



**HAL**  
open science

## **Antioxidant activity of three microalgae *Dunaliella salina*, *Tetraselmis chuii* and *Isochrysis galbana* clone Tahiti**

Ita Widowati, Muhammad Zainuri, Hermien Pancasakti Kusumaningrum, Ragil Susilowati, Yann Hardivillier, Vincent Leignel, Nathalie Bourgougnon, Jean-Luc Mouget

### ► To cite this version:

Ita Widowati, Muhammad Zainuri, Hermien Pancasakti Kusumaningrum, Ragil Susilowati, Yann Hardivillier, et al.. Antioxidant activity of three microalgae *Dunaliella salina*, *Tetraselmis chuii* and *Isochrysis galbana* clone Tahiti. IOP Conference Series: Earth and Environmental Science, 2017, 55, pp.012067. 10.1088/1755-1315/55/1/012067. hal-02115064

**HAL Id: hal-02115064**

**<https://ubs.hal.science/hal-02115064>**

Submitted on 27 May 2024

**HAL** is a multi-disciplinary open access archive for the deposit and dissemination of scientific research documents, whether they are published or not. The documents may come from teaching and research institutions in France or abroad, or from public or private research centers.

L'archive ouverte pluridisciplinaire **HAL**, est destinée au dépôt et à la diffusion de documents scientifiques de niveau recherche, publiés ou non, émanant des établissements d'enseignement et de recherche français ou étrangers, des laboratoires publics ou privés.

PAPER • OPEN ACCESS

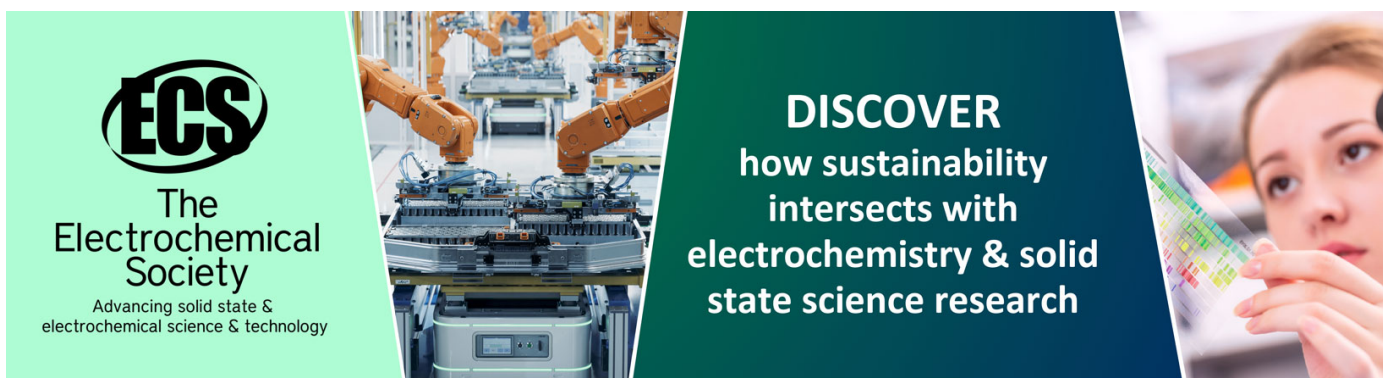
## Antioxidant activity of three microalgae *Dunaliella salina*, *Tetraselmis chuii* and *Isochrysis galbana* clone Tahiti

To cite this article: Ita Widowati *et al* 2017 *IOP Conf. Ser.: Earth Environ. Sci.* **55** 012067

View the [article online](#) for updates and enhancements.

