Egyptian Journal of Aquatic Research (2014) **40**, 251–260

HOSTED BY

ELSEVIER

National Institute of Oceanography and Fisheries

Egyptian Journal of Aquatic Research

http://ees.elsevier.com/ejar
www.sciencedirect.com

FULL LENGTH ARTICLE

Effects of short term feeding of some marine microalgae on the microbial profile associated with *Dicentrarchus labrax* post larvae

Heba S. El-Sayed [a], Hassan A.H. Ibrahim [b], Ehab A. Beltagy [b], Hanan M. Khairy [c,*]

[a] *Fish Reproduction & Larval Rearing (Marine Hatchery) Laboratory, National Institute of Oceanography and Fisheries, Anfoushi, Qayet Bay, Alexandria, Egypt*
[b] *Microbiology Laboratory, National Institute of Oceanography and Fisheries, Anfoushi, Qayet Bay, Alexandria, Egypt*
[c] *Hydrobiology Laboratory, National Institute of Oceanography and Fisheries, Anfoushi, Qayet Bay, Alexandria, Egypt*

Received 13 May 2014; revised 23 August 2014; accepted 23 August 2014
Available online 25 October 2014

KEYWORDS

Marine microalgae;
Dicentrarchus labrax post larvae;
Growth;
Antimicrobial;
GC–MS

Abstract This study investigates the microbial profile and antimicrobial activity of four marine microalgae species, *Tetraselmis chuii*, *Nannochloropsis salina*, *Isochrysis galbana* and *Chlorella salina* used in aquaculture of *Dicentrarchus labrax* in the post larval stage to estimate which was the best algal species that could be used as a green water technique and achieving the maximum rate of growth and survival of *D. labrax* post larvae. The results represented a significant increase in the length and width of *D. labrax* at $p < 0.05$ recorded in the case of enrichment with *I. galbana* followed by *N. salina*, and the most weight was recorded in the case of *N. salina* as compared with the control. Significant increase in percentage of survival of *D. labrax* was recorded in the case of *C. salina* and *T. chuii* (70% and 60.1%, respectively) as compared with the control (22%). The antibacterial activity (AU) of the different microalgal ethanolic extracts against fish indicator pathogens was determined. The results indicated that the ethanolic extracts of *C. salina* and *T. chuii* have the most positive records against the fish indicator pathogens (*Escherichia coli*, *Pseudomonas aeruginosa*, *Vibrio damsela*, *Vibrio fluvialis* and *Aeromonas hydrophila*). The current study was extended to determine the GC–MS of ethanolic extract of *C. salina* and *T. chuii*. The main constituents detected in the ethanolic extract were organic acids like hexadecanoic acid, octadecanoic acid, and an acyclic diterpene alcohol like phytol.

© 2014 Hosting by Elsevier B.V. on behalf of National Institute of Oceanography and Fisheries.
Open access under CC BY-NC-ND license.

Introduction

Overuse of antibiotics, appeared to be ineffective in aquaculture and mariculture. It not only increased bacterial resistance, damaged normal microflora of the culture environment causing microdysbiosis, as double pollution; but also caused

* Corresponding author. Tel.: +20 1006048854.
E-mail address: hanan_khairy@yahoo.com (H.M. Khairy).
Peer review under responsibility of National Institute of Oceanography and Fisheries.

http://dx.doi.org/10.1016/j.ejar.2014.08.001
1687-4285 © 2014 Hosting by Elsevier B.V. on behalf of National Institute of Oceanography and Fisheries.
Open access under CC BY-NC-ND license.

antibiotics residues to accumulate in aquatic products posing risk for human consumption. Counter measures have been taken including Integrated Disease and Pest Management (Li, 2008), especially by using beneficial microorganisms. Sustainable aquaculture or mariculture requires setting up of clean culture models by Basal medium (BM) with no toxin, no side effects, no residue, and no resistance, while it is effective in improving the environment and in raising the immunity of cultured animals, in reducing diseases and in maintaining eco-equilibrium (Bao and Shen, 2005).

As a key cost-saving step, antibiotics are commonly employed in most fish aquacultures to prevent disease. However, the risk in this practice is that antibiotic-resistant pathogens may be spread out along with wastewater to cause serious environmental pollution. To address this problem, an attempt is made to develop a safer, more effective and less expensive biological bactericide for aquaculture use. Documentation is required to evaluate the use of the biocontrol system as an alternative method for inhibition of the fish pathogenic bacteria in infected fish farms (Anne-Marie et al., 2003).

