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PHARMACOLOGICAL STUDIES ON SOME ANTIBACTERIAL DRUGS IN FISH

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ABSTRACT

The antibacterial activities of ciprofloxacin(CIP), florfenicol(FC) and oxytetracycline (OCT) were investigated either in-vitro and in-vivo against Aeromonas hydrophila in Clarias Lazera . The obtained results indicated that these drugs were effective in vitro against this microorganism with MIC 1.1 ,0.08 and 2.8 µg/ml for florfenicol , ciprofloxacin and oxytetracycline respectively . The in-vivo antibacterial activity of these drugs in their therapeutic doses showed a decline in mortality rates(%) and pathogenic clinical signs compared to that of infected non medicated group. The estimated hematological parameters showed that both ciprofloxacin and oxytetracycline caused significant decrease in all hematological parameters while florfenicol cause a slight decrease , and all these changes were reversible and returned to the normal range within 7 days of treatment . The effects of these drugs on the estimated biochemical parameters showed that, florfenicol induced non significant changes in all biochemical parameters compared to the control group, while significant changes were recorded between florfenicol treated group and that treated with either ciprofloxacin and oxytetracycline. There were also a significant changes between the infected non treated group compared to all other groups .

INTRODUCTION

More and more attention is being paid to fish farming due to the over increasing need for protein to meet the world's over population problem , especially in the developing countries where problem is acute **(Shalabi, 1992**) .Fish is often the cheapest source of

animal protein and is, therefore, important in the diets of the lowest income groups **(Allison, 2001).** Outbreaks of disease have become a critical factor which has hampered the development of aquaculture in many countries. Bacterial diseases are a major problem in aquaculture and account for significant losses of fish among these various disease agents. The Gram-negative bacteria. ***Aeromonas*** species are commonly found in a wide range of aquatic environments including fish ponds and it is the causative agent of motile aeromonas infection (MAI), which occur in a wide variety of freshwater fish species ***Aeromonas hydrophila*** and other motile aeromonads are among the most common bacteria in freshwater habitats throughout the world, and these bacteria frequently cause disease among fish (**Mirand and Zemelman , 2002).**

Burgos et al, (1990) stated that chloramphenicol, ciprofloxacin and tetracyclines were extremely active against all Aeromonas species and thus can be used effectively for therapy of infections when Aeromonas is implicated.

Recently, attention has been focused on residues in food fish, the risk of developing resistant pathogens, and environmental bioaccumulation .Morover, other deleterious effects, such as immunosuppression, nephrotoxicity, and growth retardation have been associated with the use of drugs in aquatic organisms **(Lunden et al., 1998).**

The work reported here was designated to determine the in vitro and in vivo antibacterial activities of ciprofloxacin, florfenicol and oxytetracycline . Moreover , the effects of these drugs on some hematological and biochemical parameters in clinically healthy and experimentally infected fish were also studied .

MATERIALS AND METHODS

Materials

Antibacterial agents

(1) Florfenicol : (Nor-Fenicol) ® a sterile injectable 30% solution (50 ml vial) obtained from Kim-Vet company, Cairo, Egypt.

(2) Ciprofloxacin : Ciprofloxacin (Ciprotril®) 10% solution of Veterinary and Agricultural products, Co. Cairo, Egypt .

(3) Oxytetracycline: Oxytetracycline (Oxytetracycline®) 25% powder ,The Nile Co.for pharmaceuticals.

Tested organisms-

1- Aeromonas hydrophila : They were obtained from Animal Health Research Institute .Dokky ,Cairo

2- Salmonella entertidies & E. coli : were used as a tested organism for estimation of MIC of the tested drugs ; they were obtained from Animal Health Research Institute.Dokky,Cairo.

Fish

A total number of 130 apparently healthy cat fish (*Clarias lazera*) were employed for this study.They were obtained from a private fish farm in Dakahlia governorate with average body weight of 200 gm. Fish were acclimatized for 2 weeks before beginning of experiment in order to make sure that all fish have been adapted to the same environmental condition. Fish were provided with a commercial balanced diet.

Media

Nutrient broth; Nutrient agar for bacterial growth; Muller –Henton agar for sensitivity test .

Chemicals used for hematological studies -

Heparin,Methanol, Giemsa stain, Natt & Herrik's solution and Hydrochloric acid (1\10 N)

Kits for serum biochemical analysis : kits used for determination of serum transaminases (AST&ALT), serum urea and creatinine levels and serum protein fractions .