You may also like

- [Carnegie Supernova Project: The First Homogeneous Sample of Super-Chandrasekhar-mass/2003fg-like Type Ia Supernovae](#)  
C. Ashall, J. Lu, E. Y. Hsiao *et al.*
- [PISCO: The PMAS/PPak Integral-field Supernova Hosts Compilation](#)  
L. Galbany, J. P. Anderson, S. F. Sánchez *et al.*
- [ASASSN-15hy: An Underluminous, Red 03fg-like Type Ia Supernova](#)  
J. Lu, , C. Ashall *et al.*



**ECS**  
The  
Electrochemical  
Society  
Advancing solid state &  
electrochemical science & technology

**DISCOVER**  
how sustainability  
intersects with  
electrochemistry & solid  
state science research

## Antioxidant activity of three microalgae *Dunaliella salina*, *Tetraselmis chuii* and *Isochrysis galbana* clone Tahiti

Ita Widowati<sup>1</sup>, Muhammad Zainuri<sup>1</sup>, Hermien Pancasakti Kusumaningrum<sup>2</sup>,  
Ragil Susilowati<sup>3</sup>, Yann Hardivillier<sup>4</sup>, Vincent Leignel<sup>4</sup>, Nathalie Bourgougnon<sup>5</sup>,  
Jean-Luc Mouget<sup>4</sup>

<sup>1</sup>Department of Marine Science, Faculty of Fisheries and Marine Science, Diponegoro University, Jl. Prof. Soedarto SH, Tembalang, Semarang -50275, Indonesia

<sup>2</sup>Department of Biology, Faculty of Sciences and Mathematic, Diponegoro University, Jl. Prof. Soedarto SH, Tembalang, Semarang -50275, Indonesia

<sup>3</sup>Alumny Master of Coastal Ressource Management, Faculty of Fisheries and Marine Science, Diponegoro University, Jl. Prof. Soedarto SH Tembalang, Semarang-50275, Indonesia

<sup>4</sup>Laboratoire du Mer Molécule et Santé, Université du Maine, Le Mans, France

<sup>5</sup>Laboratoire de Chimie Biologie Marine, Université de Bretagne Sud, Vannes, France

Email : ita\_jusup@yahoo.co.id

**Abstract.** Natural alternatives antioxidant source has become a trending topic in the past decades to replace synthetic antioxidant. Microalgae have been mentioned to show interesting bioactive properties and one of them is its antioxidant activity. This study aims to evaluate the potential of three microalgae *Dunaliella salina*, *Tetraselmis chuii* and *Isochrysis galbana* as new source of natural antioxidant. Proximate analysis and total phenolic content of *D. salina*, *T. chuii* and *I. galbana* were determined. Antioxidant activity of methanolic extracts of these three species prepared in different concentration (50, 100, 250, 500, and 1000 ppm) was performed through DPPH assay. *I. galbana* clone Tahiti demonstrated a highest antioxidant potential with 61.64% of inhibition at 50 ppm followed by *D. salina* with 58.45% of inhibition and *T. chuii* with 52.58% of inhibition. *I. galbana* clone Tahiti was the best antioxidant with total phenol content of 17.798 mg GAE g<sup>-1</sup> extract at 50 ppm; followed by *T. chuii* 16.868 mg GAE g<sup>-1</sup> extract and the lowest was *D. salina* with 4.672 mg GAE g<sup>-1</sup> extract. Results suggest that these microalgae possess antioxidant potential which could be considered for future applications in medicine, dietary supplements, cosmetics or food industries.

**Keywords:** antioxidant activity, DPPH, microalgae, total phenol.

### 1. Introduction

Indonesia, as an archipelagic state surrounded by ocean, presents a potential as a microalgae source because of its huge natural biodiversity. Microalgae are distributed everywhere: in salt, brackish or freshwater, in tropical to cold regions and sometimes as symbionts with other organisms, and some species are used and cultivated by farmers.



Microalgae, also known as phytoplankton, are common name of all the aquatic autotrophic organisms living in suspension in the water column. This name refers to several phyla, mostly eukaryotes but including the photosynthetic prokaryotes known as cyanobacteria. As photosynthetic organisms, this group plays an important role in the productivity of oceans and constitutes the basis of the marine food chain. Moreover, phytoplankton is responsible for half of the oxygen released in the atmosphere.

