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PHARMACOKINETICS AND TISSUE RESIDUES OF MARBOCYL IN NORMAL AND *AEROMONAS HYDROPHILIA* INFECTED CATFISH *(CLARIAS LAZERA)*

El-Sayed M. G. A.[1], Enas A. H. Farag[2] and Shimaa F. Elzoghby[3]*

[1]Department of Pharmacology, Faculty of Veterinary Medicine, Benha University, Egypt.

[2]Department of Pharmacology, Animal Health Research Institute, Benha, Egypt.

[3]Work at Vet. Hospital of Faculty of Veterinary Medicine, Benha University, Egypt.

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***Corresponding Author**
Shimaa F. Elzoghby
Work at Vet. Hospital of Faculty of Veterinary Medicine, Benha University, Egypt.
al-ghazaly@hotmail.com

ABSTRACT

The pharmacokinetics of marbocyl were studied following intravenous and intramuscular (single and repeated) administrations. Following a single intravenous injection of 10 mg/kg body weight of marbocyl in normal catfish (*Clarias Lazera*), serum concentration-time curve was best described by two compartments open model with elimination half-life ($t0.5_{\beta}$), volume of distribution (Vd_{ss}) and total body clearance (CL_{tot}) of 7.018 hours, 783.230 ml/kg and 91.819 ml/kg/min, respectively. Following a single intramuscular administration of 10 mg marbocyl /kg body weight in normal catfish (*Clarias Lazera*), the peak serum concentration (C_{max}) was 8.721 µg/ml, achieved at a maximum time (T_{max}) of 2.134 hours. The mean systemic bioavailability was 68.34%. The serum concentrations of marbocyl following repeated intramuscular administration of 10 mg/kg body weight once daily for five consecutive days in normal and experimentally *Aeromonas hydrophilia infected catfish (Clarias Lazera)* showed a lower significant value recorded in experimentally *Aeromonas hydrophilia infected catfish (Clarias Lazera)* than in normal ones. Marbocyl showed accumulative behavior in serum of fish. Results of this study indicated that marbocyl was useful for treatment of *Aeromonas hydrophilia* infections in fish. Marbocyl was assayed in serum, liver, kidney, dorsal muscle, abdominal muscle and skin after 24, 48, 72, 96 and 120 hours from the last daily dose of 10mg marbocyl /Kg body weight for five days.

KEYWORDS: *Aeromonas hydrophilia infected catfish (Clarias Lazera).*

INTRODUCTION

The world of fish pharmacology is now changing quickly as aquaculture continues to expand, there is a need for greater knowledge for medicinal treatments both for the prevention and treatment of diseases and for economic husbandry of fish. Fish has now been considered as the model organism for conducting different experimental studies, increased incidence of disease, also due to bacterial infection leading to heavy losses, from farm level to the hobbyist tank. *Aeromonas spp.* has emerged as the most common bacterial pathogen, it causes infections in wounds, and opportunistic infections following the stresses of temperature change, handling, or poor water quality. Resistances of *Aeromonas spp*.to commonly used antibacterials is an emerging problem in the fish industry.[1,2] Stated that it may necessary at this time to begin screening antimicrobial compound for efficacy against *Aeromonas spp*.

Fluoroquinolones antibacterial, are increasingly being employed in veterinary medicine for treatment of mild to severe bacterial infection.[3] Marbofloxacin is an antimicrobial fluoroquinolone, carbxylic acid derivative recently introduced for use in veterinary medicine medicine[4]**,** high plasma concentration initially is important as the drug act by concentration dependent mechanism. It has extended spectrum of activity which include mainly Gram-negative pathogen and some of Gram-positive and mycoplasma species.[5] Marbofloxacin is approved for treatment of respiratory, urinary, dermatological disease and gastrointestinal infectious disease. Similar to other fluoroquinolone, marbofloxacin is a bactericidal antibacterial. Marbofloxacine is as inhibitor of DNA gyrase which is an essential cell enzyme necessary for the supercoiling of DNA, which allows bacterial DNA to fit within the bacterial cell and leads to rapid bacterial cell death.[6]

MATERIAL AND METHODS

Drug: Marbofloxacin was used in this study under the trade name (Marbocyl 10%® solution**)**. Was purchased from Vetoquinol Ltd. (Lure, France). Each bottle contains 50 ml solution which has 100.0 mg marbofloxacin. Which was exclusive distribution in Egypt by INTERCOVA Animal health products.

