TETRACYCLINE

First draft prepared by
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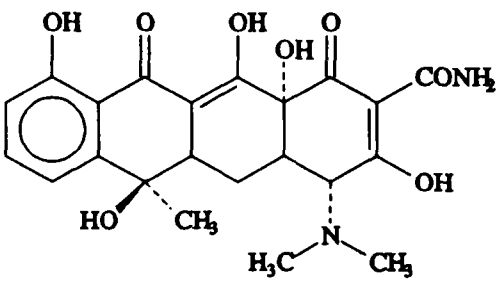
IDENTITY

Chemical names: 4-(Dimethylamino)-1,4,4a,5,5a,6,11,12a-octahydro-3,6,10,12,12a-pentahydroxy-6-methyl-1,11-dioxo-2-naphthalenecarboxamide

Synonyms: Deschlorobiomycin; Tsiklomitsin; Abricycline; Achromycin; Agromicina; Ambramicina; Ambramycin; Bio-Tetra; Bristaciclina; Cefracycline suspension; Criseociclina; Cyclomycin; Democracin; Hostacyclin; Omegamycin; Panmycin; Polycycline; Purocyclina; Sanclomycine; Steclin; Tetrabon; Tetracyn; Tetradecin.

Structural formula:



Tetracycline

Molecular formula: $C_{22}H_{24}N_2O_8$

Molecular weight: 444.43

OTHER INFORMATION ON IDENTITY AND PROPERTIES

Pure active ingredient: Tetracycline, tetracycline hydrochloride or the phosphoric acid complex of tetracycline

Major impurities: Chlortetracycline, epitetracycline (8% max.)

Appearance: Finely divided yellow powder

Melting point: Trihydrate, crystals, swell at 165°, m.p. 170-175°(dec), becomes anhydrous by drying *in vacuo* at 60° for 8 hrs.

Optical rotation: $[a]^{25}_D$ = -239°(in methanol)

Solubility: Water 1.7 g/L, MeOH >20 g/L

Stability: Tetracycline, tetracycline hydrochloride and the phosphate complex of tetracycline are stable for more than 6 months at ambient temperatures.

Manufacture: Chlortetracycline is antibacterial agent obtained by aerobic fermentation of strains of *Streptomyces aureofaciens* or *Streptomyces viridifaciens*. It is obtained commercially by large scale fermentation.

Mode of Action: All tetracyclines act by the inhibiting attachment of aminoacyl-t RNA to the A site on the 30S ribosome to prevent protein synthesis. Translation is inhibited bv one molecule of the tetracycline per ribosome. It is postulated that a tetracycline-magnesium complex is formed at the ribosome, making it less flexible and therefore unable to bind aminoacyl-t RNA. It has been found that binding of tetracyclines to the 30S ribosome is dependent on proteins S7, S14 and S19 (Sande and Mandell, 1990).

RESIDUES IN FOOD AND THEIR EVALUATION

CONDITIONS OF USE

Tetracycline hydrochloride is a broad-spectrum antibiotic used in both human medicine and veterinary medicine for treatment of infections caused by tetracycline sensitive organisms. It has been used in veterinary medicine for more than 40 years. Tetracycline has been used primarily as short-term, oral dosage formulations for the treatment of clinical diseases. This usage contrasts with that of chlortetracycline and oxytetracycline, antibiotics which have been principally used as prophylactic agents in medicated feeds.

Chickens and Turkeys

Treatment of chronic respiratory disease; treatment of infection by tetracycline sensitive organisms; control of synovitis; control of coccidiosis; treatment of blue comb, infectious enteritis and sinusitis, mud fever and hexamitiasis.

Swine

Treatment of bacterial swine enteritis; treatment of infection by tetracycline sensitive organisms.

Cattle/Sheep/Horses

Treatment of infection by tetracycline sensitive organisms.

METABOLISM AND PHARMACOKINETICS

Metabolism

There is no definitive evidence of tetracycline metabolism *in vivo*. In common with other members of the tetracycline group, tetracycline can be readily converted to microbiologically inactive isomers but these isomers are almost certainly artifacts produced by chemical rather than biological processes.

Studies concerning the metabolism of tetracycline were conducted in rats and dogs using radiolabelled tetracycline (Kelly and Buyske, 1960). One rat received an intraperitoneal dose while a second rat was given a similar oral dose. Tables 1 and 2 show the cumulative excretion of radioactivity from rats following oral and intraperitoneal doses of labelled tetracycline.

Table 1. **Cumulative Percent Radioactivity Excreted Following a Single Oral Dose of ^{14}C-labelled Tetracycline Hydrochloride (60 mg/kg) in the Rat**

	%in Urine		%in Faeces		% Total
Hour	Rat A	Rat B	Rat A	Rat B	(Average)
0-24	2.7	3.8	24.3	10.7	20.7
24-28	3.7	4.2	71.5	78.6	79.0
48-72	3.8	4.5	71.9	78.8	79.5
72-96	3.9	4.5	72.2	79.0	79.8
96-120	3.9	4.5	72.2	79.3	79.9
120-144	3.9	4.5	72.2	79.4	80.0
144-168	3.9	4.5	72.2	79.5	80.1

Table 2. **Cumulative Percent Excretion of Administered Radioactivity Following a Single Intraperitoneal Injection of 60 mg/kg Tetracycline.HCl in the Rat***

	% in Urine		% in Faeces		% Total
Hour	Rat C	Rat D	Rat C	Rat D	(Average)
0-24	28.0	59.3	8.5	23.8	59.8
24-28	37.6	61.6	52.2	25.3	88.4
48-72	40.1	62.1	52.9	25.9	90.5
72-96	41.1	62.5	53.3	26.1	91.5
96-120	42.1	62.7	53.5	26.4	92.4
120-144	42.6	62.9	53.6	26.5	92.8
144-168	43.0	63.2	53.7	26.8	93.3

*One rat received ^{14}C-labelled drug, the other tritium-labelled at the carbon number 7 position

The low urinary excretion of radioactivity following the oral administration as compared to that following the intraperitoneal dosing is to be noted. Also of interest is the surprisingly high percentage of the intraperitoneal dose eliminated in the faeces. Approximately 90% of the administered radioactivity in the rat was eliminated either by the urinary or faecal route. A significant portion of the remaining activity was bound as chelated tetracycline on the skeleton of the animal. A dilute formic acid extract of the radioactivity from the faeces, when submitted to countercurrent distribution in a butanol : formic acid : water system, resulted in a single peak which agreed with that obtained from pure tetracycline treated in a similar manner. Similar techniques applied to the unextracted urine from the rat gave identical results. These data show that with the exception of the metal chelate formation in the skeleton, tetracycline appears to be chemically unaltered by the rat.

Two beagle dogs each received a single oral dose of 25 mg of ^{3}H-labelled tetracycline hydrochloride/kg of body weight. Table 3 shows the cumulative urinary excretion of radioactivity.

Table 3. Cumulative Percent Radioactivity and Microbiological Activity Excreted in the Urine Following a Single Oral Dose of 25 mg/kg ^{3}H-labelled Tetracycline Hydrochloride in the Dog

	Radioactivity		Microbiological Activity
	Dog A	Dog B	Dog A
0-24	2.0	2.7	2.1
24-28	2.8	3.1	2.7
48-72	--	3.2	2.8

Faecal measurements were not made in this study. Direct measurement of ^{3}H-labelled tetracycline and analysis after countercurrent separation of tetracycline from the urine in an orally dosed dog showed identical curves, indicating that no chemical transformation had occurred in the body of the dog. The cumulative excretion of microbiological activity is also given. It can be seen that the values obtained by microbiological and radiological methods are in close agreement, supporting the countercurrent distribution studies that little or no biotransformation of tetracycline occurred in the body of the dog.

