Revista Brasileira de Zootecnia
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ISSN 1806-9290
www.sbz.org.br

R. Bras. Zootec., v.41, n.12, p.2458-2462, 2012

Short Communication

Reassessing flavophospholipol effects on broiler performance

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ABSTRACT - The objective of this study was to evaluate bird responses to flavophospholipol at higher doses than those recommended by the Brazilian legislation. A trial was conducted with 900 male broilers divided into five doses of flavophospholipol: 0, 2, 4, 8 and 16 mg/kg. The performance evaluation was conducted weekly until 42 days of age. The evaluated parameters were: weight gain, feed conversion corrected for mortality, feed intake and mortality. At the end of experimental period, birds supplemented with growth promoter differed from the negative control for weight gain and feed conversion in the period from 1 to 42 days. Body weight gain and feed conversion were significantly higher for birds fed 16 mg/kg of flavophospholipol, compared with treatment without the growth promoter in the period from 1 to 21 days. In the accumulated period from 22 to 42 days, all doses were different for the negative control for feed conversion and body weight gain. The performance improvement was maximized at the dosage of 10.1 mg/kg for feed conversion ratio and 10.9 mg/kg for body weight gain by regression analysis. Flavomycin can be used as a growth promoter to improve feed conversion ratio and body weight gain in broilers from 1 to 42 days of age.

Key Words: bambermycin, broiler chicken, flavomycin, flavophospholipol, growth promoter

Introduction

Antimicrobials have been added to the diets of animals raised for the production of meat at sub therapeutic levels for many years. Consistent improvements in feed efficiency have justified this practice, which is supposedly mediated through modulation of the gut microbiota. Niewold (2007) suggested that the antibiotic growth promoters (AGP) actually reduce inflammatory responses by sparing the immune system, which leads to reduced morbidity and mortality as well as subclinical and clinical diseases.

Despite the well-recognized benefits, a trend to ban AGP from animal diets is now stronger than ever in many countries. Risk of resistance to antibiotics by pathogenic microorganisms has been a main driver to influence government authorities as they change the laws on the use of this class of product. Nevertheless, the association of bacterial resistance with AGP may be linked to questionable research (Cox et al., 2003; Dibner & Richards, 2005; Cervantes, 2011). It seems that, more than any long-term research, the precautionary principle has been the main reason for AGP banning in the European Union (World Health Association, 2003, 2004).

Flavomicyn, a glycophospholipid which has a known inhibiting capacity for Gram positive bacteria (Butaye et al., 2003a), can still be legally added at subtherapeutic doses in poultry diets in many countries. This molecule has never been used as a therapeutic drug for poultry and it lacks pharmacokinetic and pharmacodynamic activities (Pfaller et al., 2006). Inhibition of the enzyme transglycosylase, necessary for the formation of the bacterial cell wall and its consequent intestinal microbiota modulation promotes the development of *Lactobacilli* and *Bifidobacteria* in the intestinal tract, which are generally considered beneficial bacteria (Bolder et al., 1999). The establishment of a population of microorganisms that supports animal growth by flavophospholipol is based on the competitive exclusion effect, which has been observed to be accompanied by lower incidence of resistant genes and lower excretion of pathogenic bacteria, such as *Salmonella*, *Clostridium* and *Campylobacter* (Kissel, 1998; Butaye et al., 2003a; Pfaller et al., 2006). Worldwide, the inclusion of flavophospholipol varies from 1 to 20 mg/kg of feed. In Brazil, the use of growth promoters is regulated by MAPA (2008), which sets the use of flavophospholipol in birds at 1 to 2 mg/kg, which is similar to the regulation in the United States by the FDA (1985).

Received: October 12, 2012 and accepted November 7, 2012.
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The purpose of this study was to reevaluate the effects of flavophospholipol in broiler diets since it has not been used in practice for many years, which may become a cheap alternative for rotating AGP programs.

Material and Methods

The procedures adopted throughout this study avoided unnecessary animal discomfort and followed the directives of the Committee of Ethics and Use of Animals of Universidade Federal do Rio Grande do Sul, Porto Alegre, Brazil.