Experimental FISH: Catfish *(Clarias Lazera)* were used in this investigation**.** The weights of fish were 300 ± 0.5.Catfish (*ClariesLazera*) were obtained from local farms in Benha city, El Qualubia government, Egypt..It were feed on pelleted feed (25% proteins, 8% fat) from El-abassa farm, El- Sharkia government twice daily The fish were apparently healthy, and

were transferred and maintained for acclimatization in glass aquaria (80 x 40 x30cm) supplied with declorinated tap water.

Experimental design

The fish were divided into 3 groups

Group (1): Group (1): It included 18 catfish (*Calrias Lazera*) which were divided into 6 glass aquaria. Each fish was injected intravenously in the caudal vein with 10 mg marbocyl /kg b.wt. These fish were left for 15 days after the intravenous injection to ensure complete excretion of marbocyl from their bodies. Then the fish were injected intramuscularly 10 mg of marbocyl /kg b.wt in dorsal muscle. to calculate bioavaibility of marbocyl in normal catfish (*ClariesLazera*).

Group (2): It included 30 catfish (*Calrias lazera*) divided into 10 glass aquaria. Each fish was injected intramuscularly into dorsal muscle with 10 mg marbocyl /kg b.wt, once daily. Serum samples were taken, and then intramuscular dose was administered every 24 hours for five consecutive days. Tissue samples were taken for assaying of drug residues after the last sampling.

Group (3): It included 30 catfish *(Calrias lazera)* divided into 10 glass aquaria. Each fish was injected intrapertonially with *Aeromonas Hydrophilia*. After the appearance of sings of infection as hemorrhage all over the skin especially under the base of the fins and erosion in fins, each fish was injected intramuscularly with 10 mg marbocyl /kg b.wt every 24 hours for five consecutive days. After that serum and tissue samples were taken for assaying of residues till disappearance of the drug from tissues.

Collection of samples

Blood samples

Blood samples were collected from caudal vein following intravenous or intramuscular administration in normal and experimentally infected cat fish. Blood samples are collected after 0.083, 0.167, 0.25, 0.5, 1, 2, 8, 12 and 24 hours of administration in single study, and after0.083, 0.167, 0.25, 0.5, 1, 2, 8, 12 and 24 hours in the first day, second, third, fourth and fifth dose in the same study in repeated intramuscular administration in normal and experimentally *Aeromonas hydrophilia* infected catfish*(Clarias Lazera)*. Serum samples were separated by centrifugation and stored at -20[7] until assay of marbocyl.

Tissue samples

Three fish were slaughtered serum, Liver, kidney, muscle with skin (dorsal and abdominal muscles) were taken from fish after repeated intramuscular injection in normal and experimentally *Aeromonas hydrophilia* infected catfish *(Clarias Lazera)* for assaying of residues of marbocyl at 24,48,72,96,120 hours after the last sampling**.**

Analytical procedures

Marbfloxacin was determined in serum, physiological saline by using microbiological method using ***Escherichia coli*** **ATCC** (American type culture collection 25922 as tested microorganism,which was obtained from Microbiological Department, Animal Health Institute Benha Egypt.[8] From the stock solution of marbofloxacin (1000µg), standard concentrations were prepared by further dilution in physiological saline, antibiotic free fish serum to obtain concentration of 0.049,0.098,0.195,0. 391,0.781,1.56,3.13,6.25,12.5, and 25 µg/ml for preparation of standard curve of marbofloxacin.

The pharmacokinetic parameters were calculated by Winnonlin program, version 1.1 and other parameters according to.[9] All statistical analysis was carried out according to.[10]

Assay of tissue samples

According to[11], three plates were used for each sample. Four pores were made, one of them were filled with reference concentration while the other three pores were filled with the supernatant of tissue samples in triplicate manner. The plates were incubated at 37°C for 18 hours then the diameters of inhibitory zones were measured. The diameter of inhibitory zones of samples determined till disappear of inhibition zones that indicate complete excretion of drug from the body of fishes.

