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An Assessment of Antioxidant Activity, Phytochemical screening and antifungal Efficiency of red sea weeds, *Gracilaria corticata* J.Ag., and *Gracilaria follifera (Forssk.) Boergs*

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ABSTRACT:-The global biodiversity can be terrestrial as well as marine. The marine diversity is seen to be higher than terrestrial one. Algae being one of the most important plant species in the sea have high photosynthetic capacity. Algae are considered to be primitive plants. In the present investigation, studies on the phytochemical, anti-fungal, antioxidant, flavonoid and phenol properties of *Gracilaria corticata* and *Gracilaria follifera* were conducted with the help of three different solvent extracts like hexane, ethyl acetate and ethyl acetate+hexane mixture. The phytochemical analysis showed the presence of alkaloids, steroids, flavonoids, phenols, coumarins, cardiac glycosides, tannins and saponins in the algae. These phytochemicals have significant application on human pathogens. These algae also have antifungal activity against *Aspergillus niger* and *Fusarium oxysporum*. Seaweeds are rich in antioxidants such as polysaccharides, carotenoids, and pigments. Phenolic compounds and flavonoids are maximum in red seaweeds. The main objective of the present study was to determine the antioxidant, phytochemical and antifungal efficiency of red seaweed *Gracilaria corticata* and *Gracilaria follifera* collected from Thikkodi coast.

Index Terms - *Gracilaria corticata* J.Ag.,Antioxidant Activity,Phytochemical screening, *Gracilaria follifera (Forssk.) Boergs* and antifungal efficiency

INTRODUCTION:-

The global biodiversity can be of terrestrial as well as marine. The marine diversity is seen to be higher than the terrestrial one. Algae being one of the most important plant species in sea have high photosynthetic capacity. In fact a major share of photosynthetic production resulting in the release of huge volumes of oxygen into the atmosphere comes from algal photosynthesis. Algae are considered to be primitive plants. Some of them are unicellular and some other multicellular

Seaweeds have high tolerance against the salinity in water. They have the capacity to maintain the osmolarity between the cytoplasm of algal cell and sea water. Seaweeds contain protein, iodine, bromine, vitamins, polysaccharides, steroids, dietary fibers, chlorophyll, xanthophyll, vitamin A , B, C and E, minerals, lipids, omega- 3 fatty acids, essential amino acids, high amount of ash and sulphates. They also contain storage polysaccharides like laminarin and floridean starch, saturated and unsaturated fatty acids.

Algae also possess antioxidant, and anti-chemical properties.The antioxidant effect varies with different species of algae like red, green and brown. Antioxidants are molecules, capable of inhibiting the oxidation of substances and protect nature (Govindasamy *et al.*, 2012). Antioxidants are used against different diseases like cancer, heart disease etc. It is one of the most important parts in nutrient supply. Antioxidants are mainly used for the removal of free-radicals. Free radicals cause diseases in the body of organisms. The use of algae is increasing day by day all over the world mainly in the medical field. In recent years broad range of studies involving algae occur in different parts of the world. Seaweeds are also used in other fields like cosmetology, industrial pharmaceutical, fertilizer, textile, paper, dairy and confectionary. They are also used for the preparation of commercial products like Agar-Agar, carrageenan, diatomaceous earth etc. The present study aims to evaluate the phytochemical analysis, anti-fungal activity and anti-oxidant activity of red algae *Gracilaria corticata* and *Gracilaria follifera* which are collected from the Thikkodi coast of Kerala.

Materials and methods:-

Study area: The algal samples were collected from Thikkodi (11^0 $29^{'}$N lat&75^0 $37^{'}$ E long) during February 2021. The station has an extensive rocky promontory with small bays of sand and possess rich algal vegetation. There is no fresh water influence.

Collection of seaweeds: Marine samples of two species of seaweeds namely *Gracilaria corticata* J. Ag., and *Gracilaria follifera* (Forssk.) Boergs (Rhodophyceae) were collected from Thikkodi.The samples were immediately washed with seawater to remove the adhered sand and brought in plastic bags to the laboratory. The samples washed thoroughly with tap water to remove attached epiphytes and adhered dirt particles. Then they were again washed thoroughly three to four times with water for removing the debris and sand particles from seaweeds.

Preparation of seaweed powder: The water was drained off and seaweeds were spread on blotting paper to remove excess water. All the samples were dried at room temperature followed by 40^0 c in the hot air oven for two days. Then they were powdered and kept in airtight plastic bottles at room temperature.

