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Research Article

Evaluation of Phytochemical Analysis, Anti Oxidant and Anti-Elastase Activity of *Hemigraphis colorata*

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ABSTRACT

Plants have always played a major role in the treatment of human diseases. Worldwide interest in the use of medicinal plants is increasing nowadays. *Hemigraphis colorata* belonging to the family Acanthaceae, was used for the treatment of various diseases in traditional system of medicine. In this study phytochemical analysis of *Hemigraphis colorata*, its anti oxidant effect and anti elastase activity were evaluated. The antioxidant effect of alcoholic and aqueous extracts of *Hemigraphis colorata* was studied by DPPH radical scavenging assay and by reducing power. The primary phytochemical screening showed the presence of carbohydrates, proteins, alkaloids, phenolic compounds, flavonoids, oils, steroids and terpenoids in alcoholic extract and the presence of alkaloids, flavonoids, and phenolic compounds in aqueous extracts. The quantitative phytochemical analysis revealed that the total phenol, flavanoids and flavanols contents were 315, 13.68 and 6.27 mg and 440,7.43 and 6.27 mg respectively for alcoholic and aqueous extract of *Hemigraphis colorata*. For both the extracts the antioxidant activity was found to be increased with increasing concentration. The effect of the plant extracts on the Porcine Pancreatic Elastase was also evaluated and showed inhibition on the activity of the enzyme. The percentage inhibition of elastase was found to be 53.31%, 58.80%, 63.23% for aqueous extracts and 39.20%, 44.80%, 50.50% of ethanolic extract with the concentration of 0.5, 1.0 and 1.5 mg/ml of extract in DMSO The 50% inhibitory concentration was found to be 0.109 and 1.485 mg/ml for aqueous and alcoholic extracts respectively. This result supports the traditional use of *Hemigraphis colorata* as an effective anti ageing, wound healing, anti-wrinkling and anti cancer agent.

Keywords: Antioxidant Activity, Anti elastase activity, wound healing, Phytochemical analysis, *Hemigraphis colorata*.

INTRODUCTION

Plant based products in health care and medicine has been in practice since times immemorial. Many of the pharmaceuticals currently available have a long history of use as herbal remedies. Medicinal plants occupy a major position in areas of cancer and infectious disease treatment. In a number of degenerative diseases like cirrhosis, diabetes, atherosclerosis, wound healing *etc*, reactive oxygen species are involved. Plant derived antioxidants such as tannins, lignin, phenolic acids, flavonoids, flavones, anthocyanin, and proanthocyanins seems to delay the onset of such diseases. Recent reports indicated that there is an inverse relationship between dietary intake of antioxidant rich foods and the incidence of human diseases[1]. Therefore, the commercial development and utilization of plants as source of antioxidants to enhance health and food preservation is of current interest. Insufficient levels of natural elastase inhibitors has been suggested as a contributing factor in a number of diseases including acute lung injury ,cystic fibrosis, ischaemic reperfusion injury, rheumatoid arthritis, atherosclerosis, psoriasis, and malignant tumors. A number of plants have been screened for therapeutic elastase inhibitors and as a source of natural antioxidants that may be of use in wound healing, anti-ageing and ant wrinkle treatments.[2,3]

Hemigraphis colorata is a tropical showy species growing in dense evergreen forests. This is a prostrate herb with rooting branches. . It is also used for the treatment of abdominal pain, glossitis, stomatitis, urolithiasis, acute wounds, diabetes, and anthelmentic [4,5,6,7,8.].and screened the volatile organic pollutant removal efficiency of the plant [9]also.This plant species has been used in folklore medicines for various skin desease. However, it is not scientifically validated. The present study investigation is done on phytochemical analysis of aqueous and alcoholic extracts of *H. colorata* and their effect on free radical scavenging and Porcine Pancreas elastase.

MATERIALS AND METHODS

Plants used in this study was *Hemigraphis colorata* ,which was collected from AMPRS, Kerala agricultural university research station, Odackali, Eranakulam and were cultivated..The plant material (leaves) were shade dried at ambient temperature and then ground in a grinder. The enzyme- Elastase Type IV from porcine pancreas, substrate-N-Succinyl-Ala-Ala-Ala-p-nitroanilide(SANA),Soybean Trypsin inhibitor, DPPH (1,1 diphenyl-2-picryl hydrazyl hydrate) were purchased

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from Sigma. Tris-HCl- buffer and ascorbic acid were supplied by Merck.

