

EFFECT OF ZINGIBER OFFICINALIS AND CYANODON DACTYLON AGAINST PH STRESS IN GIANT FRESHWATER PRAWN MACROBRACHIUM ROSENBERGII.

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ABSTRACT

To investigate the ability of *Zingiber officinalis* (*Z. officinalis*) and *Cyanodon dactylon* (*C. dactylon*) supplementation to antagonize the stressful pH condition on *Macrobrachium rosenbergii* (*M. rosenbergii*) juveniles, three groups each of 40 Juveniles in two replicates fed diets supplemented with 1.5 % *Z. officinalis* and 2 % *C. dactylon* were exposed to pH 9.5 for 5 weeks. Juveniles exposed to high pH and fed with *Z. officinalis* and *C. dactylon* had significant increase ($p < 0.05$) in Average Body Weight (ABW), Average Body Length (ABL), Specific Growth Rate (SGR), Condition Factor (CF), Weight Gain Rate (WGR), Length Gain Rate (LGR) and Feed Conversion Ratio (FCR) compared with those of control along the whole experiment. Juveniles exposed to high pH and fed *Z. officinalis* and *C. dactylon* had significantly higher ($p < 0.05$) Total Hemocyte Count (THC), Hemocyte Viability (HV), Differential Hemocyte Count (DHC), Phagocytic Rate (PR) and Phagocytic Index (PI) as compared with control at 4 and 5 weeks from the start of feeding. Microscopically, hepatopancreas and stomach of *Z. officinalis* and *C. dactylon* fed juveniles showed no marked alterations. According to the present study, it could be concluded that *Z. officinalis* and *C. dactylon* were effective in controlling the pH stress in *M. rosenbergii*.

Key words: Herbals – Growth promotor – Immunostimulant – Prawn

1. INTRODUCTION

M. rosenbergii is the most popular prawn species used for commercial farming and has been transported worldwide (New, 2002). At the global level, people have understood the bad effect of chemical products and they are now shifting to natural products (Fauci, 1993). Natural plant products have been reported to promote various activities like anti-stress, growth promotion, appetite stimulation, tonic and immunostimulation, and to have antimicrobial properties in finfish and shrimp (Citarasu *et al.*, 2001, 2002; Sivaram *et al.*, 2004).

In reared shrimp, the physico-chemical quality of water affect the metabolism, growth, survival and the immune system, therefore the study aimed to investigate this effect of *Z.officinalis* and *C. dactylon* on growth performance and immune parameters in *M. rosenbergii* as well as, their antagonism to overcome high water pH. Water with low (4.6-5.0) and high pH (9.0-9.5) levels have been reported to decrease the THC and PO activity of the fresh water prawn *Macrobrachium rosenbergii* (Chen *et al.*, 1995).

Z. officinalis is beneficial to growth and immune systems in aquatic animals (Yueh *et al.*, 2012). Ginger rhizomes contain a number of active ingredients (Govindarajan VS., 1982). *C. dactylon* plant has been used to control various diseases like diabetes, ulcer and diarrhea (Badri and Renu, 2011). The aim of this study was to evaluate the effect of *Z. officinalis* and *C. dactylon*, through measuring some growth performance, immune response parameters and histopathological changes of *M. rosenbergii* stressed juveniles.

2. MATERIALS AND METHODS

2.1. Acclimation of juveniles to 0 ppt salinity:

A total of about 300 *M. rosenbergii* Juveniles with average body weight 0.3g were obtained from Maryout Project And Company for fish farms, Alexandria, Egypt and stocked into two well prepared cement ponds for one month, ponds were supplied with brackish water 12 ppt salinity that was decreased gradually until reached 0 ppt salinity. Green house ponds were prepared and water parameters were adjusted at Temperature 30°C, pH 7 and 0 ppt salinity. Juveniles were fed with basal diet (crude protein 30%) (Maryout Company) at rate of 3% body weight (Hien, 1999). Excess feed and excreta were siphoned daily along the acclimation period. Later on, Juveniles were divided into nine groups and stocked in cement ponds at rate of 40 juveniles/pond and left for 3 days for acclimatization.