In the last decades, an increased attention has been paid to the commercial and industrial potential of microalgae. Several species are currently being studied for their ability to synthesize valuable secondary metabolites (pigments, lipids, carotenoids, etc) for biofuel production, pharmaceutical industry or aquaculture applications [1]. Other fields of investigation include nanotechnologies, environmental survey, forensic sciences and paleontology. Similarly, some micro-algae contain and/or excrete pharmacologically active compounds. For example, the dinoflagellates *Gymnodinium* sp. and *Gonyaulax* sp. produce an alkylguanidine compound that affects the central nervous system. Brominated bi-indoles of *Rivularia firma* show pharmacological activity.

Among different compounds with functional properties, antioxidants are the most widely studied since the interest in finding new, safe and powerful antioxidant from natural sources is growing nowadays. Moreover, consumers have demonstrated the important role of antioxidants in human health thus increasing the interest in such products and their demand. Most of commercially available natural antioxidants are extracted from terrestrial plants, such as cocoa, tea grape and rosemary [2]. It appears that unicellular microalgae shows to have promising potential as alternative source of antioxidants [3-5]. Microalgae have been considered as important source of bioactive compounds since they are sustainable resources easily to culture that do not require an arable land unlike the terrestrial plant. Some studies have reported the interesting and remarkable antioxidant potential of microalgae [6-8]. Additionally, microalgae species like *Dunaliella*, *Chlorella*, *Isochrysis*, *Nannochloropsis*, *Nannochloris*, *Chlamydomonas*, *Haematococcus* and *Spirulina* are available to grow in mass production [1].

In regard of these biotechnological challenges, there is a constant effort actually provided for both finding and exploiting new microalgal resources and developing their putative commercial outcomes or industrial valorizations. Thus, this research aims to characterize microalgae of interest for aquaculture for their potential biological activities, especially in antioxidant properties.

## 2. Material and Methods

### 2.1. Culture of microalgae

Three species of microalgae, *Dunaliella salina*, *Tetraselmis chuii*, and *Isochrysis galbana* clone Tahiti, were cultured in a batch culture with ratio of algae and seawater was 1:3 (volume). Walne medium was used as nutritional compound for the culture, with ratio of 1:1 (mL L<sup>-1</sup>); with luminous intensity of 1500–3000 lux and temperature of 23-25°C [9].

The density of the microalgae culture was measured daily with haemocytometer by using binocular microscope. The microalgae were homogenized before the measurement. The cell density was calculated by following formula:

$$N = \frac{(N_1 + N_2)}{2} \times \frac{1}{1.02 \text{ mm}^2 \times 0.1 \text{ mm}} \times \frac{1 \text{ mm}^2}{10^{-3} \text{ ml}}$$

Where N = cell density (cell.mL<sup>-1</sup>), N<sub>1</sub> = total cell in 80 small square (replica 1), N<sub>2</sub> = Total cell in 80 small square (replica 2), 0.2 mm = wide of haemocytometer in 80 square, and 0.1 mm = the depth liquid on haemocytometer.

Microalgae were harvested on the stationary phase of culture. The biomass of microalgae was obtained with centrifugation at 5.000 rpm for 10 min and then left to dry for a few days to obtain the dry biomass [10].

## 2.2. Extraction

Dry biomass was extracted by using methanol solvent with sonication at 50 Hertz for 15 min. The solvents were evaporated by using rotary evaporation [11].

## 2.3. Antioxidant activity

Antioxidant activity study was divided into 2 steps: first was to identify and drying the specimen. Second step was the extraction of dry specimen and bioactive compound analysis including alkaloid, triterpenoid/steroid, saponin, phenol, flavonoid, tannin [12]. The total phenol test was performed according to [13] and antioxidant activity test with DPPH method [14].

