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Detection of some Active Phenolic Compounds in Two Local Microalgae Species *Chlorella Sorokiniana* Shihira & R.W.Krauss and *Coelastrella Saipanensis* N.Hanagata

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ABSTRACT

The present study aimed to characterise some active phenolic compounds in the alcoholic extract of two types of local microalgae, *Chlorella sorokiniana* Shihira & R.W.Krauss and *Coelastrella saipanensis* N.Hanagata These compounds were detected using HPLC device. The results showed that there were four types of active phenolic compounds, which were Gallic acid, Catechine, Ferulic acid and Hydrobenzoic acid. The highest concentration of all these compounds found in *C. saipanensis*. Concerning the compounds, the highest value was for Gallic acid and it was 45. 85 ppm, while the lowest value was for Hydrobenzoic acid and it was 14. 28 ppm in *Chlorella sorokiniana*. It is that these compounds have vital activity in inhibiting different types of pathogenic bacteria and fungi as shown in many previous studies. From this study, we conclude the ability to produce these active compounds from both algae under natural conditions of growth.

Keywords: *Coelastrella saipanensis*, *chlorella sorokiniana*, Gallic acid, Catechine, Ferulic acid, Hydrobenzoic acid

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INTRODUCTION

Microalgae have garnered significant attention increasing attention because they are an environmentally friendly treatment for getting antibiotics [1]. These algae are among the most versatile autotrophic unicellular organisms that predominantly rely on photosynthesis [2]. The active bioactive compounds having diverse functional properties found in algae such as chlorella can work as antioxidant supplements against free radicals that cause oxidative stress in different organisms [3]. *Chlorella* sp. is considered a potential source of natural bioactive compounds for use the food and pharmaceutical industries [4]. Regarding to *coelastrella saipanensis*, is one of the microalgae that is nominated as a source for producing phenolic compounds with different and diverse applications. This, in turn, opens new possibilities for industries looking for natural and sustainable alternatives [5]. The alga *Coelastrell asaipanensis* has been nominated for producing active compounds that exhibit anti-cancerous tumor activity [6]. Searching for natural alternatives to synthetic antioxidants has led to extensive studies of phenolic contents and their relationship with antioxidant activities, knowing that total phenolic content plays a direct role in total antioxidant activity [7]. In addition to producing active compounds, *Coelastrell asaipanensis* was found to have the ability to produce photosynthetic pigments such as chlorophyll a and b and carotene [8].

Gallic acid it is one of the most promising phenolic compounds. It has great potential in the field of food preservation besides its antibacterial and antifungal properties [9]. This compound is one of the phenolic acids with strong antioxidant activity and it is a potential dietary supplement because of its health benefits for various functional disorders associated with oxidative stress, including diseases of the liver, kidney, heart and nervous system [10].

Catechine is also considered one of the phenolic compounds with notable antioxidant properties for its role in removing free radicals and its anti-carcinogenic. Its anti-carcinogenic potential is associated with its ability to induce apoptosis by increasing the expression of pro-apoptotic genes [11]. Catechine has been proven to have a role in treating and preventing diseases. Its role in treating diseases can be attributed to its antioxidant and anti-inflammatory properties as it possesses a chemopreventive effect [12]. The third one is ferulic acid which is also a widespread phenolic compound either in free form or combined with carbohydrates, proteins and lignin in plant cell walls. Ferulic acid exhibits various biological activities such as anti-inflammatory, hepatoprotective and antiviral [13]. Previous studies revealed that ferulic acid has protective effects against metabolic diseases such as diabetes, neuropathy, nephropathy, obesity and hypertension and it regulates the activity of inflammatory cytokines [14]. This compound has attracted great interest from researchers, as it can be considered as a biomolecule with strong prospects as a nutritional and functional ingredient [13].

Finally, hydrobenzoic acid is one of the aromatic phenolic compounds having a high chemical stability. It is widely used in the food, pharmaceutical and cosmetic industries [16]. Hydrobenzoic acid acts as an antifungal agent. It has been found that this acid has a direct role in inhibiting the growth of *Aspargillus flates* that infect kiwi, produce aflatoxins and cause serious metastatic diseases. This compound prevents the cytoplasmic division of the fungal cell and inhibits the biosynthesis of B1 and B2 which are a serious threat to human health. Treatments with hydrobenzoic acid reduced the fungus growth by 68% [17].

Production of the thioflavin T is a compound flavonoid, which was first discovered in *C. saipanensis*. The present study aimed to characterise some active phenolic compounds in the alcoholic extract of two types of local microalgae, *Chlorella sorokiniana* and *C. saipanensis* [18].

