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CYTOTOXICITY OF SEAWEEDS COUNTER TO HUMAN CARCINOMA LUNG CELLS

Iqra Rasheed, Asma Tabassum and R. Aliya

Department of Botany, University of Karachi, Karachi-75270, Pakistan.

ABSTRACT

This study was to explore the anticancer potential of marine algae. a total of fourteen seaweeds belonging to different groups (5 red, 5 brown and 4 green) were collected from Buleji coast, Karachi. Cytotoxic effects of seaweeds was determined by MTT assay, and no activity was shown by members of Rhodophycota whereas *Spatoglossum variabile* (Phaeophycota), *Enteromorpha intestinales* and *Caulerpa scalpelliformis* (Chlorophycota) revealed very promising results against NCI-H460 cells (human carcinoma lung cell).

Key words: seaweeds, cytotoxic, MTT assay, anticancer, carcinoma lung cell.

INTRODUCTION

According to Shalbay, (2011), healthy life style and intake of proper diet including fresh fruits and vegetables may helpful in detoxification of body and prevent from several diseases. Plants, animals and marine organisms are natural source of several beneficial compounds including proteins, carbohydrates, lipids and vitamins which have potential of antifungal, antibacterial, analgesic, enzyme inhibition and other important activities in biological assay (Shameel 1990; Atta-ur-Rehman, 1991).

Seaweeds have valuable natural compounds which are not commonly found in any other land plants (Carté *et al.,* 1996). These compounds mainly include alkaloids, glycosides, phytotoxins, amino acids phenols and flavonoids (Carte *et al.,* 1996; Harada *et al.,* 2002). On these basis seaweeds have been utilized in mainly cosmeceutical, pharmaceutical and food industry on commercial scale (Boukhari and Sophie, 1998; Kelman *et al.,* 2012; Berna *et al.,* 2013).

In recent era cancer has become more threatening disease for human beings (Mary *et al.,* 2012), For this, cure chemotherapy is considered as effective treatment but at the same time toxic to normal cells of the body and affects the immune system (Zandi *et al.,* 2010). It has now become necessary to discover new drugs with low side effects which intensify the patient recovery (Harada *et al.,* 2002).

Biomolecules isolated from land and marine resources are widely reported for their cytotoxic potential (Orech *et al.,* 2005). Bioactive compounds from seaweeds contain several chemical constituents which showed significant cytotoxic potential against cancer and antitumor cells (Selvin and Lipton, 2004). Previously algal compounds have been reported for their strong cytotoxic activity in different bioassay (Coombe *et al.,* 1987; Dias *et al.,* 2005), hence they have an ability to being utilized in drugs to treat several linked diseases (Smit, 2004).

Coombe *et al.,* 1987, reported the polysaccharides from seaweeds for their strong cytotoxicity against antitumor, anti-cancer and antimetastatic activities in mice. Similarly 47 algal species also showed significant cytotoxicity counter to L1210, NIH 3T3 and HDF cell line (Harada *et al.,* (2002). *Melanothamnus somalensis* and *Hypnea bryodies* also represented the Azoxymethane-induced Hepatotoxicity in rats (Waly et al., 2014, 2016). From Pakistan, Ara *et al.,* (1999) examined macro algae by brine shrimp bioassay, furthermore Ayesha *et al.,* (2010) investigated the cytotoxic activity of seaweeds on *Artemia salina*. In this context present research aimed to discover the cytotoxic potential of some other seaweeds collected from Karachi coast by MTT bioassay. Oceanic resources have been great focus of consideration for researcher and industrialist for innovation of new products (Leyman, 2002). This kind of study may contribute noble compounds to the industry.

MATERIALS AND METHODS

Collection of seaweeds

Fourteen seaweeds belonging to phylum Chlorophycota (*Valoniopsis pachynema, Codium iyengarii, Enteromorpha intestinales,* and *Caulerpa scalpelliformis*), Phaeophycota (*Padina tetrastromatica, Spatoglossum variabile, Sargassum tenerrimum, Cystoseira indica, Jolyna laminarioides*) and Rhodophycota (*Melanothamnus somalensis, Coelarthrum muelleri, Laurencia obtusa, Gracilaria corticata,* and *Gelidium pusillum*) were collected from Buleji coast, Karachi.

After washing with tap water seaweeds were dried under shade and stored in polythene bags. Each dried seaweed (100 g) was extracted thrice with 70 % ethanol for a month and extract was concentrated on rotary vacuum evaporator (Buchi rotvapor R-200) to obtain thick slurry.

Cytotoxic activity (MTT Bioassay)

The cytotoxic activity was performed according to method described previously (Dariusz *et al*., 1993) with some alterations. NCI-H460 (Lab no. 316, NRL, HEJ, ICCBS, UoK) carcinoma lung cells were seeded in 96 well plate at a density 10,000 cell per well respectively and incubated in a humidified incubator at 37˚C with 5% CO_2 for 24h.