## 2.4. DPPH antioxidant activity

DPPH antioxidant activity of methanol extract from *D. salina*, *T. chuii*, and *I. galbana* clone Tahiti were determined according to [15]. The assay was performed at 50, 100, 250, 500, and 1000 ppm. Extract solution (1 mL) (in triplicates) was mixed with 3 mL of 60  $\mu$ M methanolic solution of DPPH radicals. The mixture was put in the dark for 30 min before the absorbance was taken at 517 nm. The percent inhibition was calculated using the formula:

$$\% \text{ of inhibition} = \left\{ \left( \frac{A - B}{A} \right) \times 100 \right\}$$

Where, A = absorbance of the control (DPPH) and B = absorbance of test sample.

## 2.5. Determination of total phenol content

Total phenol content of methanol extract from *D. salina*, *T. chuii*, and *I. galbana* clone Tahiti were determined according to the Folin-Ciocalteu method [16]. 300  $\mu$ L of extract was dispensed into test tube (in triplicates) and it was added with 1.5 mL of Folin-Ciocalteu reagent (diluted 10 times with distilled water) and a 1.2 mL of Na<sub>2</sub>CO<sub>3</sub> solution (7.5 % w/v). The solution was mixed the put 30 min at room temperature before the absorbance at 765 nm. Total Plate Counted was measured in mg g<sup>-1</sup> Gallic Acid Equivalent (GAE). The calibration equation for Gallic Acid was Y = 0.0645x - 0.0034.

## 2.6. Phytochemical content

Methanol extracts prepared from all the three different microalgae were used to screen various phytochemicals such as tannins, flavonoids, steroids, saponins, and alkaloids [17].

### 2.6.1. Flavonoids

The algal extract was diluted and taken some of the water, and put into a test tube. Mg was then added and three drops of HCl into a test tube. Amyl alcohol then added and beat until separated. The reaction was positive if it forms a reddish yellow color to red.

### 2.6.2. Quinones

Five mL of extract obtained from the experiments to extract flavonoids identification was inserted into a test tube, a few drops of a solution of NaOH 1 N then added. The formation of the red color indicates the class of quinone compound.

### 2.6.3. Tannins

Algal extracts added to 2-3 drops of 1% FeCl<sub>3</sub> solution. Extracts will be positive if it contains tannins greenish-black or dark blue.

### 2.6.4. Alkaloids

Crude extract was added with 0.5 HCL 2% and was divided into two tubes. The first tube coupled with 2-3 drops of reagent Dragendorff and the second tube was added with 2-3 drops of reagent Mayer. When

an orange precipitation was formed in the first tube and yellowish precipitation was shown by second tube, it indicated the presence of alkaloid.

### 2.6.5. Saponins

Extract was introduced into a test tube and added with distilled water that has been warmed. It was shaken and allowed to stand for 10 min. Samples showed positive results when the foam was formed and did not disappear until 15 min after the drops of HCl.

### 2.6.6. Terpenoids and Steroids

The extract was added with three drops acetic anhydride and one drop of sulfuric acid (H<sub>2</sub>SO<sub>4</sub>). The red color indicates terpenoids, while blue color indicates steroid. Samples showed positive results when the foam was formed and did not disappear until 15 min after the addition of HCl.

## 3. Results and Discussion

The activity of antioxidant of methanol extracts of three microalgae *D. salina*, *T. chuii*, and *I. galbana* clone Tahiti were studied in this research at different concentrations (1000, 500, 250, 100, 50 ppm). Results showed that *I. galbana* clone Tahiti at 250 ppm showed the best result than other concentration with 67.93 % inhibition. *D. salina* gave the best result of inhibition of 62.19 % at 500 ppm. While *T. chuii* showed the best result with 71.36 % of inhibition at 1000 ppm (Table 1).

**Table 1.** DPPH Radical Scavenging Assay from methanolic extracts of three microalgae *D. salina*, *T. chuii*, and *I. galbana* clone Tahiti at different concentrations (1000, 500, 250, 100, 50 ppm).

