

Potential of liquid fertilizer obtain from *Trubinararia conoides* (J.Agardh) Kuetz and its phytochemical analysis by UV-Visible and FTIR spectroscopy

Abstract

Brown seaweed, *Turbinaria conoides*, has been used as food since ancient times and is more popular in Asia than in Europe and North America. In terms of antioxidant, anti-inflammatory, antibacterial, and anti-cancer actions, they possessed a variety of biological potentials. The *T. conoides* shadow dry powder was combined in a 1:20 (w/v) ratio with distilled water to create the seaweed liquid fertiliser (SLF), which was then heated to 70°C for an hour. Using UV-Visible and FTIR spectroscopy, SLF was employed to investigate their impact on avarai and black soya bean seed germination and phytochemical analyses. The greatest absorption peaks were found in the qualitative UV-Visible spectra to be at 200, 224, 231 and 300 nm. By using UV-VIS spectroscopic examination, it was confirmed that flavonoids were present in the SLF. The results of an FTIR analysis showed that a peak at 3351.6 cm⁻¹ indicated the presence of O-H stretching, a peak at 1630.1 cm⁻¹ suggested C=O vibration, a peak at 1022 cm⁻¹ indicated inorganic ions such silicate ions, and a peak at 654.66 cm⁻¹ indicated C-Br and C-I stretch, respectively. The percentage of seeds that germinated in the control experiment ranged from 40 to 44 percent. Germinated seeds treated with SLF yielded 72 and 84 percent at 100% of SLF concentration in avarai and black soya bean seeds, respectively. The SLF-treated seeds grew twice as quickly as the untreated seeds. Compared to avarai seeds, SLF more successfully promotes black soya bean seeds. The growth rate of germinated seeds was measured on the seventh day and quantified in terms of cm shoot and root growth, showing a similar tendency to that of seed germination. The SLF-treated seeds grew twice as quickly as the untreated seeds. SLF more successfully promotes avarai seeds than black soya bean seeds.

Keywords: *Turbinaria conoides*, SLF, seed germination, UV-Vis, FTIR

Volume 12 Issue 1 - 2023

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Received: February 10, 2023 | **Published:** February 24, 2023

Introduction

The tradition of using seaweeds as manure in farming dates back thousands of years and was widespread among the Romans, the British, the French, the Spanish, the Japanese, and the Chinese. Marine macroalgae have a long history of use as crop plant fertilizer in coastal regions all over the world. Even in the early 1900s, farmers still valued seaweed cast. In several nations, beach cast and seaweed are still utilized in gardening and agriculture.¹ The use of marine macroalgae as fertilizer has been the subject of numerous studies over the last three decades and is now widely used in modern agriculture. They can be used whole, as powdered algal manure, finely minced, or as aqueous extracts. Due to the existence of seaweed extracts (SWEs), which contain plant growth hormones, regulators, growth promoters, carbohydrates, amino acids, antibiotics, and vitamins, many researchers have recently looked at the use of these marine macroalgae in modern agriculture. The constitutions improved as a result, increasing crop output and quality, seed germination, and resilience to cold, fungal, and insect attacks. Several crops are successfully sprayed with liquid seaweed extracts as foliar sprays. According to Karthik et al.,² the value of seaweeds as fertilizers was not just a result of their richness in nitrogen, phosphorous, and potash, but also because they had trace elements and metabolites. Using seaweed extract as an organic biostimulant in agricultural and horticultural crops is quickly becoming standard practice. It has been observed that the seaweed extract has a growth-regulatory influence on seed germination as well as morphological and biochemical traits in agricultural crops.¹

However, the usage of chemical fertilizers is harmful to both plants and the helpful microorganisms present in soil since they contain carcinogens such nitrosamines, excessive levels of sodium, and potassium, all of which damage the soil's fertility, pH, and overall quality as well as human health.^{3,4} The accumulation of carcinogens in the soil was seen in plants and vegetables, and it even reached further up the food chain.⁵ Vegetable eaters consequently experience renal and lung disease, blood disorders, brain haemorrhages, and even cancer.⁶ As a result, organic fertilizer is now preferred over chemical fertilizer since it has less negative impacts on soil health, is biodegradable, provides a balanced diet of minerals, and increases food yield and quality.⁷ They are produced from a variety of biological elements, including compost of agricultural wastes (straw, decomposing wastes, etc.), green manure (young plants, legume plants, and seaweeds), and biofertilizers (*Rizobium*, *Azolla*, and *Anabena* symbiosis).⁵ Seaweeds (marine macroalgae) were among them, and they were thought to be a potential source to use as organic fertilizer.⁸ Due to its extensive use as a foliar spray to boost crop yields in Indian agriculture, SLF has recently been demonstrated to be an organic biostimulant. This is because it contains enough levels of potassium, nitrogen, growth-promoting hormones, micronutrients, and humic acids.⁹

