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Impairment induced due to chloramphenicol and ginger on growth parameters of *Channa punctatus* (Bloch)

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ABSTRACT

A maiden study was aimed to comparatively investigate the results of chloramphenicol and ginger on certain growth parameters of *Channa punctatus* (Bloch). The 96hr-LC_{50} tests have been carried out along with a control group. Data were analyzed using regression analysis to determine 96hr-LC_{50} values. The mean values for chloramphenicol and ginger were calculated 55.0 mg l^{-1} and 52.0 g kg^{-1} respectively. Phytochemical components of ginger were analyzed. *Channa punctatus* (57.5±2.0 g and 19.0±1.0 cm) were assigned to seven treatments with three replicates each. Fish had been fed twice daily with commercial feed at 5% body weight for 60 days. The samples have been investigated for various growth parameters such as final weight (FW), Weight gain (WG), daily weight gain (DWG), specific growth rate (SGR), feed conversion ratio (FCR), feed efficiency ratio (FER) and survival rate (SR). Most of growth parameters (FW, WG, DWG, SGR and FER) had been found increased even as FCR reduced than the normal. ANOVA inferred that 2.60g/kg ginger adjust the growth parameters more profoundly than 2.75mg/l chloramphenicol. Although, the mean values of growth parameters except FCR inferred their decline in chloramphenicol but elevation in ginger treated groups as compared to control. The statistical evaluation revealed a significant distinction between chloramphenicol and ginger treatments. The results indicated the chloramphenicol is harmful for fish health. Therefore, ginger may be without difficulty used in vicinity of chloramphenicol.

Keywords: *Channa puncatatus*, Chloramphenicol, ginger, toxicity, growth parameters

INTRODUCTION

Over decades, aquaculture has contributed significantly to the arena protein and food production. However, bacterial diseases have affected aquaculture production (Romero *et al.* 2012). In order to check the situation, antibiotics have been recommended (Cabello 2006). Antibiotics are synthetic compounds succesful to smash or inhibit the growth of pathogens. The use of

antibiotics in aquaculture prevents or treats fish sicknesses and promotes fish growth (Romero *et al.* 2012). The unregulated use of chloramphenicol for the prevention of diseases has been stated in aquaculture (Shukla and Pandey 2005). But, essential issues have been raised on the use of antibiotics in aquaculture as in line with its side consequences on aquaculture and aquatic ecosystems (Ramadu and Dash 2013).

Attention has being shifted to medicinal plant as a feasible opportunity to antibiotics and chemotherapeutics (Ramadu and Dash 2013). Ginger (*Zingiber officinalis*) has been reported to be an appropriate and safe herbal plant (Raa 2000). It is also reported that ginger has the faculty to increase resistance to pathogenic infections and stimulate the immune system against diseases resulting from poor management practices and bad water quality in cultured fish.

Although ginger has been said to be powerful in dealing with stress incurred by the fish during transportation, sorting and grading (Raa 2000), there is need to analyze its impact on growth parameters of the fish.

Channa punctatus is an air breathing freshwater highly priced food fish within South-East Asian countries due to their taste, salutatory and reproductive prosperities. Therefore, the current study aims at ascertaining the results of chloramphenicol and ginger on growth parameters of *Channa punctatus.*

MATERIALS AND METHODS

This work was carried out at the laboratory of the Department of Zoology, Veer Kunwar Singh University, Arrah – 802 301, Bihar, India from January 2018 to December 2019. *Channa punctatus* (57.5±2.0 g and 19.0±1.0 cm) were procured from local fish market, disinfected with dilute $KMnO_4$ solution and transported to the Departmental laboratory. Fish were acclimated for a fortnight in plastic aquaria (1.0x0.5x0.8m) before the experiment and fed with fish food.

Important physico-chemical parameters of experimental water were monitored and measured on alternate day using standard method APHA (2005). Miller and Tainter (1944) regression analysis method was used to determine LC_{50} dose of test substances as there was no partial mortality.

The antibiotic, Chloramphenicol OPF (250mg capsule) (manufactured by Jantec Pharma, New Delhi) was purchased, decapsulated and stored in an airtight container. Fresh rhizomes of ginger (*Zingiber officinale*) were purchased from an open market. The rhizomes were dried under shade for one week. The dried rhizomes were crushed into powdered form, homogenized and sieved using a hand sieve and stored in an airtight container. Phytochemical analysis of ginger and proximate analysis of the experimental feed was performed to ascertain composition of diet (AOAC 2006).

Experimental fish were randomly assigned to seven treatment aquaria. Each aquarium contained ten fish. The fish were divided into group A and B. Chloramphenicol and ginger were given in 1:1000 ratios following their 96hr-LC_{50} values. Group A received chloramphenicol (1.25, 2.50 and 5.00 mg l^{-1} of water) while group B ginger (1.25, 2.50 and 5.00 g kg^{-1} of fish) treatment at different concentrations. The control group contained neither chloramphenicol nor ginger. Water in each aquarium was replaced on alternate day to prevent fouling resulting from feed remnants. The fish were fed with experimental feed twice daily (8.00 am and 17.00 pm) at 5 % body weight for 60 days.

