Haryana Vet. (Dec., 2019) **58(2),** 185-189

Research Article

PHARMACOKINETICS OF AMOXICILLIN AND CLOXACILLIN FOLLOWING SINGLE DOSE INTRAVENOUS AND INTRAMUSCULAR ADMINISTRATION IN SHEEP

LIPI KALESHWARI, A.H. AHMAD*, DISHA PANT, MANISH KUMAR and RICHA BAFILA
Department of Pharmacology and Toxicology, College of Veterinary and Animal Sciences,
G.B. Pant University of Agriculture & Technology, Pantnagar-263153, India

Received: 15.10.2018; Accepted: 19.08.2019

ABSTRACT

Pharmacokinetics of Inimox® (Amoxicillin and Cloxacillin formulation) was conducted in sheep following single dose (10 $mg.kg^{-1}$) intravenous (i.v.) and intramuscular (i.m.) administration. Plasma concentrations of Inimox® were determined by high performance liquid chromatography (HPLC). A one and two-compartment model adequately described plasma-concentration time profile of Amoxicillin and Cloxacillin following single dose i.m. and i.v. administration, respectively. The volume of distribution (Vdarea), clearance (ClB) and area under curve (AUC) of Amoxicillin, were 2121.4 and 3246.63 $mL.kg^{-1}$; 22.79 and 24.42 $mL.kg^{-1}.min^{-1}$; 436.40 and 414.88 $mg.mL^{-1}.min$ following i.v. and i.m. administration, respectively. The Vdarea, ClB and AUC of Cloxacillin were 792.35 and 3590.80 $mL.kg^{-1}$; 10.63 and 28.51 $mL.kg^{-1}.min^{-1}$; 943.48 and 351.45 $mg.mL^{-1}.min$ following i.v. and i.m. administration, respectively. The bioavailability (F%) of Amoxicillin and Cloxacillin were 97.38 and 37.40%, respectively. Based on pharmacokinetic data, a dosage regimen of 5.5 $mg.kg^{-1}$ IV and 2.5 $mg.kg^{-1}$ IM for Amoxicillin, whereas 14.5 $mg.kg^{-1}$ IV and 15.5 $mg.kg^{-1}$ IM for Cloxacillin at 6 h interval in sheep was calculated.[1]

Key words: Amoxicillin, Cloxacillin, Intramuscular, Intravenous, Pharmacokinetics, Sheep

Amoxicillin is broad-spectrum semi-synthetic amino penicillin, which is effective against a wide range of gram-positive and gram-negative bacteria and is widely used in veterinary practice for the treatment of respiratory, urinary and skin bacterial infections (Pijpers *et al.*, 1989).

An isoxazolyl-penicillin, cloxacillin sodium is a semi synthetic penicillinase-resistant penicillin. Cloxacillin is stable to staphylococcal penicillinase and active against both penicillinase and penicillinase sensitive Staphylococci. Few reports have been cited regarding bioavailability and pharmacokinetic studies of amoxicillin in various animal species; but there is a lack of information regarding pharmacokinetics and bioavailability of cloxacillin in sheep. The purpose of the present study was to determine single dose pharmacokinetics and dosage regimen of Amoxicillin and Cloxacillin following single dose i.v. and i.m. administration in sheep.

MATERIALS AND METHODS

The pharmacokinetic study was conducted in four non-lactating female sheep (2.0 - 2.5 years of age and weighing 35±5.0 kg). The animals were procured from Department of Livestock Production Management, College of Veterinary and Animal Sciences, Pantnagar. These animals were housed in animal house of the department and were reared as per the guidelines of Institutional Animal Ethics Committee. They were kept on pre-experimental period of one month to acclimatize them to the new environment. The animals were stall-fed on green fodder and concentrate ration along with partial grazing. The animals had free access to clean fresh drinking water.

A formulation of Amoxicillin and Cloxacillin (Inimox®, Amoxicillin- 2 grams, cloxacillin-2 grams, M/s Indian Immunologicals) was injected in sheep intravenously (i.v.) @ 10 $mg.kg^{-1}$ b.wt single dose in jugular vein. After an intervening wash out period of one month, same animals were used for pharmacokinetic studies following intramuscular (i.m.) administration of Inimox® @ 10 $mg.kg^{-1}$ b.wt, single dose. The blood samples were collected in heparinized tubes through an i.v. cannula placed in the contralateral jugular vein at 0, 2, 5, 10, 15, 30, 45, 60, 90, 120 and 180 min. The blood samples were centrifuged for 15 minutes at 3500 rpm for separation of plasma. Plasma was separated and stored at –20 ºC till analysis. The procedure for analysis in HPLC for pharmacokinetic parameters and extraction was different for both the drugs in the formulation. The plasma samples were divided in two parts and further analysis was done separately.