METHODS

I- Efficacy of florfenicol, ciprofloxacin and oxytetracycline against Aeromonas hydrophila infection in Clarias lazera:

a- In-vitro : 1- Sensitivity test (disc diffusion method)

The in-vitro antibacterial effect of florfenicol , ciprofloxacin and oxytetracycline against ***Aeromonas hydrophila*** was carried out using disc diffusion method **(Bauer et al.,**

1966) . The technique was standardized by the National Committee for Clinical Laboratory Standards **(NCCLS, 1994)**

2- Determination of Minimum Inhibitory Concentrations (MICs) of florfenicol, ciprofloxacin and oxytetracycline against Aeromonas hydrophila , Salmonella entertidies and E.coli.

Determination of Minimum Inhibitory Concentrations (MICs) of florfenicol, ciprofloxacin and oxytetracycline against Aeromonas hydrophila was carried out using micro dilution technique (dilution broth method) (Elmer et al., 1988).

b- In Vivo :

To determine the in-vivo efficacy of florfenicol, ciprofloxacin and oxytetracycline against Aeromonas hydrophila infection , fifty apparently healthy Claris lazera fish (200 gm) were used . Fish were divided into five equal groups (each of 10 fish) as the following:-

1st group : fish were served as non infected non treated (negative control).

2nd group : fish were inoculated intraperitoneally (I/P) with 0.2 ml of 24 hrs broth cultures of Aeromonas hydrophila (2.5×108CFU / ml) and kept without medication (positive control).

3rd group : fish were experimentally infected similarly , and 18 hours post bacterial inoculation, (Fukui et al., 1987), fish were injected intramuscularly with florfenicol (30 mg/kg b.wt) daily for 3 successive days .

4th group : fish were experimentally infected similarly , and 18 hours post bacterial inoculation (Fukui et al., 1987), fish were injected intraperitoneally with ciprofloxacin (10 mg/kg b.wt) daily for 3 successive days .

5th group : fish were experimentally infected similarly , and 18 hours post bacterial inoculation, (Fukui et al., 1987) fish were orally administered with 50 mg/L oxytetracycline per day for 3 successive days .

Cumulative mortalities were observed at the 7th day post medication . Also any adverse signs were recorded during that period .

11-Haematological studies :

This study was conducted on 8 groups of fish (10 fish each) as follows :

1st group : fish served as non infected non treated (negative control).

2nd group: normal fish were injected with florfenicol (30 mg/kg b.wt) interamuscularly for 3 successive days

3rd group : fish were injected I/P with ciprofloxacin (10 mg/kg b.wt) daily for 3 successive days.

4th group : fish were orally dosed with 50 mg/L oxytetracycline per day for 5 successive days.

5th group : fish were inoculated intraperitoneally (I/P) with 0.2 ml of 24 hrs broth culture of Aeromonas hydrophila (total bacterial count 2.5 x 108/ml) and kept without medication (positive control).

6th group : were infected similarly , and 18 hours post bacterial inoculation, fish were injected intramuscularly with florfenicol (30 mg/kg b.wt) daily for 3 successive days as described by (Fukui et al., 1987).

7th group :were infected similarly , and 18 hours post bacterial inoculation, fish were injected I/P with ciprofloxacin (10 mg/kg b.wt) daily for 3 successive days as described by (Fukui et al., 1987).

8th group : fish were infected similarly , and 18 hours post bacterial inoculation, fish were orally dosed with 50 mg/L oxytetracycline per day for 5 successive days as described by (Fukui et al. 1987).

Blood samples were collected with anticoagulant from caudal veins of five fish from each group at 1st , 7th, and 14th days post administration of the drug .

Total erythrocytic and leucocytic counts were performed according to **Natt and Herrick (1952)** , Heamoglobin was determined according to **Wintrobe (1967) ,** while packed cell volume was determined according to **Cohen ., (1967) .**

111 - Biochemical analysis :

This study was conducted on 8 groups of fish each group contains ten fish in similar manner as in haematological studies .Serum samples were collected from each group at 1st , 7th and 14th days post drug dosing and were used for determination of serum AST & ALT (**Reitman and Frankel ,1957)** urea level (**Patton and Crouch ,1977)**,creatinine **(Henry , 1974)**,total proteins (**Doumas ,1975)** ,Albumin (**Doumas et al. ,1981)** and globulin by

subtraction of the obtained albumin level from the level of total proteins as described by **Doumas and Biggs(1972).**

– Statistical analysis :

Data were analyzed using computerized SPSS. Results of the biochemical estimations were reported as mean ± S.E. The total variation was analyzed using one – way analysis of variance (ANOVA)> Duncan test was used for determining significance probability levels of less than 0.05 were considered significant (Snedecor and Cochran (1987).