Preparation of solvent extracts:50 gm dried crushed samples were extracted using each 250 ml solvent of hexane, ethyl acetate and ethyl acetate+hexane using Soxhlet apparatus.

Qualitative phytochemical screening

Phytochemical screening was carried out by using standard procedure described by Harborne (1998).

Total phenolic content

The amount of total phenolics in the extract were determined with Folin-ciocalteu reagent according to the method of Singleton and Rossi (1965).

Total flavonoid content

Calorimetric technique was used for flavonoid estimation (Chang *et al., 2002*).

Antioxidant assay

DPPH radical scavenging assay

Scavenging effects of samples for DPPH radical were monitored according to the method of Yen & Chen (1955).

Data analysis

The data was presented as a mean standard deviation (n=3). The concentration of solvent and DPPH between the extracts was calculated using regression analysis. The significance of the variations in concentrations among the different solvents and DPPH of *Gracilaria corticata* and *Gracilaria follifera* was tested using analysis of variance (ANOVA) at 95 percent confidence level (5 percent level of significance). To determine the correlation coefficient, a linear regression analysis was done. The t-test and $P< 0.05$ were used to determine statistical significance.

Results:-

Phytochemical substances such as alkaloids, steroids, flavonoids, phenols, coumarins, cardiac glycosides, tannins, terpenoids, and saponins were determined in various extracts (Hexane, Ethyl acetate, Ethyl acetate +Hexane) of two different species of algae (*Gracilaria corticata and Gracilaria follifera*) (Table 1).

Alkaloids are very abundantly present in ethyl acetate and ethyl acetate + hexane extract of *Gracilaria corticata* and *Gracilaria follifera.* Steroids are absent in hexane extract of both the algae but abundantly present in ethyl acetate + hexane extract of *Gracilaria follifera*. Flavonoids are absent in hexane, ethyl acetate and ethyl acetate+ hexane extract of *Gracilaria corticata* and abundantly present in ethyl acetate + hexane extract of *G. follifera*. Phenols are very abundantly present in ethyl acetate + hexane extract of both seaweeds and also very abundantly present in ethyl acetate extract of *Gracilaria follifera*. Coumarins are very abundantly present in hexane, ethyl acetate and ethyl acetate + hexane extract of *Gracilaria follifera*. Cardiac glycosides are very abundantly present in hexane, ethyl acetate, ethyl acetate + hexane extract of *G.follifera* and abundantly present in hexane & ethyl acetate extract of *G. corticata*. Tannins are absent in hexane & ethyl acetate extract of *Gracilaria corticata* and abundantly present in hexane, ethyl acetate, ethyl acetate + hexane extract of *G. follifera*. Terpenoids are absent in hexane and ethyl acetate extract of *Gracilaria corticata* and very abundantly present in ethyl acetate and ethyl acetate + hexane extract of *Gracilaria follifera*. Saponins are very abundantly present in hexane, ethyl acetate and ethyl acetate + hexane extract of both seaweeds.

Antifungal activity of hexane,ethyl acetate and ethyl acetate +hexane extract of *Gracilaria corticata* and *Gracilaria follifera* were studied.(Table 2&3). The extracts were screened for antifungal activity by well-diffusion method using *Aspergillus niger* and *Fusarium oxysporum* as test organisms. No antifungal activity was shown by *Aspergillus niger* and *Fusarium oxysporum* in hexane extract of G.corticata. In the ethyl acetate extract of *G.corticata* both the fungi are grown in two wells. In the 1st well of *Aspergillus niger* the zone of inhibition is 8 mm,where as in 2nd well it is 7 mm. So the average inhibition zone of *Aspergillus niger* is 7.5 mm. In the case of *Fusarium oxysporum*, inhibition zone of 1st well is 10 mm and inhibition zone of 2nd well is 8.5 mm. So the average inhibition zone of *Fusarium oxysporum* in ethyl acetate extract is 9 mm.In the ethyl acetate + hexane extract of *G. corticata* both the fungi are grown in two wells. In the 1st well of *Aspergillus niger*, the zone of inhibition is 10.5 mm whereas of 2nd well it is 9.5mm. So the average inhibition zone is 10 mm. The inhibition zone of *Fusarium oxysporum* in 1st well is 10.mm and inhibition zone in 2n well is 12 mm. So the average inhibition zone of *Fusarium oxysporum is 11 mm. In control medium both the fungi have no inhibition zone.* Table 3, also shows the result of fungal inhibition in different extracts. In this study both the fungi are grown in hexane, ethyl acetate, ethyl acetate + hexane extract of *Gracilaria follifera*. In the hexane extract of *G. follifera*, inhibition zone of *Aspergillus niger* of 1st well is 7.5 mm and zone of

inhibition in 2nd well is 6.5mm. Average inhibition zone is 7mm.Zone of inhibition of *Fusarium oxysporum* in 1st well is 8 mm. 2nd well is 6.5mm. Average inhibition zone is 7.