Drug extraction from plasma samples for Amoxicillin and Cloxacillin was carried out as per the method of Castro and Pedrazzoli, (2003) and Briguglio and Laucam (1984), respectively. A 200 µl aliquot of each plasma sample was transferred to a 1.5 ml polypropylene tube and 400 µl of cold methanol (kept on ice) was added. After vortex mixing for 1 min, the tubes were centrifuged (14000 rpm at 4 ºC for 15 min) and supernatant was filtered through 0.22 µm filter paper. 20 µl of the sample thus obtained was injected into HPLC system for analysis.

Drug estimation in the plasma was done by high

[1]Inimox® was provided by M/s Indian Immunologicals Ltd., Hyderabad

*Corresponding author: ahahmadpharma@gmail.com

performance liquid chromatography. Separation was achieved using C 18 reverse phase column, particle size 5µm (4×150 mm) as a stationary phase. The mobile phase for estimation of amoxicillin consisted of phosphate buffer (0.001 mol/L, pH-4.8) and acetonitrile mixture in the ratio of 95:5 (v/v). The flow rate was kept at 0.8 $mL.min^{-1}$. Chromatography was performed at the temperature of 25 °C with UV detection at 229 nm. The mobile phase for estimation of cloxacillin consisted of phosphate buffer (0.001 mol/L, pH -4.7), acetonitrile and methanol in the ratio of 70:19:11 (v/v/v). The flow rate was kept at 0.6 $mL.min^{-1}$. Chromatography was performed at the temperature of 25 °C with UV detection at 225 nm.

Drug standards were prepared by dissolving 5 mg of amoxicillin & cloxacillin (Sigma Aldrich Ltd.) in 5 ml of plasma separately. Further dilutions were made from this stock solution in plasma in the concentrations of 10.0, 5.0, 2.5, 1, 0.5, 0.25 and 0.1 $mg.mL^{-1}$. 20ml of each of these concentrations were injected into HPLC under the conditions mentioned above. A standard calibration curve was obtained by plotting concentrations against the peak areas obtained for Amoxicillin and Cloxacillin. Recovery % of Amoxicillin and Cloxacillin were 81 and 74 in sheep plasma, respectively.

The pharmacokinetic analysis was done by computer software 'Winnonlin'.

RESULTS AND DISCUSSION

In the present study, the pharmacokinetics of Amoxicillin and Cloxacillin was conducted following single dose (10 $mg.kg^{-1}$) i.v. and i.m. administration in sheep. The plasma concentration-time profile following single dose i.v. administration of Amoxicillin and Cloxacillin in sheep was adequately fitted to a two compartment open model. Plasma concentration-time profile upon i.m. administration of Amoxicillin and Cloxacillin in sheep was adequately described by one compartment open model. The semilogarithmic plot of plasma concentration time profile of Amoxicillin & Cloxacillin after single dose IV and IM administration (10 $mg.kg^{-1}$) and pharmacokinetic parameters are presented in Figure 1 and Table 1, respectively.

Pharmacokinetics of Amoxicillin:

Following single dose (10$mg.kg^{-1}$) i.v. administration of the formulation, plasma concentrations of Amoxicillin observed at 2 min post administration were 14.77 $\mu g.mL^{-1}$. The concentrations observed were much lower than the concentrations reported by Craigmill *et al.* (1992) following i.v. administration of Amoxicillin in goats (60.63 $\mu g.mL^{-1}$) and sheep (44$\mu g.mL^{-1}$). This difference in the concentrations of the two studies may be attributed to species variation.