2.2. Experimental design:

2.2.1. Preparation of experimental diet supplemented with *Z.officinalis* (Suheyla *et al.*, 2003) and *Cyanodon dactylon* (Ngan *et al.*, 1988):

Rhizomes of *Z. officinalis* and whole plant of *C. dactylon* were shade-dried and ground into fine powder at Agricultural Botany Department, Faculty of Agriculture, Benha University. Three experimental diets were prepared. Fine powdered basal diet was divided

into three portions first portion incorporated with 1.5 % (w/w) *Z. officinalis*, the second portion mixed with 2 % (w/w) *C. dactylon* and the last portion kept free without any additives (control). Suitable amount of water was added to form moist dough then pelleted,

Allowed to dry at room temperature, then packed in clean dry plastic containers and kept tightly closed at 4°C till use.

2.2.2 Effect of high rearing water pH (9.5) and feeding of *Z. officinalis* and *C. dactylon* on growth performance and immune response of *M. rosenbergii*:

2.2.2.1. Acclimation of juveniles to pH 9.5:

Juveniles were divided into four three and stocked in cement ponds at rate of 40 juveniles/pond and left for 3 days for acclimatization before start of experiment. pH was adjusted at 9.5 along the period of the experiment.

2.2.2.2. Feeding trials:

Three groups were used, one control and two treatment groups in two replicates. The control group and the two treatment groups were fed with control diet and the basal diet supplied with *Z. officinalis* 1.5 % and *C. dactylon* 2%, respectively. The treated and control groups received experimental diets for period of five weeks. Feeding amount increased regularly based on body weight measurements every two weeks. Number of dead juveniles was recorded daily in all groups along the experiment.

2.2.2.3. Determination of growth parameters:

Sample of five juveniles were taken from each group at the end of 2, 3, 4 and 5 weeks after the start of feeding for evaluation of growth performance parameters.

Body weight was measured using portable electric balance and Body length was measured using graduated ruler. Weight gain rate (WGR), Length gain rate (LGR), Feed conversion ratio (FCR) were calculated according to Bo Liu *et al.*, 2010. Moreover, Specific growth rate (SGR) and condition factor (K) were determined as described by Laird and Needham, 1988.

2.2.2.4. Measurement of immune related parameters:

Hemolymph was collected and divided in two portions one was mixed with Alsever's solution according to FAO, 2002 with slight modification {Citric acid (0.055 g), Sodium citrate (0.8 g), Glucose (2.05 g), Nacl (0.64 g), EDTA (4 g) and 100 ml Distilled water, pH (7)} for estimation of Total Hemocytic Count (THC), Hemocyte Viability (HV) and Phagocytic activity, while the other portion was mixed with 10% formalin for determination of the Differential Hemocytic Count(DHC). Immune related parameters such as THC, HV, DHC and Hemocytes phagocytosis were measured in all groups at 4 and 5 weeks from start of feeding.

THC and HV were measured following the method of Cheng *et al.*, 2003:

THC = number of counted haemocytes in the 4 corners $\times 2 \times \frac{1}{4} \times 10^4 \times$ the dilution factor.

HV (%) = (total number of viable cells /total number of cells) $\times 100$

DHC was measured according to Wootton and Pipe, 2003; Abdel hameed, 2008.

Phagocytic Index (PI) was measured according to **Itami *et al.* [24]** with minor modification as following:

$PI = 100 \times (\text{number of haemocytes engulfed beads} / \text{total number of haemocytes}) \times (\text{number of beads that were engulfed by the haemocytes} / \text{total number of haemocytes})$.

Phagocytic rate (PR) was calculated according to Cheng *et al.*, 2003:

$PR = (\text{Phagocytic Hemocytes} / \text{Total Hemocytes}) \times 100$.

2.2.2.5. Sampling and histopathological examination:

Soon after scarification, samples were taken at 3, 4 and 5 weeks from start of feeding from all groups and fixed in Davidson's Fixative and then subjected to histopathological sectioning and preparation according to Banchroft *et al.*, 1996.

2.2.2.6. Statistical analysis:

Results were analyzed using SPSS (version 16.0) software. One way ANOVA and Duncan's multiple range tests were used to determine the significance of differences between groups. All the results were expressed as means \pm standard error ($M \pm SE$) and significant differences were expressed ($P < 0.05$).

3. RESULTS

3.1. Growth parameters:

Growth performance parameters including AW, AL, SGR, CF, WGR, LGR and FCR of juveniles from all groups were significantly increased compared to the control along the whole experiment period (Fig, 1). In addition the survival rate of *M. rosenbergii* among all groups in each period of time was similar (100%).