RESULTS

Following a single intravenous injection of 10 mg /kg b.wt. in normal fish, marbocyl could be detected therapeutically for 24 hours post intravenous injection. The serum concentration – time curve of marbocyl following intravenous injection showed that the drug obeyed a two compartments open model. The disposition kinetics of marbocyl following a single intravenous and intramuscular administration were recorded in tables (1) and showed in figure (1). Intramuscular administration of 10mg/kg b.wt every 24 hours for five consecutive days in normal and *Aeromonas hydrophilia* infected catfish*(Clarias Lazera)*. revealed a lower significant serum marbocyl concentration at all time sampling in *Aeromonas hydrophilia*

infected catfish *(Clarias Lazera)* than in normal fish. The pharmacokinetic parameters of marbocyl after repeated intramuscular administration in normal fish were compared to those in *Aeromonas hydrophilia* infected catfish *(Clarias Lazera)* in table (2).

Tissue residues for marbocyl in, liver, kidney, dorsal muscle and abdominal muscle with skin after repeated intramuscular administration in normal fish were compared to those in *Aeromonas hydrophilia infected catfish (Clarias Lazera)* were recorded in table (3).

Table. (1): Pharmacokinetic parameters of marbocyl following a single intravenous and intramuscular injection of 10 mg/kg b.wt. in normal fish (n=6).

Parameter	Unit	Intravenous (X ± S.E.)	Intramuscular (X ± S.E.)
C^0	µg/ml	29.446 ± 0.48	-
A	µg/ml	21.88 ± 0.395	-
A	h^{-1}	1.437 ± 0.153	-
$t_{0.5(\alpha)}$	H	0.510 ± 0.055	
K_{ab}	h^{-1}	-	0.992±0.018
$t_{0.5(ab)}$	H	-	0.699±0.012
K_{12}	h^{-1}	0.820 ± 0.097	--
K_{21}	h^{-1}	0.428 ± 0.047	--
T_{max}	H		2.134±0.019
C_{max}	µg/ml		8.721±0.038
V_{dss}	ml/kg	783.23±59.34	--
K_{el}	h^{-1}	--	0.153±0.007
B	µg/ml	7.55 ± 0.491	
B	h^{-1}	0.099 ± 0.002	
$t_{0.5(\beta)}$	H	7.018 ± 0.201	
AUC	hr/µg/mL	112.25 ± 8.31	76.71 ± 0.819
AUMC	hr/h/µg/ml	961.58±76.72	525.1 ± 15.61
MRT	H	0.820±0.097	6.84 ± 0.147
CL_{tot}	L/hr/kg	91.81±7.45	-

[1]**A & B,** Zero time serum drug concentration intercepts of biphasic intravenous disposition curve. The coefficient B is based on the terminal exponential phase (µg/ml); **α & β,** Hybrid rate constant of biphasic intravenous disposition curve values of α and β are related to the slopes of distribution and elimination phase respectively, of biexponential drug disposition curve (h-1); **AUC,** Total area under the serum drug concentration versus time curve from t = 0 to t = α after administration of a single dose; **C°,** Drug concentration in the serum at zero time immediately after a single intravenous injection (µg/ml); **C max,** Maximum serum concentration of drug in blood after extra vascular administration (µg/ml); **Cl tot,** The total clearance of a drug, which represents the sum of all clearance processes in the body (ml/kg /min); **K ab,** Apparent first order absorption rate constant (h-1); **K el,** First - order

elimination rate constant for disappearance of drug from central compartment (h-1); **K12,** First - order transfer rate constant for drug distribution from central to peripheral compartment (h^{-1}); **K21,** First order transfer rate constant for drug distribution from peripheral to central compartment (h^{-1}); **K13,** First - order elimination rate constant for disappearance of drug from central compartment (h^{-1}); **t 0.5(ab),** The absorption half- life (h); **t 0.5(α),** Distribution half - life (h); **t 0.5(β),** Elimination half - life (h); **t max,** The time at which the maximum concentration of drug was reached after extravascular administration (h); **V1 c,** The apparent volume of central compartment (ml/kg); **Vd(B),** The apparent volume of distribution Which calculated by extrapolation method (ml/kg); **Vd(area),** The apparent volume of distribution which was calculated by the area method (ml/kg); **Vdss,** The apparent volume of distribution which was calculated by Steady - state method (ml/kg).