The value of zero time intercept of distribution phase (A) of Amoxicillin was 5.18 $\mu g.mL^{-1}$, whereas zero time intercept of the elimination phase (B) was calculated as 3.24 $\mu g.mL^{-1}$ following i.v. administration. The values for A and B were lower than reported by Craigmill *et al.*, (1992) in sheep (53.5 and 1.69 $\mu g.mL^{-1}$). The variation may be attributed to the different methods of estimation used. The distribution (α) and elimination (β) rate constants were 0.04 and 0.01 min^{-1}. Fernandez *et al.* (2007), Elsheikh *et al.* (1999) and Craigmill *et al.* (1992) reported similar values for amoxicillin in sheep and 0.02 and 0.0035 min^{-1} in pigs following i.v. administration (Martinez-Larranaga *et al.*, 2004). The distribution half-life ($t_{½}\ \alpha$) of Amoxicillin was 19.89 min indicating a rapid absorption phase. The elimination half-life ($t_{½}\beta$) of 57.36 min. in our study is in accordance with earlier findings in goats of 61.22 min. by Elsheikh *et al.* (1999). Area under the plasma concentration-time curve (AUC) was 436.40 $\mu g.mL^{-1}$.min, which was lower than the findings of Elsheikh *et al.* (1999), 1832.73 and 1603.47 $\mu g.mL^{-1}$ min in goats and sheep, respectively but it can be corroborated to the value of 402.6 $\mu g.mL^{-1}$ min reported in pigs (Martinez-Larranaga *et al.*, 2004) following i.v. administration. Mean residence time (MRT) was 57.63 min. which was 28.8 min higher than reported by Fernandez *et al.*, 2007 following i.v. administration in sheep. MRT provides an estimate regarding the persistence time of the drug in the body. The apparent volume of distribution at steady state (Vd_{ss}) and apparent volume of distribution(Vd_{area}) in sheep were 1463.1 $mL.kg^{-1}$ and 2121.4 $mL.kg^{-1}$, respectively, indicating its extensive distribution in extravascular tissues. Vdss was greater than that reported in goats (390 $mL.kg^{-1}$) and sheep (460 $mL.kg^{-1}$) (Elsheikh *et al.*, 1999). Clearance was 22.79 $mL.kg^{-1}.min^{-1}$ which was faster than the clearance reported (1.5 $mL.kg^{-1}.min^{-1}$) by Carceles *et al.* (1995).

Following i.m. administration, the peak plasma concentration of 3.06 $\mu g.mL^{-1}$ was observed at 30 min. It was lower than (13.42 $\mu g.mL^{-1}$ at 21.6 min.) reported by Fernandez *et al.* (2007) in sheep. The value of zero time intercept of the elimination phase (B) calculated as 3.94 $\mu g.mL^{-1}$ was closer to the finding (1.69 $\mu g.mL^{-1}$) of Craigmill *et al.* (1992) in sheep. The absorption (K_a) and elimination (K_e) rate constants following i.m. administration in the present study were 0.02 and 0.008 min^{-1} which were lower than the findings (0.14 and 0.023 min^{-1}) in sheep (Fernandez *et al.*, 2007). The absorption half-life ($t_{½}\ K_a$) in sheep following i.m. administration was 37.43 min. The findings in the present study were close to the findings (10.5 and 11.7 min) reported by Craigmill *et*

al. (1992) in sheep. A lower absorption half life indicates a rapid absorption phase.

The elimination half-life ($t_{½}$ K_e) was 92.05 min, which is higher than the findings (33 min) of Fernandez *et al.* (2007) and (71.33 min) Elsheikh *et al.* (1999) in desert sheep. The value of Area Under Curve (AUC) for Amoxicillin was 414.88 μg.mL^{-1}.min. which was lower than the earlier reported values, 1685.86 and 1512.71 μg.mL^{-1}.min (Elsheikh *et al.*, 1999) in goats and sheep, respectively, following i.m. administration and 903.00 μg.mL^{-1} min in sheep after i.v. administration (Fernandez *et al.*, 2007). The mean residence time (MRT) in sheep was 139.75 min following i.m. administration. These findings are similar to the findings of Elsheikh *et al.* (1999) in goats and sheep (121.90 and 128.77min) following i.m. administration. The values of Vd_{ss}, Vd_{area} and clearance were 3398.48 mL.kg^{-1}, 3246.63 mL.kg^{-1} and 24.42 mL.kg^{-1}.min^{-1}, respectively. The findings in the present study are higher than the findings (Vd_{ss} and Vd_{area}) of Elsheikh *et al.* (1999) and Craigmill *et al.* (1992) in sheep and goats, respectively.

Bioavailability was 97.38% which is higher than reported in sheep (69%) by Fernandez *et al.*, 2007 and horses (67%) by Montesissa *et al.*, 1988. However, the values in the present study in goats and calves were less when compared to other workers [120% in cows, (Rutgers *et al.*, 1980), 100% in horses (Wilson *et al.*, 1988), 83% in pigs (Agerso and Friis, 1998)]. The variation in the bioavailabilty may be attributed to difference in extent of absorption of drug in different species.