Pharmacokinetics of Tetracycline

Studies involving the absorption of chlortetracycline in the presence of divalent cations such as calcium and magnesium have been extensive and have been reviewed earlier in this volume (see monograph on Chlortetracycline). It was found that the absorption of chlortetracycline was suppressed in the presence of divalent cations but that this effect could be lessened by co-administration of organic acids, such as citric acid, which were able to form complexes with these cations. Although tetracycline can also form strong complexes with divalent cations, the effect of cations on tetracycline absorption has received little mention in the literature. Some of the apparent inconsistencies in the literature reviewed below may be due to the effects of divalent cations, co-administered with tetracycline, on absorption efficiency.

Pharmacokinetics in Laboratory Animals

Tetracycline was administered to rats which had been fasted for 12 hours as single oral doses of 75 mg/kg (Berte and Vandoni, 1962). Tetracycline plasma concentrations were measured at 1, 2, 3, 4 and 6 hours after dosing. Peak levels, averaging 3.6 mg/L, occurred two hours after dosing and declined to 0.5 mg/L by 6 hours. Tetracycline tissue concentrations were highest in liver and kidney at all sampling times. Residue levels in the lung tissue and plasma were of the same order whereas very low levels were found in brain tissue. Results are summarised in Table 4.

Table 4. Tetracycline Concentrations in Tissues and Plasma of Rats Following a Single Oral Tetracycline Dose of 75 mg/kg of body weight

	Tetracycline Concentration, mg/kg				
Time After Dosing (h)	Plasma	Lungs	Brain	Liver	Kidney
1	3.1±0.7	3.7±1.4	0.12±0.02	8.5±1.1	11.0±1.6
2	3.6±1.4	4.0±0.8	0.13±0.05	10.1±2.6	12.8±2.6
3	2.1±0.77	1.7±0.9	0.02±0.01	4.0±0.7	8.7±0.5
4	2.2±0.75	1.5±0.9	0.01±0.01	3.0±0.08	4.5±0.5
6	0.5±0.45	1.2±1.1	0.01±0.0001	2.5±2.4	2.6±1.9

Beagle dogs were given single oral tetracycline doses of 25 mg/kg live weight. Peak serum levels of tetracycline, averaging 2.98 mg/L, occurred 2 hours after dosing, and declined to an average of 0.27 mg/L by 24 hours post dosing (Kanegis, 1958). Excretion in the urine accounted for 9.8% of the administered dose by 72 hours after dosing. In this same study, dogs given single IV tetracycline doses of 10 mg /kg attained average serum levels of 10.6 mg/L one hour postdosing, declining to 1.3 mg/L at 24 hours and 0.14 mg/L at 48 hr. Tetracycline levels, determined by microbiological assay, are summarised in Table 5.

Table 5. Average Blood and Urine Concentrations of Tetracycline in Dogs Given Single Oral Doses (25 mg/kg) or Single Intravenous Doses (10 mg/kg)

	Oral Dosing			Intravenous Dosing	
Hours After Dosing	Blood (mg/L)	Urine (mg/L)	Cumulative Recovery in Urine	Blood (mg/L)	Cumulative Recovery in Urine
1	2.05	63.0	0.35%	10.6	7.9%
2	2.98	301.0	1.4%	8.7	13.7%
4	2.38	340.0	2.9%	7.8	20.2%
6	1.88	201.0	4.1%	6.5	27.9%
8	1.41	320.0	5.1%	5.6	32.4%
24	0.27	36.0	8.2%	1.3	51.9%
48		7.7	9.7%	0.14	57.0%
72		1.1	9.8%		57.8%

^{3}H-labelled tetracycline hydrochloride was administered IV to two beagle dogs at 10 mg/kg to determine its distribution throughout the body (Kelly, 1964). Concentrations of tetracycline in the various tissues and fluids were determined by radioassay. Where it was possible to obtain the entire organ or tissue, the total antibiotic content within the tissue was calculated for the major tissues and is shown in summary form in Table 6. Four hours after dosing the organs containing the most tetracycline were the liver and kidney. A large proportion of the recovered antimicrobial activity was found in urine, intestinal contents and bile. With the exception of fat and spinal cord, radioactivity was found throughout all tissues and fluids examined.

Table 6. Distribution of Tetracycline in Tissues and Fluids of Dogs Four Hours After an Intravenous Dose of 10 mg TC/kg Live Weight

	Tetracycline, mg/kg		Total Tetracycline Content, μg	
Tissue or Fluid	Dog 98	Dog 78	Dog 98	Dog 78
Blood	NM	6.4	NC	NC
Liver	13.5	15.9	4,666	2,752
Kidney	14.1	75.8	776	3,024
Heart	3.3	5.7	315	297
Lungs	3.0	5.1	259	221
Brain	0.21	0.42	17	29
Skeletal Muscle	1.9	3.4	NC	NC
Femur-epiphysis	9.5	16.9	NC	NC
Femur-diaphysis	4.9	7.4	NC	NC
Bile	112.5	157.8	1,350	1,200
Spleen	3.5	5.7	56	84
Pancreas	6.0	3.0	119	35
Adrenals	0.9	2.6	NC	NC
Subcutaneous Fat	*	ND	NC	NC
Perirenal Fat	ND	*	NC	NC
Uterus	1.7	2.5	14	13
Trachea	NM	1.7	ND	12
Esophagus	1.6	2.2	41	38
Stomach	1.4	2.5	164	190
Duodenum	2.0	2.9	177	195
Jejunum	1.8	3.8	254	143
Ileum	1.9	3.5	197	186
Cecum	1.4	3.1	29	16
Colon	2.6	3.4	92	95
Rectum	1.6	2.4	54	29
Stomach Contents	1.5	0.9	14	35
Duodenal Contents	26.1	30.3	248	124
Jejunal Contents	37.9	28.2	459	592
Ileal Contents	187.0	202.0	1,552	384
Colon Contents	335.1	316.8	1,173	1,188
Rectal Contents	10.6	5.0	636	113
Thymus	2.3	2.0	NC	NC
Urinary Bladder	1.5	2.5	NC	NC
Diaphragm	3.1	3.0	88	160
Salivary Gland	1.7	3.4	NC	NC
Teeth	2.1	3.6	NC	NC
Urine	102.6	82.5	18,468	4,520
Spinal Cord	*	*		

NM = Not Measured; NC = Not Calculated; * = No detectable radioactivity present

Studies in rats, mice and rabbits have shown that tetracycline has a great affinity for bones and teeth. Under UV light, a brilliant yellow-gold induced fluorescence was observed almost instantaneously after IV administration in diffusely distributed tissues (with the exception of the brain) and within 30 minutes after IP injection (Milch et al, 1957). Fluorescence disappeared from all tissues except bone within 6 hours after a single parenteral injection. Bone fluorescence persisted throughout the 10-week period of observation following a single parenteral dose of tetracycline. Indeed, this property continues to be frequently used as a qualitative guide to previous exposure of an animal to tetracyclines.

Deposition of tetracycline in bone has also been studied quantitatively (Buyske et al, 1960). Unlabelled and ^{3}H-labelled tetracycline was administered to rats and the results of part of this work in which various ^{3}H-labelled tetracycline doses were administered to rats by single IP injections are shown in Table 7. A direct relationship was found between tetracycline dose and residue concentrations in blood and bone.

A single oral dose of 250 mg ^{3}H-labelled tetracycline was administered to male rats in a further experiment from the same publication. The results of this study showed a much lower concentrations of tetracycline in the bone (ie, 1.9 mg/kg at 24 h, 0.4 mg/kg at 4 weeks) than found in the previous study with intraperitoneal dosing.