A total of 900 one-day-old Cobb × Cobb 500 male broiler chicks originated from the University hatchery were placed in 60 floor pens, 1.70 × 1.65 m each, 15 birds per pen. Each pen had rice hulls as litter and was equipped with 3 nipple drinkers and a tube feeder. Individual one-day-old chick weight averaged 48±1 g and the birds had been vaccinated for Marek's and Gumboro disease at the hatchery. Average house temperature was 32 °C at placement, being reduced as animals aged to provide comfort throughout the study using heaters, fans and foggers whenever appropriate. Lighting was continuous until 7 d of age; 18 hours light:6 dark cycles were used afterwards. Feed and water were provided *ad libitum*.

Mash diets were provided for the birds in a 4-phase feeding program as follows: 1 to 7 d (pre-starter), 8 to 21 d (starter), 22 to 35 d (grower), and 36 to 42 d of age (finisher). Diets were formulated with corn, soybean meal and meat and bone meal as major ingredients. Treatments consisted of five graded inclusions of flavophospholipol (Flavomycin® 80, Huvepharma, Sofia, Bulgaria): 0, 2, 4, 8 and 16 mg/kg (Table 1). A pooled sample of each treatment per phase feeding was analyzed for the contents of flavophospholipol as suggested by Gallo et al. (2010).

Live performance was evaluated weekly on a pen basis through the following measurements until 42 days of age: body weight gain (BWG), feed intake (FI), feed conversion

Table 1 - Composition of experimental diets[1]

	Pre-starter					Starter					Grower					Finisher				
	Flavophospholipol																			
Ingredients, g/kg or as noted	0	2	4	8	16	0	2	4	8	16	0	2	4	8	16	0	2	4	8	16
Corn	541.5					582.6					604.8					605.2				
Soybean meal	360.7					312.8					281.0					266.2				
Meat and bone meal	50.00					50.00					50.00					50.00				
Sodium bicarbonate	10.00					1.60					1.60					2.10				
Soybean oil	20.40					31.50					44.10					60.90				
Dicalcium phosphate	7.80					1.90					0.80					-				
Limestone	4.50					6.70					6.50					6.30				
Salt	3.80					3.40					2.90					2.00				
Methionine 84%[2]	4.60					4.10					3.50					3.20				
L-lysine HCl 78%	2.00					2.00					1.70					1.50				
L-threonine 98.5%	0.70					0.70					0.50					0.40				
Mineral and vitamin suplemment[3]	1.70					1.50					1.30					1.00				
Choline chloride 60%	0.50					0.40					0.50					0.40				
Flavomycin 8%[4], g/t	0 – 200					0 – 200					0 – 200					0 – 200				
Kaolin, g/t	200 – 0					200 – 0					200 – 0					200 – 0				
Calculated composition, g/kg or as noted[5]																				
Metabolizable energy, kcal/kg	2,960					3,050					3,150					3,250				
Crude protein (analyzed)	235.0 (245.5)					211.0 (226.6)					198.1 (210.5)					191.0 (203.7)				
Digestible lysine	12.80					11.50					10.50					10.00				
Ca (analyzed)	10.0 (10.5)					9.50 (9.90)					9.0 (10.2)					8.7 (9.0)				
Total phosphorus (analyzed)	7.4 (7.5)					7.0 (6.50)					6.8 (7.0)					6.6 (6.5)				
Available phosphorus	5.00					4.70					4.50					4.20				
Na	2.30					2.20					2.00					1.80				
Dietary electrolyte balance, mEq/kg[6]	230					215					200					200				
Choline, ppm	1,700					1,600					1,600					1,500				

[1] Each treatment was supplied to 9 replicate pens of 15 broilers at the beginning of the study.
[2] Alimet®, Novus International, São Paulo, Methionine hydroxi analogue, guaranteed 84% methionine activity and 12% calcium.
[3] Provided per kg of feed: vitamin A - 9800 IU; vitamin D - 2520 IU; vitamin E - 70 IU; vitamin K - 3 mg; thiamin - 2.5 mg; riboflavin - 7 mg; pyridoxine - 4 mg; vitamin B12 - 0.025 mg; pantothenic acid - 12 mg; niacin - 50 mg; folic acid - 1.5 mg; biotin - 0 -15 mg; iron - 50 mg; zinc - 50 mg; manganese - 80 mg; copper - 10 mg; iodine - 1 mg; cobalt - 1 mg; selenium - 0.3 mg; monensin - 120 mg.
[4] Flavomycin® 80, Huvepharma do Brasil Comércio e Importação Ltda, Flavophospholipol, guaranteed levels 8%. Inclusion levels were 0, 25, 50, 100 and 200 g/t, replaced by kaolin (Sericita M-200, Mineracão Violani, Colombo, Paraná, Brazil).
[5] Minimum ratios for digestible lysine:digestible total sulfur amino acids: 75%, digestible threonine 65%, digestible valine 75%, digestible isoleucine 67%.
[6] Dietary electrolyte balance (Na + K – Cl), mEq/kg of the diet.

ratio corrected for the weight of dead birds (FCR) and mortality.

