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# Bioactive Potential of Turbinaria Conoides (J Agardh) Kuetz: *In vitro* and *In vivo*

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**Abstract**

Brown seaweeds including *Turbinaria conoides* have been used as food since ancient times which are widely consumed in Asian region than that of in Europe and America. Though, the chemical composition varies with species, habitat, maturity and environmental conditions however they are excellent sources for the bioactive phytochemicals with rich dietary fiber, minerals, non-digestible polysaccharides and capacity to absorb inorganic substances from their surroundings. The main bioactive phytochemicals are steroids, phenolics, flavonoids, reducing sugars, fucosterol, sulfated polysaccharides including fucoidan, neutral glucan, guluronic and alginic acid. These phytochemicals are responsible for the following biological properties such as antioxidant, anti-inflammatory, antimicrobial, and anti-cancer. Therefore, in the present work was clearly documented about the bioactive potential of *T. conoides* with respect to their phytochemicals in both model of *in-vitro* and *in-vivo*.

**Keywords:** Brown algae; Turbinaria conoides; Phytochemicals; Biological activities

## Introduction

It is well known that marine algae have budding source because of their numerous health-promoting effects including antioxidant, anti-inflammatory, antimicrobial, and anti-cancer [1,2]. Particularly, brown seaweeds gripped with rich novel antioxidants which are more acceptable than synthetic /chemical components [3]. Henceforth, marine macroalgae are believed to exhibit potent biological impact especially due to phenolic compounds as well as carotenoids, ascorbic acid, glutathione, sulphated polysaccharides, fucoxanthin, astaxanthin, polyphenols, phlorotannins, phospholipids, flavonoids, bromophenols and so on [4,5]. This rich bioactive phytochemicals are also there pretty in *Turbinaria*. It is brown algae belong to the family of Sargassaceae (brown algae) under the order of Fucales. It is consist of only 22 species so far described in which highest diversity was found in south west Asia akin to India, Srilanka where documented around 14 species. *T. turbinate* was only present in the Atlantic Ocean where as three species such as *T. conoides*, *T. decurrens* and *T. ornate* found in the South Pacific Ocean [6]. It traditionally been used as a fertilizer, insect repellent, pesticide, anti-bacterialcidal and also antioxidant, anti-inflammatory, and anti-cancer due to bioactive phytochemicals [7,8]. Apart, essential components digestible proteins along with mineral salts (K, Ca, and Fe) and polyunsaturated fatty acids along with wealthy source of dietary fiber and iodine content which play an immense role in enhancing the food quality and biochemical homeostasis [9]. Therefore, in the present review work was aimed to illustrate the potential biological properties of *T. conoides* with respect to their phytochemicals in both model of *in-vitro* and *in-vivo*.

## Antioxidant and its phytochemicals

Various methods are essential to give an overall idea about the broad spectrum of antioxidant activity of phyto/chemical components [10]. Radical scavenging is one of the most powerful mechanisms by which antioxidants inhibit oxidation process. Many *in-vitro* methods has been used to examine scavenging of free radicals in which most often used are ABTS Radical cation, DPPH radical, and reactive oxygen species (ROS), such as superoxide anion, hydrogen peroxide, peroxyl radicals, hydroxyl radical, singlet oxygen and peroxynitrite. ROS are responsible for oxidative damage in the human body as well as in the food samples [11]. ABTS oxidized to give the radical cation ($ABTS^{\cdot+}$) as blue in colour and decolorized by water-soluble and lipid-soluble food samples/extracts and is expressed as TEAC (Trolox equivalent antioxidant capacity) [12,13]. Using the method, total antioxidant activity (TAA) of *T. conoides* was documented in the range 46-85% in hexane, dichloromethane, ethyl acetate and aqueous fraction (Table 1). Relative antioxidant activity (RAA) was also found ≥1

in the ethyl acetate fraction than that of other fractions. Apart, the highest superoxide radical scavenging activity was reported in ethyl acetate fraction (77% at the 80µg/ml) and better than ascorbic acid (74% at 30µg/ml) [14].

