Evaluation of Anticancer Activity of Chlorella Vulgaris against Human Breast Adenocarcinoma Cell Line (MCF7)

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Abstract

Cancer is an increasing trend and effective therapies are inadequate to approach these malignancies. Normally rapidly dividing cells are controlled by anti-cancer drugs, but the normal cells are also affected and pattern in which is determined the side effects. A great number of antitumor compounds are natural products or their derivatives, mainly produced by blue-green algae. Chlorella vulgaris was shown to have chemo preventive effect in induced liver cancer and breast cancer rats and possess a glycoprotein with antitumor effects. In our present study shows that the phytochemicals of Chlorella vulgaris , may be responsible for displaying anticancer activities.

KEYWORDS - Chlorella vulgaris, MCF7, Cancer

1.INTRODUCTION

Malignant tumor incidences are in increasing trend and effective therapies are inadequate to approach these malignancies. Normally rapidly dividing cells are controlled by anti-cancer drugs, but the normal cells are also affected and pattern in which is determined the side effects. The behavior in which the disparate cells are concerned determines the side effects of the all by one lonesome drug. These side effects may be minimized by improving and new remedial preparations. These drugs could be of ethno botanical origin. Auspiciously numerous preceding readings have shown that the anticancer activities of non-toxic biological macromolecules are higher than conventional chemotherapy drugs. Marine algae is obliged as significant sources of natural bioactive substances and there has now emerged a new proclivity towards isolating and identifying such compounds and constituents from algae. This review article has poised studies about algal anticancer agents (Nadia et al,2014).

In 2016, Mohdet al, determine the anticancer activity of Chlorella vulgaris against breast cancer cell lines. The tests were carried out by means of MTTassay (a colorimetric assay) to determine cell viability by assessing the values of IC_{50}.The result showed that the MCF7 was the cancer cell lines that reach 50% of inhibition by algae. The extraction by chloroform of Chlorellavulgaris indicates that there was 50% inhibition (IC50) of cells on MCF7 much higher compare to the ethanol extraction of both spirulina and chlorella with at89 µg/ml of extract. In recently Jayashree et al (2016) also observed the anticancer activity against human breast cancer cell lines. They were observed the anticancer activity by MTT assay and 50% of cell death with 28 µg/ml of extract. Therefore, the present study is to investigate Chlorella vulgaris

(C. vulgaris), a kind of freshwater green microalgae, which evaluates the anticancer activity against MCF7.

II. MATERIALS AND METHODS

The sample microalgae species Chlorella vulgaris sp. was obtained from Royal Research Centre, Chennai. The algae was cultured on using Bold Basal Medium(BBM).The cultures were grown at 24±1°C in a thermostatically controlled room with cool white fluorescent lamps at 23weeks.After incubation algal growth was measured by using UV-VIS spectrophotometer at 680nm. Ten ml from cultures were filtered under vacuum using filter membrane (0.45ųm) and washed several times with distilled water. Then, the algae cells were dried at 80°C for 30min and weighed.

A. Preparation of algae extracts

Extraction was performed using the solvents of different polarity: acetone and Methanol. Five grams of the algal dry powder were suspended in the solvents at the ratio of 6:1 (v/w), left at room temperature for 48 h and then, were homogenized for 30 min. Finally, the suspensions were centrifuged at 3000 g for 10 min and the supernatants were filtered and concentrated. The extracts were kept in dark at 4˚C until use (Ahmad et al., 2017). Followed by the preliminary phytochemical screening was performed by (Solomon et al., 2013)

B. Cytotoxcity assay

Antitumor activity assay of the extractswas determined by Human Breast Cancer (MCF7). The cell line was obtained from the NCCS, Pune. The cells were grown in RPMI- 1640 medium supplemented with 10% inactivated fetal calf serum and 50µg/ml gentamycin. The cells were maintained at 37°C in a humidified with 5% CO_2 and were subcultures two to three times a week, according to (MohdSyahril and Roshani Othman, 2011).

Calculation: % cell viability = A540 of treated cells / A540 of control cells × 100