The experimental design was completely randomized. Mortality data was analyzed after arcsine transformation ($\sqrt{x}/100$). Linear regressions were run using formulated concentrations of flavophospholipol with the PROC REG procedure of SAS (2001). Significance was accepted as $\alpha = 0.05$, and mean differences were separated using Tukey's test.

Results and Discussion

Analyzed crude protein, calcium and total phosphorus for all diets and feed phases were as expected from feed formulation (Table 1). Analyzed flavophospholipol concentrations in all feeds were in accordance with expected levels from feed formulation; therefore, regression analysis was performed with formulated flavophospholipol in feeds.

Significant differences ($P<0.05$) occurred for BWG and FCR throughout the study and for FI from 1 to 21 days of age (Table 2). Adjustments obtained by linear regression analysis demonstrated improvements in BWG and FCR as flavophospholipol increased in the diet. Optimal responses obtained with flavophospholipol supplementation were calculated whenever a significant quadratic adjustment was possible. For BWG and FCR optimal responses were of 14.68 and 12.17 mg/kg of feed from 1 to 21 days (Figure 1), 9.83 and 10.31 mg/kg of feed from 22 to 42 days (Figure 2), and 10.10 and 10.90 mg/kg of feed from 1 to 42 days (Figure 3). Feed intake was adjusted by a quadratic model only from 1 to 21 days when the optimal response was achieved at 16 mg/kg (Figure 4).

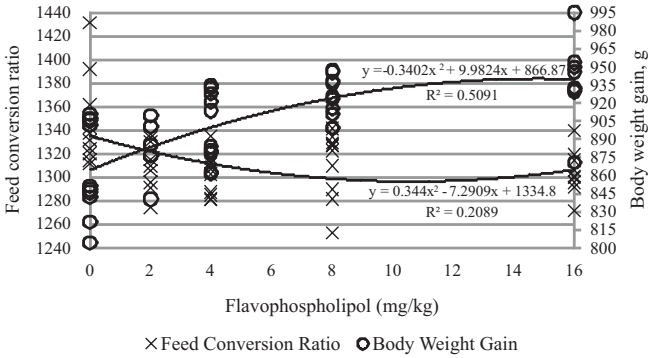

Figure 1 - Feed conversion ratio and body weight gain from 1 to 21 days of age.

Table 2 - Live performance of male broilers fed diets with graded increases in flavophospholipol

Flavophospholipol[1], mg/kg	0	2	4	8	16	Linear, r^2	Quadratic, R^2	Plateau[5]	P (regression)	Mean	SEM, %	P (ANOVA)
							1- 21 days					
Feed intake, g[2]	1,104b	1,112b	1,113b	1,154ab	1,168a	0.2648	0.2720	16.00	0.0008	1,187	11.69	0.0039
Feed conversion ratio[3]	1.279b	1.247ab	1.238a	1.249ab	1.244a	0.0791	0.1853	12.17	0.0296	1.315	0.005	0.0031
Body weight gain, g[4]	864c	892bc	899 bc	924ab	940a	0.3902	0.4250	14.68	0.0001	903	7.17	0.0001
							22-42 days					
Feed intake, g	4,081	4,105	4,097	4,096	4,142	--	--	--	NS	4,097	33.61	0.9859
Feed conversion ratio	1.764b	1.713a	1.695a	1.684a	1.701a	0.1692	0.5305	10.31	0.0001	1.711	0.008	0.0001
Body weight gain, g	2,313b	2,396a	2,422a	2,433a	2,409a	0.0818	0.2966	9.83	0.0004	2,394	19.68	0.0011
							1-42 days					
Feed intake, g	5,243	5,273	5,275	5,308	5,321	--	--	--	NS	5,283	39.04	0.6315
Feed conversion ratio	1.651b	1.604a	1.588a	1.582a	1.589a	0.2213	0.5326	10.90	0.0001	1.602	0.006	0.0001
Body weight gain, g	3,177b	3,288a	3,321a	3,356a	3,349a	0.2390	0.4163	10.10	0.0001	3,298	23.79	0.0001

Means followed by different letters differ by Tukey's Test ($P>0.05$).
NS - not significant; SEM - standard error of the mean.
[1] Formulated concentrations of flavophospholipol using Flavomycin® 80, Huvepharma do Brasil Comércio e Importação Ltda, Flavophospholipol, guaranteed levels 8 g/kg. Inclusion levels were 0, 25, 50, 100 and 200 g/t.
[2] Individual feed intake.
[3] Individual feed conversion ratio corrected for mortality.
[4] Individual body weight gain.
[5] Optimal response based on calculated values of flavophospholipol in the predicted equation.