Preparation of extracts: Alcohol and aqueous extracts of *Hemigraphis colorata* were prepared by soxhlet extraction method. 10 g of dried finely powdered shade dried leaves were extracted in 150 ml distilled water and ethanol for 72 hours till the extraction was completed. The crude extracts were obtained by using rotary evaporator.

Qualitative Phytochemical Screening: The alcoholic and aqueous extracts of *Hemigraphis colorata* were subjected to qualitative phytochemical screening, using the methods given in the standard books [10,11]

Quantitative Phytochemical Analysis[12,13]

Total Phenolic compounds: The amount of phenol in the aqeous and alcoholic extracts of *Hemigraphis colorata* was determined by the following method. Two hundred microlitres of sample (100µg/ml) were taken in test tubes (3 replicates); 800 µl of Folin-Ciocaleu reagent (diluted ten-fold) & 2 ml (7.5%) sodium carbonate was added. The tubes were mixed & keep in dark for 2 hrs and absorbance of the samples was measured at 765nm using UV/visible light against a blank, which contained 200 µl of water in place of sample. The total phenolic content was expressed as mg Gallic acid equivalents /g of plant extract and calculated as C = c. V/ m, C - Total content of phenolic compound, mg/g plant extract in GAE, c- The concentration of gallic acid established from the calibration curve, mg/ml,V-volume of extract, ml, m-The weight of pure plant methanolic extract, g

Total flavonol content: The content of flavonols was determined by the following method in rutin equalents. The standard calibration curve was prepared by mixing 2ml of 0.5, 0.4, 0.3, 0.2, 0.166, 0.1, 0.05, 0.025 and 0.0166 mg/ml rutin in ethanol with 2 ml(20g/l) Aluminium Trichloride and 6 ml (50 g/l) sodium acetate. The absorption at 440 nm was read after 2.5 hours at 20^{o} C. The same procedure was carried out with 2ml of plant extract (10 g/l) instead of rutin solution. All determinations were carried out in duplicates. The content of flavonols, in rutin equivalents (RE) was calculated by the following formula: X=C.V/m Where: X-flavonol content, mg/g plant extract in RE; C-the concentration of rutin solution, established from calibration curve, mg/ml; V, m-the volume and the weight of plant extract, ml/g.

Determination of flavonoid content: The content of flavonoids was determined using rutin as a reference compound. One ml of plant extract in methanol (10 g/l) was mixed with 1 ml aluminium trichloride in ethanol (20 g/l) and diluted with ethanol to 25 ml. The absorption at 415 nm was read after 40 min at 20^{o}C.Blank samples were prepared from 1 ml plant extract and 1drop acetic acid, and diluted to 25ml.The absorption of rutin solutions was measured under the same conditions. Standard rutin solutions were prepared from 0.05 g rutin. All determinations were carried out in duplicate. The amount of flavonoids in plant extracts in rutin equivalents (RE) was calculated by the following formula: $X=(A.m_0.10)/(A_0.m)$ Where: X-flavonoid content, mg/g plant extract in RE; A-the absorption of plant extract solution; A_0-the absorption of standard rutin solution; m-the weight of plant extract, g;m_0-the weight of rutin in the solution.

Evaluation of *In Vitro* Antioxidant Activities of Plant Extracts

Assay of reducing power [14]: Substances, which have reduction potential, react with potassium ferricyanide (Fe^{3+}), which then reacts with ferric chloride to form ferric ferrous complex that has an absorption maximum at 700nm. Various concentrations of extract (50, 100, 150µg/ml) as well as Ascorbic acid in 1 ml of 5% DMSO were mixed with 2.5 ml phosphate buffer (0.2M, pH 6.6) and 1 % potassium ferricyanide (2.5m).The mixture was incubated at 50^{0}C for 20min. Aliquots of Trichloro acetic acid (2.5ml, 10%) were added to the mixture, which was then centrifuged at 3000 rpm for 10 min. The upper layer of solution (2.5 ml) was mixed with distilled water (2.5ml) and a freshly prepared $FeCl_3$ solution (0.5 ml 0.1%). The absorbance was measured at 700 nm. Increased absorbance of the reaction mixture indicated