Microalgae	Concentration (ppm)				
	1000	500	250	100	50
<i>D. salina</i>	53.17	62.19	58.43	59.01	58.45
<i>T. chuii</i>	71.36	66.18	52.38	42.19	52.58
<i>I. galbana</i> clone Tahiti	53.77	55.87	67.93	64.02	61.64

Note: The results in % inhibition DPPH.

**Table 2.** Total Phenol Content from methanolic extracts of three microalgae *D. salina*, *T. chuii*, and *I. galbana* clone Tahiti at 4 concentrations (1000, 500, 250, 100, 50 ppm).

Microalgae	Concentration (ppm)				
	1000	500	250	100	50
<i>D. salina</i>	0.539	0.839	1.223	2.336	4.672
<i>T. chuii</i>	0.466	1.015	2.918	8.95	16.868
<i>I. galbana</i> clone Tahiti	0.678	1.408	3.167	8.02	17.798

Note : The results in mg GAE g<sup>-1</sup> extract

Phytochemical screening using methanol of three marine microalgae strains was evaluated. The phytochemicals present in the microalgae strains were identified as flavonoids, tannin, alkaloids, saponin and steroids; while quinon was absent (Table 3).

**Table 3.** Screening of phytochemicals of methanol extracts of three microalgae

Microalgae	Flavonoid	Tannin	Alkaloid	Saponin	Steroid	Quinon
<i>D. salina</i>	+	+	+	+	+	-
<i>T. chuii</i>	+	+	+	+	+	-
<i>I. galbana</i> clone Tahiti	+	+	+	+	+	-

The use of methanol as solvent on extract of *D. salina* and *T. chuii* has been studied before [18]. Furthermore, on the screening on antioxidant and phytochemical contents of different solvent extracts (acetone, methanol, ethanol, chloroform) of five different microalgal strains including *Tetraselmis sp.*, *Dunaliella sp.*; showed that the maximum antioxidant activity were found in acetone extracts, followed by methanol extracts of *Tetraselmis sp.* and *Dunaliella sp.*[18]. The study on screening on nine Moroccan microalgae by using the ethanolic extracts showed that the antioxidant activity of *Dunaliella sp.* (IC<sub>50</sub> 283 ± 0.09 µg mL<sup>-1</sup>), *Tetraselmis sp.* (IC<sub>50</sub> 247 ± 0.01 µg mL<sup>-1</sup>) were highest antioxidant potentials among nine microalgae studied. *D. salina* and *T. chuii* were two potentials microalgae on antioxidant activities and DPPH was the simple method to elucidate the activity by using different solvents [4].

The total phenolic content of methanolic extracts of *D. salina*, *T. chuii*, and *I. galbana* clone Tahiti was evaluated using the Folin–Ciocalteu method. The phenolic content varied from 4.672 to 17.798 mg GAE g<sup>-1</sup> extract at concentration of 50 ppm. Among five concentrations tested (1000, 500, 250, 100 and 50 ppm), 50 ppm gave the best result. The highest phenolic content was found on *Isochrysis galbana* clone Tahiti (17.798 mg GAE g<sup>-1</sup> extract) followed by *Tetraselmis chuii* 16.868 mg. g<sup>-1</sup> GAE and the lowest was found in *Dunaliella salina* (4.672 mg GAE g<sup>-1</sup> extract) (Table 2).