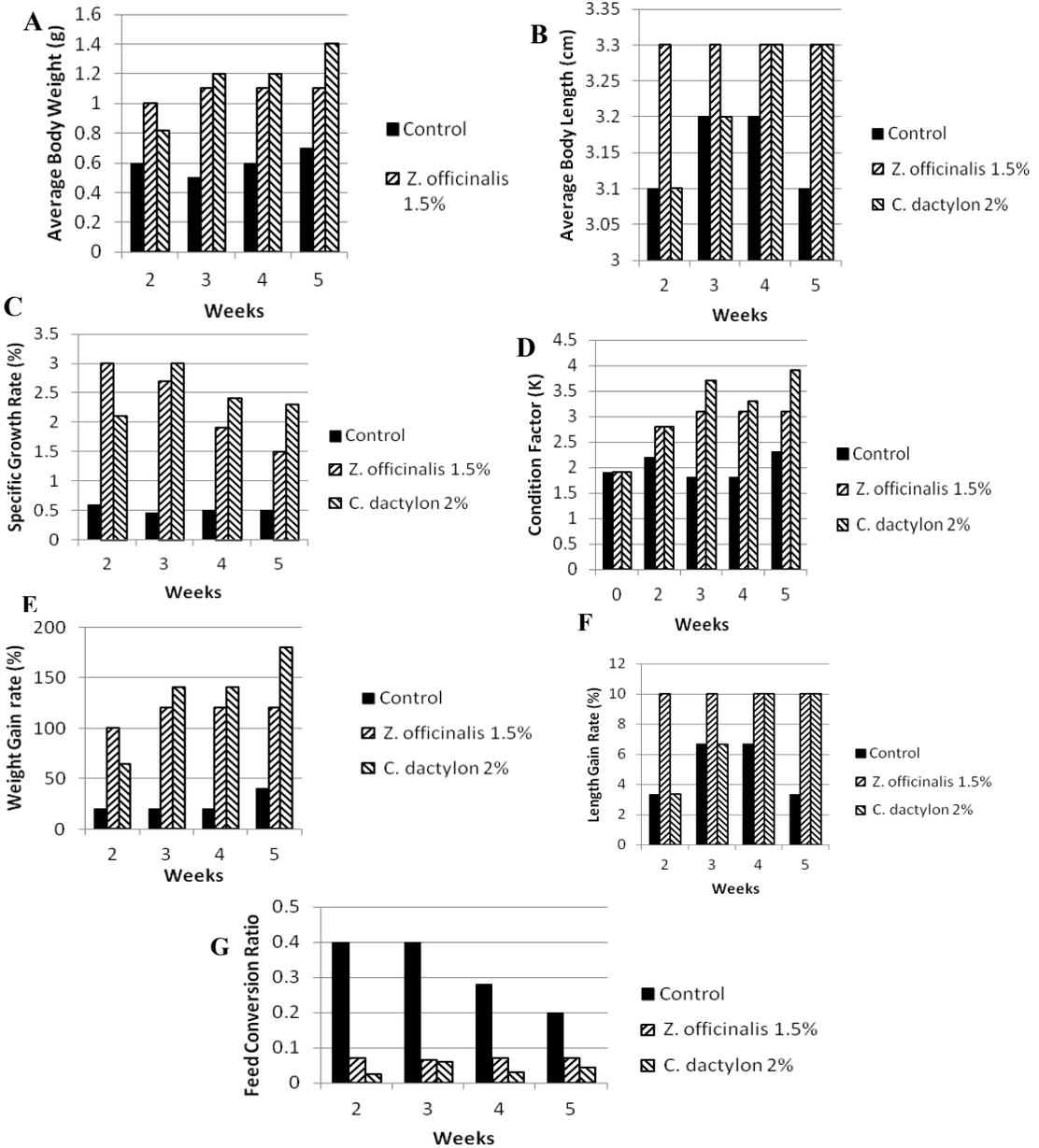


Fig 1 Effect of *Zingiber officinalis* and *Cyanodon dactylon* incorporated diets on Average body weight (A), Average body length (B), Specific growth rate (C), Condition factor (D), Weight gain rate (E), Length gain rate (F) and Feed conversion ratio (G) of stressed *M. rosenbergii* for period of 5 weeks.

3.2. Immune parameters:

THC and HV of juveniles from all groups were significantly increased compared to the control at 4 and 5 weeks from start of feeding (Table, 1).

Moreover, DHC all groups were significantly increased compared to the control at 4 and 5 weeks from start of feeding. It was clear that small granular hemocytes were the most affected cell type by *Z. officinalis* and *C. dactylon* treatment (Fig, 2A). In addition, PI and PR of all groups were significantly higher than those of control (Fig, 2B).