Mortality was not affected by the treatments and it was considered in an acceptable range for male broilers grown to the age used in this study (0.56±0.08).

Data from the present study show that birds experienced improvements in live performance whenever flavophospholipol was added to the diets. It was also demonstrated that benefits of adding flavophospholipol in the feeds could be obtained using levels much higher than those presently allowed to be used in commercial broiler production (maximum of 2 mg/kg of feed in Brazil (MAPA, 2008) and the USA (FDA, 1985).

Flavophospholipol is not absorbed throughout the gastrointestinal tract (GIT) of birds (Bauer & Dost, 1965; Wasielewski et al., 1965). Other studies have demonstrated that live performance improvements were obtained with broilers receiving flavophospholipol added in the feeds at doses higher than 2 mg/kg (Esteve-Garcia et al., 1997; Dibner & Richards, 2005; Demir et al., 2008).

Benefits of the inclusion of AGP in broiler diets on live performance have long been demonstrated (Miles et al., 2006; Cervantes et al., 2008; Hossain et al., 2008; Demir et al., 2008; Vieira et al., 2010). The mechanism of action of these compounds, however, is not easily explained. Flavophospholipol is a phosphoglycolipid antimicrobial produced by various strains of *Streptomyces*, active against Gram positive bacteria, such as *Staphylococcus* spp and *Enterococcus* spp (Aarestrup et al., 1998; Riedl et al., 2000; Butaye et al., 2003b). Therefore, it is expected that as harmful microorganisms are suppressed, the gut microbiota will come into balance towards a more benefic population. A well-balanced intestinal microbiota is considered to be an effective barrier against pathogenic bacteria, which besides positively affecting animal performance, may also result in positive effects for public health (Pfaller, 2006). For example, flavophospholipol was able to reduce the colonization and shedding of *Salmonella* spp and *Clostridium perfringens* (Cox et al., 2003; Bolder et al., 1999). Despite antimicrobial effects, live performance improvements with the use of AGP such as flavophospholipol, inhibition of inflammation effects have also been proposed as a mean of action for this type of compound (Niewold, 2007).

In the present study, including flavophospholipol in the diets led to improvements in FCR and BW gain. These are benefits that can be translated into important amounts of feed saved in the process of animal production for human consumption. Therefore, the use of flavophospholipol as an additive in broiler nutrition is still an important tool in maintaining competitive production of broiler meat.

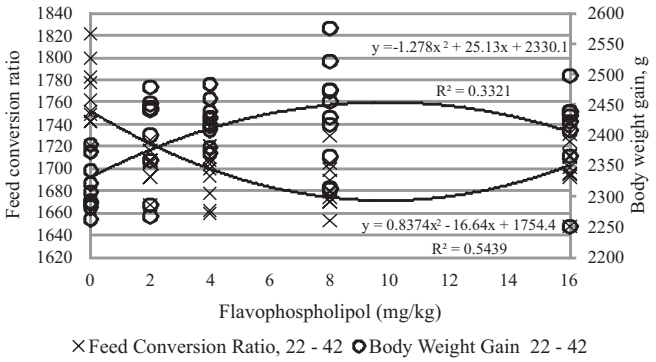

Figure 2 - Feed conversion ratio and body weight gain from 22 to 42 days of age.

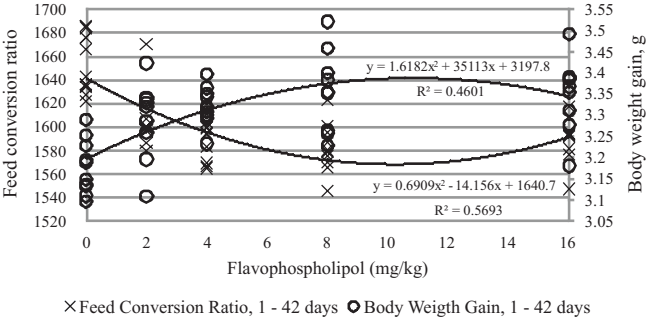

Figure 3 - Feed conversion ratio and body weight gain from 1 to 42 days of age.