Table 7. Concentration of Tetracycline in the Serum and Femora of Male Rats Following a Single Intraperitoneal Dose

	Tetracycline Concentrations, mg/L or mg/kg				
	Serum				Bone
Dose, mg/kg	2 h	4 h	6 h	24 h	24 h
150	48	54	47	35	186
100	38	38	31	15	140
65	28	17	21	4	112
45	21	18	15	1.4	91
30	9	7	4	0.2	17
20	8	6	3	0.1	12
10	6	3	1.9	<0.1	10

Single IV doses of ^{3}H-labelled tetracycline (15 mg/kg) were given to two rats with cannulated bile ducts and indwelling catheters in the ureters to measure the relative importance of bile in the excretion of tetracycline from the body. Radioactivity was measured in urine, bile and gastrointestinal tract contents for 24 hours after dosing. A summary of the results are presented in Table 8.

In another study, two rats and one beagle dog were given single intravenous doses of ^{3}H-labelled tetracycline hydrochloride (Eisner and Wulf, 1962). The results of this work, presented in Table 9, shows the percentage of the dose excreted via the urine and faeces.

Table 8. Excretion of Radioactivity by the Rat and the Dog Following Intravenous Administration of ^{3}H-labelled Tetracycline Hydrochloride

Species	Number of Animals	Dose mg/kg	Hours After Dose	Urine	Faeces	Total
Rat	2	15	0-72	69.2	19.5[a]	88.7
Dog	1	4	0-168	71.0	9.0	80.0

a = 0-48 hours

Table 9. **Excretion of Radioactivity from ^{3}H-labelled Tetracycline by Rats with Chronic Bile Duct Canulae for 24 Hours Following an IV Dose of 15 mg/kg**

	Volume, mL			% of Dose		
Rat Number	Urine	Bile	Recovery of Dose, (%)[a]	Urine	Bile	GI Tract
1	9.3	18.5	69.5	67.8	29.8	2.4
2	20.5	14.5	85.0	88.3	8.9	2.7

a = % of radioactivity from dose recovered in urine, bile and gastrointestinal tract contents over 0-24 hours after dosing

Although the results from the two rats did not agree well with each other, this experiment did indicate that relatively small amounts of tetracycline were excreted into the intestine by means other than the biliary route. A simpler experiment was also conducted in which the bile ducts of three rats were ligated. Whole blood, plasma and gastrointestinal tract contents were measured for radioactivity and microbiological activity following a single IV dose of 15 mg/kg of ^{3}H-labelled tetracycline. The results are summarised in Table 10.

Table 10. **Blood Concentration and Urinary Excretion of Tetracycline by Rats with Ligated Bile Ducts Following an Intravenous Dose of 15 mg/kg ^{3}H-labelled Tetracycline**

	Whole Blood, mg/L		Plasma, mg/L	
Hours	Microbiological	Radioactivity	Microbiological	Radioactivity
4	3.96	6.44	6.62	7.36
8	3.05	3.80	4.54	5.28
24	1.66	2.65	2.54	3.03
	% Recovery in Urine		% Recovery in GI Tract	
Hours	Microbiological	Radioactivity	Radioactivity	
0-4	35.3	36.3	1.7	
0-8	38.8	40.3	1.9	
0-24	47.6	49.1	4.8	

Less than 5% of the administered radioactivity was recovered from the gastrointestinal tract contents over the 24-hour period following dosing, while 49.1% of the administered dose was recovered in the urine. Tissues of these animals were not examined, so no conclusions can be made regarding total recovery of the administered dose. There is good agreement between the microbiological and radioactivity results for urine and blood, supporting the conclusions of the metabolism studies (Kelly and Buyske, 1960) which showed that little or no biotransformation of the tetracycline molecule had taken place in the body of rats and dogs which were used in the studies.

The absorption of tetracycline hydrochloride excreted in the bile of rats was evaluated using an *in situ* intestinal preparation (Adir, 1975). For comparative purposes, the absorption of the drug from an aqueous solution having the same pH as that of the bile was also determined. After 4 hours, the amounts of tetracycline absorbed from the bile and aqueous solutions were 72.9 and 77.3% respectively. There was no significant difference in the amount of drug accumulated in the gut tissue. The disappearance of the drug from the intestinal lumen was biexponential and the kinetic parameters appeared to be similar. It was concluded that tetracycline excreted in the bile is readily absorbed from the rat intestine. Accordingly, biliary excretion does not seem to account for

a significant elimination of this antibiotic from the body. The findings of this investigation raise questions regarding the other routes of elimination of tetracycline in addition to excretion in urine.

Pharmacokinetics in Pigs

Five Yorkshire x Hampshire crossbred pigs averaging 95 pounds body weight were given drinking water containing 600 mg of tetracycline hydrochloride/gallon for five consecutive days (Schumacher, 1968). Average daily intake of tetracycline during the 5-day medication period was 13.8 mg/kg. Average tetracycline levels in the blood and urine, measured periodically during and after medication, are shown in Table 11.

Table 11. Average Concentrations of Tetracycline in Blood and Urine of Pigs During and After Consumption of Drinking Water Containing 600 mg Tetracycline per Gallon for Five Consecutive Days

	Treatment Phase on Medication			Off Medication	
	Tetracycline, mg/L			Tetracycline, mg/L	
Time (h)	Blood	Urine	Time (h)	Blood	Urine
6	0.29	NS[1]	6	0.16	NS
12	0.33	NS	12	0.08	NS
24	0.33	9.6	24	ND[2]-0.06 (2/5)[3]	2.87
48	0.14	16.8	48	ND-0.05 (1/5)	2.21
72	0.16	19.8			
96	0.22	9.7			
120	0.37	13.4			

[1]NS = Not Sampled; [2]ND = Not Detected, less than 0.06 mg/L; [3](2/5) = Number of pigs with detectable value of tetracycline

It is apparent from these data that tetracycline is rapidly absorbed by pigs when administered via drinking water, reaching a peak of 0.33 mg/L of blood by 12 hours on medication. The concentrations of tetracycline are much higher in the urine than in the blood, and tetracycline continues to be found in the urine after it is no longer detectable in the blood.

Two studies were conducted to assess the bioavailability and pharmacokinetics of tetracycline fed to swine (Kniffen et al, 1989). In the first of these, four Yorkshire x Chester White gilts weighing approximately 32 kg were used in a 2 x 2 crossover design. In the first phase, two gilts were dosed with 22 mg/kg of an aqueous solution of tetracycline hydrochloride by stomach tube after being fasted for 16 to 20 hours, while the remaining two gilts were administered 11 mg/kg via an indwelling arterial cannula. Blood samples were obtained at predetermined intervals over the 72 hours following dosing. One week after completion of each of the initial phases of the trial, the gilts which had been dosed orally were dosed intra-arterially and the gilts which had been dosed intra-arterially were dosed orally. The results of the area under the curve measurements, shown in Table 12, indicate that tetracycline was poorly and variably absorbed after oral administration. The mean F value was 0.23, which corresponded to a bioavailabiltiy of 23%.

Table 12. Area Under the Plasma Tetracycline Concentration-time Curve (AUC) Values after Intravascular (IV) and Oral Administration of Tetracycline Hydrochloride to Swine at the Dosage of 11 and 22 mg/kg, respectively

	Gilt Number					
Variable (units)	1	2	3	4	Mean	SD
AUC IV (h·mg/L)	50.20	59.40	65.00	66.00	60.15	7.24
AUC Oral (h·mg/L)	10.60	32.40	37.40	32.60	28.25	11.99
F*	0.11	0.27	0.29	0.25	0.23	0.08

*F (fraction absorbed) was calculated as the dose-corrected ratio of the oral to-intravascular area under the curve

Pharmacokinetic variables describing the disposition of tetracycline in gilts indicated that after intravascular administration of tetracycline hydrochloride, plasma concentration of tetracycline decreased in a triexponential fashion (Table 13).