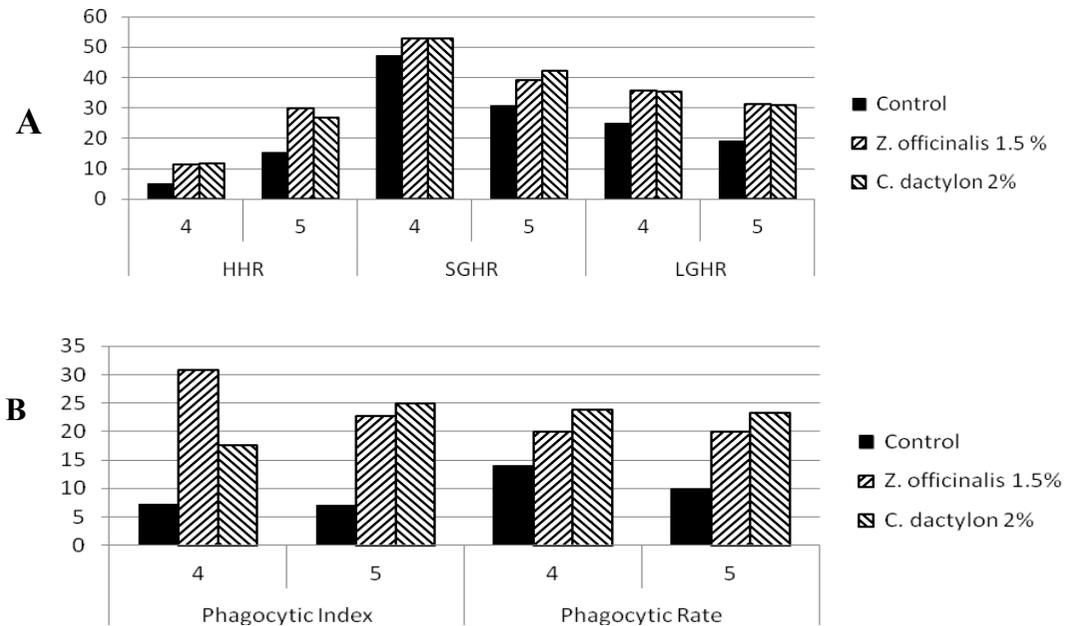


Fig 2 Effect of *Zingiber officinalis* and *Cyanodon dactylon* incorporated diets on Differential Hemocytic Count and phagocytic activity of stressed *M. rosenbergii* at week 4 and 5.

3.3. histopathological examination:

Hepatopancreas and stomach of *Z. officinalis* and *C. dactylon* fed juveniles of all groups showed similar and healthy histological structures without any detrimental effect. (plate, 1).

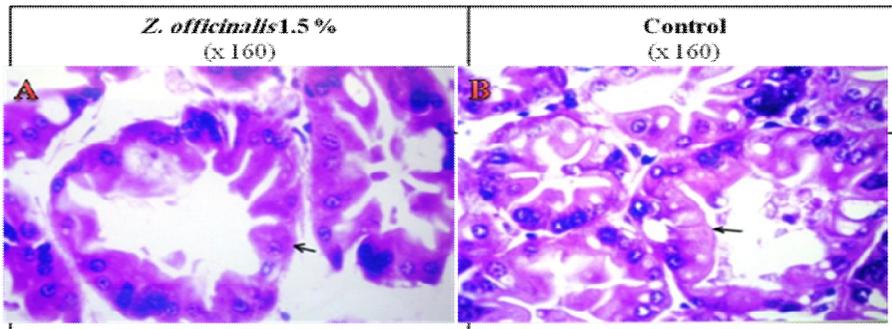


Plate 1 Hepatopancreas of *M. rosenbergii* in control (B) and group fed diets incorporated with *Z. officinalis* 1.5 % (A) at 4 weeks from start of feeding, showing normal histological structure of the tubules lined by different cells (arrow) (H&E).