III RESULTS

A.Preliminary phytochemical analysis

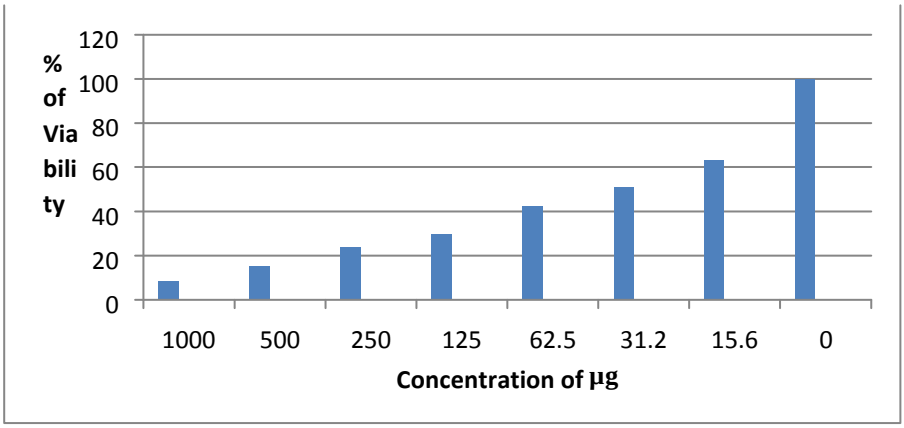
In this study, the crude methanol extract revealed the presence of alkaloids, Carbohydrate, Saponins, Terpinoids and Quinines but Flavonoids Phenols, Steroids Tannins and proteins show negative results. Anticancer activity of plant extract

The methanol extract of Chlorella vulgariswas reported to possess anticancer effects against human tumor cell lines. The results showed the inhibition of in vitro proliferation of human tumor cell lines against MCF7 cell lines. This parameter was carryout with Methyl thiazolyl tetrazolium (MTT) assay. Totally seven types of Concentration of plant extracts were subjected to anticancer activity, among them 7 concentrations were exhibited anticancer activity, the fifty percentage of cell death occur when using 31.2µg of plant extract. The result was tabulated in table.2

Table 1. Anticancer activity of methanol extract of Chlorella vulgaris

S.No	Concentration µg/ml	Dilution	Absorbance 540nm	% cell Viability
1	1000	Neat	0.06	8.4
2	500	1:1	0.11	15.4
3	250	1:2	0.17	23.9
4	125	1:4	0.21	29.5
5	62.5	1:8	0.30	42.2
6	31.2	1:16	0.36	51.0

7	15.6	1:32	0.45	63.3
9	Control	-	0.71	100



Graph.1. Percentage of cell viability of MCF7 cells

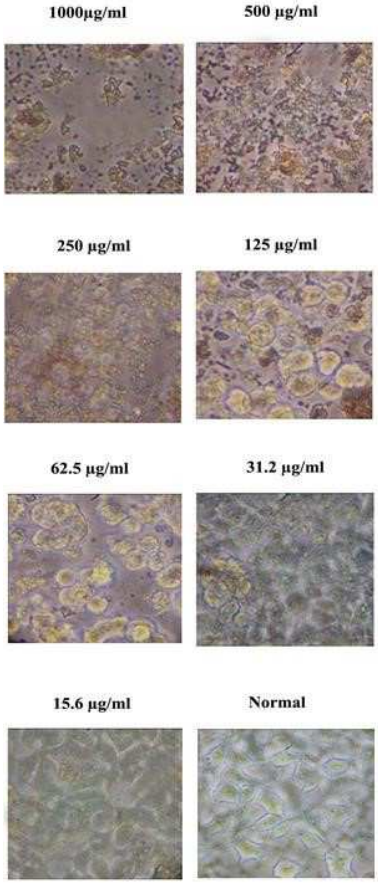


Figure 1.Microscopic examination of Anticancer activity

IV DISCUSSION

Chemotherapy is one of the main treatments used to cure cancer. Besides that, a group of drugs are used to kill or inhibit the growth of cancer cells. These drugs are associated with toxicity and side effects of chemotherapeutic drugs include hair loss, mouth sores, diarrhea, nausea and vomiting, loss of appetite and fatigue. Hence new

anticancer agents should be investigated from various resources. A great number of antitumor compounds are natural products or their derivatives, mainly produced by blue-green algae (Patterson et al, 1991, 1993). Chlorella vulgaris was shown to have chemo preventive effect in induced liver cancer and breast cancer rats and possess a glycoprotein with antitumor effects (Sulaiman 2006;Amin, 2009).In this current study, the methanol extract of Chlorella vulgaris was subjected to the anticancer activity against the MCF7 cancer cell line. The fifty percentage of cell death occurs when using 31.2µg of plant extract. The previous report emphasized the importance of strain selection as activity varied between strains of the same species (Ordoget al., 2004). Similar results were observed from previous study of Jayashree et al., (2016). They were also observed the 50% of cell death from 28.1 µg of Methanolic Chlorella vulgaris extract against MCF 7. In conclusion, phytochemicals were present in the algae, may be responsible for displaying antimicrobial and anticancer activities. It can be pointed out from this study that microalgae have the potential to be further developed as an anticancer agent taking into account that it is a product of nature. However, further studies need to be performed to fully exploit its anticancer properties such as determination of the components of cell death caused by the extracts or visual detection and confirmation of apoptosis.

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