Pharmacokinetics of Cloxacillin:

Following single dose (10 mg kg^{-1}) i.v. administration of the formulation, plasma concentrations of cloxacillin observed at two minutes post administration were 25.06 μg.mL^{-1}. There are limited reports regarding pharmacokinetics of cloxacillin in animal species following i.v. administration.

The values for A and B in sheep were 35.75 and 5.23 μg.mL^{-1} and elimination half-life ($t_{½}$ β) and elimination rate constant (β) were 51.84 min. and 0.04 min.$^{-1}$, respectively. The $t_{½}$ β reported in the present study was near to that reported by Khargariya *et al.* (2013) (0.82 ± 0.09 h) in black bengal goats but more than the $t_{½}$ β (19.5 min.) reported in calves by Daigneault *et al.* (1990), following i.v. administration. The area under the concentration-time curve (AUC) was 943.48 μg.mL^{-1}.min.

Table 1

Pharmacokinetic parameters of Amoxicillin and Cloxacillin in plasma following its single dose (10 mg/kg) i.v. and i.m. administration in sheep (n=4)

Parameters	Unit	Amoxicillin IV Mean ± S.E.	Amoxicillin IM Mean ± S.E.	Cloxacillin IV Mean ± S.E.	Cloxacillin IM Mean ± S.E.
A	μg.mL^{-1}	5.18±0.29	-	35.75±0.55	-
B/B'	μg.mL^{-1}	3.24±0.38	3.94±0.76	5.23±1.33	1.85±0.11
α/ K_a	min^{-1}	0.04±0.01	0.02±0.0004	0.07±0.02	0.11±0.02
β/K_e	min^{-1}	0.01±0.0008	0.008±0.0008	0.04±0.03	0.01±0.0003
$t_{1/2\,á}/K_a$	min	19.89±2.46	37.43±6.24	7.51±0.72	6.77±0.92
$t_{1/2\,á}/K_e$	min	57.36±2.90	92.05±5.95	51.84±7.68	87.17±2.07
C_{max}	μg.mL^{-1}	-	3.08±0.05	-	4.15±0.10
T_{max}	min	-	30.00±0.00	-	30.00±0.00
AUC	μg.mL^{-1}.min	436.40±24.39	414.88±28.54	943.48±28.74	351.45±9.50
AUMC	μg.h^2/ml	27205.85±1721.90	58248.25±5357.76	35748.73±1642.8	33692.30±1080.28
MRT	min	57.63±3.08	139.75±3.20	37.43±1.71	95.88±1.51
$V_{d(area)}$/ V_d/F	mL.kg^{-1}	2121.4±216.65	3246.63±300.56	792.35±116.63	3590.80±165.12
Vd_{ss}	mL.kg^{-1}	1463.1±163.61	3398.48±145.73	403.70±24.33	2732.95±83.40
Cl_B	mL.kg^{-1}.min^{-1}	22.79±1.31	24.42±1.56	10.63±0.33	28.51±0.77
F	%	-	97.38±5.78	-	37.40±1.09

A: Zero time intercept of distribution slope in two compartmental model; B/B': Zero time intercept of elimination phase; α/K_a Absorption/ distribution rate constant; β/K_e : Elimination rate constant; $t_{1/2á}$ / $t_{1/2}k_a$: Absorption/ distribution half-life; $t_{1/2}$β / $t_{1/2}k_e$: Elimination half-life; AUC: Total area under the time concentration curve; AUMC: Total area under the first moment curve; MRT: Mean residence time; C_{max} : Peak plasma concentration; T_{max} : Maximum time required to attain peak plasma concentration; $V_{d\,(area)}$/ V_d/F: Apparent / relative volume of distribution; Vd_{ss} : volume of distribution at steady state Cl_B Total body clearance; F: bioavailability

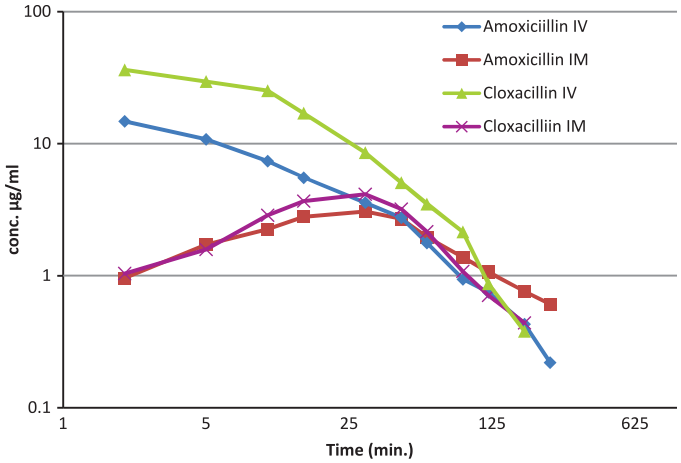

Fig. 1. Semilogarithmic plot of plasma concentration time profile of amoxicillin and cloxacillin following single dose (10 mg/kg) i.v. and i.m. administration in sheep (n=4)

