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Antioxidant And Spore Germination Inhibition Activities Of *Spirogyra Majuscule* Of Fresh Water Ponds In Koppa Taluka Of Chickmagaluru District

Prathima Mathias D.A.

Associate Professor, Department of PG studies in Industrial Chemistry and Chemistry,
Sir M.V. Government Science College, Bommanakatte, Bhadravathi – 577 302,
Shivamogga, Karnataka, India

Abstract: Oxidative stress is a result of variation in levels of antioxidants that act as protective shields. Lower levels of antioxidants lead to deterioration of healthy cells exposing them to reactive oxygen species, peroxyl radicals, peroxy nitrite, hydroxyl radicals, singlet oxygen and allied species. Many diseases associated with lack of antioxidants are Parkinson's and Alzheimer's, ischemic injury, cancer and premature ageing.

Spirogyra majuscule found in Koppa of Chickmagaluru district was investigated for its antioxidant and spore germination inhibition activities. Dry mass of algae had 30.01 % moisture and 6.2 % of ash. Extractions were carried out using hot and sonicated methods. Sohxlet extract gave 21 % pet ether, 3.6 % protein, 25 % reducing sugar fractions. The sonicated extract had 13 % pet ether, 3.2 % protein and 31.3 % reducing sugar concentrations.

The hot: sonicated extracts showed phenol (590 mg: 450 mg), Flavanoid (250:360), Antioxidant (54.8:50.7) and Reducing powers (0.050:0.045).

The spore germination inhibition of both the extracts by cavity slide method, in comparison with standard fluconazole, exhibited an average of 95.5 % activity against *Fusarium oxysporum* and 90.5 % against *Curvularia lunata.*

Index Terms - Antioxidant activity, reducing power assay, bio-composition, *Spirogyra majuscule*, spore germination inhibition.

I. INTRODUCTION

Algal bioenergy is cost effective method for production of renewable energy. It does not require special cultivation lands or specific growth conditions, thereby making it easy to handle and acquire in large quantities. Alga is sustainable sources of bio energy production and a promising subject in many developed countries for microalgae bio energy research projects.

The fast growth rate, complexity of structure and useful phytochemicals especially oils make algae most suitable subject for many types of energy related studies. Carbon neutral biodiesel, ethanol and crude oil can be generated effectively from different kinds of algae, thereby making it a smarter choice for future bio energy production sciences (Patil and Tran, 2008).

More than 400 species of Spirogyra belonging to Zygnematophyceae (Class), Zygnematales (Order), Zygnemataceae (Family), are commonly found in fresh water areas, across the world.

Measuring up to 10 to 100µm in width, it grows on shallow warm water forming vibrant green mats that appear to float with bubbles emerging from their surfaces due to production of large quantities of oxygen. They are therefore called as 'water silk'. Suring spring the mats are found just below the water surface.
There are few reports on the antioxidant properties of Spyrogyra and other similar kinds of algae (Mayalen Zubia 2007). Various scientists have detected presence of few dyes and metals like Cu, Ni etc in algae due to their absorption in polluted water sources (Khataee et al. 2013, Ulker, Meltem 2013, Dhananjay, 2012, Alpana et al. 2007, K. Suresh Kumar et. al. 2015, Fabíola V. Hackbarth et al. 2014).
In continuation of our similar kind of work, we have collected the available species of Spirogyra from Koppa of Chickmagaluru District, used two methods of extraction carried out basic analysis along with its anti-oxidant and spore germination inhibition studies.

II. MATERIALS AND METHOD

Moisture content, Total Ash analysis, crude extraction, protein analysis, etc were all done by reported methods and are mentioned in Table-1.

Table -1: Table parameters estimated with the methods adopted for each

Sl. No.	Parameters Estimated
1	Moisture : Dry weight
2	Total Ash: Anaerobic combustion
3	Extraction : a) Hot method- Sohxlet b) Sonication
4	Protein: Lowry's method
5	Total Phenol: Spectrophotometry
6	Redusing sugar: DNS
7	Flavanoid assay: Reduction
8	Antioxidant assay: DPPH
9	Reducing power assay: Ferrocyanide
10	Antifungal activity : Spore germination inhibition method using cavity slide technique

III. RESULTS AND DISCUSSION

The algae had 30.01 % moisture content and 6.2 % ash content. The collected species of *Spirogyra majuscule* yielded 8 % higher pet ether extract by sonication method. Both methods gave almost same percentage of protein content (3.3). Sonicated extract showed higher percentage of reducing sugar (31.3). These results are as shown in Table-2.

Spirogyra majuscule		Table -2: Table of results for biochemical analysis	
Moisture	30.01 %		
Ash	6.2 %		
Extract	Pet ether (%)	Protein (%)	Reducing sugar (%)
Sohxlet	21	3.6	25.0
Sonication	13	3.2	31.3

Antioxidant activities are usually influenced by the percentage of phenols and flavonoids present in the sample under consideration. The phenol, flavanoid values along with antioxidant assay results are tabulated in Table-3.

Both methods of extraction sowed nearly equal values of phenolic content, the sohxlet method showed 590 mg GAE/100 g DW and sonication method gave 450 590 mg GAE/100 g DW.
Flavanoid content was slightly higher in sonication method by 110 units.

Antioxidant activity was on an average of 52.6 ± 2. Sohxlet extract showed the reducing power of 0.050 whereas sonicated extract showed 0.045.

Table 3: Phenol, Falvanoid and antioxidant assays of *Spirogyra majuscule*

Extract	Total Phenol (mg GAE/100 g DW)	Flavanoid	Antioxidant	Reducing power assay
Sohxlet	590	250	54.8	0.050
Sonication	450	360	50.7	0.045

From Table-4 it is clear that the sonicated extract of *Spirogyra majuscule* showed 96 ± 0.04 and 91 ± 0.04 higher antifungal activities against *Fusarium oxysporum* and *Curvularia lunata* respectively.

Table 4: Spore germination inhibition activity of *Spirogyra majuscule*

Extract (0.05 g)	Standard (Fluconazole)	Control	*Fusarium oxysporum* NCIM 1281	*Curvularia lunata* NCIM 1131
Sohxlet	97% ± 0.04	Distilled water – 0% DMF– 0%	95.3 ± 0.04	90 ± 0.04
Sonication			96 ± 0.04	91 ± 0.04

Key: $p < 0.05$ - Significant

IV. CONCLUSION

The easy availability, maintenance and antioxidant assay of Spirogyra makes it a best suited candidate as antioxidant. Both sonicated and hot methods of extraction did not affect the biological activity. The sonicated sample showed higher antifungal activities against *Fusarium oxysporum* and *Curvularia lunata.*Isolation of the active component and further testing can throw further light on the obtained results.

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