In the ethyl acetate extract of *G. follifera* both the fungi are also grown in two wells. In the 1st and 2nd well inhibition zone *Aspergillus niger* is 12 mm. So average zone is 12 mm. The inhibition zone of *Fusarium oxysporum* in both wells are 10.5 mm. So average zone is 10.5mm.In the ethyl acetate + hexane extract of *Gracilaria follifera* both the *fungi* are grown in two wells. Inhibition zone of *Aspergillus niger* in1st well is 12.5 mm and in 2nd well is 14mm. So the average inhibition zone is 13 mm. In the 1st well inhibition zone of *Fusarium oxysporum* is 11 mm and 2nd well as 12 mm. Average inhibition zone is 11.5 mm. In control medium, both the fungi have no inhibition zone. In *Gracilaria follifera* the highest activity was shown by *Aspergillus niger*(13mm) and *Fusarium oxysporum*(11.5) in ethyl acetate+hexane extract. The antifungal activity of extracts were compared with standard antibiotic Ampicillin, antibiotic showed inhibition zone on all two organisms. Highest activity was shown against *Fusarium oxysporum* (29mm) (Table-4). In all the experiments, the petriplates which were kept as control showed no zone of inhibition. Abundant fungal growth were present in all the plates. control showed no zone of inhibition. Abundant fungal growth were present in all the plates.

The phenol content of various extracts of seaweed varied from 9.97±0.21mg/g to 5.75 ± 0.21mg/g. In *Gracilaria corticata* the maximum phenol content was recorded in ethyl acetate + hexane mixture extract (9.97 ± 0.21) and minimum in hexane extract (5.75 ± 0.21mg/g). But no flavonoid contents were recorded in the hexane, ethyl acetate, ethyl acetate+hexane extract of *Gracilaria corticata*.In *Gracilaria follifera*, the maximum phenol content was recorded in ethyl acetate + hexane extract (14.77 ±0.19mg/g) and minimum in hexane extract (10.25 ± 0.25mg/g).The flavonoid content when analysed ranged from 44.4 ± 0.36 mg/g to 24.18 ± 0.28 mg/g. The maximum content was recorded in ethyl acetate extract (44.4 ± 0.36 mg/g) and minimum in ethyl acetate + hexane extract (24.18 ± 0.28 mg/g).A positive relation has been documented between antioxidant capabilities and total flavonoid content for *Gracilaria follifera,* but not with phenol content. (Table 5-7)

DPPH free radical scavenging activity of hexane seaweed extract of *Gracilaria corticata* reported highest value of 37.43 ± 0.52% at 500 µg/ml concentrations and lowest value of 32.29 ± 0.30% at 100 µg/ml concentrations. In *Gracilaria follifera*, the reported highest value was 30.49 ± 0.51% at concentration 500 µg/ml and lowest was 20.94 ± 0.06% at concentration 100 µg/ml. (Table 8).

DPPH free radical scavenging activity of ethyl acetate seaweed extract of *Gracilaria corticata* reported highest value of 31.33 ± 0.46% at 500 µg/ml concentrations and lowest value of 24.46 ± 0.45% at 100 µg/ml concentrations. In *Gracilaria follifera*, the reported highest value was 28.58 ± 0.54% at concentration 500µg/ml and lowest value was 21.38 ± 0.54% at concentration 100µg/ml. (Table 9).