MATERIAL AND METHODS

Pure samples of *Chlorella sorokiniana* Shihira & R.W.Krauss and *C. saipanensis* N.Hanagata were obtained from the Faculty of Science, University of Baghdad. In the room growth of the Textile Agriculture Laboratory, the samples were kept at 25±2°C under an 8/16-hour light-dark cycle with a light intensity of 3000 lux.

The Preparation of Media Culture

After sterilizing by the autoclave, Bg-11 culture medium was prepared. The algae were grown under intensive conditions (118°C and 121 bar) To obtain the required amount of extraction.

The Preparation of the Moss Extract

The algal alcoholic extract was prepared by placing 1g of dried powder of *Chlorella sorokiniana* and *C. saipanensis* in the thimble of the Soxhlet apparatus with 150 cm^3 of ethanol solvent in a conical flask of 250 cm^3. The device was connected to a condenser and the process was carried out for 6-8 hs with 7 cycles per sample [19].

The Assessment of Compounds in HPLC

Quantification of distinct phenolic compounds was performed using reversed-phase high-performance liquid chromatography (HPLC) analysis, employing a SYKAMN HPLC chromatographic apparatus that includes a UV detection system, Chemstation software, and a Zorbax Eclipse Plus-C18-OSD column measuring 25 cm in length and 4. 6 mm in diameter. The operational temperature of the column was maintained at 30 ºC, and the gradient elution technique was executed, utilizing eluent A (methanol) and eluent B (1% formic acid in an aqueous solution, v/v), as delineated: initial period of 0-4 minutes at 40% B; 4-10 minutes at 50% B; with a flow rate established at 0. 7 ml/min. Samples and standard injection volumes were 100 µL, and this procedure was executed automatically via an autosampler. The spectral data were acquired at a wavelength of 280 nm [20].