**Table 1:** Summary of biological properties of T conoides.

| Extracts | Assay | Expression | Reference |
|---|---|---|---|
| **Antioxidant** | | | |
| Polysaccharides | DPPH | % | Chattopadhyay et al. [8] |
| Crude polysaccharide | DPPH | % | Ananthi et al.[28] |
| Aqueous extract | DPPH, ABTS, NO, LPO | % | Ananthi et al. [28] |
| Ethyl acetate (EtOAc) fraction | DPPH,ABTS | % | Chakraborty et al. [10] |
| | $H_2O_2$ scavenging | | |
| | LPO (TBARS) | mg/ml | |
| | Reducing ability | | |
| | Phenolic content | (mg GE/g) | |
| Ethanol fractions | ABTS | % inhibition, mg/g | Arumugam et al. [14] |
| | Superoxide radical | | |
| | Iron chelation | | |
| | Reducing Power | | |
| | uric acid formation inhibitory | | |
| **Cytotoxicity/Anticancer** | | | |
| Fucosterols | Various cancer cell lines by | % | Jyh-Horng et al. [7] |
| | MTT assay | | |
| cyclohexane extracts | Human embryonic lung cells, microscopy assay | minimal cytotoxic concentration (MCC) | Shanmugam et al. [23] |
| Steroid from Ethyl acetate extract | HeLa cells | % | Sadish Kumar & Jayendra [29] |
| Ethyl acetate fraction | Hep G2 cells by MTT assay | % | Arumugam et al. [14] |
| | Cell cycle and Apoptosis by Flowcytometry | | |
| **Antimicrobial/Antibacterial activity** | | | |
| Cyclohexane extract | Disc diffusion method | Zone of inhibition | Shanmugam et al. [23] |
| Ethanol extracts | Disc diffusion method | Zone of inhibition | Manivannan et al. |
| Methanolic extract | Disc diffusion method | Zone of inhibition | Vijayabaskar & Shiyamala [22] |
| Solvent extract | Disc diffusion method | Zone of inhibition | Sridharan & Dhamotharan [20] |
| Ethanol extract | Disc diffusion method | Zone of inhibition | Senthilkumar & Sudha [21] |
| **Anti-inflammatory** | | | |
| crude polysaccharide | Carrageenan-induced paw edema and vascular permeability test | % inhibition | Ananthi et al. [31] |
| Aqueous extract | Carrageenan induced paw edema | % inhibition | Ananthi et al. [28] |
| Ethyl acetate fraction | Carrageenan induced paw edema | cm | Arumugam et al. [27] |
| | Acetic acid induced writhing Tail immersion methods | Sec<br>Min | |
| Fucoidan like sulphated polysaccharide | cotton pellet induced Granuloma | % inhibition | Ananthi et al. [31] |
| **Antigenotoxic** | | | |
| Ethyl acetate fraction | Micronucleus assay | % | Arumugam et al. [27] |



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In general, metal ions ($Fe^{2+}$) can also kindle and speed up oxidative damage later on leads to lipid peroxidation which is inhibited by chelators either decreasing metal reactivity or by physically partitioning the metal away from the lipids [15]. It is well know that bioactive compounds like phenolic acids, flavonoid, quercetin, and phenolic glycosides are potentially participate to chelate metal ions which is determine by Spectrophotometry [10]. Iron chelation and uric acid inhibition activities of ethyl acetate fraction of *T. conoides* were reported in the range of 15-70% [14]. The reducing capacity assay is one of the methods to reduce the oxidative damage by inhibiting the peroxidation process. This process can be done by low molecular weight antioxidants which is able to donate electrons to reactive oxygen species. It can be determined by simple methods:

a) The FRAP assay, based on the reduction of the $Fe^{3+}$/ tripyridyltriazine complex [16],

b) The direct reduction of $Fe^{3+}$ ferricyanide complexes and

c) Electrochemical methods [17,18]. Reducing power exhibited by solvent extracts of *Turbinaria spp*. was comparatively higher than α-tocopherol [19].

## Antimicrobial and its phytochemicals

Antimicrobial properties of *T. conoides* were determined by disc diffusion method which was well documented by many research articles (Table 1). Arumugam P et al. [14] reported that among the four solvent fractions of *T. conoides,* ethyl acetate fraction exhibited highest antibacterial activity which was comparable to the standard, streptomycin against *Bacillus subtilis, Enterococcus faecalis* and *Pseudomonas aeruginosa.* Similarly, out of four solvent fractions, petroleum ether extract reported to be showed effective antibacterial activity [20]. The antimicrobial activity of *T. conoides* was also reported in dose dependent activity of all four extracts and highest activity exhibited at 500µg/mL [21]. It is mainly due to secondary metabolites like phenolic compounds which may explore inhibiting effect on microbial growth based on their chemical constitutions and concentrations [22]. Consecutive extraction of *T. conoides* with *n*-hexane, cyclohexane, methanol and ethanol: water (1:1) was also documented with their antibacterial and antifungal activities by disc diffusion method. In which, cyclohexane extract was possessed a broad array of antibacterial activity and exhibited remarkable antifungal property over the other extracts [23]

