Study of disease resistance in *Mystus vittatus* against some pathogens fed by dietary kiwi fruit *Actinidia deliciosa*

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ABSTRACT:

In present investigation attempts had been made to detect dietary effect of kiwi fruit on growth performance and immunology of *Mystus vittatus* Fruits are an important part of a healthy diet in our daily routine. All the fishes were put in portable plastic pools each portable plastic pools stacked with 10 infected fish averaging 11.60 ± 0.80 g and those were fed at maintenance level for 60 days prior to the experiments.. the pathogens were isolated, characterized and identified as *Aeromonas sp.,Escherichia coli, Edwardsiella sp. Mycobacterium sp and Vibrio sp.*. The various concentration of soxlet extract of kiwi fruit was used as 20 ml/kg,(Dk-2) , 30 ml/kg (Dk-2), 50 ml/kg (Dk-3). 60 ml/kg(Dk-4) and Dk-1 as control. These feeds selected for further antagonistic activity against all the pathogens by formation of zone of inhibition.. Present study suggests that the kiwi fruit can used with its antibacterial potential in fish culture.

Index Terms: Kiwi fruit , *Mystus vittatus*, fish fe

INTRODUCTION

The health of the fish thus can be improved by the elimination of pathogens or at least by minimizing their effects in aquaculture. Due to outbreak of disease in aquaculture industry in the last 20 years, use of antibiotics have lead to the development of drug-resistant strains which results depletion of natural defense system in aquatic animals. To overcome these problems there is need to research on eco friendly alternatives like use of natural fruits having medicinal properties In present investigation dietary effect of *Actinidia deliciosa* Kiwi fruit on disease resistance of *Mystus vittatus.* The review study by David P. Richardson (1) he shows that usefulness of kiwifruit as nutritional and health promoter. Kiwi fruit has become terribly popular during the past two decades due to its various medicinal properties. It is also known as Macaque peach and Mihoutau and Chinese gooseberry. It is said that the fruit got its name Kiwi from the land of New Zealand. It is native to Chang Kiang valley (Yang Tao) of northern China. In the early 1900, kiwi fruit was first exported to Asia as an ornamental vine. It has more potassium than a banana or citrus fruits. Many studies have shown that it contains many medicinally useful compounds, which may be beneficial in the treatment of sleep disorders. Commonly some varieties of kiwi fruit found in China have a strong anti-mutagenesis effect.

Fish is an important aquatic organism, fish products are an important source of protein for human consumption. (2) *Mystus vittatus* is classified as economically important fish species that has great commercial demands.

MATERIALS AND METHODS

1. Preparation of experimental Diet: This study was carried out at the RSML laboratory, to investigate the effects of kiwi fruit on biochemical parameters and growth performance of *Mystus vittatus*. Adult male *Mystus vittatus* of initial weight 164.78 g were collected from local fish market, fish was acclimated to the experimental conditions for a period of two weeks. (See Table 1 and 2, Figure I and II)

2. Antagonistic effect: Here we used the soxlet extract (3, 4) of kiwi fruit for in vitro study against isolated pathogens from diseased *Mystus vittatus*

3. Experimental Setup and Feeding Trial

Collected fish *Mystus vittatus* were stocked for acclimation in 60 liter portable plastic pools for one week. Fishes were fed twice daily, at 09:00h and at 15:00h. the feeding rate was at 6 % body weight day^{-1} for the whole rearing period of 60 days, and the amount of feed was adjusted every tenth day following a bulk weighing of each group of fish.

Antagonistic activity

5 sets of experiment, *Aeromonas* sp.*, Mycobacterium sp.,E-coli, Edwardsiella sp* and *Vibrio* sp and were conducted for 3 week For each bacterium 10 conical flasks (100 ml) have culture medium (50 ml) containing pure strain of bacterial fish pathogens. In this 5 ml of Kiwi Soxlet Extract was added. The control flask contains only pathogens. The entire flasks were incubated at 37^{o}c for 2 week. Starting from first day, the number and growth of organism was monitored using standard dilution technique.