At the end of experiment, weight of the fish was determined with an electronic balance and following growth parameters were calculated.

a. Weight (g) gain : Mean final weight (g) – mean initial weight (g)

b. Weight gain (%) : $\frac{\text{Mean final weight} - \text{mean initial weight}}{\text{mean initial weight}} \times 100$

c. Average daily gain (ADG) : $\frac{\text{Mean final weight} - \text{mean initial weight}}{\text{Time in days}}$

d. Daily growth co-efficient (DGC): $\frac{(\text{Mean final body weight}^{0.3333} - \text{mean Initial body weight}^{0.3333})}{\text{Time in days}} \times 100$ (Cowey 1992)

e. Specific growth rate (SGR) (% per day): $\frac{\text{log mean final body weight} - \text{log mean initial body wieght}}{\text{Time at final sampling} - \text{Time at initial smapling}}$ x 100

(Castell and Tiews 1980)

f. Feed conversion ratio (FCR): $\frac{\text{Total dry feed consumed (g)}}{\text{Total wet weight gained (g)}}$

g. Survival rate (SR) (%): $\frac{\text{No.of harvested fish}}{\text{No.of stocked fish}}$ x 100

Values obtained after the experiment were subjected to F-test using SPSS (version 20 for Windows XP) software.

RESULTS AND DISCUSSION

The physico-chemical parameters of water were found within in range for favourable growth performances as documented by Boyd (1981). Constant values of carbon-dioxide in all the treatments of both chloramphenicol and ginger are indicative of non suffocative conditions which confirm that mortality of fishes due to toxicant exposure. Similarly, the values of pH are not indicative of and acidic or alkaline conditions in aquarium (Table 1). For optimum growth achieved in juvenile fishes, there has to be appropriate water parameters along with inherent factors of age and species differences (Milikin 1982).

The results of acute toxicity of chloramphenicol and ginger are presented in Tables 2 and 3. LC_{50} values recorded in this study were attributed to size of fishes with potentially immune system for biotransformation of test substances from the body. Moreover, the rapid distribution of test substances in the body of fishes lead to faster alterations in behaviour than the normal for the uptake of a toxicant is directly dependent on the size of fishes. Similar results of LC_{50} for chloramphenicol have been reported by Nwani *et al.* (2013) on *Clarias gariepinus*, but much higher dose 305 mg l^{-1} by Snaderson and Thomsen (2009) in a fish. On the other hand, Abdulrazaq *et al.* (2012) calculated 24hr-LD_{50} dose of ginger in 4.5 g kg^{-1} in rats.

The nutritional and phytochemical composition of ginger has been given in Table-4. Ginger contains various mineral elements, vitamins and phytochemicals (alkaloid, flavonoid, saponin, tannin). The variation in composition of ginger depends on the type, variety, agronomic conditions, curing methods, drying and storage conditions (Govindarajan 1982; Gugnani and Ezenwanze 1985). Ingredients and proximate composition of experimental diets has been arranged in Tables 5 and 6. Result on proximate composition of diet revealed that crude protein has the highest percentage (37.39±0.4), followed by fiber (11.23±0.3) and fat (8.93±0.4). Similar observations were also reported by Iheanacho *et al.* (2017).

Effects of chloramphenicol and ginger on growth parameteres of *Channa punctatus* are presented in Table 7. Both the rest substances were found to alter the growth parameters of fish, yet chloramphenicol was found to have more adverse effects than ginger. The ginger treated group had the highest mean values for final weight (233.12±7.21g), weight gain (174.92±6.40g and 300.49±7.49%), daily weight gain (2.91±0.11), specific growth rate (2.31±0.02%), feed efficiency ratio (58.82±1.45%) and survival rate (99.33±0.94%) but lowest feed conversion ratio (1.27±0.92) followed by control and lowest for chloramphenicol treated group respectively. Application of 2 way ANOVA considering the effect of chloramphenicol and ginger concludes that both duration of exposure as well as their doses has highly significant effect on final weight and feed conversion ratio of this fish. On the other hand, that variation in the durations of test materials only has a significant effect on specific growth rate and feed conversion ratio of this fish.

Table 1. Physico-chemical parameters of experimental water.