Turbinaria is rich in phytochemicals and has a lovely disposition. It is a species of brown algae that belongs to the fucales suborder's sargassaceae family. There are just 22 species that have been identified thus far, with the most diversity being found in south-west Asia, from India to Sri Lanka, where 14 species have been recorded. *T. turbinata* was solely found in the Atlantic Ocean, but the South Pacific Ocean was home to three species, including *T. conoides*, *T. decurrens*,

and *T. ornate*.^{10,11} Due to a range of bioactive phytochemicals, it has historically been used as a fertiliser, insect repellent, pesticide, anti-bacterial, anti-inflammatory, and anti-cancer.^{12,13} In addition to being essential, digestible proteins, mineral salts (K, Ca, and Fe), polyunsaturated fatty acids, rich sources of dietary fibre, and iodine content all contribute significantly to the improvement of food quality and biochemical balance.¹⁴⁻¹⁶ The purpose of the current study was to use UV-Visible and FTIR spectroscopy to investigate the impact of seaweed liquid fertilizer of *T. conoides* on seed germination and phytochemical analyses.

Materials and methods

Seeds and chemicals

Black soya bean (*Glycine max*) and avarai (*Lablab purpureus*) seeds are both members of the Fabaceae family and were both commercially acquired in April 2022 at Harur Market in the Tamilnadu district of Dharmapuri. In this investigation, class clothing, cotton, and tissue paper were autoclave-sterilized at 121°C, 15 lbs/sq. inch, for 30 minutes. The investigation only employed analytical-grade solvents. Analysis of phytochemicals was done by using FTIR and UV-Visible spectroscopy.

Seaweed collection

Brown seaweed known as *T. conoides* was found in the subtidal zone off the coast of Mandapam (Lat. 09017'N; Long. 79007'E, Gulf of Manner, India). To get rid of salt, sand, and epiphytes, seaweeds were washed in sweater. They were then shade-dried for five days in a room with fresh air. The shade-dried sample should be chopped and powdered using a home blender (Figure 1).

Preparation of seaweed extract

The distilled water was added to the shade-dried seaweed powder in a 500 mL beaker at a ratio of 1:20 (w/v), and the mixture was cooked at 70°C for an hour on a hotplate. A double-layered cheese cloth was used to filter the hot aqueous extract, which was then allowed to cool to room temperature 15 minutes were spent centrifuging the filtrate at 10,000 rpm. The supernatant was regarded as containing only liquid seaweed extracts. Different percentile concentrations of seaweed liquid extracts, including 25, 50, 75, and 100, were created along with double-distilled water as the control.

UV-visible and Fourier transform infrared (FTIR) spectroscopy

Turbinaria conoides seaweed liquid fertilizer was placed in a cuvette and subjected to UV-visible spectroscopy using a Hitachi double beam spectrophotometer (UH5300) in the wider range of 200 to 800 nm (Figure 1). Similar to this, an FTIR spectrometer paired with a TGS (Tri-glycine sulphate) detector was used to record the FTIR analysis of seaweed liquid fertilizer from *T. conoides* in the mid-IR region (4000e400 cm⁻¹) at a resolution of 4 cm⁻¹. To make salt-discs, 1 mL of water extract sample was combined with 100 mg of potassium bromide (KBr) (3mmdia). An infrared spectrophotometer using the fourier transform was used to analyse these discs (Perkin Elmer model 10.6.2).

Biostimulant assays

Seed germination

Black soya bean and avarai seeds were separated into five groups, each with 25 seeds. Out of the five groups, one was used as the control and spent up to 12 hours in the double-distilled water. Similar to this,

the other four groups received varied concentrations of *T. conoides* liquid seaweed fertilizer, including 25, 50, 75, and 100%. On the third day, the number of seeds from each group that had germinated was counted, and the growth rate was reported as a percentage using numbers in triplicate.