The investigators concluded that for all gilts, a triexponential equation provided a better fit to the data than did a biexponential equation. The pharmacokinetic behaviour of the drug can best be described by a 3-compartment open model. The terminal elimination phase (g) rate constant of 0.043 $\pm$ 0.01 /h corresponded to a biological $t_{1/2}$ of approximately 16 hours. The large Vd_{area} of 4.5 + 1.06 L/kg suggested that tetracycline was widely distributed in swine tissues. After oral administration of tetracycline hydrochloride, the resulting plasma concentrations of tetracycline were too variable to be fitted to a rational absorption curve.

These authors also fed a ration containing 550 mg/kg of tetracycline hydrochloride to three gilts. Tetracycline plasma concentrations were determined at selected times during 96 hours after exposure to the medicated feed and were found to reach a fairly constant value of 0.3-0.5 mg/L after 4 hours after start of medicated feeding.

Table 13. Pharmacokinetic Variable for the disposition of Tetracycline in Gilts after Intra-arterial Administration of Tetracycline Hydrochloride (11 mg/kg)

	Gilt Number					
Variable (units)	1	2	3	4	Mean	SD
A (mg/L)	78.10	69.80	95.50	90.20	83.40	11.63
B (mg/L)	7.52	6.89	7.80	8.78	7.75	0.79
C (mg/L)	0.861	0.750	1.60	0.623	0.959	0.439
a (/h)	18.50	10.00	11.70	13.70	13.48	3.68
b (/h)	0.307	0.221	0.227	0.243	0.250	0.039
g (/h)	0.0436	0.0382	0.0579	0.0321	0.0430	0.0110
Cl (L/kg/h)	0.219	0.185	0.169	0.167	0.185	0.024
Vd_{area} (L/kg)	5.03	4.85	2.92	5.19	4.50	1.06
K (/h)	1.78	1.34	1.50	1.60	1.56	0.18
Vp (L)	0.127	0.142	0.105	0.110	0.121	0.017

A, B, and C = zero time plasma drug concentrations; a, b, and g = disposition rate constants; Cl = clearance; Vd_{area} = volume of distribution; K = elimination rate constant; Vp = volume of central compartment.

Six Yorkshire pigs, weighing 46 to 83 kg and fitted with jugular vein catheters, were given drinking water containing 400 mg/L tetracycline for 3 days (Luthman et al, 1989). After a resting period of one week, the study was repeated with drinking water containing 800 mg/L tetracycline. Serum levels, measured at 2-hour intervals during the day, were highest in each animal in the late evening, followed by a decrease during the night. The highest serum levels with 400 mg/L tetracycline were 0.30 to 0.47 mg/L, while the corresponding tetracycline levels with the 800 mg/L treatment were 0.65 to 1.15 mg/L. Six other pigs with a mean weight of 25 kg were given feed containing 1000 mg/kg tetracycline for nine days. The pigs were fed twice daily at 07.00 and 16.00. Blood samples, taken at 2-hour intervals between 09.00 and 19.00, showed mean serum levels ranging from 0.13 to 0.20 mg/L. In a third experiment, seven pigs were given single oral doses of 40 mg/kg tetracycline. Five of the pigs were fasted overnight prior to dosing while two pigs were fed 2 hours prior to tetracycline administration. Feeding was not allowed during the 24-hour sampling period after dosing. Tetracycline serum levels were highest 2 to 4 hours after dosing and peak levels were less than 1 mg/L for the fed pigs compared to about 1.5 to 5.0 mg/L for the fasted pigs. It is clear that feeding significantly reduced the bioavailability of orally administered tetracycline in these pigs.

Groups of six pigs weighing approximately 15 kg received drinking water containing 147 mg/kg of tetracycline continuously for 1, 3 and 7 days (Rooney, 1990). Resulting average blood and lung concentrations are shown in Table 14.

Table 14. Average Tetracycline Levels in blood and Lung Tissues from Pigs Receiving Drinking Water Containing 147 ppm of Tetracycline

	Tetracycline, mg/L	
Time (days)	Blood	Lung
1	0.262	0.372
3	0.268	0.373
7	0.256	0.366

Concentrations of tetracycline in the lung tissues averaged 39 to 43% higher than those found in the blood.

Pharmacokinetics in Cattle and Sheep

Calves approximately one month of age were used to study the bioavailability of tetracycline (Luthman et al, 1989). The calves were fed 2 litres of a conventional milk replacer twice daily and had free access to hay. In addition, about 0.3 kg of a milk replacer was offered shortly after the first daily milk feeding, most of which was consumed in 3-4 hours. The calves were fed 25 mg/kg bw mixed into the milk replacer twice daily. The calves were treated in this manner for five days. Blood was sampled on days 1, 2 and 5 as shown in Table 15.

Table 15. Mean Serum Concentrations (mg/L) of Tetracycline in Calves Fed 25 mg/kg b.w. Twice Daily of Tetracycline in the Milk Replacer

	Time of Day				
	10.00	12.00	16.00	18.00	20.00
Day 1	1.78±0.26	1.85±0.27	1.35±0.39	2.25±0.30	
Day 2	0.95±0.18				
Day 5	2.27±0.32	2.10±0.32		2.23±0.30	2.10±0.34

The same authors also conducted a second study in which calves were given 50 mg /kg bw of tetracycline in

the milk replacer at the first morning feeding. After one week the same dose was given in 1 litre of water 4 hours after the morning feeding. The calves were fed concentrate and had access to hay during the whole sampling period. Blood was sampled after 1, 2, 4, 6, 8 and 10 hours. Serum tetracycline levels were significantly higher for the calves receiving tetracycline in the milk replacer than after the same dose given in water 4 hours after the first daily milk feed. C_{max} was achieved after 4 hours in all animals. It was postulated that the 0.3 kg of concentrate consumed prior to the dose of tetracycline in water may have been responsible for the lowered bioavailability of the tetracycline.

Tetracycline was administered by a rapid IV injection into the jugular vein of six Israeli-Friesian cows and four Awassi ewes at a dose of 20 mg/kg of bw, and periodic blood samples were obtained over the 72 hours following dosing (Ziv and Sulman, 1974). As concentrations of tetracycline in the blood serum (determined by microbiological assay) were similar for cows and ewes, the data were presented as the combined data. Venous blood concentration curves of all cows and sheep in this experiment were biexponential, and the data appeared to be adequately described by the 2-compartment open model. Pharmacokinetic values calculated from the data are presented in Table 16.