DPPH free radical scavenging activity of ethyl acetate + hexane seaweed extract of *Gracilaria corticata* reported highest value of 26.92 ± 0.06% at 500 µg/ml concentration and lowest value of 22.50 ± 0.70 % at 100 µg/ml concentration . In *Gracilaria follifera*, the reported highest value was 25.88 ± 0.01% at concentration 500 µg/ml and value was 17.45± 0.63 % at concentration 100 µg/ml.(Table 10). In Gracilaria corticate a linear relationship was obtained when a graph was plotted for the concentration of the solvent and the DPPH inhibition with a correlation coefficient value ($r2$) of GC-H, GC-EA and GC-EA+H are 0.991, 0.989 and 0.999 respectively, it can be seen that the coefficient for DPPH-H,DPPH-EA and DPPH-EA+H are 0.013, 0.016, 0.011 respectively which is significant at 0.0003,0.0005 and 2.97 level. In this case two p-values are below 0.05 level .Smaller the P-values the greater the probability Therefore a rectilinear regression coefficient equations were GC-H(y=0.0132x + 31.073,$R2$ =1),GC-EA(y=0.0168x + 22.753,$R2$ =1, and GC-EA+H(y=0.0111x + 21.431, $R2$ =1) respectively. When a graph was plotted for the concentration of the solvent and the DPPH inhibition in Gracilaria corticate, a linear relationship was obtained with a correlation coefficient value (r2) of 0.991, 0.989, and 0.999, respectively. It can be seen that the coefficient for DPPH-H, DPPH-EA, and DPPH-EA+H are 0.013, 0.016, 0.011, respectively, which is significant at 0.0003,0.0005 Two p-values are below the 0.05 level in this example. The higher the likelihood, the lower the P-values. GC-H(y=0.0132x + 31.073,R2 =1), GC-EA(y=0.0168x + 22.753,R2 =1), and GC-EA+H(y=0.0111x + 21.431, R2 =1) were the rectilinear regression coefficient equations, respectively. (Table 11)

DISCUSSION:-

This Phytochemical analysis revealed that alkaloid, phenol, coumarin, cardiac glycosides and saponins are present in all the three tested extract of *Gracilaria corticata* and *Gracilaria follifera*. But steroids, flavonoids,tannins and terpenoids are not present in all the three extracts.

Flavonoids were present all the three extracts of *Gracilaria follifera*. Phenols were present in higher amounts only in ethyl acetate +hexane mixture of *Gracilaria corticata* and *Gracilaria follifera*.

These phytochemical compounds are found present in a variety of medicinal plants and have significant applications against human pathogens, including those that cause enteric infections. Thus the above study reveals that phytochemicals like alkaloids, phenols, coumarins, cardiac glycosides and saponins are present in both the algae *Gracilaria corticata* and *Gracilaria follifera.*

The zone of inhibition obtained from the ethyl acetate extracts of both seaweed *Gracilaria corticata and Gracilaria follifera* against fungal pathogens *Aspergillus niger* and *Fusarium oxysporum* were comparatively very less when compared to ethyl acetate+ hexane mixture solvent extract. The antifungal activity was compared with the antifungal activity of standard antibiotic ampicilin.. Ampicilin has highest activity against *Fusarium oxysporum* than *Aspergillus niger*. *Aspergillus niger* is an asexual saprophytic fungus which contain several toxins, some harmless and others harmful to certain people, occassionaly resulting death. *Fusarium oxysporum* is a common inhabitant of soil. The control medium hexane, ethyl acetate, ethyl acetate +hexane (EH) have no sensitivity against the ampicillin.

Gracilaria corticata has no flavonoid content in all three solvent extracts. Phenol content was high in mixture and low in hexane. In *Gracilaria follifera* flavonoid was high in ethyl acetate and low in mixture.

In *Gracilaria corticata* and *Gracilaria follifera* more phenolic content was observed in ethyl acetate +hexane extract and lower in hexane extract flavonoids and phenol level of *Gracilaria follifera* is seen to be relatively high when linked to other observed activities. Both the algae possess high total phenolic content and show good free radical scavenging activity The free radical scavenging activity increased as the concentration increased in all the extracts.

A positive relation was seen between antioxidant and flavonoid content. There was an inverse relationship between phenolics and antioxidant activity. No direct relationship was found between antioxidant activity and total phenolic content .Suggesting that phenol play a minor role in antioxidant activity. And the other compounds such as polysaccharides, organic acid was found to contribute in the overall antioxidant activity.

The bioactive potentials of *Gracilaria* species collected from Thikkodi region were evaluated. All the two species were found to have good antioxidant and antifungal capacities and thus could be potential rich source in various medical fields.

CONCLUSION:-

In conclusion,the aim of the present study was to evaluate the phytochemical,antifungal and antioxidant activities of two different species of *Gracilaria.*In this study, phytochemicals of *Gracilaria corticata* and *Gracilaria follifera* were analyzed qualitatively with different solvents like hexane, ethyl acetate and ethyl acetate +hexane mixture.