Root and shoot length

A seed germination experimental setup was utilized to measure the shoot and root length of seeds that had sprouted on the seventh day. This configuration included a control group as well as treatment groups that received various concentrations of liquid seaweed fertilizer (25, 59, 75, & 100%). All germination seeds' shoot and root lengths were measured, expressed in centimeters, and the results were triple-checked.²

Results and discussion

Since ancient times, *Turbinaria conoides*, a member of the brown seaweed group of algae, has been consumed in greater quantities in Asia than in Europe and North America. Even though their chemical makeup varies depending on the species, habitat, stage of development, and environmental factors, they are excellent sources of bioactive phytochemicals and have high levels of dietary fiber, minerals, and non-digestible polysaccharides in addition to the ability to absorb inorganic substances from their environment. Steroids, phenolics, flavonoids, reducing sugars, fucosterol, and sulfated polysaccharides such fucoidan, neutral glucan, guluronic acid, and alginic acid are the primary bioactive phytochemicals. The biological characteristics of antioxidant, anti-inflammatory, antibacterial, and anti-cancer are all a result of these phytochemicals. In order to assess the impact on seed germination, avarai and black soya bean seeds were treated with seaweed liquid fertilizer (SLF) made from *Turbinaria conoides*. The *T. conoides* shadow dried powder was combined in a 1:20 (w/v) ratio with distilled water and heated to 70°C for an hour to create the seaweed liquid fertilizer. SLF was centrifuged after being filtered twice through cheese cloth. In the current investigation, the supernatant was employed as an SLF and was expressed as percentiles of 25, 50, 75, and 100. To get the sharpness of peaks and correct baseline, SLF of *T. conoides* was exposed under UV-visible spectroscopy using a Hitachi double beam spectrophotometer (UH5300). Maximum absorbance peaks were discovered in the qualitative UV-Visible spectrum profile of SLF of *Turbinaria conoides* at 200, 224, 231, and 300 nm (Figure 1). According to the UV-Visible spectrum profile, flavonoids and phenolics are commonly found between 190 and 800 nm.¹⁸ The outcome of UV-Visible spectrum analysis supports the discovery of flavonoids in *T. conoides* SLF.

It is widely known that the peak value in the infrared region of the FTIR spectrum was used to determine the molecular arrangement of various functional groups of the active components. When the SLF of *T. conoides* passed through the FTIR, the FTIR spectrum results in terms of its peak values were observed. The functional groups of the components were separated based on their peak ratios, and their major peak was observed at 3351.6, 2096.8, 1630.1, 1022, 654.66, 597.28, and 559.02 cm⁻¹ (Figure 2-4). The results of the FTIR study showed that 1630.1 cm⁻¹ peak value suggested C=O vibration due to the presence of a carboxylic acid group whereas 3351.6 cm⁻¹ peak value demonstrated O-H stretching due to the presence of a free hydroxyl group. Aside from that, the peak value at 1022 cm⁻¹ was typical of common inorganic ions such the silicate ion. The peak values of 654.66, 597.28, and 559.02 cm⁻¹ were associated with either the C-Br or C-I stretch vibration, which might be, respectively, an aliphatic bromo chemical or an aliphatic iodo molecule.

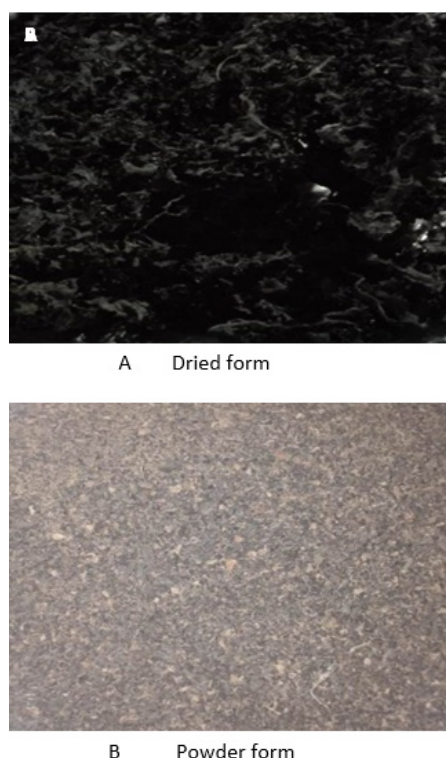


Figure 1 Dried and powder form of Brown seaweed of *Trubinaria conoides*.