RESULTS

1. Identification and characterization of microorganisms

among the various biochemical assays studied the positive results were observed in all five pathogens isolates, such as methyl red, nitrate reduction, Catalase, Oxidase And Maltose, Mannitol, Rhamnose for sugar assimilation. Whereas the negative results were obtained in Indole, Vogus Proskur, Citrate Utilization, Urease And Arabinose for sugar assimilation. Arabinose, Fructose, Glucose, Lactose were fermented by isolated *Vibrio* Sp.*, E. Coli, Edwardsiella SP* Sp. and *Mycobacterium Sp* produce acid and gas (Table 4,).

Based on the morphological characteristics and biochemical properties, the isolated fish pathogens are identified as *aeromonas* sp.*, vibrio* sp.*, e. coli, edwardsiella* sp.*, mycobacterium sp.*

2.Antagonistic effect

To evaluate the antagonistic effect of *lactobacillus* sp. isolate against the fresh water fish pathogens, *vibrio* sp.*, aeromonas* sp.*, e. coli, edwardsiella* sp. and *mycobacterium sp* isolates were isolated from the *Mystus vittatus*. The antagonistic

activity of *lactobacillus* sp. isolate was done against fish pathogens by Cross streak method. The zone of inhibition showed against *aeromonas* sp.(15.21±1.09) which is more than 10 mm , followed by *mycobacterium sp (*13.03±0.9), *e. coli (*10.01±1.0), *vibrio sp (*8.2±0.5) and *edwardsiella sp(*9.1 ±1.7) (table 3 Fig-III).

the size and weight of the *Mystus vittatus* increased nearly 21 g when compared to that of control fish, here total weight recorded was 17.03 g. it shows that the kiwi fruit reach feed showed greater growth improvement.

Among the various biochemical assays studied the positive results were observed in *lactobacillus* isolates such as urease, catalase, fructose, glucose, lactose, maltose, mannitol and rhamnose whereas the negative results were indole, methyl red, vogus proskur, citrate utilization, nitrate reduction, oxidase and arabinose. fish pathogens, such as *Aeromonas* sp.*, e. coli, edwardsiella sp Mycobacterium sp* and *vibrio* sp were isolated from the gut, gill, muscle region of infected fishes. these five pathogens were morphologically characterized as *Aeromonas* Sp.*, E. Coli, Edwardsiella* Sp. *Mycobacterium Sp* And *Vibrio Sp* they were physically gram negative, motile and rod shape.among the various biochemical assays shown in table 4 .

DISCUSSION:

From the present investigation, we can concluded that the antagonistic effects of Kiwi Extract And Kiwi Feed against the fresh water fish pathogens is effective and safe which helpful in the bacterial disease control in *Mystus vittatus*. We seen here no attempts have been taken on fishes .Some literature related to similar experiment shows significance of utilization of kiwi fruit.

Kiwi fruit vitamins (mainly C + E vitamins) work as antioxidant enzymes (5). These vitamins amid the significant nutrients persuade inner immune system and their supply can reduce fish mortality and improve performance. Vitamin C is taking spotlight in intensive aquaculture due to which there is note about high growth performance and maximum survival occurs in fish community (6). Further the most importat part in kiwi fruit is Vitamin C which play role of immune modulators(7) , not only vitamine rich but also serve as good antioxidant (8) in terms of reproduction it responsible to produce excellence gametes (9). So above evidences support for kiwi fruit use. Though it is costly but useful instead of chemical additives. In aquaculture if there is scarcity or deformity of Vitamin C was reported by Fracalossi (10) in case of fishes with structural deformities viz.Scoliosis, abnormalities in the cartilage, eyes gills or fins, abnormal pigmentation, increased fragility of the capillaries, reduce the immune response and reproductive performance.

