I, III, V and VI were euthanized. The live weight (after 16 h fasting), the weight of plucked carcass without head and legs (under the tarsal joints), the weight of plucked carcass without viscera, the weight of viscera and bones with fasciae were determined. Specimens from the white and red muscles of 6 birds were collected for determination of moisture, protein, fat and mineral content (according to Pozhariskaja et al., 1964). For those parameters, pooled samples were used.

The residues of FPL and SLM in musculature, some viscera (liver and kidneys) and skin (with fat tissue) were determined. The samples were collected at day 56 of the experiment – immediately after the cessation of the treatment and thereafter – in 3 consecutive days and 6 days after the cessation of treatment (for detection of SLM residues). For that purpose, immediately after the end of treatment, 2 birds from groups I, III, V and VI were euthanized and at post treatment days 1, 2, 3 and 6 – 6 birds from the respective groups. The FPL residues were determined using a microbiological method (modification of the method of Nesemann, 1969) by agar gel diffusion with *Bacillus cereus ATCC 19637* as test organism. The limit of quantitation was 0.5 μg/g tissue.

The detection of SLM residues was done by the bioautographic method using *Bacillus stearothermophilus var. calidolactis C 593* as test-organism with a limit of quantitation of 0.05 μg/g.

The statistical analysis was performed by the non-parametric Mann-Whitney test and by one-way analysis of variance (ANOVA).

RESULTS

It was found out that the body weight of chickens from group I (negative control) at the end of the experiment was 1058.0 ± 23.1 g (Table 1). For group VI (treated with FLS) it was higher by 2.8% ($p>0.05$). The body weights of positive controls from groups II, III and V (treated with HLF, FPL and SLM, respectively) were not statistically different from that of the negative control. There was a decrease in group IV (FPL+HLF) by 6% ($p<0.05$) and a tendency towards increase in group VII (FLS+HLF) by 4.1%.

The total live weight gain of the negative control (group I) was 1018.4 ± 22.8 g (Table 1). The parameter tended to increase in group VI (FLS) by 2.9%, better

[5] The concentration refers to the active substance(s).

[6] Chickens received 0.6% HLF up to the age of 3 weeks and thereafter – 12% HLF.

Table 1. Effect of the combination of flavophospholipol and salinomycin (formulation Flavosalin-premix) upon the body weight (g) and the live weight gain (g) (mean ± SEM) in broiler chickens treated for 56 days

Group	n	Body weight				Average live weight gain			
		initial	%	final	%	total	%	daily	%
I (control)	20	39.6 ± 0.7	100.0	1058.0 ± 23.1	100.0	1018.4 ± 22.8	100.0	18.1 ± 0.4	100.0
II (HLF)	20	39.6 ± 0.7	100.0	1049.5 ± 33.2	99.2	1009.8 ± 32.8	99.2	18.0 ± 0.6	100.0
III (FPL)	20	39.6 ± 0.7	100.0	1066.0 ± 30.3	100.8	1026.4 ± 30.4	100.8	18.3 ± 0.5	101.1
IV (FPL+SLM)	20	39.6 ± 0.7	100.0	994.8 ± 44.3*	94.0	955.1 ± 44.1*	93.4	17.1 ± 0.8*	94.4
V (SLM)	20	39.6 ± 0.7	100.0	1068.2 ± 31.3	101.0	1028.6 ± 31.2	101.0	18.4 ± 0.6	101.7
VI (FLS)	20	39.6 ± 0.7	100.0	1088.0 ± 30.5	102.8	1048.4 ± 30.3	102.9	18.7 ± 0.5	103.3
VII (FLS+HLF)	20	39.6 ± 0.7	100.0	1101.2 ± 42.2	104.1	1061.6 ± 42.3	104.2	19.0 ± 0.8	105.8

FLS=flavosalin; HLF=halofuginone; FPL=flavophospholipol; SLM=salinomycin; * $p<0.05$ vs group I (control).

expressed in female birds compared to males – by 4.8% and 2.6% respectively. For the other groups, the values were similar and the pattern of changes – very close to that of body weight. The ANOVA test showed no statistically significant differences among groups.

The feed consumption in group I at the end of the experiment was 2953 g. In groups II (HLF) and VI (FLS) the feed consumption was lower by 0.8% and 1.1% respectively, while in FPL, FPL+HLF, SLM and FLS+HLF groups (III, IV, V and VII) it was increased by 0.3%, 0.9%, 1.3% and 0.8%, respectively.

The feed conversion ratio per 1 kg live weight gain was lower by 4% after the FLS treatment (group VI) as well as after FPL (by 0.5%) and FLS+HLF (by 3.4%) treatments (groups III and VII). A minor over-expenditure was noticed in positive controls from groups II (HLF, by 0.1%) and V (SLM, by 0.3%) and a more pronounced one – in group IV (FPL+HLF, by 7.6%).

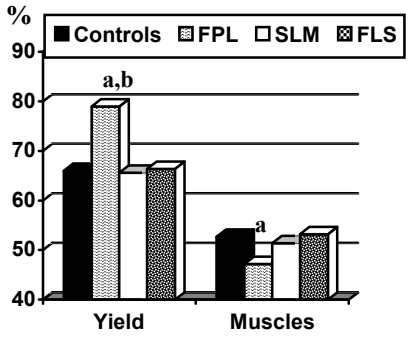

Fig. 1. Slaughtering data in broiler chickens treated orally with the combination of flavophospholipol and salinomycin (formulation Flavosalin-premix) at 65 mg/kg forage for 56 days – yield and muscle mass. Level of significance: [a] $p<0.05$ vs controls; [b] $p<0.05$ vs SLM (group V) and vs FLS (group VI).