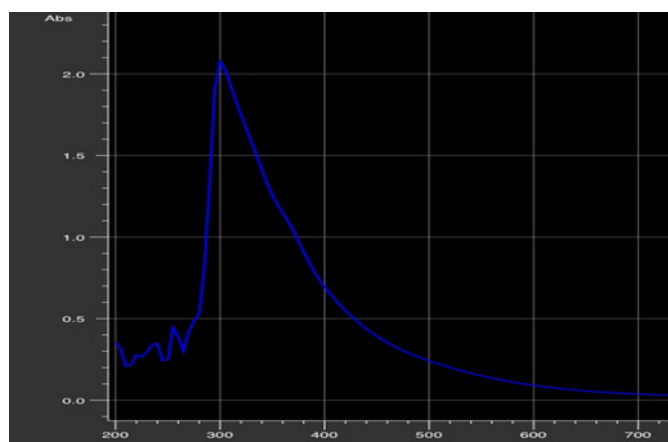


Figure 2 Absorbance spectrum of seaweed liquid fertilizer of *Trubinaria conoides*.

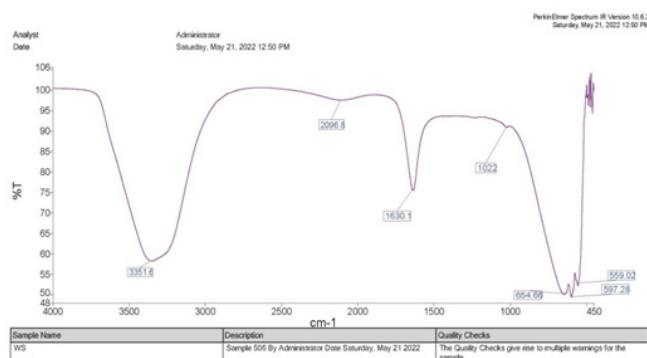


Figure 3 FTIR spectrum of seaweed liquid fertilizer of *Trubinaria conoides*.

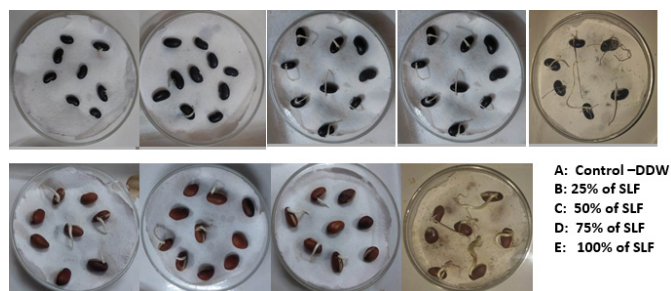


Figure 4 Control and seaweed liquid fertilizer of *Trubinaria conoides* on seed germination.

To assess their capacity for seed germination, the varying percentile concentrations of SLF were administered to Avarai and black soya bean seeds. In the experimental regime of seed germination, 25 numbers of each seed were soaked for up to 12 hours in the respective control and varied percentile concentrations of SLF (25, 50, 75, and 100) (overnight). The seeds were then carefully placed on the glass petriplate with sterile cotton underneath the tissue paper at a distance of 5 mm. Every day for seven days, the seed germination was observed, and on the third day, the germination of the seeds was recorded (Table 1). Percentile of seed germination was used to describe the total number of seeds that germinated out of 25 seeds. In the control experiment, the number of grown seeds was recorded as 10 and 11, respectively, out of 25 seeds of avarai and black soybeans, and they exhibited up to 40–44% germination. Out of each 25 seeds of avarai and black soya beans, it was discovered that 7, 12, 15, and 18 and 9, 15, 17, and 21 grown seeds, respectively, were treated with SLF at varied concentrations (25, 50, 75, and 100%). Similar results were found for avarai and black soya beans, where the germination rates were 28, 48, 60, and 72%, and 46, 60, 68, and 84%, respectively (Table 1). The seeds fed with seaweed liquid fertilizer grew twice as well as the untreated ones. The findings made it abundantly evident that SLF successfully increases the germination of avarai and soya bean seeds relative to controls, and that black soya bean seeds also exhibit superior germination than avarai seeds. With 15 mL L⁻¹ of SLF derived from *Ascophyllum nodosum*, marigold seed germination and growth of the shoots and roots morphology were improved.¹⁹ Other seaweed, including *Padina minor*, *Sargassum crassifolium*, *Sargassum cristaefolium*, and *T. decurrens*, were treated to soybean vegetables in concentrations of 0.1%, 0.2%, 0.3%, and 0.4% SLF. The findings showed that *P. minor* treatment at 0.4% concentration considerably enhances all measures of soybean vegetative development.²⁰