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Tables and figures

Table (1): Ingredients composition of the basal diet

Ingredients (%)	**Experimental Fish Feeds**				
Ingredients	**control**	**Dk-1**	**Dk-2**	**Dk-3**	**Dk-4**
Soybean	60	60	60	60	60
Wheat	10	10	10	10	10
Rice bran	10	10	10	10	10
Ground nut oil cake	20	20	20	20	20
Kiwi	00	20*	30*	50*	60*
Freeze dried tubifex worms	5	5	5	5	5
* Milliliter Per 1 Kg of Diet					

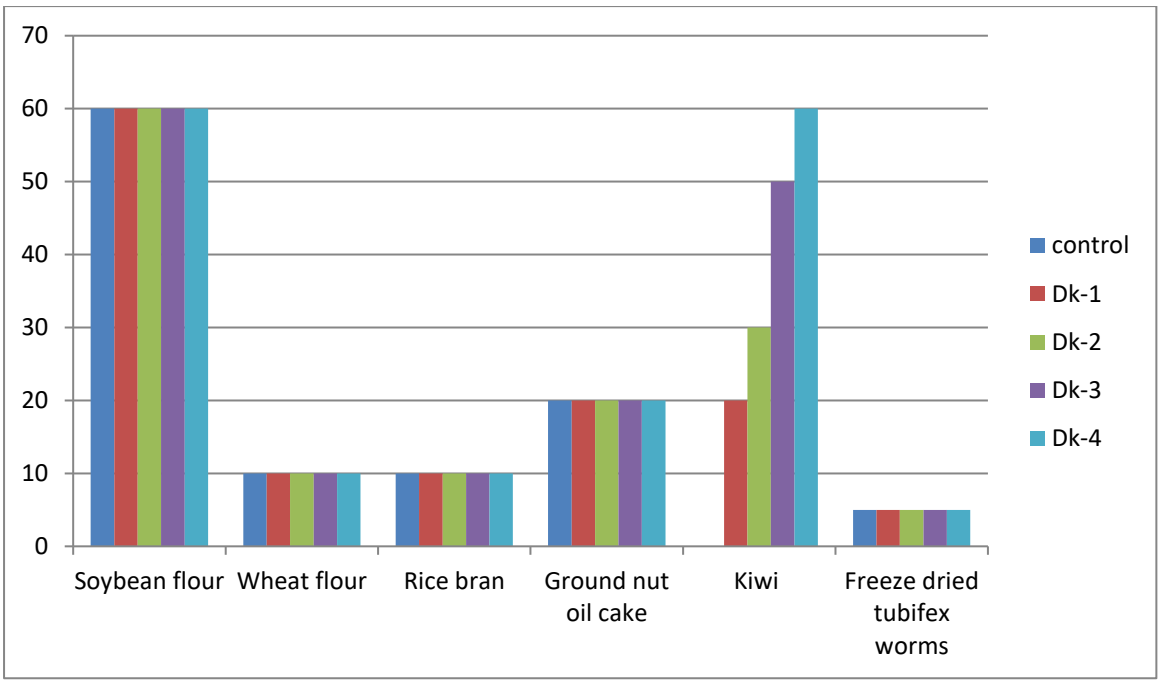

Fig I: Ingredients of the basal diet

Table (2): Chemical analysis of experimental (basal) diet

Nutrient composition	% on dry matter basis
Dry matter (DM)	91.50
Crude protein (CP)	26.41
Ether extract (EE)	7.90
Crude fiber	8.22
Ash	8.24
Total carbohydrate	41.17