RESULTS AND DISCUSSION

***In-vitro* activity**

1- ***In-vitro*** sensitivity test of ***A.hydophila*** strain against florfenicol (FF), ciprofloxacin (CF) and oxytetracycline (OT) using agar disc diffusion method showed that ***A.hydophila*** was highly susceptible to the tested drugs with clear zone of inhibition (Table 1). Sensitivity studies of ***A . hydrophila*** isolates against florfenicol indicated its high sensitivity to florfenicol . This result is similar to that obtained by **Aoki and Egusa, (1971)** and **Fukui et al. (1987)** who reported that ***A . hydrophila*** was sensitive to florfenicol in vitro .

2- The minimum inhibitory concentration (MIC) of florfenicol for ***A. hydrophila*** organism that cause septicemia in ***Clarias lazera*** was 1.1 µg/ml . This value is nearly similar to that of **Robert and Jean (1981)**, who recorded that MIC of chloramphenicol against ***A.hydrophila*** was in range of 0.5 – 4 µg/ml.. This low value of MIC indicates that florfenicol is highly active in vitro against ***Aeromonas hydrophila*** pathogen as shown in Table (2) .

The decreased in vitro activity of oxytetracycline against ***A.hydrophila*** could be attributed to the development of drug resistance , since this drug is already in veterinary use for many years ago compared with the recently used florfenicol and ciprofloxacin

In vivo activity

1- mortality (%):

Our results revealed that experimentally infected ***Clarias lazera*** with ***A. hydrophila*** organism responded to the treatment with florfenicol , as the recorded clinical signs were declined after 5 days post treatment .

The mortality started at the third day and reached (80%) at the 7th day post infection in the infected non treated group while the medicated groups with therapeutic dose of florfenicol , ciprofloxacin and oxtetracycline showed reduction in mortality rates (40 & 30 and 50%), respectively, compared to non medicated group . (Table 3)

2- Pathogensity :-

- The effect of florfenicol , ciprofloxacin and oxytetracycline on pathogencity of experimentally infected *Clarias Lazera* inoculated with *A. hydrophila* .

Two days after inoculation of ***A. hydrophila*** the infected fish showed some clinical signs manifested by sluggish movement , swimming near the water surface , progressive erosion allover the body , erythema at the base of the fins and some fish showed ulcer formation . (Fig 1 , 2 and 3).

Treatment with florfenicol (30 mg/kg b.w.) intramuscular daily for 3 successive days in Group (3) showed decline in clinical sings (Table) at 5th day post medication. This result is nearly similar to the finding of **Nada (2006)** who stated that florfenicol is effective for the control of mortality in catfish due to enteric septicemia.

Our results revealed that experimentally infected ***Clarias lazera*** with ***A. hydrophila*** organism responded to the treatment with ciprofloxacin where the clinical signs appear to be recovered at the 3rd day treatment .These results were compatible with those obtained by **Amer and Zaki (1999)** ,who reported that there were variable decline in the mortality rates in medicated groups with ciprofloxacin compared to that of infected non medicated control one .

On the other hand oral administration of oxytetracycline to the infected group showed moderate recovery at the 7th day post treatment (Fig.6).This result was not in harmony with that of **Mayer (1964)** who stated that oxytetracycline has been the drug of choice for treating motile Aeromonas septicemia in fish.

Hematological results :

Our results showed that florfenicol caused slight decrease in all hematological parameters manifested by decrease in RBCs count , Hb and PCV volume . This effect may be attributed to depression of florfenicol on mitochondrial synthesis of protein in bone marrow . Inhibition of mitochondrial protein synthesis ultimately disrupts mitochondrial function , cellular function and cellular proliferation **(Yunis, 1988)** .

In contrast we found that ciprofloxacin induced hemolytic anemia manifested by a significant decrease in erythrocytic count , Hb and PCV % and these were in a agreement with **Cynthia et al., (2006)** who reviewed that hemolytic anemia may be associated with fluoroquinolones administration .

While oxytetracycline caused reversible decrease in RBCs count, Hb and PCV values . These results were in accordance with those of **Rijkers et al. (1981)** who concluded that oxytetracycline caused significant decrease in all hematological values.

