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KINETIC DISPOSITION, SYSTEMIC BIOAVAILABILITY AND TISSUE DISTRIBUTION OF APRAMYCIN IN BROILER CHICKENS

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SUMMARY

Apramycin was administered to chickens orally, intramuscularly and intravenous to determine blood concentration, kinetic behaviour, bioaviability and tissue residues. The drug was given through intracrop, i.m. and i.v. routes in a single dose of 75 mg kg^{-1} body weight. The highest serum concentrations of apramycin were reached 0.20 and 0.76 hours after a single oral i.m. dosage with an absorption half-life [$t_{1/2}(ab)$], 0.10 and 0.19 hours and elimination half-life [$t_{1/2}(\beta)$]1.21 and 0.53 hours, respectively. The systemic bioavailability percentage 2.03 and 57.96 percent after intracrop and i.m. administration, respectively, indicating the very lower extent of apramycin absorption from the oral route in chickens. Following i.v. injection, the kinetic of apramycin can be described by a two-compartment open model with a $t_{1/2}(\alpha)$ 1.5 hours, (volume of distribution) Vd(ss) was 4.82 litre kg^{-1} and Cl(B) (total body clearance) was 1.88 litre kg^{-1} h^{-1}. The serum protein-binding tendency of apramycin as calculated in vitro was 26 per cent.

The highest concentration of apramycin residues were present in the kidneys and liver after a successive daily intracrop and i.m. administration for 5 days. No apramycin residues were detected in tissues after 6 hours except in the liver and kidneys and that disappeared completely by 12 and 24 hours after intracrop and i.m administration, respectively.

INTRODUCTION

Apramycin is a broad spectrum , aminocyclitol antibiotic used for systemic and enteric infections in a variety of species and is not well absorbed from the gastrointestinal tract of animals (Thomson et al., 1991). Extensive studies concerning the rate of absorption, distribution and elimination of many aminoglycosides in veterinary practice have been carried out (Regamy et al., 1973; Podkopaev, 1974; Beech et al., 1977; Baggot, 1978; Riviere and Coppoc, 1981; Atef et al., 1986 and Aziz et al., 1988).

The extensive use of aminoglycosides such as apramycin for treatment of many systemic and enteric infectious diseases in poultry and problems appeared from their residues in meat, encouraged studying of disposition of this drug in chickens. Few literature were recorded about the kinetic behaviour of apramycin in the chickens.

The present study was thus initiated to describe the kinetic disposition, bioavailability, tissue distribution pattern and residue of apramycin in chickens when given after oral i.m. and i.v. administration.

MATERIALS AND METHODS

Drug:

Apramycin sulphate (Apra 200®) was supplied by Eli Lilly, Italia.

Birds:

Seventy clinically healthy Hubbard chickens with body weights of 1.65 to 1.75 kg, 45 to 50 days old, were obtained one week before the study began. During acclimatisation and subsequent treatment periods, they were fed antibacterial-free balanced commercial ration and drinking water was freely available. The birds were housed in groups of five birds in each cage.

Experimental design

Pharmacokinetic studies:

Ten chickens were classified into two equal groups. Chickens of the first group were administered a single dose of apramycin (75 mg kg-1 boody weight) orally via intracrop, whereas those of the second group intramuscularly. Blood samples were obtained from the wing vein before and at five, 15, 30 minutes, one, two, three, four, five, six, eight, 12 and 24 hours after administration for estimation of the drug concentration in serum. One ml of blood was collected at each sampling time. Two weeks later (to ensure complete clearance of the drug from their bodies) apramycin was injected intravenously in these 10 chickens with the same dose as before and the same way of sampling was applied to reveal the bioavailability of the tested drug. Samples were centrifuged to separate serum to determine the apramycin concentration and serum protein binding tendency on the same day as sample collection.

Tissue distribution:

Sixty birds were divided into two equal groups of 30 birds each. Birds of the first and second groups were administered apramycin (75 mg kg^{-1} body weight) daily for 5 successive days via intracrop and i.m. routes, respectively. Five chickens were slaughtered from each goup at 15 minutes, one, three , six, 12 and 24 hours after the last dose administration. Blood and tissue samples (liver, kidney, lung, brain, intestine and breast muscle) were taken from the slaughtered birds. Blood samples were centrifuged to separate serum to determine apramycin concentration.

Analyatical methods:

Apramycin in blood and tissue samples was estimated by the microbiological method described by Bennett et al., (1966) using Bacillus subtilis (ATCC 6633) as test organism.

Statistical analysis:

The obtained data were statistically analysed and the results are given as mean ± s.e.m. The pharmacokinetic parameters of apramycin in chickens were calculated according to the method of Baggot (1978).