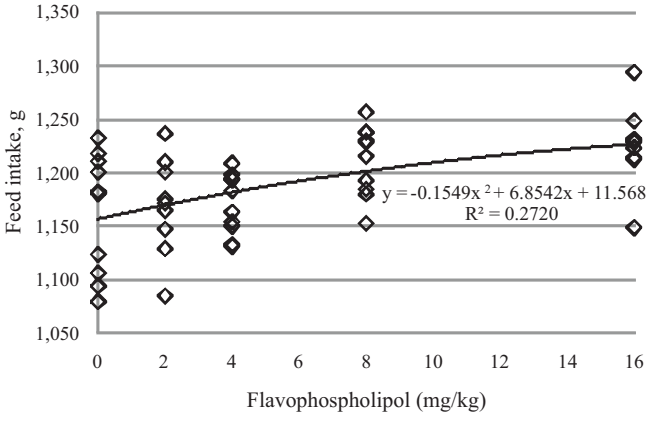

Figure 4 - Feed intake from 1 to 21 days of age.

Conclusions

The use of flavophospholipol improves broiler live performance from 1 to 42 days of age. Optimal benefits vary with flavophospholipol concentrations from 9.83 to 16.00 mg/kg of feed.

References

AARESTRUP, F.; BAGER, F.; JENSEN, N.E. et al. Surveillance of antimicrobial resistance in bacteria isolates from food animals to antimicrobial growth promoters and related therapeutic agents on Denmark. **Acta Pathologica, Microbiologica et Immunologica Scandinavica**, v.106, n.6, p.606-622, 1998.

BAUER, F.; DOST, G. Moenomycin in animal nutrition. **Antimicrobial Agents Chemotherapy**, v.5, p.749-752, 1965.

BOLDER, N.M.; WAGENAAR, J.A.; PUTIRULAN, F.F. et al. The effect of flavophospholipol (Flavomycin) and salinomycin sodium (Sacox) on the excretion of Clostridium perfringens, Salmonella enteritidis, and Campylobacter jejuni in broilers after experimental infection. **Poultry Science**, v.78, p.1681-1689, 1999.

BUTAYE, P.; DEVRIESE, L.A.; HAESEBROUCK, F. Antimicrobial growth promoters used in animal feed: effects of less well known antibiotics on Gram-positive bacteria. **Clinical Microbiology Review**, v.16, p.175-188, 2003a.

BUTAYE, P.; DEVRIESE, L.A.; HAESEBROUCK, F. Influence of different medium components on the in vitro activity of the growth-promoting antibiotic flavomycin against enterococci. **Journal of Antimicrobial Chemotherapy**, v.46, p.713-716, 2003b.

CERVANTES, H.M.; BAFUNDO, K.W.; PESTI, G.M. et al. Live and processing performance responses of broilers fed low and extra-low nutrient density withdrawal diets supplemented with virginiamycin. **Journal of Applied Poultry Research**, v.17, p.87-92, 2008.

CERVANTES, H.M. O Futuro dos antimicrobianos em produção animal. In: CONFERÊNCIA FACTA, 29., 2011, São Paulo. **Anais…** São Paulo: Facta, 2011. p.17-38.

COX, N.A.; CRAVEN, S.E.; MUSGROVE, M.T. et al. Effect of sub-therapeutic levels of antimicrobials in feed on the intestinal carriage of campylobacter and Salmonella in turkeys. **Journal of Applied Poultry Research**, v.12, p.32-36, 2003.

DEMIR, E.; KILINC, K.; YILDIRIM, Y. et al. Comparative effects of mint, sage, thyme and flavomycin in wheat-based broiler diets**. Archiva Zootechnica**, v.11, p.54-63, 2008.

DIBNER, J.J.; RICHARDS, J.D. Antibiotic growth promoters in agriculture: history and mode of action. **Poultry Science**, v.84, p.634-643, 2005.

ESTEVE-GARCIA, E.; BRUFAU, J.; PEREZ-VENDRELL, A. et al. Bioefficacy of enzyme preparations containing beta-glucanase and xylanase activities in broiler diets based on barley or wheat, in combination with flavomycin. **Poultry Science**, v.76, p.1728-1737, 1997.