4. DISCUSSION:

Recently, Several studies was directed to use Natural plants products for various activities like anti-stress, growth promotion, appetite stimulation, tonic and immunostimulation, and to have antimicrobial properties in finfish and shrimp (Citarasu *et al.*, 2001). In the present study *M. rosenbergii* juveniles efficiently tolerated the exposure to pH (9.5). The results revealed that 1.5 % *Z. officinalis* and 2 % *C. dactylon* fed juveniles tolerate high pH stress effectively and this was expressed in increasing growth performance and immune response of stressed juveniles exposed to pH (9.5) compared to those of control. On the same instance, *M. rosenbergii* post-larvae fed on *Artemia* enriched with a herbal medicinal diet tolerated stress efficiently (Citarasu *et al.*, 2002). In addition, the products Stresstol (Citarasu *et al.*, 2002; Cheng and Chen, 2000; Cheng and Jiann, 2008) and Tefroli (Bindhu *et al.*, 2007; Rani, 1999) had a good influence on shrimp post-larvae as the post-larvae fed with the above products were resistant to osmotic pressure, temperature and pH stress. This tolerability can be attributed to the ability of herbal compounds to inhibit the generation of oxygen anions and to scavenge free radicals (Bindhu *et al.*, 2007).

The present study showed that juveniles fed with 1.5 % *Z. officinalis* and 2 % *C. dactylon* had significant increase in ABW, ABL, SGR, CF, WGR, LGR and FCR compared to the control. This increase can be attributed to that herbals and spices active principles in the diets are reported to improve animal performance by stimulating secretion of the digestive enzyme that can result in improvements in digestibility, stimulating the appetite and increasing food consumption and efficiencies. In addition, they shorten the feed transit time which is more prominent in the case of *Z. officinalis*. This reduction in transit time might have a beneficial influence on digestive enzymes and could accelerate the overall digestive process (Cheng and Jiann, 2008). These results indicated that juveniles fed *Z. officinalis* and *C. dactylon* grow much better (El Desouky *et al.*, 2012) compared to previous studies that fed shrimp herbal plant enriched *artemia* (Cheng and Chen, 2000) and

herbal plant active ingredient (Chen *et al.*, 1995) and herbal plant extract (Bindhu *et al.*, 2007). This can be attributed to the active ingredients of *Z. officinalis* (Govindarajan, 1982) and, *C. dactylon* (Grzanna *et al.*, 2005; Paranjpe, 2001). Those active ingredients are suspected to enhance appetite, stimulate digestive enzymes and improve the overall digestive process. Similarly, a better growth and production were observed in cultivable fishes fed Livol (IHF-1000, which is a herbal growth promoter) and in postlarvae fed *Artemia* enriched with stresstol (Venketramalingam *et al.*, 2007). The present study showed that the survival rate among all groups was similar (100%). In a parallel studies, Postlarvae of *P. indicus* fed with the herbal products, stresstol II and stressol I enriched *Artemia* showed an increase in survival, growth and consumption (Citarasu *et al.*, 2002).

Since immunostimulants can increase non-specific immunity by promoting phagocytosis, bactericidal activity, PO activity, and respiratory bursts (Immanuel *et al.*, 2009; Shadakshari, 1993; Jayaprakas and Euphrasia, 1997; Jian and Wu, 2004; Hsieh *et al.*, 2008). Rutin extract from *Toona sinensis* administered by injection for *L. vannamei* significantly improved survival rates (Jayaprakas and Euphrasia, 1997). The current study showed that Juveniles fed with 1.5 % *Z. officinalis* and 2 % *C. dactylon* had significantly higher THC and HV compared to the control. On the same respect, The THC of *M. japonicus* that were fed with MACH at 0.2% and 0.05 showed a significant and rapid increase compared with the control group ($P < 0.05$) over the entire feeding period (Amel *et al.*, 2010). The previous increase can be attributed to accelerated maturation of haemocyte precursors in the haematopoietic tissue followed by release of new cells into the circulation (Amel *et al.*, 2010; Cheng *et al.*, 2004). In contrary, there was no significant difference in THCs between shrimp fed the zingerone diets and control shrimp (Chen *et al.*, 1995).

The present study indicated that small granular hemocytes were the most affected cell type by *Z. officinalis* and *C. dactylon* treatment. On the same instance, SGH was found to be the most predominant cell in *M. rosenbergii*, representing about 54% from total circulating hemocytes (Sequeira *et al.*, 1996). Versasly, Hyaline hemocytes were the most predominant cell in *P. japonicas* fed with MACH (Amel *et al.*, 2010) and *p. japonicas* injected with 1, 3-glucan (Cheng *et al.*, 2004). This difference can be attributed to difference in shrimp species, water salinity and plant species.

In the present study, 1.5 % *Z. officinalis* and 2 % *Cynodon dactylon* fed *M. rosenbergii* showed a significant increase in Pahogocytic index (PI) and Pahogocytic rate (PR) compared to the control. Nearly the same results were observed by several authors (Amel *et al.*, 2010; Sequeira *et al.*, 1996; Sörderhäll *et al.*, 2003).