which was lower than the findings of Levy *et al.* (1990) (1977.8 µg.mL^{-1}.min.) and Spino *et al.* (1984) (1126.4 µg. mL^{-1}.min.) in human beings following i.v. administration. The difference in values may be due to species variation moreover, there is paucity of data in animal species. The mean residence time (MRT) in sheep following i.v. administration was 37.43 min. Khargariya *et. al.* (2013) has reported MRT of 1.16 ± 0.13 h in adult Black bengal goats following single dose i.v. administration at 10 mg/kg b.wt.

The apparent volume of distribution at steady state (Vd_{ss}) and the apparent volume of distribution (Vd_{area}) following i.v. administration in sheep were 403.70 and 792.35 mL.kg^{-1}, respectively. The higher value of Vd (> 1 L) indicates that the drug was extravasated into the tissues. The body clearance in sheep following i.v. administration in the present study was 10.63 mL.kg^{-1}.min^{-1}.

Following i.m. administration, peak concentration in plasma at 30 min. post administration was 4.14 µg.ml^{-1} in sheep which was lower than reported by Paton, 1986 in humans (11.9 µg.ml^{-1} at 48 min. post oral administration). Absorption half-life ($t_{½}$ k_a) for cloxacillin in sheep was 6.77 min. The values for elimination half-life ($t_{½}$ k_e) and elimination rate constant (β) in sheep following i.m. administration were 87.17 and 0.01 min.$^{-1}$, respectively. These values are higher than those obtained following i.v. administration of Cloxacillin in the present study and two fold higher in a study conducted by Paton (1986) following oral administration in human beings. The difference in half-lives could be attributed to the differences in absorption of Cloxacillin in animals and humans. The area under the concentration-time curve (AUC) in sheep following i.m. administration were 351.45 µg.mL^{-1}.min. The findings in the present study were lower (1116 µg.mL^{-1} min.) than reported in humans (Paton, 1986).

The mean residence time (MRT) was 95.88 min. indicating that cloxacillin persisted for a longer duration in the body following i.m. administration. The apparent volume of distribution at steady state (Vd_{ss}) and the apparent volume of distribution (Vd_{area}) following i.m. administration in sheep were 2732.95 and 3590.80 mL.kg^{-1}, respectively. The higher values of V_d explains good extravascular distribution and here cloxacillin seems to show good distribution in the body indicating the better clinical advantage. Volume of distribution was higher following i.m. administration of cloxacillin than by i.v. administration of cloxacillin, showing higher distribution in tissues following i.m. administration. Clearance for cloxacillin following i.m. route of administration was 28.51 mL.kg^{-1}.min.$^{-1}$ in sheep. The clearance for cloxacillin, following i.m. administration was faster than that by intravenous administration in the same study indicating fast elimination of drug from the body following i.m administration.

Bioavailability following i.m. route of administration in sheep was 37.40%. This value is comparatively low which indicates that incomplete absorption.

Based on the pharmacokinetic data, dosage regimen with a priming dose of 10.3 mg.kg^{-1} followed by a maintenance dose of 10.22 mg.kg^{-1} at every 6 h interval, while an intramuscular dosage regimen with a priming dose of 6.04 mg.kg^{-1} followed by a maintenance dose of 5.9 mg.kg^{-1} at every 6 h interval was calculated and recommended for Amoxicillin in sheep. Dosage regimen with a priming dose of 14.44 mg.kg^{-1} followed by a maintenance dose of 14.32 mg.kg^{-1} at every 6 h interval, while an intramuscular dosage regimen with a priming dose of 15.5 mg.kg^{-1} followed by a maintenance dose of 14.93 mg.kg^{-1} at every 6 h interval was calculated and recommended for cloxacillin in sheep.

ACKNOWLEDGEMENTS

The authors would like to place thanks to the Dean, College of Veterinary & Animal Sciences and Directorate Experiment Station, Govind Ballabh Pant University of Agriculture and Technology, Pantnagar for providing necessary facilities in carrying these experiments.