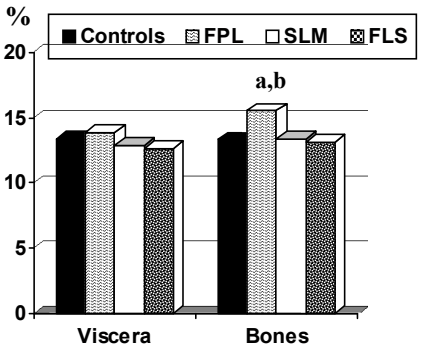

Fig. 2. Slaughtering data in broiler chickens treated orally with the combination of flavophospholipol and salinomycin (formulation Flavosalin-premix) at 65 mg/kg forage for 56 days – viscera and bones. Level of significance: [a] $p<0.05$ vs controls; [b] $p<0.05$ vs SLM (group V) and vs FLS (group VI).

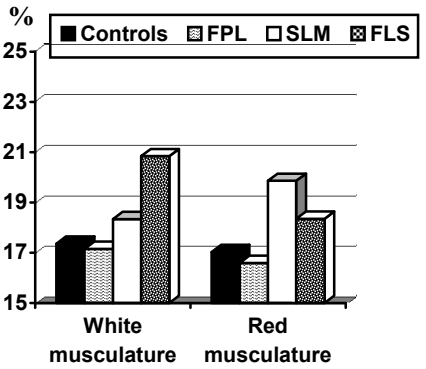

Fig. 3. Protein content (%) in the musclulature of broiler chickens treated perorally with the combination of flavophospholipol and salinomycin (formulation Flavosalin-premix) at 65 mg/kg forage for 56 days.

The slaughtering data showed that the application of FLS for 56 days (group VI) resulted in statistically insignificantly increased yield compared to the negative control – by 0.4% and increased muscle

mass by 0.5% (Fig. 1), accompanied by a decreased weight of viscera and bones+fasciae (by 0.8% and 0.2% respectively) (Fig. 2).

The yield of group III (FPL) was higher and that of the group V (SLM) – lower than the negative control. Both antibiotics resulted in increased bone mass with significant differences only for group III. The single FPL administration resulted in decreased muscle mass by 5.5% ($p<0.05$). The ANOVA results showed a statistically significant differences between FPL and controls and between FPL and birds that received SLM and FLS.

The chemical analysis of meat showed a tendency towards increased protein content compared to negative control (by 1.32% for red musculature and 3.48% for white musculature). Similar changes were also observed in group V, while in group III, a tendency towards decreased protein content was noticed (Fig. 3). The changes in mineral, fat and moisture contents were negligble and statistically insignificant in all experimental groups. Following the application of FLS (65 mg/kg forage), no residues of FPL in musculature and viscera were detected. Salinomycin was detected in minor quantities (0.05 μg/g at day 0 in some musculature, liver, kidneys and fat specimens). A complete depletion was registered 6 days after the cessation of treatment.

DISCUSSION

Our data suggest that the application of FLS for 56 days with the forage (65 mg/kg forage) resulted in a tendency towards growth promotion in chickens, more expressed than when SLM and FPL were independently used.

The data for FLS evidencing the presence of unidirectional, favourable tendencies and effects (compared to the independent administration of both substances) by respect to the decreased forage consumption, feed conversion ratio per 1 kg live weight gain, the persistance of those effects during all periods of the study, the increase in the muscle mass, the decrease in viscera and bone with fasciae weights, the increase of protein content in white musculature and in red musculature (caused by FPL) could be assessed as advantageous.

There are literature data about the more favourable effects of FPL upon the live weight gain (Krüger & Volrath, 1969; Vogt, 1969; Vogt & Gürocak, 1970) – an increase by 9-11% and better feed conversion ratio by 5%. This could be due to the different experimental design (different housing, dosage etc.). Similar results for the effect of FPL upon the chemical composition of musculature are reported by Droumev et al. (1978).

The lower live weight gain in the group treated with FPL+HLF was probably due to the predominance of female chickens whose body weight was smaller than that of males.

The positive changes in the live weight gain in the FLS+HLF group (a tendency for increase) could be explained by the better resistance to coccidiae when both drugs were simultaneously used.

The lack of FPL residues in the musculature and viscera following the treatment corresponded to the data of Bauer & Dost (1969), that reported the lack of measurable levels in the organs of birds, rats and calves following FPL application for weeks, months and one year at a dose of 0.5-250 mg/kg forage. This could be due to the limited absorption (following p.o. administration FPL was practically not absorbed) (Bauer & Dost, 1965).

CONCLUSION

Our studies upon the drug interaction and in particular upon the ergotropic effect of FPL and SLM showed that their simultaneous administration in broiler chickens was not accompanied by negative effects. The changes in chickens development, observed under the influence of FLS could be evaluated as unidirectional, positive effects and a tendency towards increased live weight gain, improved feed conversion ratio, increased yield and improved quality of meat.

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