FOOD AND DRUG ADMINISTRATION - FDA. **Registered number for use: NADA nº 44.759**. 1985. Available at: <http://www.fda.gov/ora/compliance_ref/cpg/cpgvet/cpg689-100.html>. Accessed on: Sept. 4, 2011.

GALLO, P.; FABBROCINO, S.; SERPE, L. et al. Determination of the banned growth promoter moenomycin A in feed stuffs by liquid chromatography coupled to electrospray ion trap mass spectrometry. **Rapid Communications in Mass Spectrometry**, v.24, p.1017-1024, 2010.

HOSSAIN, M.H.; BULBUL, S.M.; NISHIBORI, M. et al. Effect of different growth promoters on growth and meat yield of broilers. **The Journal of Poultry Science**, v.45, p.287-291, 2008.

KISSEL, A. In vitro Efficacy of salinomycin sodium and flavophospholipol against bacterial isolates from the gastrointestinal tract of different target species. **Hoechst Roussel Vet**, v.1, p.1-17, 1998.

MILES, R.D.; BUTCHER, G.D.; HENRY, P.R. et al. Effect of antibiotic growth promoters on broiler performance, intestinal growth parameters, and quantitative morphology. **Poultry Science**, v.85, p.476-485, 2006.

MINISTÉRIO DA AGRICULTURA, PECUÁRIA E ABASTECIMENTO - MAPA. Departamento de Fomento e Fiscalização da Produção Animal. Secretaria de Apoio Rural e Cooperativismo.. **Antimicrobianos, anticoccidianos e agonistas autorizados para uso em animais de produção em âmbito Nacional**. 2008. Available at: <http://www.abiquifi.org.br/legislacao/outras/Tabela%20de%20Antimicrobianos,%20Anticoccidianos%20e%20Agonistas%20Autorizados%20MAPA.xls>. Accessed on: Nov. 23, 2010.

NIEWOLD, T.A. The nonantibiotic anti-inflammatory effect of antimicrobial growth promoters, the real mode of action? A hypothesis. **Poultry Sciencen**, v.86, p.605-609, 2007.

PFALLER, M.A. Flavophospholipol use in animals: Positive implications for antimicrobial resistance based on its microbiologic properties. **Diagnostic Microbiology and Infectious Disease**, v.56, p.115-121, 2006.

RIEDL, S.; OHLSEN, K.; WERNER, G. et al. Impacto f flavophospholipol and vancomycin on conjugating transfer of vancomycin resistance plasmids. **Antimicrobial Agents and Chemoterapy**, v.44, p.3189-3192, 2000.

ROSTAGNO, H.S.; ALBINO, L.F.T.; DONZELE, J.L. et al. **Tabelas brasileiras para aves e suínos:** Composição de alimentos e exigências nutricionais. Viçosa, MG: Universidade Federal de Viçosa, 2000. 141p.

SAMBETH, W., NESEMANN, G; BAUER, F. et al. Investigations of the excretion and retention of flavomycin. In: FLAVOMYCIN SYMPOSIUM, 1969, Washington, D.C. **Proceedings…** Washington, D.C., 1969. p.133-139.

VIEIRA, S.L.; FAVERO, A.; BERRES, J. et al. Live performance and processing yields of broilers fed diets with tiamulin and salinomycin combinations. **Revista Brasileira de Ciência Avícola**, v.12, p.35-39, 2010.

WASIELEWSKI, E.; MUSHAWECK, R.; SCHÜTZE, E. Moenomycin, a new antibiotic. III. Biological properties. **Antimicrobial Agents Chemotherapy**, v.5, p.743-748, 1965.

WORLD HEALTH ORGANIZATION. **First joint FAO/OIE/WHO expert workshop on non-human antimicrobial usage and antimicrobial resistance: Scientific assessment**. Geneva, Switzerland, 2003. Available at: <http://whqlibdoc.who.int/hq/2004/WHO_CDS_CPE_ZFK_2004.7.pdf>. Accessed on: Sept. 17, 2011.

WORLD HEALTH ORGANIZATION. **Second joint FAO/OIE/WHO expert workshop on non-human antimicrobial usage and antimicrobial resistance: Management options.** Oslo, Norway, 2004. Available at: <http://whqlibdoc.who.int/hq/2004/WHO_CDS_CPE_ZFK_2004.8.pdf> Accessed on: Sept. 17, 2011.