The highest phytochemical activity was shown by GF_E and GF_M, lowest in GC_H. *Gracilaria follifera* showed highest antifungal activity than *Gracilaria corticata*. The highest content of total phenols were present in ethyl acetate +hexane extract of *Gracilaria follifera*, while the lowest content of total phenols were present in hexane extract of *Gracilaria Corticata.* The marine macroalgae collected from Thikkodi coast exhibited antifungal activities against the fungus *Aspergillus niger* and *Fusarium oxysporum*. The result shows that *Gracilaria corticata* and *Gracilaria follifera* are promising species for antifungal and antioxidant properties. Both of the algae show DPPH free radical scavenging activity. Hence they can be used in different medical applications.

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Table 1. Qualitative phytochemical analysis of various extracts of *Gracilaria corticata and Gracilaria follifera*

Solvents	**Hexane**		**Ethyl acetate**		**Ethyl acetate + Hexane(EH)**	
Seaweeds	GC_H	GF_H	GC_E	GF_E	GC_{EH}	GF_{EH}
Alkaloids	+ +	+ + +	+ + +	+ + +	+ + +	+ + +
Steroids	_	_	+	+	+	+ +
Flavonoids	_	++	_	+++	_	+
Phenols	+	+	+ +	+ + +	+ + +	+ + +
Coumarins	+ +	+ + +	+ + +	+ + +	+	+ + +
Cardiac glycosides	+ +	+ + +	+	+ + +	+ +	+ + +
Tannins	_	+ +	_	+ +	+	+ +
Terpenoids	_	+	_	+ + +	+	+ + +
Saponins	+ + +	+ + +	+ + +	+ + +	+ + +	+ + +

GC – *Gracilaria corticate* ,GF – *Gracilaria follifera*

(+) – Present (trace amount), (++) – Abundant , (+++) – Very abundant

Table 2. Zone of inhibition(mm) in different extracts of *Gracilaria corticata* against two fungal pathogens**.**

Name of organism	Hexane extract Zone diameter(mm)			Ethyl acetate extract zone diameter (mm)			Ethyl acetate + hexane extract zone diameter (mm)			Control Hexane	Control Ethyl Acetate	Control Ethylacetate+hexane
Aspergillus niger	Well 1 Nil Nil	Nil	Average	Well 1 8 8	8	Average 7.5	Well 1 11 10	10.5	Average 10	Nil	Nil	Nil
	Well 2 Nil Nil	Nil	Nil	Well 2 7 7	7		Well 2 10 9	9.5		Nil	Nil	Nil
Fusarium oxysporum	Well 1 Nil Nil	Nil	Nil	Well 1 10 10	10	9	Well 1 10 11	10.5	11	Nil	Nil	Nil
	Well 2 Nil Nil	Nil		Well 2 9 8	8.5		Well 2 12 12	12		Nil	Nil	Nil

Table 3. Zone inhibition (mm) in different extracts of *Gracilaria follifera* against two fungus pathogens

Name of organism	Hexane extract zone diameter(mm)			Ethyl acetate extract zone diameter (mm)			Ethyl acetate & hexane extract zone diameter (mm)			Control Hexane	Control Ethyl acetate	Control Ethyl acetate & Hex-ane
Aspergillus niger	Well1 7 8	7.5	Average 7	Well 1 12 12	12	Average 12	Well 1 12 13	12.5	Average 13	Nil	Nil	Nil
	Well2 6 7	6.5		Well 2 12 12	12		Well 2 15 13	14		Nil	Nil	Nil
Fusarium oxysporum	Well1 8 8	8	7	Well 1 11 10	10.5	10.5	Well 1 11 11	11	11.5	Nil	Nil	Nil
	Well2 7 6	6.5		Well2 11 10	10.5		Well 2 12 12	12		Nil	Nil	Nil

Table 4: Shows sensitivity of the test organisms to Ampicillin

Name of organism	Name of Antibi-otic	Zone of inhibition	Average
Aspergillus niger	Ampicillin	26 26	26
Fusarium oxysporum	Ampicillin	29 29	29
Control Hexane		Nil	Nil
Control Ethyl acetate		Nil	Nil
Control Ethyl acetate +hexane		Nil	Nil

Table 5. Total Phenol and Flavonoid content of various extracts of *Gracilaria corticata* and *Gracilaria follifera*.