Table (2):Pharmacokinetic parameters of marbocyl in normal (N) and experimentally *Aeromonas hydrophilia infected catfish(Clarias Lazera)*. (I) during repeated intramuscular injections of marbocyl 10 mg /kg b.wt. once daily for five consecutive days (n=3).

Paramater	Unit	1st dose		2nd dose		3th dose		4th dose		5th dose	
		N – (X ± S.E.)	I – (X ± S.E.)	N – (X ± S.E.)	I – (X ± S.E.)	N – (X ± S.E.)	I – (X ± S.E.)	N – (X ± S.E.)	I – (X ± S.E.)	N – (X ± S.E.)	I – (X ± S.E.)
K_{ab}	h^{-1}	1.040±0.018	0.778±0.018**	1.147±0.040	1.094±0.027	1.322±0.022	5.682±4.366	1.369±0.028	1.278±0.014*	1.497±0.027	1.465±0.027
$t_{0.5(ab)}$	H	0.666±0.011	0.890±0.021***	0.605±0.020	0.634±0.016	0.524±0.008	0.367±0.160	0.506±0.010	0.542±0.006*	0.463±0.008	0.473±0.008
T_{max}	h	2.109±0.020	2.345±0.009***	2.135±0.038	2.194±0.016	1.988±0.017	1.397±0.604	1.964±0.018	2.038±0.007*	1.924±0.012	1.947±0.016***
C_{max}	µg/ml	8.781±0.039	7.629±0.067***	9.701±0.115	8.547±0.063***	10.868±0.028	12.618±2.839	12.409±0.069	11.414±0.026***	14.04±0.063	12.483±0.032***
K_{el}	h^{-1}	0.136±0.001	0.156±0.002***	0.118 ±0.00	0.115 ±0.001	0.111±0.002	0.117±0.001*	0.103±0.002	0.115±0.001**	0.102±0.003	0.109±0.002
$t_{0.5(\beta)}$	h	5.093±0.046	4.422±0.066***	5.847±0.028	5.978±0.052	6.231 ±0.015	5.919±0.084*	6.680±0.162	5.985±0.071**	6.803±0.239	6.348±0.174**
AUC	hr/µg/mL	80.709±0.568	64.303±0.557***	102.639±0.97	91.065±0.347***	119.644 ±0.337	108.66±1.398**	141.42±1.61	125.89±0.811***	172.753±2.21	150.16±1.86**
AUMC	hr/hr/µg/ml	592.651±10.074	423.831±8.638***	876.83±8.053	791.19 ±3.340***	1070.62 ±5.670	928.66±6.934***	1351.17±47.34	1105.57±13.820**	1704.37±71.2	1415.975± 50.67*
MRT	H	7.342±0.080	6.589±0.077**	8.542±0.004	8.688±0.019**	8.948 ±0.0232	8.549±0.146*	9.549±0.223	8.781±0.053*	9.855±0.284	9.424±0.217

*P<0.05 ** P<0.01 *** P<0.001

Table. (3): Serum (µg/ml) and tissue(µg/g) concentrations of marbocyl (µg/ml) in normal (N) and experimentally *Aeromonas hydrophilia* infected catfish*(Clarias Lazera)* (I) during repeated intramuscular injections of 10 mg /kg b.wt. once daily for five consecutive days (n=3).