It is very important to use histological methods to assess the effects of feed on the digestive tract of fish Božidar *et al.*, 2011; Miriam and Menon, 2005). In our study, *Z. officinalis* and *C. dactylon* fed *M. rosenbergii* juveniles showed almost similar and healthy histological structures of hepatopancreas and stomach without detrimental effect] on their structure. On the same respect, *P. semisulcatus* PL fed on mannan oligosaccharides (MOS) showed healthy histological structures, and none of the dietary levels of MOS resulted in any detrimental effect on the hepatopancreatic tissue (Genc *et al.*, 2007) .

In conclusion, the present study documents that *Z. officinalis* and *C. dactylon* increase tolerability to pH stress in *M. rosenbergii* and can be used as an appetizer, growth promotor and immunostimulant to effectively enhance growth and immunity parameters.

REFERENCES

- New, M.B. (2002).** Farming Freshwater Prawns ,A Manual for the Culture of the Giant River Prawn (*Macrobrachium rosenbergii*). FAO Fisheries Technical Paper 428, FAO, Rome.
- Fauci, A.S. (1993).** Multifactorial nature of human immunodeficiency virus: implications for therapy. *Science* (262):1011–1018.
- Citarasu T, M.M Babu., S.M.J. Punitha, K Venket Ramalingam. and M.P Marian (2001).** Control of pathogenic bacteria using herbal biomedicinal products in the larviculture system of *Penaeus monodon*. International Conference on Advanced Technologies in Fisheries and Marine Sciences, MS University, India.
- Citarasu T, R.R Sekar., MM Babu and MP Marian (2002).** Developing Artemia enriched herbal diet for producing quality larvae in *Penaeus monodon*. *Asian Fish Sci* 15:21–32.
- Sivaram V, MM Babu, T Citarasu, G Immanuel, S Murugadass and MP Marian (2004).** Growth and immune response of juvenile greasy groupers (*Epinephelus tauvina*) fed with herbal antibacterial active principle supplemented diets against *Vibrio harveyi* infections. *Aquaculture* 237:9–20.
- Chen. J.C., M.N Lin, Y.Y Ting and J.N Lin (1995).** Survival, haemolymph osmolality and tissue water of *Penaeus* juveniles acclimated to different salinity and temperature levels. *Biochem. Physiol.* 110:253-258.
- Govindarajan VS. (1982).** Ginger—chemistry, technology, and quality evaluation: part 2. *Crit Rev Food Sci Nutr*;17:189-258.
- Badri P. N. and Renu S. (2011).** *Cynodon dactylon* (L.) Pers.: A Valuable Medicinal Plant. *Research Journal of Medicinal Plant* 5 : 508-514.
- Grzanna R , L Lindmark and C Frondoza (2005).** Ginger—an herbal medicinal product with broad anti-inflammatory actions . *J Med Food.* 8(2):125-132.
- Paranjpe, P.(2001).** *Durva*. In: *Inhan Mehcinal Plants: Forgotten Healers.* 1st Edn., Chaukhamba Sanskrit Pratishthan, Delhi, pp: 75-76.
- Yueh, P. C. , C Liu. , C. Wu, C. Chiang, J. Lian and S. Hsieh (2012).** Dietary administration of zingerone to enhance growth, non-specific immune response, and resistance to *Vibrio alginolyticus* in Pacific white shrimp (*Litopenaeus vannamei*) juveniles. *Fish & Shellfish Immunology* 32 :284-290.
- Hien, T.T.T. (1999)** Effects of feeding types and feeding regism on the growth and spawning of freshwater prawn (*Macrobrachium rosenbergii*). Master Thesis. Nha Trang Fisheries University, Vietnam. 84p.
- Suheyla, K. D., A Nazh and C. Akin, (2003).** Some medicinal plants as Immuno-stimulants for fish. *J. Ethnopharmacol.* 88, 99- 106.
- Ngan F., R.S. Chang, H.D. Tabba and K.M. Smith (1988).** Isolation, purification and partial characterization of an active anti-HIV compound from the Chinese medicinal herb *Viola yedoensis* *Journal of Antiviral Research,* (10): pp. 107–116.
- Bo L. , G. Xianping, H Yanhui., X. Jun, X. Pao, H. Yijin, Z. Qunlan, P. Liangkun and C. Ruli (2010).** Effects of anthraquinones extracted from *Rheum officinale* Bail on