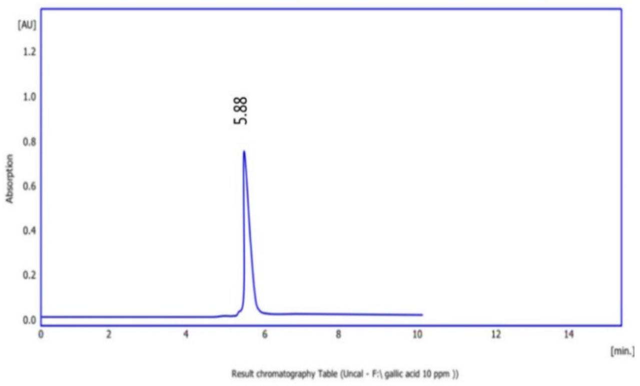

Fig. 1 Standard curve of gallic acid

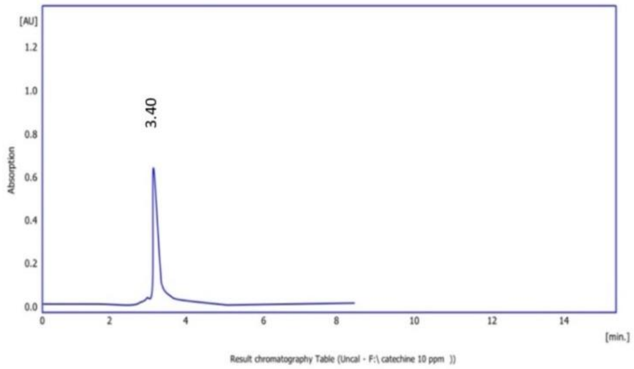

Fig. 2 Standard curve of catechine

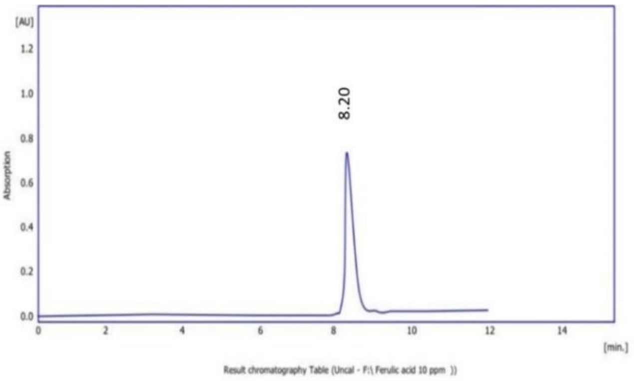

Fig. 3 Standard curve of ferulic acid

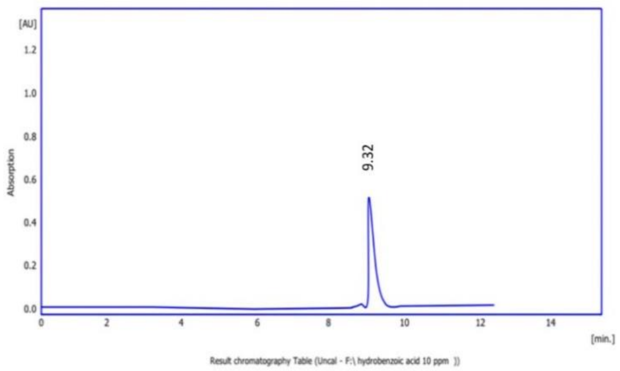

Fig. 4 Standard curve of hydroxybenzoic acid

RESULTS AND DISCUSSION

Results shown in Table (1) Shows that the highest values of phenolic compounds represented by Gallic acid, Catechine, Ferulic acid and Hydrobenzoic acid in the ethanolic extract of *C. saipanensis* were 45.85, 33.69, 28.49 and 22. 10 ppm respectively. In contrast, The values of these compounds were lower in *Chlorella sorokiniana*; they were 30.57, 21.59, 18.58 and 14.28 ppm respectively. These compounds appear within the results explained by measurements of HPLC as shown in Figures (1) and (2).

Scientific research has shown that the concentration of phenolic compounds can vary greatly between different genes and even within species [21]. The difference between species in the same location and identical environmental conditions leads to a tendency towards similar biochemical characteristics [22]. Primary metabolism processes are considered an important source of raw materials for the synthesis of secondary metabolites [23]. Also, biomass, cell number, carbohydrates and proteins may influence the phenolics production. [6] reported that these characteristics increased in *C. saipanensis* compared to *Chlorella* sp and the productivity of the alga varies with volume-to-surface area (V/S) ratios. Microalgae convert light energy via photosynthesis to synthesise many primary metabolites such as polysaccharides, proteins and lipids which in turn are converted into secondary metabolites [24].

[25] found that the concentrations of carbohydrates, proteins, and total chlorophyll increased in C. *saipanensis* when compared to *Chlorella* sp. It has been emphasized that high-value secondary metabolites can be produced from algal biomass and *Coelastrella* sp is considered an important pillar in this regard because it has a high biomass and lipid productivity [26]. Examining four strains of *Coelastrella*, it was found that they had high potential for growth under laboratory conditions besides being promising strains for producing flavonoid compounds due to their flavonoid content which reached 84. 3 mg/g of the dry weight [27].

CONCLUSION

We conclude from this study that microalgae are an important source for the production of bioactive compounds. that have medicinal and pharmaceutical value. They represent sustainable alternatives to various plant species of nutritional and economic value. Accordingly, further research is needed to enhance the concentration of active compounds within algal cells and to optimize methods for their pure extraction.

Table 1 The effect of genus differences in the production of active compounds

Name (ppm)	*Chlorella sorokiniana*	*C. saipanensis*
Gallic acid	30.57	45.85
Catechine	21.59	33.69
Ferulic acid	18.58	28.49
Hydrobenzoic acid	14.28	22.10

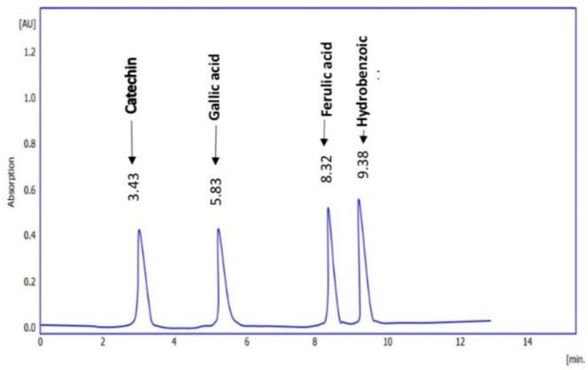

Fig. 5 Curve compound in *Chlorella sorokiniana*

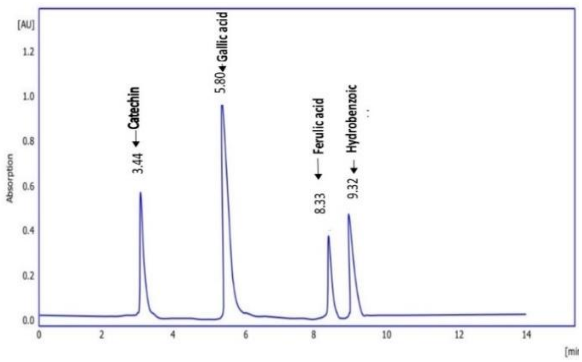

Fig. 6 Curve compoend in *C. saipanensis*

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