Table 16. Pharmacokinetic Constants for Tetracycline Administered Intravenously to Cows and Ewes at 20 mg/kg of Body Weight

Value	Definition and Method of Calculation	Mean	SD
A μg/ml	Extrapolated zero-time serum drug concentration of the α-phase	20.5	3.6
B μg/ml	Extrapolated zero-time serum drug concentration of the β phase	12.5	1.7
C_p° μg/ml	A + B	33.0	4.8
α h^{-1}	Slope of the initial phase of serum TC concentration	1.236	0.14
β h^{-1}	Slope of the second phase of serum TC concentration	0.054	0.01
k_{12} h^{-1}	First-order distribution rate constant between the central compartment and the peripheral compartment = $AB(\beta-\alpha)^2/ A\beta+B\alpha(A+B)$	0.655	0.21
k_{21} h^{-1}	First-order distribution rate constant between the peripheral compartment and the central compartment = $(A\beta)+(B\alpha)/A+B$	0.502	0.08
k_{el} h^{-1}	Overall rate constant for TC elimination by various routes = $(\alpha\beta)(A+B)/A\beta+B\alpha$	0.121	0.04
$t^1/_2k_{el}$ h	Half-life of overall rate constant for drug elimination = $0.693/k_{el}$	5.73	0.6
V_p % bw	Specific total volume of distribution = V_p/kg of body weight 100	332.2	23.5
F_c	Fraction of the drug in the body located in the central compartment = β /k_{el}	0.45	0.06

In another study, a solution containing a mixture of 100 mg of unlabelled tetracycline and 0.8 mCi of ^{3}H-labelled tetracycline (3 mCi/mg) was injected into the right front quarter of an Israeli-Friesian dairy cow after a normal morning milking (Ziv et al, 1974). Milk samples and blood samples were taken at hourly intervals for 8 hours after treatment. The concentration of tetracycline in the treated quarter decreased exponentially during the first 7 to 8 hours after treatment. The rates of decrease of radioactivity and of tetracycline by microbiological assay were essentially parallel. Antibiotic activity was not detectable in the blood serum or in the milk of the untreated quarters after treatment. However, radioactivity counts, 3 to 6 times greater than the background counts, were recorded in the serum and in milk from the nontreated quarters from 1 to 8 hours after the radioactive-labelled tetracycline was injected into the treated quarter. It appears that the principal mode of transfer of tetracycline from treated to untreated quarters was via the bloodstream.

Pharmacokinetics in Poultry

Two experiments by Anadon, et al. (1985) studied the pharmacokinetics of tetracycline in chickens. Forty to sixty-day-old broiler chickens weighing 1.5 to 2.0 kg were used. In the first experiment, tetracycline hydrochloride was injected into the left brachial vein of each of a group of six chickens at a single dose of 65 mg/kg bw. Plasma tetracycline concentrations, measured by HPLC analysis, are shown in Table 17. The pharmacokinetic parameters which describe the distribution and elimination phases of tetracycline after the 65 mg/kg dosage are given in Table 18.

Table 17. Plasma Concentrations (mg/L) of Tetracycline in Chickens Receiving an Intravenous Dose of 65 mg/kg of Body Weight

Time after Administration	Tetracycline, mg/L	
	Mean	SEM
15 min	690	17
30 min	359	12
1 hour	81	5
2 hours	52	6
4 hours	29	4
8 hours	12	2.3
12 hours	3.8	0.9

Table 18. Pharmacokinetic Parameters for Tetracycline Administrated IV at 65 mg/kg of Body weight to Chickens

Parameters	Mean
A, mg/L	2000±450[1]
B, mg/L	82±6
α, h^{-1}	4.274±0.489
ß, h^{-1}	0.252±0.009
$t_{1/2}\alpha$, h	0.162
$t_{1/2}$ß, h	2.772
VC=Central compartment, L/kg	0.037
Peripheral compartment, L/kg	0.137
Total vol of distribution, L/kg	0.174
K_{12}, h^{-1}	1.497
K_{21}, h^{-1}	0.408
K_{10}, h^{-1}	2.614
Conc. time curve, mg/h/kg	796.38
Total body clearance, L/h/kg	0.0979
K_{12}/K_{21}	3.667
K_{12}/K_{10}	0.573
K_{21}/K_{10}	0.156

[1]Mean ± standard error for 6 chickens

In the second experiment, bile ducts were cannulated with polyethylene tubing. Two groups of 3 chickens each were given a single IV injection of tetracycline at doses of 10 mg/kg and 15 mg/kg, respectively. Two other groups of chickens received single oral doses of 100 mg/kg and 200 mg/kg of tetracycline. Tetracycline was excreted into the bile following both IV and oral administration. The biliary excretion data after IV administration are shown in Table 19.

Table 19. Biliary Excretion Rates (μg/h) of Tetracycline (mean ± SEM) After Intravenous Administration in Chickens

	Tetracycline Dose	
Time after Administration (h)	10 mg/kg	15 mg/kg
0.5	225.0±21.0	329.0±17.3
1	407.0±10.8	606.0±36.2
2	308.6±36.2	275.3±13.8
3	44.8±5.6	118.0±13.1
4	24.0±3.8	70.4±8.5
5	15.7±1.9	39.1±2.9
6	8.0±1.7	20.5±4.9

With both of the doses there was a log-linear decline of bile concentration indicative of a one compartment model. The investigators calculated that with the IV doses used, the median rate constants for drug biliary excretion were 0.834 and 0.665 h^{-1}, for the 10 and 15 mg/kg doses, respectively. The corresponding median half-lives were calculated to be 0.831 and 1.05 hours. During the 6 hour period following IV administration, 7% of the administered tetracycline was excreted into the bile. After oral administration, the percentages of the total doses recovered from the bile were much lower than following IV administration. The average maximum cumulative amount of tetracycline excreted into the bile with 8 hours was 260 and 1480 μg after oral administration of 100 and 200 mg/kg respectively, which corresponds to 0.2 and 0.5% of the total doses administered.

The pharmacokinetics of tetracycline was studied in turkeys (Gonzelman a et al., 1985). In the first of these experiments, five Broad-breasted White turkeys weighing 3 to 4 kg were used. Feed was withheld for 12 hours before dosing, but the birds had free access to water. After collection of pretreatment blood samples, 25 mg/kg bw of tetracycline hydrochloride, buffered with ascorbic acid, was injected intravenously. Blood samples, taken periodically after dosing, were assayed microbiologically for tetracycline content. Results of this work are shown in Tables 20 and 21.

Table 20. Mean (± SD) Concentration (mg/L) of Tetracycline in Serum Samples Collected After a Single Intravenous Injection of Tetracycline Hydrochloride at 25 mg/kg Body weight

Time After Injection (min)						
5	15	30	60	90	127	232
47.4±9.0	27.4±7.2	16.7±1.8	12.9±2.2	9.5±2.4	6.7±2.6	5.0±2.5

Kinetic parameters calculated from these data are shown in Table 21.

Table 21. Pharmocokinetics of Tetracycline in Turkeys Following Intravenous Administration of a Single 25 mg/Kg Dose of Tetracycline Hydrochloride

Kinetic Parameter	Units	Measurement	Kinetic Parameter	Units	Measurement
A	mg/L	62.8±25	k_{el}	min^{-1}	0.0230±0.0115
B	mg/L	15.8±4.3	V_c	ml/kg	345±136
α	min^{-1}	0.1210±0.057	$t_{0.5}$	h	2.41±1.03
β	min^{-1}	0.0054±0.002	$Vd_{(area)}$	ml/kg	1377±373
k_{12}	min^{-1}	0.0742±0.04	Cl_B	ml/min-kg	7.70±1.9
k_{21}	min^{-1}	0.0293±0.0125	$AUC_{(IV)}$	$\mu g \cdot h/ml$	59.74

In a second experiment, nine turkeys of similar weight to those used in the first experiment were given a single oral dose of tetracycline hydrochloride at 25 mg/kg. The drug solution (5 mg/ml) was delivered into the proventriculus with a feeding tube. Table 22 shows tetracycline content of blood samples taken by venipuncture over 12 hours after dosing.