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- the growth, non-specific immune response of *Macrobrachium rosenbergii*. *Aquaculture* 310:13–19.
- Laird, L. M. and Needham, T. (1988):** Growth, nutrition and feeding, Salmon and trout farming, England, Ellis Horwood Limited, 202-216.
- FAO, 2002.** Food and Agriculture Organization of the United Nations.
- Chen W., C. Su-Mei, W Feng-I., H. Pei-I, L. Chun-Hung, C. Jiann-Chu, (2003).** Effects of temperature, pH, salinity and ammonia on the phagocytic activity and clearance efficiency of giant freshwater prawn *Macrobrachium rosenbergii* to *Lactococcus garvieae*. *Aquaculture*. 219, 111– 121.
- Wootton, W.C. and Pipe R.K. (2003).** Structural and functional characterization of the blood cells of bivalve mollusk, *Scrobicularia plana*. *Fish & Shellfish Immunology* 15 :249-62.
- Abdelhamed H. (2008).** Effect of some environmental conditions on growth rate, survival rate and immune status of *Macrobrachium rosenbergii*. Master Thesis. Benha University. Moshtohour.
- Itami, T., Y Takahashi, E. Tsuechihira, H. Igusa, and M Kondo, (1994).** Enhancement of disease resistance of Kuruma prawn *Penaeus japonicus* and increase in phagocytic activity of prawn hemocytes after oral administration of 1,3 glucan. The 3rd Asian Fisheries Forum. Asian Fisheries Society, Manila, Philippines, 375-378.
- Banchroft , J.D. ; A. Stevens , And D.R. Turner, (1996).** Theory and practice of histological techniques. Fourth Ed. Churchill Livingstone , New York , London , San Francisco, Tokyo.
- Citarasu T, G Immanuel and MP Marian (1998).** Effects of feeding Artemia enriched with stressol and cod liver oil on growth and stress resistance in the Indian white shrimp *Penaeus indicus* post larvae. *Asian Fish Sci* 12:65–75.
- Cheng, W. and Chen, J.C. (2000).** Effects of pH, temperature and salinity on immune parameters of the freshwater prawn *Macrobrachium rosenbergii*. *Fish Shellfish Immunol.* 10, 387–391.
- Chang-Che L. and Jiann-Chu C. (2008).** The immune response of white shrimp *Litopenaeus vannamei* and its susceptibility to *Vibrio alginolyticus* under low and high pH stress. *Fish & Shellfish Immunology* .25, 701-709.
- Bindhu V., E.V. Radhakrishnan, P. Abinash (2007).** Effect of environmental parameters on immune response of the Indian spiny lobster, *Panulirus homarus* (Linnaeus, 1758). *Fish & Shellfish Immunology*. 23, 928-936.
- Rani, T.V. (1999).** Fourth year annual report (CSIR Research Associateship) submitted to Council of Scientific and Industrial Research, New Delhi.
- Hiam El Desouky, A M El Asely, A A Shaheen, Amany A Abbass (2012).** Effects of *Zingiber officinalis* and *Cyanodon dactylon* on the growth performance and immune parameters of *Macrobrachium rosenbergii*. *World journal of fish and marine sciences*. 4 (3) :301-307.
- Venketramalingam K, JG Christopher and T Citarasu (2007).** *Zingiber officinalis* as a herbal appetizer in the tiger shrimp *Penaeus monodon* (Fabricius) larviculture. *Aquac Nutr* 13(6):439–443.
- Amel M El Asely, A A Shaheen, Amany A Abbass, R Sudhakaran, N T H Linh, T Yoshida, Y Tachikawa, S Yoshida and T Itami (2010).** Immunomodulatory effect