Biochemical results :

In the present study we found that there were non significant changes in the level of AST and ALT enzymes following florfenicol administration .These results were nearly similar to that recorded by **Ali et al., (2003).** while there were a slight elevation in both urea and creatinine levels in normal fish and marked elevation in experimentally infected group. This may be due to kidney damage caused by bacterial inoculation. This finding was confirmed by **Saba et al ., (2000)** who showed that chloramphenicol administration resulted in elevated serum urea and creatinine levels. There were also significant decrease in urea and creatinine in infected group of fish treated with a therapeutic dose of ciprofloxacin compared to the infected non treated fish which returned back to the nearly normal control value at 14^{th} day post medication.

Moreover, we found that serum AST and ALT were increased during oxyteteracycline administration. These results were in complete agreement with those of **Shaddad et al (1985)** who showed that, oxytetracycline tended to increase both AST and ALT activities and induced a significant increase in serum urea and creatinine levels . These results were confirmed by those of **Shils (1963)** who stated that tetracycline increase serum urea and creatinine levels, due to increased protein catabolism rather than nephrotoxicosis.

On the other hand , fish infected with ***A.hydrophila*** and treated with florfenicol , ciprofloxacin and oxytetracycline showed a moderate significant improvement in serum urea and creatinine levels when compared with infected non treated groups

CONCLUSION

We may conclude that ciprofloxacin was the powerful drug both in vitro and in vivo followed by florfenicol and lastly oxytetracycline although reversible adverse effects on hematological and biochemical parameters were recorded .

Table (1): Interpretation chart for the size of growth inhibition zones according to Quinn et al.(1994):

Drug	Diameter of inhibition zones (mm)	Interpretation
Florfenicol	22	Susceptible
Ciprofloxacin	30	Susceptible
Oxytetracycline	20	Susceptible

"Susceptible" indicates the pathogen was inhibited by generally achievable blood levels.

Table (2) : Minimum inhibitory concentrations (MICs) of the tested drugs on *A. hydophila ,Salmonella entertidis ,and E.coli.*

MIC (µg / ml)			
Drug	***A. hydrophila***	***Salmonella entertidies***	***E. coli***
Florfenicol	1.1	0.09	0.28
Ciprofloxacin	0.08	0.28	0.04
Oxytetracycline	2.8	0.8	4.0

Table (3): The effect of florfenicol , ciprofloxacin and oxytetracycline on mortality rate (%) of experimentally infected *Clarias lazera* inoculated with *A. hydrophila* .

Fish grouping	Total number	Number of dead fish	Mortality (%)
Control (non infected non treated)	10	0	0%
Infected non treated	10	8	80 %
Infected treated with florfenicol	10	4	40%
Infected treated with ciprofloxacin	10	3	30%
Infected treated with oxyteteracycline	10	5	50%

Table (4): The effect of florfenicol , ciprofloxacine and oxytetracycline on erythrocytic count (10^6 / mm) of clinically healthy and experimentally infected *Clarias Lazera* with *Aeromonas hydrophila* microorganism. (M±S.E) (n=5).

Group	Erythrocytic count (10^6 / mm)		
	1st day	**7th day**	**14th day**
G1 (control) Non infected non treated	2.480 ± 0.081^{a}	2.700 ± 0.025^{a}	2.706 ± 0.072^{a}
G2 Non infected florfenicol treated	1.700 ± 0.065^{b}	2.322 ± 0.070^{b}	2.576 ± 0.043^{ad}
G3 Non infected ciprofloxacin treated	1.374 ± 0.020^{c}	1.840 ± 0.089^{c}	2.226 ± 0.031^{b}
G4 Non infected oxytetracyclin treated	1.684 ± 0.062^{b}	2.572 ± 0.082^{a}	2.798 ± 0.053^{ad}
G5 Infected non treated	0.644 ± 0.06^{d}	0.744 ± 0.061^{d}	0.772 ± 0.022^{c}
G6 Infected florfenicol treated	1.660 ± 0.084^{b}	1.930 ± 0.061^{c}	2.570 ± 0.027^{ad}
G7 Infected ciprofloxacin treated	0.690 ± 0.087^{d}	1.264 ± 0.041^{c}	2.440 ± 0.114^{d}
G8 Infected oxytetracyclin treated	1.364 ± 0.030^{b}	1.634 ± 0.067^{c}	2.176 ± 0.025^{b}

The different letter at the same column means that there was a significant changes at ($p<0.05$) .