Microalgae are commonly used in the rearing of marine fish larvae. They are either added directly to water in the rearing tanks, when applying the "green water" technique (Reitan et al., 1997), or used as food for rotifers. Addition of microalgae often has a positive effect on the survival rates of fish larvae (Oie et al., 1997). Fishes are susceptible to a wide variety of bacterial pathogens (Schmidt et al., 2000). Many of these bacteria capable of causing disease are considered by some to be saprophytic in nature (Toranzo et al., 2005). These bacteria only become pathogens when fishes are physiologically unbalanced, nutritionally deficient, or there are other stressors, i.e., poor water quality, overstocking, which allow opportunistic bacterial infections to proceed (Anderson, 1995).

Antimicrobial activity of microalgae cultures has been shown in earlier studies (Duff et al., 1966; Austin et al., 1992). This antimicrobial activity could be explained by the bacteria present in the microalgae cultures (Dopazo et al., 1988), or by antimicrobial substances produced by the microalgae cells (Marshall et al., 2003; Guedes et al., 2011). KoKou et al. (2012) examined the antimicrobial activity for five cultures of microalgae (*Chlorella minutissima*, *Tetraselmis chuii*, *Nannochloropsis* sp., *Arthrospira platensis* and *Isochrysis* sp.) with no culturable bacteria for testing their ability to inhibit the growth of six *Vibrio* bacterial strains (*Vibrio parahaemolyticus*, *Vibrio anguillarum*, *Vibrio splendidus*, *Vibrio scophthalmi*, *Vibrio alginolyticus* and *Vibrio lentus*.

Furthermore, recent advances in algal culture techniques have placed microalgae in a unique position over many other marine organisms, as algae can be cultured under conditions that maximize the production of the desired compound (Chisti, 2007). Thus, the investigation of marine microalgae for their antimicrobial activity is likely to find compounds that can be used in the aquaculture industry. Microalgae are known to produce compounds intracellularly and extracellular, however, as a large proportion of these compounds is not excreted but remains within the cells (Das and Pradhan, 2010; Guedes et al., 2011).

Majority of hatchery protocols in larval rearing systems aim to stop using the addition of algae in the post larval rearing water starting from the age of one month after hatching for saving algae consumption and increasing the cost of their culturing and labor. So, the aims of this study were (i) Determination of microbial profile associated with the cultures of selected microalgae species during 6 weeks of batch cultures, (ii) Evaluation the using different microalgae species as green water on the growth of enriched *Dicentrarchus labrax* post larvae during 6 weeks (iii) Determination of the antagonism effect of different microalga ethanolic extracts in vitro against some pathogenic bacterial strains and identification of the most potent ones using GC–MS.

Materials and methods

Reference bacterial strains

The indicator bacteria used during this work were; *Aeromonas hydrophila*, *Staphylococcus aureus* ATCC 6538, *Vibrio damsela*, *Vibrio fluvialis*, *Streptococcus faecalis* and *Escherichia coli* provided by the culture collection of the Microbiology laboratory, National Institute of Oceanography and Fisheries, Alexandria, Egypt.

Chemicals and media

Ingredients of media were all of analytical grade and obtained from recognized chemical suppliers (mainly Oxoid Co.). Media used for isolation and enumeration of the different bacterial groups were; Seawater agar (Zobell, 1946) for determining viable aerobic heterotrophic bacteria, Mannitol salt agar (Abo-Elela and Farag, 2004) for isolating *Staphylococcus* spp., Thiosulfate citrate bile salt sucrose agar (TCBS) (Kobayashi et al., 1963) for isolating *Vibrio* spp., m-endo-les agar medium (ISO9308/1,1990) for isolation of total coliform, mFC agar medium (ISO9308/1,1990) used for isolation of *E. coli* and m-*Enterococcus* agar medium (ISO9308/2, 1984) used for isolation of *S. faecalis*.

Counting and isolation of bacterial isolates

Serial dilutions from 10^{-2} to 10^{-6} were made using filtered sterilized seawater. A portion (0.1 ml) from each appropriately diluted sample was used to inoculate plates prepared with seawater agar for total bacterial counting. Plates of six selective media were inoculated with 0.1 ml of dilution sample for counting the different bacterial groups: TC (total coliform), EC (*E. coli*), SF (*S. faecalis*), Vibrio spp., *S. aureus* ATCC 6538 and *A. hydrophila*. Purification of bacterial colonies was obtained by streaking a single colony on agar plates of the same medium. The pure colonies obtained were transferred to fresh prepared slants. Sub-cultures were kept at 4°C.

Larvae culture

Four green water techniques plus control (clear water) were carried out using post larvae obtained from eggs derived from induced spawning of *D. labrax* broadstock kept at the marine hatchery of National Institute of Oceanography and Fisheries (NIOF), Alexandria, Egypt. After hatching of eggs, the newly hatched larvae were reared in a conventional way till the post larva stage. Once, the age reached 30 DAH (one month), the

Download English Version:

https://daneshyari.com/en/article/4493158

Download Persian Version:

https://daneshyari.com/article/4493158

Daneshyari.com