S. No.	Physico-Chemical Parameters	Value	S. No.	Physico-Chemical Parameters	Value
1.	Temperature	22.0±2.0^{0}C	5.	Chloride	16.7±0.2 mg L^{-1}
2.	pH	7.08±0.24	6.	Hardness	150.6±5.2mg L^{-1}
3.	Dissolved oxygen	6.4±0.8 mg L^{-1}	7.	Total alkalinity	76.0±4.5mg L^{-1}
4.	Free Carbon dioxide	2.50 ± 0.2 mg L^{-1}			

Table 2. Statistical relationship between dose of Chloramphenicol (mg L^{-1}) and mortality of *Channa punctatus* (body weight: 57.5g).

Sl. No.	Exposure period(hrs)	Regression equation y =bx +a	Lethal Concentration (mg L^{-1})		Toxicity Factor (Ayoola et al, 2008)	t value (df=5)	F value (u = 1; v = 4)	95% Confidence limit	
								Lower	Higher
1	24	y= 0.0425x – 0.958	LC_{10}	46.071	1.000	2.270 (p>0.05)	5.153 (p>0.05)	-10.225	515.465
			LC_{50}	140.188				32.116	937.718
			LC_{90}	234.306				74.457	1359.970
2	96	y = 0.1325x – 1.958	LC_{10}	21.072	2.587	4.14 (p<0.01)	16.91 (p<0.01)	-11.642	272.561
			LC_{50}	54.183				5.504	398.863
			LC_{90}	81.449				22.649	525.166

Table 3. Statistical relationship between dose of ginger (mg $fish^{-1}$) and mortality of *Channa punctatus* (body weight: 57.5g).

Sl. No.	Exposure period (hrs)	Regression equation y =bx +a	Lethal Concentration (mg $fish^{-1}$)		Toxicity Factor (Ayoola et al, 2008)	t value (df=5)	F value (u = 1; v = 4)	95% Confidence limit	
								Lower	Higher
1	24	y= 0.000715x – 0.918	LC_{10}	2682.517	1.000	0.6946 (p>0.05)	4.095 (p>0.05)	-756.213	19225.56
			LC_{50}	8276.923				1610.65	937.718
			LC_{90}	13871.3293				3977.51	1359.970
2	96	y = 0.1325x – 1.958	LC_{10}	1232.088	2.768	2.61 (p<0.05)	12.51 (p<0.05)	-743.167	17614.38
			LC_{50}	2990.330 (52.0g/kg)				241.812	2578.431
			LC_{90}	4748.571				1226.79	3395.42

Table 4. Nutritional and phytochemical composition of powdered ginger (per 100g).

Constituents	Unit	Value	Constituents	Unit	Value	Constituents	Unit	Value
Moisture	%	15.20	Vitamin B Complex	µg	9.64	Manganese	mg	10.20
Fiber	%	58.00	Vitamin C	mg	11.92	Potassium	mg	1.80
Carbohydrate	g	38.50	Magnesium	mg	10.50	Sodium	mg	1.10
Protein	g	6.00	Calcium	mg	102.60	Chromium	µg	80.40
Fat	g	4.40	Phosphorus	mg	200.80	Alkaloid	mg	0.6
Ash	g	4.60	Iron	mg	9.50	Flavonoid	mg	0.01
Vitamin A	µg	3.20	Zinc	mg	0.90	Saponin	mg	0.03
Vitamin D	ng	17.60	Copper	mg	0.68	Tannin	mg	0.01

Table 5. Ingredients of experimental diets.

Ingredient (g kg^{-1})	D0	D1	D2	D3	Ingredient (g kg^{-1})	D0	D1	D2	D3
Ginger	0	1.25	2.50	5.00	Rice flour	50	50	50	50
Fish meal	320	320	320	320	Maize flour	80	80	80	80
Soya flour	400	400	400	400	Ground nut oil cake	70	70	70	70
Wheat flour	80	80	80	80					

Table 6. Proximate composition of experimental diets.

Proximate (%) composition	D0	D1	D2	D3	Proximate (%) composition	D0	D1	D2	D3
Ash	8.07	8.05	8.11	8.02	Dry matter	90.95	90.90	90.96	90.99
Fat	8.93	8.90	8.97	8.93	Protein	37.38	37.41	37.45	37.32
Fibre	11.21	11.24	11.28	11.20	Nitrogen free extract	45.62	45.64	45.47	45.73
Moisture	9.05	9.10	9.04	9.01					

Table 7. Growth pattern of *Channa punctatus* (n = 40) under different doses of Chloramphenicol and ginger after 60 days.