Table (5): The effect of florfenicol , ciprofloxacin and oxytetracycline on total leucocytic count (10^3 / mm^3) of clinically healthy and experimentally infected *Clarias lazera* with *Aeromonas hydrophila*. (M±S.E) (n=5).

Group	Days post treatment		
	1st day	7th day	14th day
G1(control) Non infected non treated	7.66±0.40[a]	7.76±1.50[a]	7.20±1.22[a]
G2 Non infected florfenicol treated	5.32±1.49[b]	5.56±1.60[b]	6.64±1.94[b]
G3 Non infected ciprofloxacin treated	4.16±0.74[c]	6.44±1.60[b]	4.64±1.16[c]
G4 Non infected oxytetracyclin treated	3.32±1.20[d]	5.64±3.60[b]	7.16±2.93[a]
G5 Infected non treated	5.16±1.50[b]	5.96±0.79[b]	6.18±1.74[c]
G6 Infected florfenicol treated	6.88±1.36[c]	6.48±2.06[c]	7.16±0.99[a]
G7 Infected ciprofloxacin treated	1.68±0.48[f]	4.40±1.67[d]	6.52±2.15[b]
G8 Infected oxytetracyclin treated	4.56±1.47[g]	5.48±3.77[b]	8.52±5.88[d]

The different letters at the same column means that there was a significant changes at (p<0.05) .

Table (6): The effect of florfenicol , ciprofloxacine and oxytetracycline on total Hb (gm / dl) of clinically healthy and experimentally infected *Clarias lazera* with *Aeromonas hydrophila* microorganism. (M±S.E) (n=5).

Group	Days post treatment		
	1st day	7th day	14th day
G1(control) Non infected non treated	9.92±0.45[a]	9.48±0.37[a]	10.28±0.66[a]
G2 Non infected florfenicol treated	9.16±0.17[a]	9.24±0.20[a]	9.48±0.64[b]
G3 Non infected ciprofloxacin treated	9.20±0.64[b]	9.38±0.39[a]	9.60±0.59[b]
G4 Non infected oxytetracyclin treated	9.44±0.20[a]	9.16±0.26[a]	9.08±0.36[b]
G5 Infected non treated	6.84±0.12[c]	5.72±0.36[b]	4.88±0.23[c]
G6 Infected florfenicol treated	7.60±0.21[b]	7.68±0.60[c]	7.60±0.76[d]
G7 Infected ciprofloxacin treated	8.48±0.19[b]	9.00±0.70[a]	10.92±0.40[a]
G8 Infected oxytetracyclin treated	6.98±0.10[c]	7.40±0.20[c]	8.60±0.18[d]

The different letters at the same column means that there was a significant changes at (p<0.05) .

Table (7): The effect of florfenicol , ciprofloxacine and oxytetracycline on total PCV % of clinically healthy and experimentally infected *Clarias lazera* with *Aeromonas hydrophila* microorganism. (M±S.E) (n=5).

Group	Days post treatment		
	1st day	**7th day**	**14th day**
G1 (control) Non infected non treated	20.76±1.33^{a}	22.44±2.45^{a}	24.84±1.94^{a}
G2 Non infected florfenicol treated	18.48±0.52^{ab}	20.52±1.01^{a}	22.44±1.92^{b}
G3 Non infected ciprofloxacin treated	18.60±1.90^{ab}	19.80±1.79^{a}	22.64±1.37^{b}
G4 Non infected oxytetracyclin treated	19.28±0.62^{ab}	20.88±1.06^{a}	21.24±1.08^{b}
G5 Infected non treated	17.00±0.75^{b}	15.48±0.22^{b}	13.92±0.67^{c}
G6 Infected florfenicol treated	13.80±0.63^{c}	16.68±0.79^{c}	22.92±2.70^{b}
G7 Infected ciprofloxacin treated	10.44±0.56^{d}	14.76±0.13^{b}	23.08±1.00^{a}
G8 Infected oxytetracyclin treated	12.24±0.31^{cd}	16.00±2.52^{c}	18.60±0.46^{d}

The different letters at the same column means that there was a significant changes at (p<0.05) .