Table 22. Mean (±SD) Concentrations (mg/L) of Tetracycline in Serum Samples Taken After A Single Oral Dose of Tetracycline HCl at 25 mg/kg

Time After Administration (hours)											
0.25	**0.50**	**0.75**	**1.0**	**1.5**	**2**	**3**	**4**	**6**	**8**	**10**	**12**
2.4	3.3	3.4	3.8	3.3	3.0	2.4	1.7	1.0	0.55	0.34	0.17
±.45	±.71	±1.0	±.96	±1.0	±.96	±.80	±.54	±.68	±.32	±.12	±.12

The pharmocokinetics parameters are shown in Table 23.

Table 23. Pharmacokinetics of Tetracycline in Turkeys Following a Single Oral Dose of Tetracycline Hydrochloride at 25 mg/kg of Body Weight

Kinetic Parameter	Units	Measurement
C_0	$\mu g/ml$	5.07±1.4
(hypothetical) k_a	h^{-1}	3.0963±1.189
$t_{0.5(a)}$	h	0.265±0.14
k_d	h^{-1}	0.3086±0.087
$t_{0.5(apparent)}$	h	2.44±0.77
AUC_{po}	$\mu g \cdot h/ml$	5.29
F^1	%	9

$^1F = (AUC)_{po}/(AUC)_{iv}$

The authors stated that the pharmacokinetic parameters determined above suggest that effective serum levels of tetracycline may be difficult to attain by admixture of the drug in drinking water. Although the rate of absorption is rapid, the extent of the absorption is incomplete (9% of the administered dose), and the elimination rate is rapid. The authors calculated that a concentration of 8.5 g/L of drinking water would be expected to

provide serum concentrations of 0.5-1.0 mg/L.

Summary of Pharmacokinetics Across Species

The studies reported above show that tetracycline is rapidly absorbed following oral administration in all species, but the degree of absorption is poor. Fasting of the animal overnight prior to administering an oral dose of tetracycline has been shown to increase absorption. Numerous studies with chlortetracycline and oxytetracycline have shown that the presence of calcium in a medicated feed will cause a decrease in the bioavailability of these tetracyclines, and it would be expected that calcium in the dosing medium or in the stomach of the animal would have the same effect on the bioavailability of tetracycline. Tetracycline is widely distributed throughout the body soon after dosing, and with the exception of those residues which are bound to bone, is rapidly excreted. The half-life of a dose of tetracycline in poultry appears to be shorter than in calves and swine. The primary route of excretion of tetracycline from the body is via the urine, but considerable amounts are also excreted via the faeces. It is believed that excretion via the bile accounts for the main route for absorbed tetracycline to get into the faeces. However, studies have shown that tetracycline excreted in the bile is readily available for reabsorption back into the body.

TISSUE RESIDUE DEPLETION STUDIES

General

Data from pharmacokinetic studies in laboratory animals and in target species, and metabolism studies in laboratory animals have shown that tetracycline is rapidly, but poorly, absorbed following oral administration. It is widely distributed throughout the body and, with the exception of those residues which are bound to bone, is rapidly excreted. Metabolism studies in rats and dogs indicate that little or no biotransformation of the tetracycline molecule occurs within the body. Unchanged tetracycline is eliminated primarily via the urine, but considerable amounts are also excreted in the faeces. Most of the residue data have been obtained using microbiological assay procedures. Recently, the development of HPLC methods has made available an alternative method for residue monitoring. The available residue data for the use of tetracycline hydrochloride in target species are summarised in the following section.

Depletion of Residues from Edible Tissues of Swine

Yorkshire x Hampshire crossbred pigs averaging about 20 kg in weight were offered drinking water containing 750 mg/gallon of tetracycline hydrochloride for 14 consecutive days (Berger, 1972). The calculated average daily intake of tetracycline was 24.2 mg/kg over the 14-day medication period. Tissue samples taken periodically after withdrawal of medicated drinking water were assayed microbiologically for residues of tetracycline. Results are summarised in Table 24.

Table 24. Average Residue Levels of Tetracycline in Blood and Tissues of Swine at Various Withdrawal Times After Receiving Drinking Water Containing 750 mg/gallon of Tetracycline HCl for 14 Consecutive Days

	Tetracycline Residues (mg/L or kg)				
Withdrawal Day	Blood	Muscle	Liver	Kidney	Fat
0	0.89	0.71	1.76	3.43	ND-0.078
4	ND-0.055	0.096	ND-0.131	0.241	ND
7	ND[1]	ND-0.085	ND-0.097	0.147	ND
10	ND-0.05	ND	0.10	0.170	ND
14	ND	ND	ND-0.076	ND-0.089	ND

[1]ND = Not Detected, less than sensitivity of microbiological assay method (0.06 mg /L)

Depletion of Residues from Edible Tissues of Cattle

Jersey heifer calves, average weight 76 kg, were treated by oral drench with 400 mg of tetracycline hydrochloride per head daily for 5 consecutive days (Berger, 1967). The calves were sacrificed at 0, 2, 5 and 7 days following withdrawal of treatment for assay for tetracycline content of tissues and fluids by microbiological assay. The results of these assays are summarised in Table 25.

Table 25. Average Residues of Tetracycline in Tissues and Fluids from Calves at Various Withdrawal Times after Receiving 400 mg Tetracycline/Calf/Day for Five Consecutive Days

	Tetracycline Residue (mg/kg)					
Withdrawal Day	Muscle	Liver	Kidney	Fat	Blood	Urine
0	ND-0.17	0.38	0.71	ND	0.10	10.47
2	ND[1]	ND	ND-0.12	ND	ND	0.91
5	ND	ND	ND	ND	ND	ND
7	ND	ND	ND	ND	ND	ND

[1]ND = Not Detected, less than sensitivity of microbiological assay method (0.06 mg/L for blood and 0.1 mg/kg for tissues); Bile was found to give false positive results in the microbiological assay

In another study bull calves with an average weight of 68 kg were treated with tetracycline hydrochloride by oral drench daily for 10 consecutive days at the dose of 22 mg/kg b.w. per day (Berger, 1975). Tissue and blood samples were assayed for tetracycline residue content by microbiological assay at various withdrawal times following cessation of treatment. The results of these assays are summarised in Tables 26 and 27.

Table 26. Average Residues of Tetracycline in Tissues of Calves at Various Withdrawal Times After Cessation of Treatment with Tetracycline at 22 mg/kg bw for 10 Consecutive Days

	Tetracycline, mg/kg			
Withdrawal Day	Muscle	Liver	Kidney	Fat
2	ND[1]	0.21	0.33	ND
4	ND	ND-0.12	0.17	ND
8	ND	ND	ND-0.12	ND
12	ND	ND	ND-0.13	ND
15	ND	ND	ND	ND

[1]ND = Not Detected, less than sensitivity of microbiological assay method (0.10 mg/kg)

Table 27. Average Residues of Tetracycline in Blood of Calves at Various Times Following Cessation of Treatment at 22 mg/kg bw for 10 Consecutive Days

Time Following End of Treatment	Tetracycline Average, mg/L	Time Following End of Treatment	Tetracycline Average, mg/L
Hours		Days	
2	1.03	2	0.09
4	0.79	3	0.06
8	0.62	5	ND-0.04
12	0.45	7	ND-0.03
24	0.20	9	ND-0.03
		11	ND[1]
		13	ND
		15	ND

[1]ND = Not Detected, less than sensitivity of microbiological assay method (0.0375 mg/L)

In another experiment, Holstein bull calves approximately 2 weeks of age, average weight 41.6 kg, were treated for ten consecutive days with tetracycline HCl oral boluses to furnish 22 mg/kg bw daily (Eggert, 1978a). The calves were fed a nonmedicated milk replacer twice daily during the experiment. The boluses were administered each day following the morning feeding. Three calves each were sacrificed at 0, 3, 7 and 10 days following completion of the treatment for analysis of residues of tetracycline in blood and tissue samples. The average results are summarised in Table 28.