Parameters	Control	Dose of test material					
		Chloramphenicol (mg L^{-1})			Ginger (g kg^{-1})		
		1.25	2.50	5.00	1.25	2.50	5.00
Initial weight (g)	57.46±1.16	56.97±1.72	56.66±1.20	55.58±0.98	59.18±1.56	58.37±1.16	57.04±1.91
Final weight (FW) (g)	222.77±7.74	220.86±4.75	222.69±6.66	206.75±10.29	238.27±9.54	238.17±11.67	222.92±9.49
		F value (c=2 and r=2) c=28.15** and r =19.91**			F value (c=2 and r=2) c=149.3*** and r =247.1***		
Weight gain (WG) (g)	165.31±6.58	163.89±3.03	166.03±5.46	151.17±9.31	179.09±7.98	179.80±10.51	165.88±7.58
Weight gain (WG) (%)	287.69±5.67	287.67±1.76	293.03±4.56	271.98±9.50	302.62±5.12	308.03±9.06	290.81±3.97
Daily weight gain (g)	2.57±0.11	2.73±0.05	2.77±0.09	2.52±0.16	2.98±0.13	3.00±0.18	2.76±0.13
Specific growth rate (SGR) (%)	2.27±1.90	2.27±1.02	2.28±1.70	2.18±1.68	2.32±1.80	2.33±1.46	2.28±1.60
		F value (c=2 and r=2) c=0.089^{NS} and r =38.90**			F value (c=2 and r=2) c=0.07^{NS} and r =269.6**		
Feed Conversion ratio (FCR)	1.31±0.80	1.31±0.90	1.30±0.94	1.37±0.54	1.27±0.86	1.25±0.54	1.30±0.81
		F value (c=2 and r=2) c=0.209^{NS} and r =41.87**			F value (c=2 and r=2) c=0.064^{NS} and r =54.94**		
Feed Efficiency ratio (FER) (%)	56.61±19.0	56.64±7.68	57.35±8.70	53.38±9.86	59.18±9.73	60.40±10.68	56.89±9.84
		F value (c=2 and r=2) c=11.30* and r =192.9***			F value (c=2 and r=2) c=33.33** and r =1060.0***		
Survival rate (SR) (%)	98.0±2.0	98.0±2.0	96.0±4.0	96.0±4.0	100.0±0	100.0±0	98.0±2.0

The maximum mean increase in final weight of 174.92g after feeding of ginger based diet is due to presence of carbohydrates, vitamins, carotenoids, minerals, tannin, fiber, alkaloids, flavoids and saponin (Shirin and Prakash 2010). Ginger has been reported to favour various activities like growth, enhancement of immune stimulation, maturation of fish species and antipathogenic properties due to the active phytochemicals (Romero *et al.* 2012). Similar results were recorded by Shalaby *et al.* (2006) and Mahmoud *et al.* (2019) in *Oreochromis niloticus and* Ogueji *et al.* (2017) in *Clarias gariepinus*. On the other hand, the use of chloramphenicol is reported to enhance digestibility by preventing the irritation of the intestinal lining and effective utilization of the nutrients from the intestine by thinning of the mucosal layer of the fish (Dafwang *et al.* 1987; FAO 2005) that results in better growth than the normal. Such observations based on antibiotics have also been obtained by Lawal *et al.* (2012) and Adewole (2016) in *Clarias gariepinus* with different levels of oxytetracycline, amoxicillin and furasol.

Specific growth rate was developed by Iwama and Tautz (1981) to eliminate the problem with the decline in specific growth ratio with increasing body weight. It is saturation constant and depends on the concentration of limiting substrate. It indicates that if all nutrients are present in a substance, maximum specific growth will be possible. Its maximum mean increase of 2.31% after feeding of ginger based diet in this work is also related with the ingredients. Our results are not in accordance with the findings reported by Mahmoud *et al.* (2019) in *Oreochromis niloticus and* Ogueji *et al.* (2017) in *Clarias gariepinus*. Further, the present findings are in agreement with Shalaby *et al.* (2006) in *Oreochromis niloticus* and Adewole (2016) in *Clarias gariepinus* with different antibiotics.

Feed conversion ratio is the ratio of input to output and a function of feed intake. Its value depends on feed intake and temperature. The work inferred that 1.27kg ginger supplemented feed while 1.33kg chloramphenicol supplemented feed can increase 1kg body weight of test fish. On the other hand, Ivlev (1939) proposed feed efficiency ratio as the ratio of output to input and temperature independent. It is a growth parameter related with percentage of utilized percentage of feed. A high feed efficiency ratio with 58.82% ginger supplemented feed in this work shows larger growth rate of fish and a lower excretion rate. The present findings were agreement with Jahanjoo *et al.* (2018) in *Sparidentex hasta* and Mahmoud *et al.* (2019) in *Oreochromis niloticus*. Further, with the same line, Shalaby *et al.* (2006) in *Oreochromis niloticus* and Adewole (2016) in *Clarias gariepinus* with antibiotics such as amoxicillin etc.

CONCLUSION

These results inferred that the ginger stimulate fish growth in relation to ginger supplementation in a dose dependent manner. The findings also revealed that ginger increased growth as compared to chloramphenicol and control group. Therefore, the use of ginger in aquaculture as phytobiotics should be encouraged.

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Conflict of Interest
The author declares that there is no conflict of interest.

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