Table (8) : The effect of florfenicol, ciprofloxacin and oxytetracycline on serum AST (U/ml) and ALT (U/ml) of clinically healthy and experimentally infected Clarias lazera with A. hydrophila . (M ± S.E) (n = 5)

Group	Day post treatment					
	1st day		7th day		14th day	
	AST	ALT	AST	ALT	AST	ALT
G1 (control) Non infected non treated	44.9 ± 1.0a	60.4±0.18a	46.18±0.64a	65.54±0.2a	46.43±0.38a	69.79±0.17a
G2 Non infected florfenicol treated	46.57±0.52a	62.83±0.38a	46.2±0.95a	69.03±0.1a	45.16±0.49a	60.16±0.59a
G3 Non infected ciprofloxacin treated	69.24±0.97b	69.64±1.93b	56.45±2.46b	69.46±0.61b	56.45±2.46a	62.46±0.61b
G4 Non infected oxytetracycline treated	54.96±0.68c	83.86±0.59c	44.52±0.85a	36.01±1.11c	44.69±1.07a	31.49±1.14a
G5 Infected non treated	64.81±1.04d	65.83±1.6b	83.44±1.12c	90.1.16d	116.8±0.77b	128.02±0.7c
G6 Infected florfenicol treated	52.59±1.86c	85.19±2.15d	46.32±1.57a	76.72±0.88e	46.43±0.38a	69.75±0.76a
G7 Infected ciprofloxacin treated	83.13±0.97e	74.78±1.05e	66.02±1.36d	89.79±0.65f	50.76±1.07c	43.93±1.19d
G8 Infected oxytetracycline treated	76.88±0.58f	129.4±2.04f	41.0±6.36a	61.95±0.55b	45..79±0.78a	40.08±1.02b

The different letters at the same column means that there was a significant changes at P ≤ 0.05

Table (9) : The effect of florfenicol, ciprofloxacin and oxytetracyclin on serum urea and creatinine of clinically healthy and experimentally infected Clarias lazera with A. hydrophila . (M ± S.E) (n = 5)

Group	Day post treatment					
	1st day		7th day		14th day	
	Urea (mg/dl)	Creatinine (mg/dl)	Urea (mg/dl)	Creatinine (mg/dl)	Urea (mg/dl)	Creatinine (mg/dl)
G1 (control) Non infected non treated	24.2±0.95a	0.44±0.01a	26.51±1.63a	0.45±0.01a	24.18±1.38a	0.45±0.36a
G2 Non infected florfenicol treated	29.96±1.7ab	0.74±0.01a	30.84±2.4ab	0.52±0.01b	28.54±1.7ab	0.52±0.01b
G3 Non infected ciprofloxacin treated	47.73±4.19c	0.55±0.01b	28.41±2.23a	0.42±0.01b	31.49±3.8bd	0.5±0.01b
G4 Non infected oxytetracyclin treated	33.6±1.55bd	0.52±0.01c	35.32±0.9bc	0.62±0.01c	33.38±1.1bd	0.66±0.01b
G5 Infected non treated	67.28±2.78e	0.73±0.01d	70.26±3.86d	0.76±0.01d	71.31±2.82e	0.65±0.06c
G6 Infected florfenicol treated	39.58±2.51d	0.65±0.01c	37.37±3.0bc	0.62±0.01c	30.5±2.83bd	0.61±0.04
G7 Infected ciprofloxacin treated	46.2±0.26c	0.7±0.01d	32.99±0.88c	0.54±0.01be	26.6±0.78ab	0.45±0.01a
G8 Infected oxytetracyclin treated	47.16±2.85c	0.63±0.01c	45.84±1.38f	0.65±0.01e	45.43±1.57c	0.43±0.01a

The different letters at the same column means that there was a significant changes at $P \leq 0.05$

Table (10) : The effect of florfenicol, ciprofloxacin and oxytetracycline on protein fractionation of clinically healthy and experimentally infected Clarias lazera with A. hydrophila . (M ± S.E) (n = 5)