The result of this study on *I. galbana* clone Tahiti was higher than those found by [4] by using the ethanolic extracts of *Isochrysis sp.* (13.4 ± 0.16 mg GAE g<sup>-1</sup> extract) and by [2] on extracts of *Isochrysis sp.* (4.6 mg GAE g<sup>-1</sup> extract). The total phenol content of methanolic extract of *Tetraselmis sp.* in this study (16.868 mg. g<sup>-1</sup> GAE) was higher than showed by [2] (3.8 mg GAE g<sup>-1</sup> extract); meanwhile the result of this study was lower than those showed by [4] using the ethanolic extracts (25.5±1.5 mg GAE g<sup>-1</sup> extract). In addition, total phenol content of methanolic extract of *D. salina* of this study (4.672 mg GAE g<sup>-1</sup> extract) was also lower than those showed by [4] by using the ethanolic extracts (14.0 ± 0.43 mg GAE g<sup>-1</sup> extract). It is suggested that the different results obtained between the present study and that of by [4] was due to the type of solvent. Indeed, different solvent used will generate different content of bioactive compound. Methanol is commonly used to extract bioactive compound from any organisms as this solvent dissolves various polar compounds and certain non-polar as well. Phenolic compounds are often extracted using polar solvent yet the most suitable solvents are aqueous mixtures containing ethanol, methanol, acetone and ethyl acetate [19][20]. In general, methanol is favourable for lower molecular weight phenolic compounds [21].

## 5. Conclusion

The methanolic extracts of three-tested microalgae *I. galbana* clone Tahiti, *T. chuii* and *D. salina* showed high antioxidant activities. The higher antioxidants potentials were obtained in *Dunaliella sp.*, *Tetraselmis chuii*. These microalgae possess antioxidant potential, which could be considered for future applications in medicine, dietary supplements, cosmetics or food industries. In the future however it is crucial to perform characterization of phenolic compounds from *D. salina*, *T. chuii*, and *I. galbana* clone Tahiti.

## Acknowledgement

The authors wish to thank the Ministry of Research Technology and Higher Education, for funding this research in accordance with the Implementation Agreement Assignment Research Collaboration and International Publication, No. DIPA: -023.04.1.673453/2015, Date of 14 November 2014, DIPA Revision 01, Date of 29 February 2015.

## References

- [1] Talero E, García-Mauriño S, Ávila-Román J, Rodríguez-Luna A, Alcaide A, Motilva V. 2015. Bioactive Compounds Isolated from Microalgae in Chronic Inflammation and Cancer. *Mar Drugs* 13(10):6152–6209.
- [2] Goiris K, Muylaert K, Fraeye I, Foubert I, De Brabanter J, De Cooman L. Antioxidant Potential of