RESULTS

Pharmacokinetics:

Serum concentrations of apramycin after a single oral, i.m. and i.v. administration of 75 mg kg^{-1} body weight were illustrated in Fig. 1.

Values for kinetic constants describing the absorption and disposition of the drug in chickens after oral, i.m. and i.v. administration are incorporated in Table 1. Following i.v. injection of the drug in chickens in a single dose of 75 mg

Table 1: Pharmacokinetic parameters of apramycin in chickens after a single i.v., i.m. and oral administration of 75 mg kg^{-1} body weight.

Parameter	Unit	I.V. (n = 10)	Oral (n = 5)	I.M. (n = 5)
C^0	ug ml^{-1}	16.00±0.32		
A	ug ml^{-1}	9.62±0.25		
α	h^{-1}	0.46±0.01		
$t_{1/2(\alpha)}$	h	1.50±0.02		
$K_{(ab.)}$	h^{-1}		6.66±0.06	3.60±0.07
$t_{1/2(ab.)}$	h		0.10±0.001	0.19±0.004
$t_{max.}$	h		0.20±0.01	0.76±0.03
$C_{max.}$	ug ml^{-1}		0.79±0.02	11.06±0.31
B	ug ml^{-1}	6.38±0.53	0.41±0.01	6.46±0.26
β	h^{-1}	0.34±0.01		
K_{el}	h^{-1}	0.40±0.003	1.21±0.01	0.53±0.004
$t_{1/2(\beta)}$	h	2.10±0.01	1.22±0.01	2.31±0.02
K_{12}	h^{-1}	0.01±0.002		
K_{21}	h^{-1}	0.39±0.01		
V_c	Litre kg^{-1}	4.70±0.09		
$V_{d(area)}$	Litre kg^{-1}	5.62±0.14		
$V_{d(\beta)}$	Litre kg^{-1}	5.62±0.14		
$V_{d(B)}$	Litre kg^{-1}	12.09±1.01		
$V_{d(ss)}$	Litre kg^{-1}	4.82±0.08		
$Cl_{(B)}$	L kg^{-1} h^{-1}	1.88±0.05		
A.U.C.	Ug ml^{-1} h^{-1}	39.93±1.02	0.81±0.02	23.18±1.08
F	%		2.03±0.02	57.96±1.57

Table 3: Serum and tissue concentrations of apramycin (ug ml^{-1} / g^{-1}) after a multiple i.m. dose of 75 mg kg^{-1} body weight for 5 successive days in chickens (n = 5).

Tissue	Time of slaughter after the last dose (h)					
	0.25	1	3	6	12	24
Serum	3.93 ± 0.09	9.95 ± 0.24	4.50 ± 0.09	1.88 ± 0.05	0.72 ± 0.02	--
Liver	1.65 ± 0.02	5.05 ± 0.08	2.25 ± 0.08	0.89 ± 0.02	0.29 ± 0.01	--
Kidney	7.06 ± 0.21	15.99 ± 0.44	8.68 ± 0.27	3.74 ± 0.10	1.16 ± 0.05	0.63 ± 0.01
Lung	1.38 ± 0..07	4.18 ± 0.08	1.75 ± 0.04	0.75 ± 0.02	0.23 ± 0.01	--
Brain	0.51 ± 0.02	1.50 ± 0.02	0.65 ± 0.01	0.27 ± 0.01	--	--
Intest-ine	0.65 ± 0.01	0.96 ± 0.01	0.71 ± 0.03	0.38 ± 0.02	--	--
Breast muscle	0.72 ± 0.01	2.08 ± 0.07	0.86 ± 0.03	0.38 ± 0.004	0.11 ± 0.001	--

-- not detectable.

Table 2: Serum and tissue concentrations of apramycin (ug ml^{-1} or g^{-1}) after a multiple intracrop dose of 75 mg kg^{-1} body weight for 5 successive days in chickens (n = 5).

Tissue	Time of slaughter after the last dose (h)					
	0.25	1	3	6	12	24
Serum	0.86 ± 0.02	0.37 ± 0.01	0.24 ± 0.01	0.07 ± 0.002	--	--
Liver	0.31 ± 0.01	0.14 ± 0.01	0.09 ± 0.002	0.05 ± 0.001	--	--
Kidney	1.32 ± 0.11	1.07 ± 0.02	0.72 ± 0.01	0.48 ± 0.003	--	--
Lung	0.27 ± 0.01	0.14 ± 0.01	0.09 ± 0.001	--	--	--
Brain	0.10 ± 0.003	0.09 ± 0.002	0.06 ± 0.001	--	--	--
Intest-ine	0.18 ± 0.02	0.12 ± 0.01	0.08 ± 0.01	--	--	--
Breast muscle	0.14 ± 0.004	0.10 ± 0.003	0.07 ± 0.003	--	--	--