On the seventh day after germination, the growth rate of the seeds was also assessed in terms of shoot and root length and quantified as centimeters (cm) of shoot and root length. The control seeds measured 1.4 and 1.7 cm in shoot length whereas the avarai and black soya beans measured 1.1 and 1.3 cm in root length. As opposed to this, SLF of *T. conoides* treated seeds revealed shoot length (1.3, 2.1, 2.7 & 3.5 cm) and root length (0.5, 1.2, 1.9 & 2.3 cm) of avarai seeds, while shoot length (1.5, 2.5, 3 & 4 cm) and root length (0.9, 1.4, 2 & 2.7 cm) of black soya bean seeds with respect to their different concentration (25, 50, 75 & 100%) of SLF (Table 2). *Raphanus sativus* (radish), *Phaseolus vulgaris* (green pea), and *Vigna radiata* (mung) seeds were evaluated with an SLF (20, 40, 60, 80, and 100%) produced from the marine algae *T. ornata* and *Ulva reticulata*, according to Karthik et al. (2020). It has been observed that 100 percent SLF from *T. ornata* and *U. reticulata* significantly increased growth rates when compared to artificial fertilizers that only contained 80 percent SLF. On tobacco, pea, and cotton seeds, SLF of brown algae such

Sargassum plagiophyllum, *T. conoides*, *Padina tetrastromatica*, and *Dictyota dichotoma* has demonstrated potential. Chili and turnip seeds treated with SLF grew more quickly than seeds treated with greater concentrations at lower concentrations. As previously reported, seaweeds like *Ulva fasciata*, *Enteromorpha intestinalis*, *Dictyota dichotoma*, *Sargassum wightii*, *Padina boergesenii*, *Amphiroa anceps*,

Avanthopleura spicifera, and *Spyridia hypnoides* have the ability to promote plant growth in agriculturally significant crop plants like Green gramme, Black gramme, Mustard, and Paddy.²¹ Additionally, Fourier transform infrared (FT-IR) was used to analyse the chemical composition of SLF and study its content in terms of carbon, nitrogen, phenolic acids, and hormones.²²

Table 1 Seaweed liquid fertilizer of *Turbinaria conoides* on seed germination

S. No	Concentration of Seaweed liquid fertilizer (%)	No of seed germinated		Germinated seeds (%)	
		Avarai seed	Black Soya beans seed	Avarai seed	Black Soya beans seed
1.	Control (Distilled water)	10	11	40	44
2.	25	7	9	28	36
3.	50	12	15	48	60
4.	70	15	17	60	68
5.	100	18	21	72	84

Table 2 Seaweed liquid fertilizer of *Tubinarina conoides* on shoot and root growth

S. No	Concentration of Seaweed liquid fertilizer (%)	Shoot length (cm)		Root length (cm)	
		Avarai seed	Black soya beans seed	Avarai seed	Black soya beans seed
1.	Control (Distilled water)	1.4	1.7	1.1	1.3
2.	25	1.3	1.5	0.5	0.9
3.	50	2.1	2.5	1.2	1.4
4.	70	2.7	3	1.9	2
5.	100	3.5	4	2.3	2.7

Conclusion

In order to test the effect of seed germination, avarai and black soya bean seeds were treated with a seaweed liquid fertilizer made from brown *Turbinaria conoides* seaweed. The results of the UV-VIS spectroscopic examination provide proof that the seaweed extract contains flavonoids. The primary peaks of the FTIR spectra were measured at 3351.6, 2096.8, 1630.1, 1022, 654.66, 597.28, and 559.02 cm⁻¹. The seeds fed with seaweed liquid fertilizer grew twice as well as the untreated ones. Compared to avarai seeds, SLF successfully promotes black soya bean seeds.

Acknowledgments

This research work was achieved due to in the presence of basic equipment's like UV ad FTIR provided by Sri Vidya Mandir Arts & Science College (Autonomous) is gratefully acknowledged.

Conflicts of interest

There was no any conflict of interest.

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