Group	Day post treatment								
	1st day			7th day			14th day		
	TP	AL	GL	TP	AL	GL	TP	AL	GL
G1 (control) Non infected non treated	4.26 ± 0.11a	2.32 ± 0.01a	1.90 ± 0.11a	4.28 ± 0.06a	2.28 ± 0.03a	2.0 ± 0.07a	4.42 ± 0.12	2.30 ± 0.19a	2.14 ± 0.17a
G2 Non infected florfenicol treated	4.32 ± 0.08a	2.34 ± 0.05a	1.93 ± 0.19a	4.44 ± 0.59a	2.38 ± 0.04	2.04 ± 0.16	4.64 ± 0.08a	2.42 ± 0.04ab	2.20 ± 0.08ac
G3 Non infected ciprofloxacin treated	5.22 ± 0.06b	2.98 ± 0.05	2.32 ± 0.09b	5.06 ± 0.06b	2.84 ± 0.01b	2.22 ± 0.07b	4.47 ± 0.12ac	2.42 ± 0.01ab	2.14 ± 0.12ac
G4 Non infected oxytetracyclin treated	3.25 ± 0.11c	2.01 ± 0.06c	1.28 ± 0.11c	3.77 ± 0.07c	2.20 ± 0.02c	1.49 ± 0.05c	4.35 ± 0.11ae	2.28 ± 0.01a	2.08 ± 0.11a
G5 Infected non treated	3.73 ± 0.09d	2.25 ± 0.05d	1.52 ± 0.14d	4.01 ± 0.07d	2.09 ± 0.23	1.96 ± 0.25c	4.34 ± 0.15d	2.20 ± 0.37b	2.05 ± 0.29b
G6 Infected florfenicol treated	3.95 ± 0.04cd	2.58 ± 0.24d	1.80 ± 0.96d	4.33 ± 0.10a	2.23 ± 0.28a	2.09 ± 0.29a	4.47 ± 0.12ae	2.40 ± 0.01a	2.11 ± 0.12a
G7 Infected ciprofloxacin treated	5.20 ± 0.21b	2.59 ± 0.05b	2.60 ± 0.25b	5.12 ± 0.05b	3.00 ± 0.07b	2.18 ± 0.04d	4.94 ± 0.06c	2.76 ± 0.01a	2.05 ± 0.18a
G8 Infected oxytetracyclin treated	3.01 ± 0.10e	1.94 ± 0.02e	1.11 ± 0.13e	3.20 ± 0.08e	1.60 ± 0.01e	1.20 ± 0.07e	4.29 ± 0.07e	2.16 ± 0.06b	1.70 ± 0.20c

The different letters at the same column means that there was a significant changes at P ≤ 0.05

Fig (1): ***Clarias lazera*** infected with ***Aeromonas hydrophila*** showed progressive erosion allover the body .

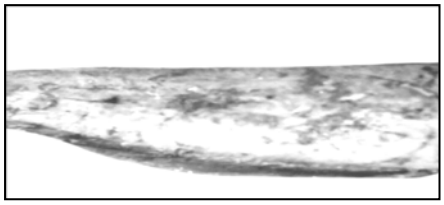


Fig (2): ***Clarias lazera*** infected with ***Aeromonas hydrophila*** showed erythema at the base of the fins .

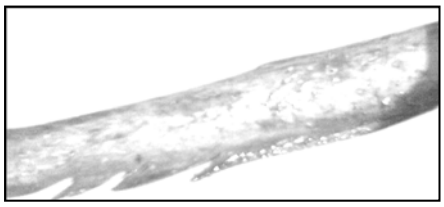
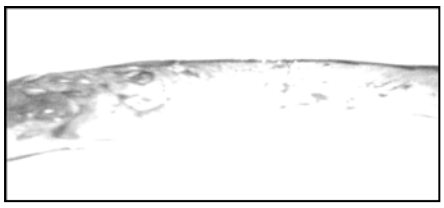
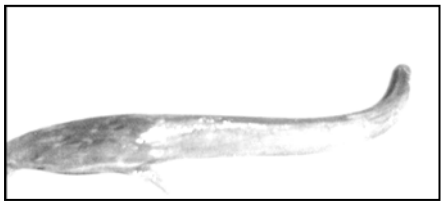


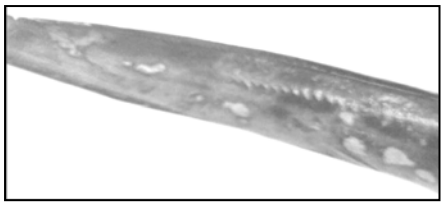
Fig (3): ***Clarias lazera*** infected with ***Aeromonas hydrophila*** showed ulcer formation .



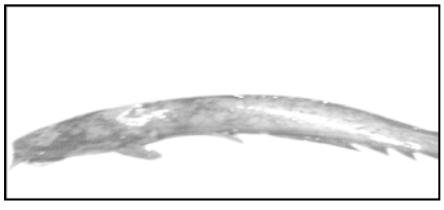
Fig(4): ***Clarias lazera*** infected with ***Aeromonas hydrophila*** and treated with florfenicol on 5^{th} day post treatment .



Fig(5): ***Clarias lazera*** infected with ***Aeromonas hydrophila*** and treated with ciprofloxacin on 3^{th} day post treatment .



Fig(6): ***Clarias lazera*** infected with ***Aeromonas hydrophila*** and treated with oxytetracycline on 7^{th} day post treatment .