Algae	Solvent	Phenol(mg/g)	Flavonoid(mg/g)
Gracilaria corticata	Hexane	5.75 ± 0.21	Nil
	Ethyl acetate	7.85 ± 0.07	Nil
	Ethyl acetate+ Hexane	9.97 ± 0.21	Nil
Gracilaria follifera	Hexane	10.25 ± 0.25	31.75 ± 0.5
	Ethyl acetate	12.71 ± 0.20	44.4 ± 0.36
	Ethyl acetate + Hexane	14.77 ± 0.19	24.18 ± 0.28

Table-6. Student t-test and Anova for DPPH scavenging and Phenol of *G.corticata* and *G. follifera*

One-way Anova(concentration of extracts and DPPH variations.

Seaweed	Multiple R	R-square	df	Slope	Y-intercept	t-value	P-value	95%Confidence level	
								Lower	Upper
G.corticata	0.972	0.946	3	-0.389	19.212	-4.193	0.149	-1.567	0.789
G.follifera	0.726	0.528	3	-0.965	35.71	-1.056	0.482	-12.57	10.640

Table-7 Student t-test and Anova for DPPH scavenging and Flavonoid of *G. follifera*

Seaweed	Multiple R	R-square	df	Slope	Y-intercept	t-value	P-value	95% Confidence level	
								Lower	Upper
G.follifera	0.888	0.782	3	5.3176	93.97	1.895	0.309	- 30.323	40.959

One-way Anova(concentration of extracts and DPPH variations.

Table 8. DPPH free radical scavenging activity of Hexane seaweed extract.

No	Concentration (µg/ml)	% Activity (± SD)		
		Standard ascorbic acid	*Gracilaria cor-ticata*	*Gracilaria follifera*
1	100	63.45 + 0.64	32.29 ± 0.30	20.94 ± 0.06
2	200	65.49 + 0.69	33.76 ± 0.32	22.95 ± 0.08
3	300	65.95 + 0.06	34.98 ± 0.03	23.25 ± 0.40
4	400	67.45 + 0.55	36.63 ± 0.53	25.49 ± 0.69
5	500	70.05 + 0.47	37.43 ± 0.52	30.49 ± 0.51

Table 9.DPPH free radical scavenging activity of Ethyl acetate seaweed extract

No	Concentration (µg/ml)	% Activity (± SD)		
		Standard ascorbic acid	***Gracilaria corticata***	***Gracilaria follifera***
1	100	56.65 + 0.34	24.46 ± 0.45	21.38 ± 0.54
2	200	56.92 + 0.12	26.39 ± 0.55	23.57 ± 0.58
3	300	58.49 + 0.69	27.33 ± 0.46	25.72 ± 0.09
4	400	60.49 + 0.69	29.44 ± 0.62	26.94 ± 0.06
5	500	62.6 + 0.57	31.33 ± 0.46	28.58 ± 0.54

Table 10.DPPH free radical scavenging activity of Ethyl acetate+Hexane seaweed extract

No	Concentration µg/ml	% Activity (± SD)		
		Standard Ascorbic acid	***Gracilaria corticata***	***Gracilaria follifera***
1	100	52.51 ± 0.67	22.50 ± 0.70	17.45 ± 0.63
2	200	54.59 ± 0.42	23. 66 ± 0.26	20.44 ± 0.62
3	300	54.95 ± 0.05	24.78 ± 0.13	22.53 ± 0.54
4	400	57.96 ± 0.13	25. 87 ± 0.15	23.85 ± 0.06
5	500	58.45 ± 0.63	26.92 ± 0.06	25.88 ± 0.01

Table-11.Student t-test and Anova for DPPH scavenging and concentration of the

extracts of *G.corticata* and *G. follifera*

Parameters	MultipleR	R-square	df	slope	Y-intercept	t-value	P-value	95%Confidence level	
								Lower	Upper
DPPH&H-GC	0.996	0.991	4	0.0132	31.073	18.59	0.0003	0.011	0.015
DPPH&EA-GC	0.994	0.989	4	0.168	22.753	16.15	0.0005	0.013	0.020
DPPH&EA+H-GC	0.999	0.999	4	0.0111	21.431	90.53	2.97	0.010	0.011
DPPH&H-GF	0.936	0.877	4	0.0216	18.132	4.62	0.019	0.006	0.037
DPPH&EA-GF	0.993	0.988	4	0.0178	19.907	15.64	0.0005	0.014	0.021
DPPH&EA+H-GF	0.990	0.980	4	0.0203	15.949	12.39	0.0011	0.015	0.025

H-Hexane, EA-Ethyl acetate, GC-*Gracilaria corticata, Gracilaria follifera*

One-way Anova(concentration of extracts and DPPH variations.Significant (P< 0.05)