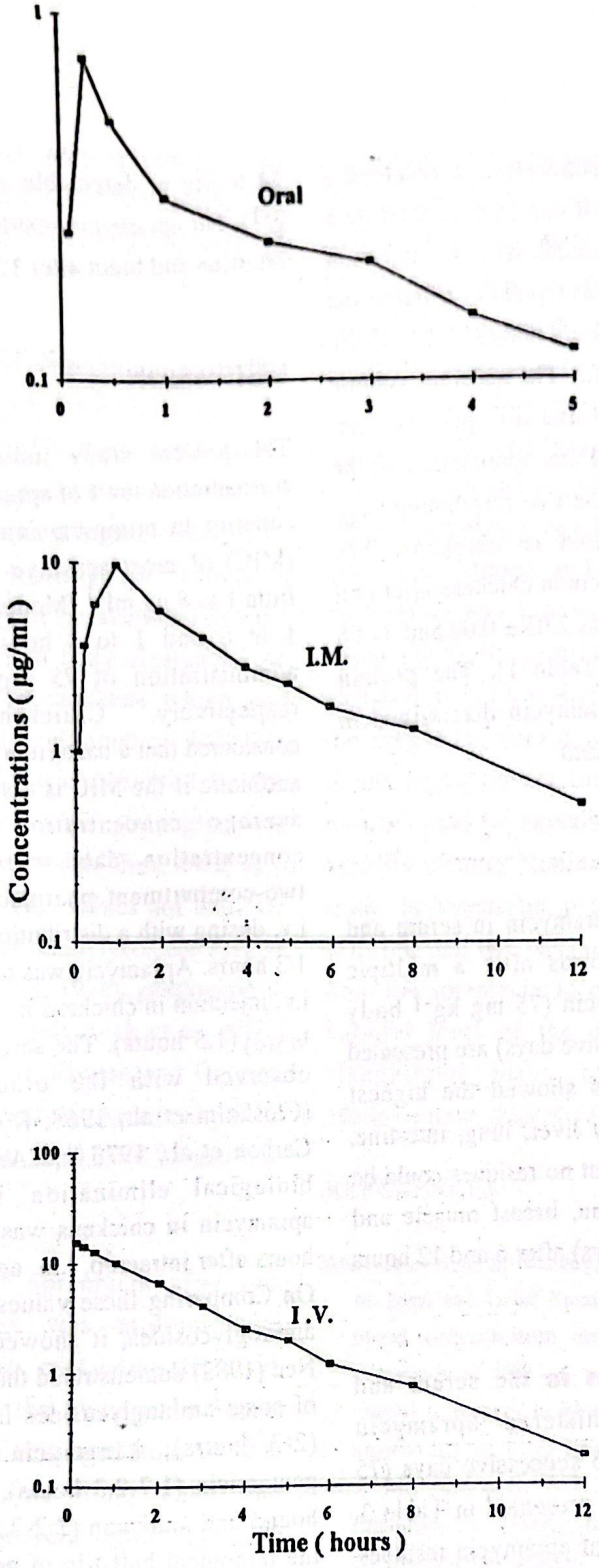

Fig. 1 : Semilogarithmic graph depicting the time concentration course of apramycin in serum of chickens after a single oral, I.M. and I.V. administration of 75 mg/kg body weight.

kg-1 body weight, its concentration revealed a biexponential decline that can be described by a two-compartment open model. After i.v. injection of apramycin, the drug was rapidly distributed and eliminated from the chicken's bodies with half-life value of 2.10 ± 0.01 hours. The apparent volume of distribution exceeded one litre per kilogram body weight. By oral and i.m. administration, the absorption half-life and their corresponding t_{max} revealed rapid absorption in chickens. The bioavailability of apramycin in chickens after oral and i.m. administration was 2.03 ± 0.02 and 57.96 per cent, respectively (Table 1). The protein binding tendency for apramycin determined *in vitro* was 26.0 ± 2.82 per cent.

Tissue distribution:

The concentrations of apramycin in serum and tissues of slaughtered birds after a multiple intracrop dose of apramycin (75 mg kg^{-1} body weight, daily for 5 successive days) are presented in Table 2. The kidneys showed the highest concentration followed by liver, lung, intestine, breast muscle and brain, but no residues could be detected in (lung, intestine, breast muscle and brain) and (liver and kidneys) after 6 and 12 hours by the intracrop route.

Apramycin concentrations in the serum and tissues of birds administered apramycin intramuscularly daily for 5 successive days (75 mg kg^{-1} body weight) are presented in Table 3. The highest concentration of apramycin residues were present in the kidneys followed by liver, lung, breast muscle, intestine and brain, but no residues could be detected in either serum or tissues (except in kidneys) after 34 hours by the i.m. route. It was found in the kidneys only until 24 hours at detectable concentrations (0.63 ug g^{-1}). No apramycin residues could be detected in intestine and brain after 12 hours.