Table 28. Average Residues of Tetracycline in Blood and Tissue Samples from Calves Following Cessation of Treatment with Tetracycline·HCl-Boluses at 22 mg/kg bw for 10 Days

	Tetracycline, mg/kg				
Days Withdrawal	Muscle	Liver	Kidney	Fat	Blood
0	0.92	1.98	3.46	0.48	0.84
3	0.51	1.27	2.39	0.41	0.63
7	0.20	0.36	0.96	0.13	0.21
10	0.08	0.17	0.43	0.06	0.08

Since all calves showed measurable residues of tetracycline after a ten-day withdrawal period on the above experiment, another study was conducted with Holstein calves (average weight 54.3 kg) to study longer withdrawal times (Eggert, 1978b). The calves were given tetracycline oral boluses to provide 22 mg/kg bw daily for ten consecutive days. The calves were fed a nonmedicated milk replacer twice daily, plus a commercial nonmedicated calf starter grain mix *ad libitum*. Three calves each were sacrificed at 0, 10, 14, 17, 20 and 24 days after cessation of tetracycline treatments for microbiological assay of residues in blood and tissues. The results of these assays are shown in Table 29.

Table 29. Average Residues of Tetracycline in Blood and Tissues Samples from Calves Following Cessation of Treatment with Tetracycline Boluses Furnishing 22 mg/kg Daily for 10 Days

	Tetracycline, mg/kg				
Days Withdrawal	Muscle	Liver	Kidney	Fat	Blood
0	0.27	0.90	1.65	0.17	0.36
10	ND[1]	ND	ND-0.09	ND	ND
14	ND	ND	ND	ND	ND
17	ND	ND	ND	ND	ND
20	ND	ND	ND	ND	ND
24	ND	ND	ND	ND	ND

[1]ND = Not Detected, less than sensitivity of microbiological assay method, 0.0375 mg/L for blood and 0.1 mg/kg for tissues

The results of this experiment differed from those obtained in the earlier experiment in two respects. First the residue levels found at zero day-withdrawal in the present experiment were lower for all tissues than for the earlier experiment. Secondly, no detectable residues were found in any of the tissues taken at 10-day withdrawal with the exception of one kidney sample which showed an apparent residue of 0.09 ppm (which is less than the validated limit of the assay method), while in the earlier experiment all tissue samples taken at the 10-day withdrawal period showed positive residues. The only difference in the procedures used in the two experiments is that in the second experiment a commercial calf starter gratin ration was fed in addition to the milk replacer diet. This difference in feeding management may be responsible for the disparity between the two experiments. Bradley et al. (1982) found in studies with calves given oral doses of chlortetracycline that milk fed calves absorbed a larger fraction of the chlortetracycline dose, had a smaller volume of distribution and a smaller overall body clearance rate than did conventionally fed calves. A similar effect might be anticipated with tetracycline.

Depletion of Residues from Edible Tissues of Chickens

A study was conducted in which broiler chickens were given drinking water containing 2830 mg tetracycline hydrochloride/gallon of water for five consecutive days. Tetracycline residues were measured by microbiological assays for blood and tissues during medication and after cessation of treatment (Hewell, 1987). The results of this study are summarised in Table 30.

Depletion of Residues from Eggs

Roudaut, et al (1989) treated laying hens for five consecutive days with drinking water containing 250 and 500 mg tetracycline/litre and for seven consecutive days with feed containing 300 and 600 ppm of tetracycline. Residues were measured separately in the yolk and albumin fractions of the egg. A summary of the residue data, measured by microbiological assay, is shown in Tables 31 and 32.

Table 30. **Average Tetracycline Residues in Blood and Tissues of Broiler Chickens During and After Supply with Drinking Water Containing 2830 mg/gallon of Tetracycline for Five Days**

	Tetracycline, mg/kg				
During Treatment	Sex	Blood	Muscle	Liver	Kidney
0 hour	Male	ND[1]	NM[2]	NM	NM
	Female	ND	NM	NM	NM
6 hours	Male	0.773	NM	NM	NM
	Female	0.590	NM	NM	NM
12 hours	Male	0.765	NM	NM	NM
	Female	0.214	NM	NM	NM
24 hours	Male	0.383	0.793	1.870	6.225
	Female	0.702	0.908	2.380	6.450
36 hours	Male	0.649	NM	NM	NM
	Female	0.704	NM	NM	NM
After Treatment					
0 hour	Male	0.433	0.821	1.845	6.113
	Female	0.578	0.978	1.973	6.210
12 hours	Male	0.083	0.198	0.368	1.450
	Female	ND	ND-0.118	ND-0.175	0.681
24 hours	Male	ND-0.068	0.140	0.232	1.390
	Female	ND-0.042	ND-0.099	ND-0.150	0.605
48 hours	Male	ND	NM	NM	NM
	Female	ND	NM	NM	NM

[1]ND = Not Detected, less than sensitivity of method, 0.0375 mg/L for blood and 0.1 mg/kg for tissues; [2]NM = Not Measured

Table 31. Average Tetracycline Residues (±SD) in Albumin and Yolk Fractions of the Egg and the Whole Eggs Following a Five Day Treatment with 250 and 500 mg/L Tetracycline in Water

	Tetracycline, mg/kg					
	Albumin		Yolk		Whole Egg	
Dose	250 ppm	500 ppm	250 ppm	500 ppm	250 ppm	500 ppm
Day of treatment						
5	0.11±0.03	0.19±0.07	0.52±0.19	0.81±0.44	0.25±0.08	0.40±0.19
Withdrawal Day						
1	0.10±0.03	0.17±0.03	0.52±0.14	1.02±0.52	0.25±0.07	0.44±0.19
2	<0.07	0.07±0.02	0.46±0.13	1.11±0.48	0.17±0.05	0.40±0.17
3		<0.07	0.42±0.12	0.91±0.46	0.17±0.06	0.29±0.15
4			0.31±0.10	0.80±0.37	0.11±0.04	0.27±0.14
5			0.22±0.08	0.53±0.19	0.08±0.03	0.18±0.07
6			0.20±0.07	0.40±0.17	0.07±0.02	0.12±0.06
7			<0.15	0.32±0.18		0.10±0.06
8				0.21±0.16		0.07±0.05
9				0.19±0.15		0.06±0.05
10				<0.15		

Table 32. Average Tetracycline Residues (±SD) in Albumin and Yolk Fractions of the Egg and the Whole Eggs Following a Seven Day Treatment with 300 and 600 ppm of Tetracycline in Feed

	Tetracycline, mg/kg					
	Albumin		Yolk		Whole Egg	
	300 ppm	600 ppm	300 ppm	600 ppm	300 ppm	600 ppm
Day of Treatment						
7	0.19±0.02	0.30±0.08	0.87±0.07	1.41±0.40	0.41±0.04	0.61±0.15
Withdrawal Day						
1	0.12±0.03	0.20±0.06	0.98±0.09	1.51±0.45	0.41±0.05	0.60±0.15
2	<0.07	0.08±0.02	1.12±0.18	1.58±0.46	0.40±0.07	0.54±0.15
3		<0.07	0.78±0.18	1.10±0.25	0.27±0.07	0.37±0.08
4			0.63±0.15	0.94±0.30	0.21±0.06	0.31±0.09
5			0.49±0.15	0.64±0.19	0.16±0.05	0.22±0.06
6			0.31±0.06	0.48±0.17	0.10±0.02	0.15±0.06
7			0.27±0.06	0.39±0.15	0.09±0.02	0.13±0.05
8			0.20±0.04	0.37±0.15	0.07±0.02	0.11±0.04
9			<0.15	0.28±0.11		0.09±0.03
10				0.24±0.08		0.07±0.02
11				0.18±0.11		0.05±0.03
12				<0.15		

Residues in Milk

Tetracycline is generally little used for intramammary infusion for control of mastitis because it tends to cause too much tissue irritation. However tetracycline residues can result from other therapeutic uses of tetracycline in lactating animals. A study was conducted using six lactating dairy cows which had two 500-mg tetracycline boluses placed in the uterus two to six days following parturition (Brown, 1961). Milk samples were taken from each cow prior to treatment and at 12-hour intervals following treatment. Microbiological assays showed no antibiotic activity in any of the pretreatment samples. The results of the assays of the post treatment samples is shown in Table 33.