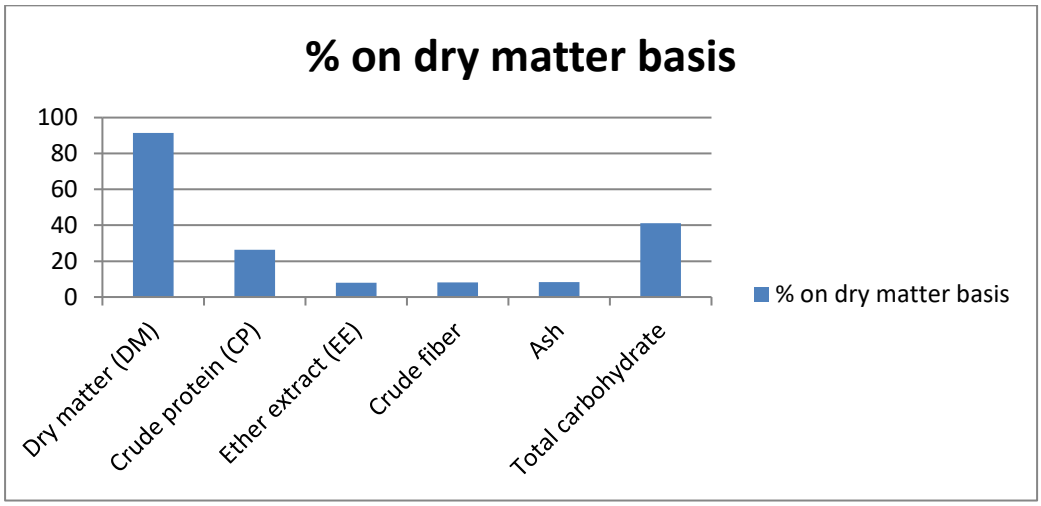

Fig.II. Graphic Representation of Chemical Analysis of Experimental feed

Table 3: Antibacterial Activity of Soxlet Kiwi Fruit Extract *against* Fish Pathogens.

Sr.No	Isolated pathogens	Zone of Inhibition Day -7	Zone of Inhibition Day-14	Zone of Inhibition Day-21
1.	*Aeromonas Sp.*	13.05±0.11	14.07±1.07	15.21±1.09
2	*Mycobacterium Sp*	10.02±0.2	11.01±0.4	13.03±0.9
3.	*E. Coli*	9.02±0.1	10.03±1.3	10.01±1.0
4.	*Edwardsiella Sp*	6.3 ±0.2	7.0±0.3	8.2±0.5
5.	*Vibrio Sp*	5.4 ±1.42	6.1 ±1.51	9.1 ±1.70

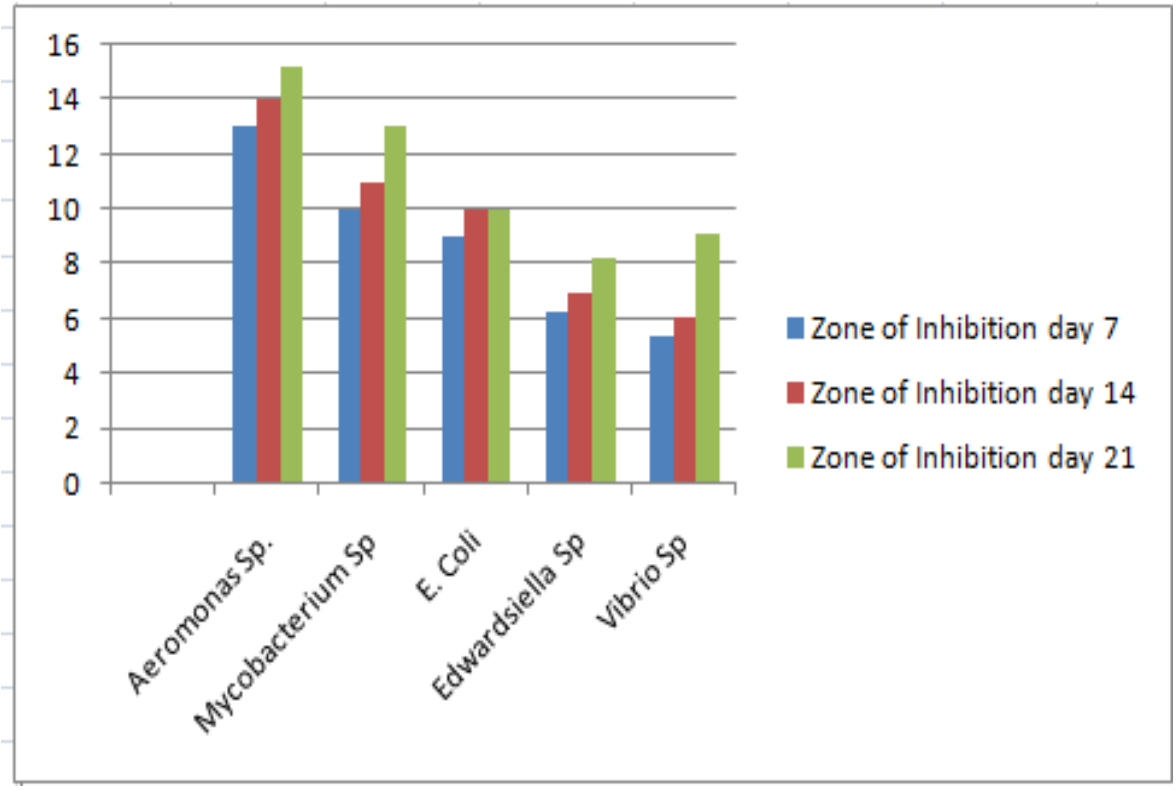

Fig. III - Antibacterial Activity of Soxlet Kiwi Fruit Extract

Table 4: Basic biochemical test for pathogenic organism isolated from *Mystus vittatus*