Time / Tissue	After first day		After second day		After third day		After forth day		After fifth day	
	N ($\bar{X}$± S.E.)	I ($\bar{X}$± S.E.)	N ($\bar{X}$± S.E.)	I ($\bar{X}$± S.E.)	N ($\bar{X}$± S.E.)	I ($\bar{X}$± S.E.)	N ($\bar{X}$± S.E.)	I ($\bar{X}$± S.E.)	N ($\bar{X}$± S.E.)	I ($\bar{X}$± S.E.)
Serum	1.3±0.214	0.930± 0.142**	-	-	-	-	-	-	-	-
Liver	12.7±0.014	5.94± 0.467***	6.3±0.231	0.43± 0.329***	0.83±0.114	-	-	-	-	-
Kidney	12.3±0.324	5.233± 0.653***	5.7±0.328	0.37± 0.268***	0.68±0.073	-	-	-	-	-
Dorsal muscle	8.8±0.236	2.6± 0.426***	4.3±0.122	0.13± 0.423***	0.25±0.286	-	-	-	-	-
abdominel muscle	6.4±0..0125	1.63± 0.591***	1.8±0.278	0.45± 0.356***	-	-	-	-	-	-

(*): Represent the significance in comparison with data of normal group.

** P<0.01 *** P<0.001

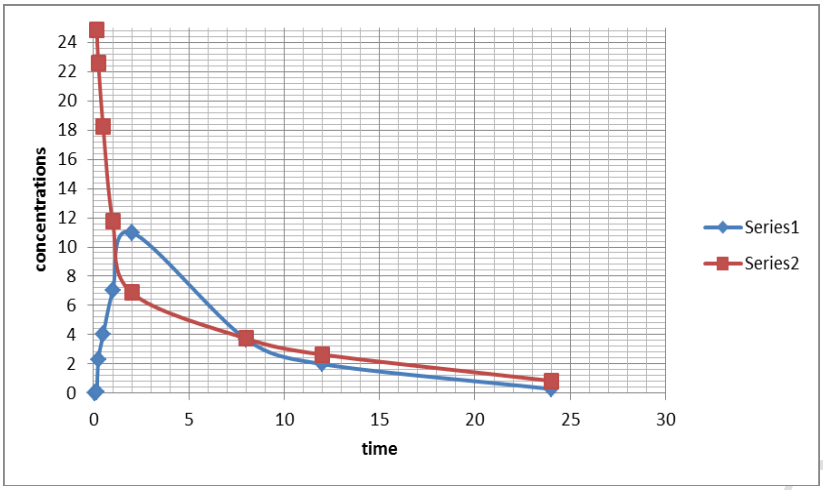

Figure. (1): Arithmetic plot of serum of marbocyl concentrations in normal fish following a single intramuscular injection of 10 mg/kg bwt.(●—●) in fish previously given the same dose by a single intravenous injection (■….■) (n=6).

DISCUSSION

In the present investigation, the intravenous injection of 10 mg marbocyl / kg.b.wt. In normal catfish *(Clarias Lazera)*, showed that the drug disposition best fitted a two-compartments-open model, a compartment of plasma and rapid equilibrating tissues, and a deeper slower compartment. The obtained result was consistent with those reported for for enrofloxacin in fingerling rainbow trout[12] for oxolinic acid in Atlantic Salmon[13] for enrofloxacin in brown trout[14] and for levofloxacin in sheep.[15]

Marbofloxacin was distributed after intravenous injection with a short distribution half life [$t_{0.5\ (\alpha)}$] equal to 0.510 hour. The shorter distribution half life of Marbofloxacin in catfish *(Clarias Lazera)* suggested the rapid distribution of the drug in body tissue and fluid and that nearly similar to that recorded for oxolinic acid in Atlantic Salmon (1 hour)[13] for enrofloxacin in brown trout (0.88 hour)[14] and for levofloxacin in sheep (0.33 hour)[15] and it is smaller than that is reported in broiler chicken by[16] ($t_{0.5\ (\alpha)}$ is equal to 30.49 minutes) and by[17] $t_{0.5\ (\alpha)}$ is equal to 0.92 the short period of distribution half-life might be attributed to a larg proportion of the administrated drug which is inside the cell also to the higher metabolic rate. The rapid distribution of Marbofloxacin in catfish *(Clarias Lazera)* is supported by high rate constant of transfer of the drug from central to peripheral compartment [$K_{12} = 0.820\ h^{-1}$] as compared to the rate constant of transfer of the drug from peripheral to central compartment [$K_{21} = 0.428\ h^{-1}$].