REFERENCES

Agerso, H. and Friis, C. (1998). Bioavailability of amoxycillin in pigs. *J. Vet. Pharmacol. Therap.* **21(1)**: 41–46.

Briguglio, G.T. and Lau-Cam, C.A. (1984). Separation and identification of nine penicillins by reverse phase liquid chromatography. *J. assoc. off. Anal. Chem.* **67**: 228.

Carceles, C.M., Escudero, E. and Baggot, J.D. (1995). Comparative pharmacokinetics of amoxicillin/clavulanic acid combination after intravenous administration to sheep and goats. *J. Vet. Pharmacol. Therap.* **18**: 132–136.

Castro, S.C. and Pedrazzoli J.J. (2003). HPLC determination of amoxicillin comparative bioavailability in healthy volunteers after a single dose administration. *J. Pharm. Pharmaceut. Sci.* **6(2)**: 223-230.

Craigmill, A.L., Pass, M.A. and Wetzlich, S. (1992). Comparative pharmacokinetics of amoxicillin administered intravenously to sheep and goats. *J. Vet. Pharmacol. Therap.* **15(1)**: 72–77.

Daigneault, J., George L.W. and Baggot, J.D. (1990). Ocular and serum disposition kinetics of cloxacillin after topical administration of benzathine cloxacillin and intravenous administration of sodium cloxacillin to calves. *Am. J. Vet. Res.* **51(3)**: 381-385.

Elsheikh, H.A., Taha, A.A., Khalafalla A.E., Osman, I.A.M. and Wasfi, I.A.. (1999). Pharmacokinetics of amoxicillin trihydrate in desert sheep and Nubian goats. *Vet. Res. Com.* **23**: 507-514.

Fernandez, C., Modamio, P., Mestorino, N., Errecalde, J.O. and Marino. E.L. (2007). Pharmacokinetics of sodium and trihydrate amoxicillin in sheep after intravenous and intramuscular administration. *J. Vet. Pharmacol. Therap.* **30**: 263–266.

Khargharia, S., Chakraborty, A.K., Bhattacharyya, A. and Mandal, T.K. (2013). Disposition kinetic of cloxacillin in healthy and nephropathic goats with immunological and residual level in blood and tissues. *J. Appl. Biopharm. Pharmacokinet.* **1**: 24-30.

Levy, M., Egersegi, P., Strong, A., Tessoro, A., Spino, M., Bannatyne, R., Fear, D., Posnick, J.C. and Koren, G. (1990). Pharmacokinetic analysis of cloxacillin loss in children undergoing major surgery with massive bleeding. *Antimicrob. Ag. Chemother.* **34(6)**:1150-1153.

Martinez-Larranaga, M. R., Anadon, A., Martinez, M.A., Diaz, M.J., Frejo, M.T., Castellano, V.J., Isea, G. C.O. and De La Cruz. (2004). Pharmacokinetics of amoxicillin and the rate of depletion of its residues in pigs. *Vet. Rec.* **154**: 627-632.

Montesissa, C., Carli, S., Sonzogni, O. and Garlappi, R. (1988). Pharmacokinetics of sodium amoxicillin in horses. *Res. Vet. Sci.* **44**: 233-236.

Paton, D.M. (1986). Comparative bioavailability and half lives of cloxacillin and flucloxacillin. *Int. J. Clin. Pharm. Res.* **6(5)**: 347-349.

Pijpers, A., Van Klingeren, B., Schoevers, E.J., Verheijden, J.H.M. and Van Miert, A.S.J.P.A.M., (1989). *In vitro* activity of five tetracycline and some other antimicrobial agents against four porcine respiratory tract pathogens. *J. Vet. Pharmacol. Therap.* **12**: 267-276.

Rutgers, L.J.E., Van Miert, A.S.J.P.A.M., Nouws, J.F.M. and Van Ginneken, C.A.M. (1980). Effect of the injection site on the bioavailability of amoxycillin trihydrate in dairy cows. *J. Vet. Pharmacol. Therap.* **3**:125-132.

Spino, M., Chai, R.P., Isles, A.L., Thissen, J.J., Gold, R. and Macleod, M. (1984). Cloxacillin absorption and disposition in cystic fibrosis. *J. Pedriat.* **105**: 829-835.

Wilson, W.D., Spensley, M.S., Baggot, J.D. and Hietala, S.K (1988). Pharmacokinetics and estimated bioavailability of amoxicillin in mares after intravenous, intramuscular and oral administration. *Am. J. Vet. Res.* **49**: 1688-1694.