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الملخص العربي

دراسات فارماكولوجية على بعض مضادات البكتريا فى الأسماك

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استهدفت هذه الدراسة معرفه كفاءة كلا من دواء السيبروفلوكساسين و الفلورفينيكول و الاوكسي تيتراسيكلين في علاج اسماك القراميط المصابة بميكروب الايروموناس هيدروفيلا معمليا . وقد أعطي دواء السيبروفلوكساسين عن طريق الحقن البريتوني بجرعة علاجيه قدرها ١٠مجم/ كيلو جرام وزن حي يوميا لمده ثلاث أيام متتالية . كما أعطي دواء الفلورفينيكول عن طريق الحقن العضلي بجرعة علاجيه قدرها ٣٠مجم/ كيلو جرام وزن حي يوميا لمده ثلاث أيام متتالية . في حين أعطي دواء الاوكسي تيتيراسيكلين عن طريق الفم بجرعة علاجيه قدرها ٧٥مجم/ لبتر ماء لمدة ٥ أيام متتالية .

أجريت هذه الدراسة علي عدد ١٣٠ سمكة تزن الواحدة ٢٠٠ جرام تقريبا وقد تم إجراء هذه الدراسة علي ثلاث مراحل :المرحلة الأولي أجريت علي ٥٠ سمكة قسمت إلي خمس مجموعات متساوية كلا منها عشر سمكات و ذلك لمقارنه كفائه هذه الأدوية معمليا وعلي الأسماك المصابة وعلي نسب النفوق أما المرحلة الثانية فقد استخدم فيها٠ ٨سمكة قسمت علي ثمان مجموعات كلا منها ١٠ سمكات لدراسة تأثير هذه الأدوية علي صوره الدم و أيضا علي وظائف الكبد و الكلي و البروتينات الكلية و الألبيومين و الجلوبيلين

وقد أوضحت الدراسة ما يلي :-

١- عقار السيبروفلوكساسين أعطي أعلي تأثير مثبط على ميكروب الايروموناس هيدروفيلا معمليا و كذلك في الأسماك الحية و كان هناك تناقص ملحوظ في عدد الأسماك النافقة في المجموعة التي تلقت العلاج بهذا العقار عند مقارنتها بالمجموعة التي لم تعالج و المجوعتين التي تم علاجهما بكلاً من عقار الفلورفينيكول و الاوكسي تيتراسيكلين حيث كانت نسبه النفوق في الأسماك الغير معالجه(٨٠%) و انخفضت إلي(٢٠ &٣٠ &٤٠%) في الأسماك المعالجة يكلا من عقار السيبروفلوكساسين و الفلورفينيكول و الاوكسي تيتراسيكلين علي التوالي .

٢- بدراسة تأثير هذه العقاقير علي صورة الدم لوحظ أن كل هذه العقاقير تحدث تغيرات سلبيه في صوره الدم بدرجات متباينة ولكن تلك التغيرات رجعت إلي معدلها الطبيعي ، وأن هناك تناقص معنوي في هذه القياسات بالنسبة للمجموعة المعداه بالميكروب بالمقارنة بالمجموعة الضابطة .

كما وجد أيضا زيادة معنوية في تلك القياسات في الأسماك المعداه و التي تم علاجها بالمقارنة بالأسماك الغير معداه و هذه الزيادة أعادت مستوي هذه القياسات إلي معدلها الطبيعي .

٣- بدراسة تأثير هذه العقاقير علي مستوي أنزيمي الأسبرتات أمينو ترا نسفيريز و الألانين أمينو ترانسفيريز و علي مستوى البولينا و الكرياتينين وعلي مستوي البروتينات الكلية و الالبيومين و الجلوبيلين في مصل الأسماك المستخدمة وجد أن عقار الفلورفينيكول أحدث تغيرات غير معنوية بالمقارنة بالمجموعة الضابطة و تغيرات معنوية بباقي المجموعات . وأن كل من عقار السيبروفلوكساسين و عقار الاوكسي تيتراسيكلين أحدثا نقصا معنويا في هذه القياسات بالمقارنة بباقي المجموعات.

كما وجد أن هناك تغيرات معنوية في هذه القياسات بالنسبة للمجموعة المعداه بالميكروب بالمقارنة بالمجموعة الضابطة .و أن هذه التغيرات في تلك القياسات عادت إلي معدلها الطبيعي في الأسماك المعداه والتي تم علاجها.