The volume of distribution of Marbofloxacin at steady-state ($V_{dss)}$ were 783.23 ml/kg. These values were higher than those recorded by[18] who found that V_{dss} for enrofloxacin in European cuttlefish was 385 ml/kg,.On the other hand, these result were differ from those recorded for flumequine in haibut (2.99 l/kg)and turbo (3.75 l/kg) by[19], for oxolinic acid in Atlantic Salmon (5.71 l/kg) by[13] and for enrofloxacin in brown trout (3.40 l/kg) by[16] and for levofloxacin in sheep was 0.86 l/kg by.[15]

High values of the volume of distribution suggesting extensive pentration power Drug with volume of distribution greater than one liter /kilogram indicate wide distribution or extensive tissue binding or both.[20]

Marbofloxacin was eliminated in the current study following a single intravenous injection with elimination half –life[$t_{0.05(\beta)}$] equal to 7.018 hours. This observation was nearly agreed with the data recorded for elimination half –life for ciprofloxacin in African catfish (14 hours) by[21], for oxolinic acid in Atlantic salmon (15 hours) by.[13] On contrast, this value was shorter than that recorded for enrofloxacin in fingerling rainbow trout (24.4 hours) by[12], for enrofloxacin in Atlantic Salmon (34.2 hours) by[22] and for flumequine in eel (314 hours) by.[19] On the other hand, it was longer than that recorded for enrofloxacin in European cuttlefish (1.81 hours) by[18] and for levofloxacin in sheep (3.29 hour) by.[15] These variations in pharmacokinetic parameters were relatively common and frequently related to method used, healthy status of animal and specific interspecies variation.[23]

The rate of total body clearance of Marbofloxacin following intravenous injection was 91.81 L/hr/kg. This value was differ from that recorded for oxolinic acid in Atlantic Salmon (0.40 l/kg/h) by[13], for that recorded for oxolinic acid in rainbow trout (1.2 l/kg/h) by[24] and for enrofloxacin in European cuttlefish (4.71 ml/kg/min) by.[18] The variation in total body clearance might be attributed to specific interspecies variation.[25]

The difference between values calculated for pharmacokinetic parameters may be attributed to animal species, the drug formulation employed, the age, the size or sex of the animal or even inter individual variation and also due to method of analysis of the drug.

Following IM administration, marboflxacin was rapidly absorbed in catfish (absorption half-life $\mathbf{t_{0.5ab}}$**:** 0.699 h). This value was higher than danofloxacin and marbofloxacin (0.31, 0.27 h) in Muscovy ducks.[26,27] and this value lower than levofloxacin in turkeys t0.5ab: 1.02 h)[28]

Rapid oral absorption is also reflected by low MAT (mean absorption time) value (1.48 h), similar to danofloxacin (1.35 h) in Muscovy ducks[27] but lower than sarafloxacin (4.40 h) in chickens.[29] This value was lower than that reported by[30] after oral administration of 5 mg/kg of flumequine in channel catfish [$t_{0.5(ab)}$= 4.94 hours] and also lower than that reported by[14] after oral administration of 10g/kg of enrofloxacin in brown trout [$t_{0.5(ab)}$= 3.03 hours].

Elimination half-life (t0.5el: 4.5 h) in catfish is agreed with Elimination half-life (t0.5el: 4.60 h) in turkeys.[29] And it was lower than difloxacin (5.64 h) in chickens.[31] and in lactating cows 5.9 h,[32] but higher than both danofloxacin and marbofloxacin (2.91, 2.82 h) in Muscovy ducks.[26,27]

maximum serum concentrations (C_{max}) were 8.721 µg/mL achieved at (T_{max}) 2.134 hours, the obtained results were higher than those reported in turkeys The Cmax was 5.59 µg/ml, achieved at (Tmax) 2 h[28], marbofloxacin (3.11 µg/ml at 1.02 h) in Muscovy ducks[26], and also These results were higher with those recorded for enrofloxacin in Atlanitic Salmon (C_{max} =1.54 µg/ml at t_{max} = 6 hours) by[22] and those for enrofloxacin in red pacu (C_{max} =1.64 µg/ml at t_{max} = 4 hours) by[33] by oral rout. In pigs1.17 µg/mL[34] These values were lower in animals reported in desert sheep and Nubian goats, 1.29 and 1.33 µg/mL, respectively[35], and 2.8 µg/mL reported in goats.[36]