- of plant-mixed feed in kuruma shrimp, *Marsupenaeus japonicus*, and its protective efficacy against white spot syndrome virus infection. *Journal of Fish Diseases*.33, 859–863.
- Immanuel G, RP Uma, P Iyapparaj, T Citarasu, SM Peter and MM Babu (2009)**. Dietary medicinal plant extracts improve growth, immune activity and survival of tilapia *Oreochromis mossambicus*. *J Fish Biol*;74:1462-75.
- Shadakshari, G.S. (1993)** Effect of Bioboost forte, Livol and Amchemin AQ on Growth and Body Composition of Common Carp *Cyprinu carpio* (Linn). MFSc Thesis, University of Agricultural Sciences, Bangalore, India.
- Jayaprakas, V. and Euphrasia, J. (1997)** Growth Responses of Indian Major Carp *Cirrhinus mrigala* to Livol (IHF-1000) – an Herbal Product, Proceedings of Indian National Science Academy, Bangalore, India.
- Jian J and Wu Z (2004)**. Influences of traditional Chinese medicine on nonspecific immunity of Jian carp (*Cyprinus carpio* var. Jian). *Fish Shellfish Immunol*;16:185-91.
- Hsieh TJ, J Chyi Wang, Hu CY, Li CT, K Ching-Ming and SL Hsieh (2008)** Effects of Rutin from *Toona sinensis* on the immune and physiological responses of white shrimp (*Litopenaeus vannamei*) under *Vibrio alginolyticus* challenge. *Fish Shellfish Immunol* 25(5):581–588.
- Cheng W, CH Liu, ST Yeh and JC Chen (2004)**. The immune stimulatory effect of sodium alginate on the white shrimp *Litopenaeus vannamei* and its resistance against *Vibrio alginolyticus*. *Fish Shellfish Immunol* ;17:41-51.
- Sequeira T., D Tavares. and M. Arala-Chaves (1996)** Evidence for circulating haemocyte proliferation in the shrimp, *Penaeus japonicus*. *Developmental and Comparative Immunology* 20, 97–104.
- So"derha"ll I., E. Bangyeekhun, S. Mayo and K. So"derha"ll (2003) Haemocyte production and Rogerio G. and Margharita A. B., (1998)**. Hemocytes of the Palaemonids *Macrobrachium rosenbergii* and *M. acanthurus*, and of the Penaeid *Penaeus paulensis*. *JOURNAL OF MORPHOLOGY* 236:209–221.
- Božidar S. B. Marko, Z. Zoran and D. Vesna (2011)**. histological methods in the assessment of different feed effects on liver and intestine of fish. *Journal of Agr. Sc.*..56:87-100
- Miriam P. and Menon N. R. (2005)**. Histopathological changes in the hepatopancreas of the penaeid shrimp *Metapenaeus dobsoni* exposed to petroleum hydrocarbons. *J. Mar: Biol. Ass. India*, 47 (2) : 160 – 168.
- Genc M.A., M. Aktas, E. Genc and E. Yilmaz (2007)**. Effects of dietary mannan oligosaccharide on growth, body composition and hepatopancreas histology of *Penaeus semisulcatus* (de Haan 1844). *Aquaculture Nutrition* 13; 156–161.

Table (1) Effect of *Zingiber officinalis* and *Cyanodon dactylon* incorporated diets on Total Hemocyte Count ($10^6/\text{ml}$) and Hemocyte Viability (%) of stressed *M. rosenbergii* at 4 and 5 weeks.

Treatments	Total Hemocyte Count		Hemocyte Viability	
	4 (w _s)	5 (w _s)	4 (w _s)	5 (w _s)
Control	5.7±0.057 ^d	3.9±0.057 ^d	43.9±0.057 ^c	58.24±0.005 ^d
<i>Z. officinalis</i> 1.5%	15.9±0.057 ^a	22.2±0.057 ^a	50.0±0.577 ^b	70.28±0.005 ^c
<i>C. dactylon</i> 2%	10.2±0.057 ^c	10.2±0.057 ^c	50.6±0.057 ^b	83.67±0.005 ^a

Mean values (\pm SE) at the same exposure time with different superscript letters are significantly different at level of $P < 0.05$.

تأثير الجنزبيل والنجيله البلدى لمقاومة إجهاد الاس الهيدروجينى على جمبرى المياه العذبه

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لدراسة قدرة الجنزبيل و النجيله البلدى على مواجهة الاس الهيدروجينى كعامل ضاغط على جمبرى المياه العذبه. ثلاثه مجموعات كلا منها تتكون من 40 زريعة تمت تغذيتها على الجنزبيل بتركيز 1.5% و النجيله البلدى بتركيز 2% وتم تعريضها لاس هيدروجينى 9.5 لمده خمس اسابيع. وقد اظهرت جميع المجموعات زياد ملحوظة فى معدلات النمو والاستجابيه المناعية مقارنة بالمجموعات الحاكمه. و اوضحت الصورة الهستولوجيه للمجموعات المغذاه على كلا من الجنزبيل والنجيله البلدى عدم وجود تغيرات ملحوظه فى التركيب الهستولوجى للمعدن والهيپاتوبنكرياس.