After incubation seaweed extracts were serially diluted to the plates in incomplete medium and incubated for 48h. After incubation media was aspirated and 3-[4, 5-dimethylthiazol-2-Y1]-2, 5-diphenyltetrazolium bromide (MTT) dye was added at a concentration of 0.5mg/ml and further incubated for 4 h.

Finally the Formazan crystals formed by mitochondria dehydrogenase, were dissolved in 100µL DMSO per well after aspiration of media and absorbance was measured at 570nm

RESULT AND DISCUSSION

According to Atta-ur-Rehman (1991), Seaweeds enclosed several beneficial compounds in their thalli which are classified on the basis of their chemical nature and bioactivity. Almost 7000 natural compounds have been isolated from marine organisms and 25% of them are contributed from algal bases (Kijjoa and Swangwong 2004; Newman and Cragg 2007). Different biological assay have been designed to signify the potential of isolated natural chemicals, among them cytotoxicity test by MTT bioassay elucidate their role in medicinal application (Mary *et al.,* 2012).

Among the fourteen members of seaweeds only three species found active against human carcinoma lung cells (NCI-H460 cells) (Fig.1). These are *Enteromorpha intestinales* with IC_{50} value 188.600 ± 0.039 µg/mL, *Spatoglossum variabile* with IC_{50} value 187.182 ± 2.517 µg/mL and *Caulerpa scalpelliformis* with IC_{50} value 149.283 ± 3.326 µg/mL showed remarkable cytotoxicity. Though other seaweed represent non-significant activity in this bioassay, and this distinction might occur due to impact of environmental traits mainly temperature, light, wind velocity and nutrients which have ability to cause variations in physiological processes of an organism (Schmitt *et al.,* 1995). According to Asma *et al.,* (2008) periodically long term analysis of biological activity of seaweeds is required for their commercial innovation.

Now a days cancer has become a challenging disease and its treatment proceed further side effects to other organs of body. To lower the damaging effects, worldwide research is continue for cancer therapy from natural sources (Leyman, 2002). Ayyad *et al.* (2003) isolated six hydroazolen diterpenes from *Cystoseira myrica*, which showed active inhibition of NIH 3T3, SSVNIH 3T3, and KA3IT cells. Similarly, Coombe *et al.,* (1987) also described the cytotoxic activity of different seaweeds. From Pakistan, Ara *et al.,* (1999) described the promising results from brown seaweeds for their brine shrimp lethality bioassay. Furthermore cytotoxic activity of brown, red and green seaweeds from Karachi coast, were examined against *Artemia salina* specie, the significant lethality confirm the importance of phytochemical compounds (Ayesha *et al.,* 2010).

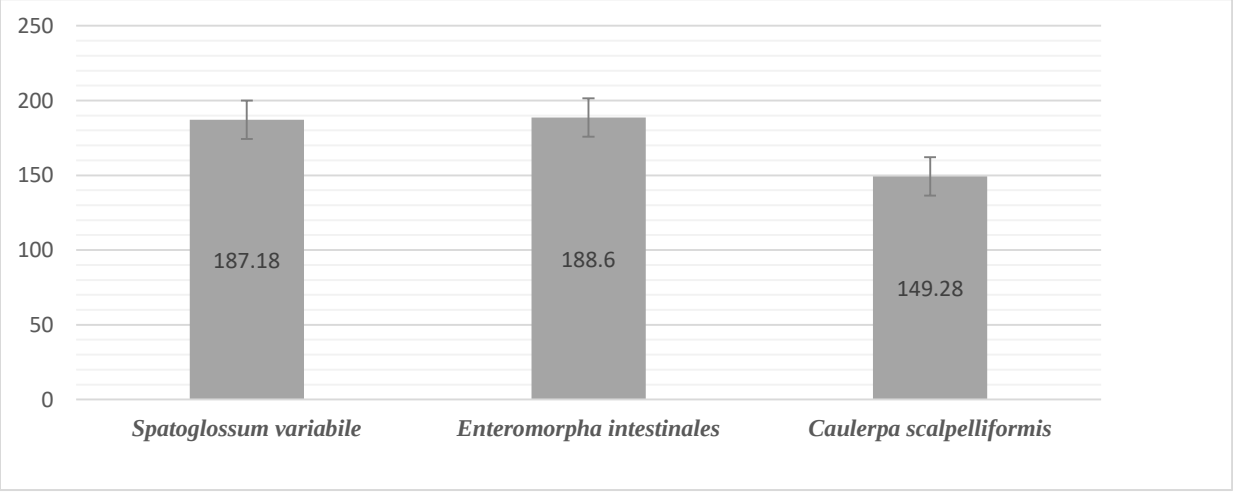

Fig.1. Cytotoxic activity of seaweeds against NCI-H460 cells.
Figure inside bar represent IC_{50}; Incubation Period = 4 h; Temperature = 37˚C.

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