Table 33. Residues (mg/L) in Milk from Cows Following Intrauterine Treatment with 1000 mg of Tetracycline in Bolus Form

	Tetracycline, mg/L					
Hours Postmedication	Cow 1	Cow 2	Cow 3	Cow 4	Cow 5	Cow 6
12	0.26	0.14	0.28	0.13	0.06	0.39
24	0.22	0.08	0.23	0.05	0.06	0.41
36	0.16	0.13	0.14	0.20	0.12	0.36
48	0.14	0.05	0.12	0.05	0.11	0.28
60	ND[1]	ND	ND	ND	ND	0.25
72	ND	ND	ND	ND	ND	0.11
84	ND	ND	ND	ND	ND	ND
96	ND	ND	ND	ND	ND	ND

[1]ND = Not Detected (LOD not given)

Another study was conducted in which the Delvo-test P was used to check milk samples for antibiotic residues (Haaland et al, 1984). In this test, a solution of tetracycline was infused into the uterus, generally as a treatment for retained placenta, but also for other reasons. Milk samples were tested from each cow at 12 hour intervals following treatment until the milk was clear of all detectable antibiotic activity. The results obtained for cows treated with tetracycline are shown in Table 34.

Table 34. Results with Cows Given Intrauterine Infusions of Tetracycline for Treatment of Retained Placenta or Other Uterine Infections.

Tetracycline Treatment	No. of Cows	Hours to Clear Antibiotic Residues
1 infusion of 3,000 mg	9	72-84
1 infusion of 500 mg	4	60
3 infusion of 500 mg	4	72-84

The nine cows receiving the single infusions of 3,000 mg of tetracycline for retained placenta required 72 to 84 hours to clear antibiotic residues from the milk. More than 60 hours were required for the milk to clear in 4 cows treated once with a 500-mg dose. Three treatments, each containing 500 mg of tetracycline given at a 24-hour interval, resulted in persistence of milk residues for the same period of time as one 3,000-mg infusion.

METHODS OF ANALYSIS FOR RESIDUES IN TISSUES

Analytical procedures for tetracyclines based on both microbiological and chemical assays have been discussed earlier (see monograph on Chlortetracycline in this volume). In general, chemical and microbiological methods measure the three commonly used tetracyclines, tetracycline, oxytetracycline and chlortetracycline together as a group. The chemical and microbiological procedure give similar quantitative results for the same antibiotic with the modern chemical testing having at least a ten fold sensitivity advantage while being less cost effective.

APPRAISAL

Tetracycline was last evaluated by JEFCA at the 12th Meeting in 1968, together with oxytetracycline and chlortetracycline. At that time, for tetracycline maximum residue levels were recommended of 0.5 mg/kg in meat, 0.3 mg/kg in eggs and 0.1 mg/kg in milk, calculated as base.

The predominant use of tetracycline is as a therapeutic drug. It is rapidly but only moderately well absorbed from the GI tract and is eliminated in both urine and faeces either unchanged or in a microbiologically inactive form. There is no evidence that tetracycline is significantly metabolised *in vivo* although some isomerisation of the drug can occur, either in the animal and/or during isolation. Microbiological assay is therefore a satisfactory method to acquire depletion of tetracycline from tissues. Tissue depletion measurements conducted both by microbiological and chemical assay give very similar results. Differences in microbiological potency of individual tetracyclines, however, requires a preference for chemical determination of residues for regulatory purposes. Since tetracycline undergoes minimal metabolism, it is the appropriate marker compound for determination of residues in tissues.

Serum level and residue studies indicate that tetracycline is both rapidly absorbed and quickly cleared from edible tissues following oral administration. Kidney and liver tissues in all species show the largest concentration of tetracycline both at the withdrawal from medication, and at any time point during the withdrawal period. When liver and kidney tissues are in compliance with any designated MRL. residues in muscle will be less than 10% those found in kidney and fat should not cause any problem. Either liver or kidney (or both) could be considered as target tissue. It is recommended that kidney is the preferred target tissue.

In pigs given tetracycline at 24 mg per kg body weight per day for 14 days in drinking-water, mean residue levels were 0.2 and 0.1 mg/kg in kidney and liver, respectively, 4 days after withdrawal of medication. Calves on the same dose had average residue levels of 0.4 and 0.17 mg/kg in kidney and liver, respectively, 10 days after withdrawal of medication. In chickens given 620 mg/L of tetracycline in drinking water for 5 days, average residue levels in kidney and liver were 1.4 and 0.2 mg/kg, respectively, 24 hours after withdrawal of the drug, while residues in whole egg averaged 0.27 mg/kg at 4 days withdrawal, declining to <0.06 mg/kg 10 days after withdrawal. A dose of 3 g given to a lactating cow by intrauterine infusion led to residues of <0.10 mg/kg in milk 84 hours after dosing.

Maximum Residue Limits

In reaching its decision on MRLs for tetracycline (and chlortetracycline) the Committee considered the following:

- MRLs were recommended for oxytetracycline at the 36th meeting of the Committee for all species of 600 μg/kg in kidney, 300 μg/kg in liver, 100 μg/kg in muscle, 100 μg/kg in milk, 200 μg/kg in eggs, and 10 μg/kg in fat. These levels were the lowest detectable by validated antimicrobial methods;

- chlortetracycline and tetracycline have been allocated a group ADI of 0-3 μg per kg of body weight with oxytetracycline;

- modern analytical techniques allow much more sensitive and specific assays than those provided by

antimicrobial assays;

- the recommended target tissues for residue analysis in cattle, pigs and poultry are kidney and muscle. Based on limited data, the kidney is the recommended target tissue for sheep; and

- the marker residue for all three substances is parent drug.

The Committee recommended the following temporary MRLs for (both) tetracycline (and chlortetracycline) in cattle, pigs and poultry, expressed as parent drug:

Muscle - 100 μg/kg
Liver - 300 μg/kg
Kidney - 600 μg/kg
Eggs (poultry) - 200 μg/kg

The Committee also recommended temporary MRLs for sheep liver and kidney of 300 μg/kg and 600 μg/kg, respectively, expressed as parent drug.

The following information is required for evaluation in 1996:

- The results of residue depletion studies in cattle, sheep, pigs and poultry to determine the rate of depletion of residues in milk (cows), fat (all species) and in muscle, liver, kidney and fat (sheep), treated in accordance with approved uses of these substances; and

- new and validated methods of analysis for chlortetracycline, oxytetracycline and tetracycline residues in tissues and milk.

ADI and MRLs allocated to chlortetracycline, and tetracycline are the same as those previously allocated to oxytetracycline at the 36th meeting for the given tissues and species. Although the Committee realized that it is unlikely that tetracyclines will be used in combination, the MRLs allocated to tetracyclines were defined as applying both to residues of individual tetracyclines and to the sum of combined tetracycline residues, including chlortetracycline, oxytetracycline and tetracycline.

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