## Anti-inflammatory, anti-genotoxicity and its phytochemicals

During the oxidative damage of cell or tissue, several inflammatory mediators such as histamine, bradykinin, serotonine, and prostaglandins are released and stimulate the inflammation and nociceptors by the induction of pain [24]. These mediators are occupied in tissues with high content of water and plasma during arachidonic acid metabolism via cyclo-oxygenase and lipo-oxygenase enzyme pathways [25]. The first phase of inflammation begins immediately up to an hour after injection of carrageenan by the release of histamine and serotonin whereas the second phase started after one hour and up to three hours by the release of bradykinin, protease and prostaglandins [26]. Anti-inflammatory effect of ethyl acetate fraction of *T. conoides* were reported to be significantly ($P < 0.05$) better than that of control and indomethacin (Table 1). The reduction of paw volume was found to be dose dependent. The acetic acid induced abdominal writhes in mice were recovered significantly from all the tested doses of *T. conoides*. Ethyl acetate fraction of *T. conoides* might have capable of reduce inflammation through stabilizing the lysosomal membrane. In addition, the analgesic effect of *T. conoides* on the tail immersion-test in mice was found to be dose dependent and significantly reduce the pain response by the increase of reaction [27]. Recent report on *T. ornate* extract revealed their better anti-inflammatory and free radical scavenging property due to fucoidan like sulfated polysaccharides [28]. *T. conoides* reported to have better antipyretic activity by restoring many hematological and biochemical parameters under toxic environment [29]. In general, exposure of any environmental toxin/genotoxin leads to genetic damage which is determined by mouse bone marrow micronucleus assay described by [30]. For each bone marrow/peripheral blood cells (experimental/control), 2,500 polychromatic erythrocytes (with or without micronuclei) and a corresponding number of normal chromatic erythrocytes (NCEs) were scored under a light microscope. Ethyl acetate fraction of *T. conoides* reported to be 72% anti-genotoxic activity against 4-NQO induced genotoxicity [27]. These biological activities of *T. conoides* are mainly due to rich source of bioactive compounds such as fucosterol, sulfated polysaccharides fucoidan, neutral glucan, guluronic and alginic acid [28,31].

## Anticancer and its phytochemicals

Cytotoxicity assay has been considered as the cell killing property of a chemical compound which is independent mechanism from the programmed cell death pathway [32]. Cytotoxicity of ethyl acetate fraction of *T. conoides* was reported to be significant and comparable to the standard of quercetin (Table1). Hence, cytotoxicity and antioxidant of *T. conoides* were well correlated and concentration dependent. The anticancer analysis exhibited that the number of accumulated cancer cells was significantly ($p < 0.05$) higher in the proliferative G0/ G1 phase and a significant decrease in the S phase, after 48h of treatment with ethyl acetate fraction of *T. conoides.* Similarly, *T. conoides* showed (43%) statistically ($p < 0.05$) significant increase of apoptotic cells than that of quercetin standard (32%, 80 µg/ml) [15]. Other reports revealed that cell cycle arrest leads to increase in sub-G0/G1 cell population after treatment with increasing doses of linalool terpenoid [33]. Cyclohexane extracts of *T. conoides* exhibited effective cytotoxicity in human embryonic lung cells [24]. Anticancer activity of fucosterols obtained from *T. conoides* was explored in various cancer cell lines [8]. Steroid from ethyl acetate extract of *T. conoides* reported to be effective cytotoxic in HeLa cells [34]. Various extract obtained from a variety algae


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collectively demonstrated that the brown algae have a potential source of phytochemicals exhibiting biological activities on tumor cells [35].

## Conclusion

It is well know that *T. conoides* possess large content of dietary fiber, minerals, steroids, phenolics, flavonoids, reducing sugars, fucosterol, sulfated polysaccharides including fucoidan, neutral glucan, guluronic and alginic acid. As results, it exhibited different biological properties such as antioxidant, antimicrobial, anti-inflammatory and anti-cancer. Many articles revealed that cyclohexane and ethyl acetate extracts/fraction of *T. conoides* have potential bioactive phytochemicals with different biological properties.

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