Sr.No	Basic Characteristics	Pathogen
1	***Aeromonas sp***	
	Arbutin	+ve
	Arginine Dehydrolase	+ve
	Elastase	+ve
	Esculin Hydrolysis	+ve
2	***Mycobacterium sp***	
	Growth at 25°C	-ve
	Growth on P-Nitrobenzoic Acid	-ve
	Growth on TCH (10 mg/ml)	-ve
	Iron Uptake	-ve
3	***E. coli***	
	Indole	+ve
	Motility	Motile
	MR (Methyl Red)	+ve
	Nitrate Reduction	+ve
4	***Edwardsiella sp***	
	Lysine Decarboxylase	+ve
	ONPG (β-galactosidase)	-ve
	Ornithine Decarboxylase	+ve
	Phenylalanine Deaminase	-ve
5	***Vibrio sp***	
	Hemolysis	Beta Hemolysis
	Indole	+ve
	Motility	Motile
	MR (Methyl Red)	-ve

REFERENCES

1. David P. Richardson,1 Juliet Ansell,2 and Lynley N. (2018) The nutritional and health attributes of kiwifruit: a review, Eur J Nutr. 57(8): 2659–2676.
2. Duran, A. and Talas, Z. S. (2009), Biochemical changes and sensory assessments on tissues of carp (cyprinus carpio) during sale conditions, fish physiol. Biochem., 35, 709-714
3. Azwanida NN.(2015;) A review on the extraction methods use in medicinal plants, principle, strength, and limitation. *Med Aromat Plants.*4:196

4. Doughari JH.(2012.) Phytochemicals: Extraction methods, basic structures, and mode of action as potential chemotherapeutic agents, phytochemicals––a global perspective of their role in nutrition and health. In: Venketeshwer R, editor. *A Global Perspective of Their Role in Nutrition and Health.*
5. Keleştemur, G. T., & Özdemir, Y. (2013). Effects of dietary vitamin a and e on growth performance and antioxidant status in blood of juvenile rainbow trout (*Oncorhynchus mykiss*, w. 1792) exposed to flow Fishe stress. Japs, Journal of Animal and Plant Sciences, 23(3), 821-827.
6. Verlhac, V., Gabaudan, J., Obach, A., Schüep, W., & Hole, R. (1996). Influence of dietary glucan and vitamin C on non -specific and specific immune responses of rainbow trout (*Oncorhynchus mykiss*). Aquaculture,143(2), 123-133.
7. Chen, R., Lochmann, R., Goodwin, A., Praveen, K., Dabrowski, K., & Lee, K. J. (2003). Alternative complement activity and resistance to heat stress in golden shiners (*Notemigonus crysoleucas*) are increased by dietary vitamin C levels in excess of requirements for prevention of deficiency signs. The Journal of nutrition, 133(7), 2281-2286.
8. Dabrowski, K., & Ciereszko, A. (2001). Ascorbic acid and reproduction in fish: endocrine regulation and gamete quality. Aquaculture research, 32(8), 623-638.
9. Zhou, X., Xie, M., Niu, C., & Sun, R. (2003). The effects of dietary vitamin C on growth, liver vitamin C and serum cortisol in stressed and unstressed juvenile soft-shelled turtles (*Pelodiscus sinensis*). CompaFishive Biochemistry and Physiology Part A: Molecular & IntegFishive Physiology, 135(2), 263-270.
10. Debora Fracalossi ,Mary E. Allen ,Donald K. Nichols, Olav T Oftedal. (1998) Oscars, *Astronotus ocellatus*, have a Dietary Requirement for Vitamin C, *Journal of Nutrition* 128(10):1745-51