DISCUSSION

The present study indicated that the blood concentration level of apramycin in chickens were superior to minimum inhibitory concentrations (MIC) of most sensitive germs, which ranged from 1 to 8 ug ml^{-1} (Moore and Ryden, 1977) for 1 to 6 and 1 to 8 hours after i.v. and i.m. administration of 75 mg kg-1 body weight, respectively. Cruickshank et al., (1975) considered that a bacterium may be sensitive to an antibiotic if the MIC is not more than 1/4-1/2 its average concentration in blood. Serum concentration data were best fitted to a two-compartment pharmacokinetic model after i.v. dosing with a distribution phase completed by 1/3 hours. Apramycin was rapidly distributed after i.v. injection in chickens indicated by the value of $t_{0.5(\alpha)}$ (1.5 hours). The same result criteria were observed with the other aminoglycosides (Chisholm et al., 1968; Rodriguez et al., 1971; Carbon et al., 1978 and Aziz et al., 1988). The biological elimination half-life $t_{0.5(\beta)}$ of apramycin in chickens was 1.22, 2.31 and 2.10 hours after intracrop, i.m. and i.v. administration. On Comparing these values with those of other aminoglycosides, it showed relative similarity. Neu (1982) demonstrated the elimination half-life of some aminoglycosides in man, streptomycin (2-3 hours); kanamycin (2. 1-2.4 hours); gentamicin (1.7-2.3 hours), tobramycin (2.1-27 hours) and amikacin (2.2-2.5 hours). In animals, the biological half-life of gentamicin was 30.35 minutes (Luft and Kleit, 1974) and 75 minutes in dogs (Baggot, 1978), 60.9 minutes in juvenile dogs (Riviere and Coppoc, 1981), 2.54 hours in horse (Pedersoli et al., 1980), 11.55 hours in camel (Abdel-Aziz et al., 1986) and 1.61 hours in calves (Aziz et al., 1988). The variation in

elimination half-life of aminoglycosides in different species of animals could be explained by variation in their protein binding capacity of (Gorden et al., 1972).

The apparant volume of distribution at steady state of a drug ($V_{d(ss)}$) is an indication of its diffusion in body tissues (Goodman and Gilman, 1980). The mean ($V_{d(ss)}$) value in chickens was 4.82 litre kg^{-1}). The relatively higher values of V_c and $V_{d(ss)}$ were indicative for extensive distribution of the during in extravascular tissues. Apramycin showed a high body clearance rate (1.88 litre kg^{-1} h^{-1}) in chickens which was confirmed with its short elimination half-live value. This phenmenon was observed by the higher $Cl_{(B)}$ values for some aminoglycoside antibiotics in dogs in relation to their lower $t_{1/2}$ values (Baggot, 1978). This values not coincide well with values for other minoglycosides such as kanamycin in dogs (Baggot, 1978); gentamicin in man, horse and dogs (Gyselycneck et al., 1971; Pedersoli et al., 1980 and Riviere and Coppoc, 1981) and apramycin in calves (Aziz et al., 1988) which their $Cl_{(B)}$ were 0.24, 0.25, 0.35 and 0.88 litre kg^{-1}, respectively.

The bioavailability of apramycin after i.m. injection in chickens was midium with approximately 57.97 per cent being absorbed. This value was similar to that observed by Aziz et al., (1988) in calves, which ranged from 59.79 to 66.09 per cent. On the other hand, the bioavilability of apramycin after intracrop administration was very low with approximately 2.03 per cent being absorbed. Similar results were reported by Thomson et al (1991) who found that oral absorption of apramycin is normally not well absorbed from the intestinal tract of most species, of animals and was significantly increased in chickens with an induced coccidial infection. The tendency of apramycin to bind with serum protein of chickens was 26 per cent. This finding explained that apramycin is not extensively bound to serum protein in chickens. Apramycin was detected in that tissues 6 hours (except lung, brain, intestine and breast muscle) and 12 hours (except brain and intestine) after repeated daily administration of 75 mg kg^{-1} body weight. It was more concentrated in kidneys and liver. This observation was supported by that recorded by Thomson et al., (1991). The high volume of distribution and low protein binding tendency of this drug in chickens is supported by its existence in the organ tissues for a longer time. These results could be explained also by the shorter half-life of drug elimination determined in this study. In conclusion, poultry farms must give at least one and two days premarketing withdrawal time for apramycin to ensure than there is no harmful level of the drug in the tissues of slaughtered birds after oral and i.m. administration, respectively.

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