- Microalgae in Relation to Their Phenolic and Carotenoid Content. *J Appl Phycol* [Internet]. 2012 Dec 22 [cited 2017 Jan 26];24(6):1477–1486. Available from: <http://link.springer.com/10.1007/s10811-012-9804-6>
- [3] Shanab SMM, Mostafa SSM, Shalaby EA, Mahmoud GI. Aqueous Extracts of Microalgae Exhibit Antioxidant and Anticancer Activities. *Asian Pac J Trop Biomed* [Internet]. 2012 Aug [cited 2017 Jan 26];2(8):608–615. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/23569980>
- [4] Maadane A, Merghoub N, Ainane T, El Arroussi H, Benhima R, Amzazi S, et al. Antioxidant activity of some Moroccan marine microalgae: Pufa profiles, carotenoids and phenolic content. *J Biotechnol* [Internet]. 2015 Dec 10 [cited 2017 Jan 26];215:13–9. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/26113214>
- [5] Lee S-H, Lee J-B, Lee K-W, Jeon Y-J. Antioxidant Properties of Tidal Pool Microalgae, *Halochlorococcum porphyrae* and *Oltamanssiellopsis unicellularis* from Jeju Island, Korea. *ALGAE* [Internet]. 2010 Mar 1 [cited 2017 Jan 26];25(1):45–56. Available from: <http://koreascience.or.kr/journal/view.jsp?kj=JORHBK&py=2010&vnc=v25n1&sp=45>
- [6] Elsayed H, Ali A, Mahmoud S, Shanab M, Aly M, Abo-State M, et al. Screening of Microalgae for Antioxidant Activities, Carotenoids and Phenolic Contents.
- [7] Guedes AC, Gião MS, Seabra R, Ferreira ACS, Tamagnini P, Moradas-Ferreira P, et al. Evaluation of the Antioxidant Activity of Cell Extracts from Microalgae. 2013. *Mar Drugs*. 11(4):1256–1270.
- [8] Ahmed F, Fanning K, Netzel M, Turner W, Li Y, Schenk PM. Profiling Of Carotenoids and Antioxidant Capacity of Microalgae from Subtropical Coastal and Brackish Waters. *Food Chem* [Internet]. 2014 Dec 15 [cited 2017 Jan 26];165:300–6. Available from: <http://www.ncbi.nlm.nih.gov/pubmed/25038679>
- [9] Andersen R. 2005. *Algal Culturing Techniques*, 1<sup>st</sup> Edition. Academic Press. 596 pp.
- [10] Mishra, S. K, A Shrivastav, R Maurya, K Shailesh, Patidar S, Haldar S. Mishra. 2012. Effect of Light Quality on the c-phycoerythrin Production in Marine Cyanobacteria *pseudanabaena sp.* Isolated from Gujarat Coast, India. *Protein Expression and Purification* (80): 234-238. doi: 10.1016/j.pep.2011.08.011.
- [11] Trianto, A, Hermawan, I, Suzuka, T, Tanaka, J. 2011. Two New Cytotoxic Candidaspongiolides from an Indonesian Sponge. *Int'l Scholarly Res Network*. doi: 10.5402/2011/852619.
- [12] Harborne, J.B. 1973. *Phytochemical Methods*. London. Chapman and Hall, Ltd. pp. 49-188.
- [13] Yangthong M, Hutadilok-Towatana N, Phromkunthong W. 2009. Antioxidant Activities of Four Edible Seaweeds from The Southern Coast of Thailand. *Plant Foods Hum Nutr*. 2009 Sep;64(3):218-23. doi: 10.1007/s11130-009-0127-y.
- [14] Aranda RS, Lopez LAP, Arroyo JL, Garza BAA, Torres NW. 2009. Antimicrobial and Antioxidant Activities of Plants from Northeast of Mexico. *Evidence-Based Complementary and Alternative Medicine* 20(11): 1-6.
- [15] Saranya, C, Hemalatha, A, Parthiban, C, Anantharaman, P. 2014. Evaluation of Antioxidant Properties, Total Phenolic and Carotenoid Content of *Chaetoceros calcitrans*, *Chlorella salina* and *Isochrysis galbana*. *Int. J. Curr. Microbiol. App. Sci*. 3 (8): 365-377.
- [16] Irondi AE, Oboh G, Akintunde JK. 2012. Comparative And Synergistic Antioxidant Properties Of Carica Papaya And Azadarichta Indica Leaves. *Int J Phar Sci Res*. Vol. 3 Issue 12.
- [17] Sanjeet K, Kabi M, Kumari M. 2010. Study on phytochemicals analysis from leaves of *Bixaorellana*. *Emerging Science* 2:5.
- [18] Rajendran N, K Selvan B, S Piriya P, V Logeswari, Kathiresan E. Tamilselvi A, J.Vennison S 2014. Phytochemicals, Antimicrobial and Antioxidant Screening from Five Different Marine Microalgae. *J Chem and Pharmaceut Scie.* : 78 - 85.
- [19] Do QD, Angkawijaya AE, Tran-Nguyen PL, Huynh LH, Soetaredjo FE, Ismadji S, et al. 2014. Effect of Extraction Solvent on Total Phenol Content, Total Flavonoid Content, and Antioxidant Activity of *Limnophila aromatica*. *J Food Drug Anal* 22(3):296–302.
- [20] Hadiyanto H. , Sutanto A A, Suharto Y. 2014 Ultrasound assisted extraction of antioxidant from *Coleus tuberosus* peels. *Carpathian Journal of Food Science and Technology*.6(1),58-65
- [21] Dai J, Mumper RJ. 2010. Plant Phenolics: Extraction, Analysis and Their Antioxidant and Anticancer Properties. *Molecules* 15(10):7313–7352.