The bioavailability was 68.34% in catfish this value referred to a better absorption of marbofloxacin from its site of intramuscular administration, similar to that reported in danofloxacin in goats 65.70 %.[37] And it was nearly similar to to those recorded for norfloxacin in lambs 73.51 %, and pefloxacin in lactating goats 70.63 %.[38,39]

On other hand this value was higher than the bioavailabilities recorded for norfloxacin in donkeys 31.5 % and in rabbits 45 %.[40,41] On contrast this value was lower than horse 87.9%.[42]

Repeated intramuscular administration of marbofloxacin in a dose level of 10 mg/kg b.wt every 24 hours for five doses in normal and *Aeromonas hydrophila* infected catfish *(Clarias Lazera)* induced blood levels in *Aeromonas hydrophilia* infected catfish (*(Clarias Lazera)* significally lower than those in normal catfish (*(Clarias Lazera)*). These lower blood concentration in infected catfish (*(Clarias Lazera)* might be attributed to the higher penetrating power of marbofloxacin to the diseased tissues.[25]

This phenomenon was similar to data recorded by[43] who found that enrofloxacin concentrations in plasma of infected birds were lower than those of healthy ones.

The maximum [C_{max}] and minimum [C_{min}] serum concentration of marbocyl during multiple regimen in normal (14.04,8.78 µg/ml) respectively and experimentally *Aeromonas hydrophilia* infected catfish *(Clarias Lazera)* (12.48,7.62) respectively, this indicated that dose regimen of 10 mg/kg b.wt every 24 hours for five days would provide effective and safe concentration of antibacterial effect. Serum concentration of marbofloxacin in normal and *Aeromonas hydrophilia* infected catfish *(Clarias Lazera*) could be detected in therapeutic level for 24 hours following repeated intramuscular injection and exceeded MIC_{90} of marbofloxacin against *Aeromonas hydrophilia* (0.0312 µg/mL)**.** Being the time of exposure to constant concentrations of marbofloxacin an important factor which determined the final efficacy.

Following repeated intramuscular injection of 10 mg marbofloxacin /kg b.wt every 24 hours for five doses in normal and experimentally *Aeromonas hydrophilia* infected catfish *(Clarias Lazera*),the drug couldn't be detected by microbiological assay in blood, liver, kidney, dorsal and abdominal muscle with skin after 96 hours post last dose. The high clearance of marbofloxacin indicated the reduced possibility of finding residues of antimicrobial in catfish (*Clarias Lazera*) a few days after treatment and necessity of shorter withdrawal time for this antimicrobial (4 days). Results showed that the concentration in liver,kidney and muscle with skin (6.3,5.7 and 1.8 µg/g respectively) after two days from last dose of injection.this value is nearly agreed with[44] recorded the highest concentration in liver (6.2 µg/g) and the lowest concentration was in muscle with skin (1.8 µg/g) at 24 hour. and higher than which recorded in The residue testing revealed concentrations of marbofloxacin in kidney (1.74±0.04µg/g) and liver (1.52±0.04µg/g) 24 hours after the last dose. These levels were higher than that previously reported in broiler chickens kidney 0.985µg/kg and liver; 0.735 µg/kg[44] who administered a dose of 2mg/kg which is less than half the used dose in the present study. Therefore, a 4 days withdrawal time of marbofloxacin is suggested. Similar conclusion has been previously suggested.[45] This result was shorter than that recorded for difloxacin in crucian carp (15 days) after last dosing by[46] and for norfloxacin in Japanese sea perch and black sea bream following multi oral administration (14 -32 days post dosing respectively by.[47]

CONCLUSION

● The bioavailability of Marbocyl is good, so it is recommended to be used against *Aeromonas hydrophilia* infected catfish *(Clarias lazera).* The repeated intramuscular administrations of 10 mg/kg b.wt. of marbocyl once daily for five consecutive days had a cumulative effect and would provide an effective concentration against gram+ve and gram-ve infection in catfish (*Clarias Lazera).*

● Treated fish must not be slaughtered before 4 days from last dose of repeated administration of marbocyl to withdraw the drug residues from all tissues of treated fish.

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