Table.1 : Phytochemical Analysis of Extracts of *Hemigraphis colorata*

SI No	Phytochemical test	*Hemigraphis colorata*	
		Ethanolic extracts	Aqueous extracts
1	Carbohydrate		
	a)Benedict's Test	+	−
	b)Molisch's Test	+	−
	c)Barfoed's Test	+	−
	d)Fehling's Test	+	−
2	Alkaloids		
	a)Wagner's Test	+	+
	b)Mayer's Test	+	+
3	Flavonoids	+	+
4	Protein		
	a)Biuret Test	+	−
	b) Millon's Test	+	−
5	Phenolic compounds & Tanins		
	a)Ferric chloride Test	+	+
	b)Lead acetate Test	+	+
6	Oil	+	−
7	Steroids	+	−

Table 2. Quantitative Phytochemical Analysis extracts of *Hemigraphis colorata*

Extracts of *Hemigraphis colorata*	Total phenol content (mg /g plant extract in gallic acid equivalent)	Flavonoid content (mg /g plant extract in rutin equivalent)	Flavonol content (mg /g plant extract in rutin equivalent)
Ethanol	315± 0.72	13.6± 0.08	6.2± 0.07
Aqueous	440±0.51	7.3± 0.13	3.0± 0.15

Table 3: Determination of antioxidant activity

Plant/ standard	Extract/Standard	Concentration (µg /ml)	Reducing power (%)	Radical scavenging activity (%)
Ascorbic acid (Vitamin C)		50	70.5967±0.05840	94.9233 ±.01202
		100	87.2833±0 .58601[b]	94.9700±.00577
		150	94.1133±0.51385[bd]	95.0267±.01453
	Ethanolic extracts	50	62.4800±1.14752[ac]	70.64±.01453[a]
		100	70.1967±1.22129 [abc]	74.82±.0115[ab]
		150	75.620±0.60531 [abcd]	78.246±.0145[abd]
Hemigraphis colorata		50	55.703±0.62269 [a]	73.96±.011[ab]
	Aqueous extracts	100	60.0233±0.02848 [ab]	77.44±.0115[abc]
		150	63.3933±0.40317 [abd]	82.63±.0115[abcd]

Values are expressed as Mean±SE P<0.05, a-signiificantly differ with std of same concentration ,b- with lower concentration, c- significantly differ between extracts & d- with preceeding concentration at the level of significance 0.05

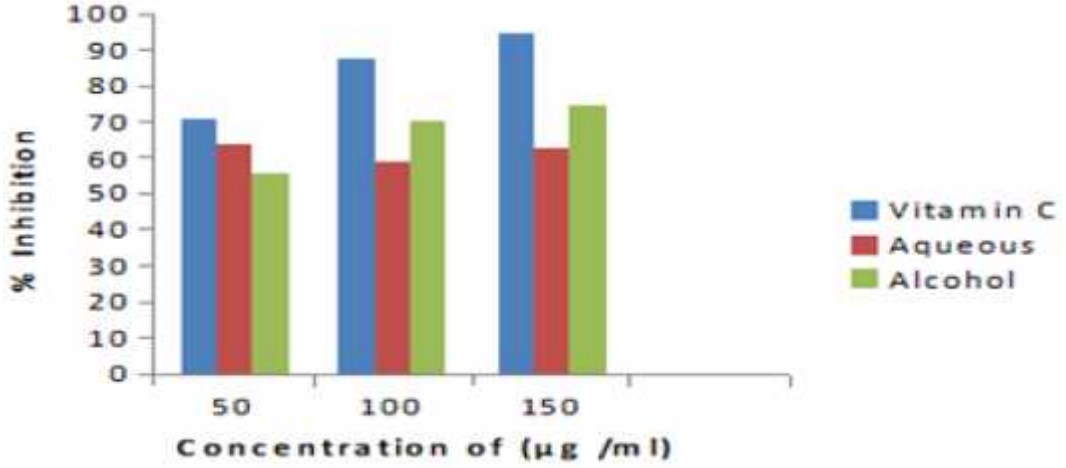

Fig .1: *Reducing power of aqueous & ethanolic extract of Hemigraphis colorata and standard ascorbic acid (Vitamin C)*

increased reducing power. Reducing power is given in ascorbic acid equivalent. The antioxidant activity of the extract was compared with the standard ascorbic acid % increase in reducing power= (A_{test} / A_{blank})-1x10; Where A_{test} is absorbance of test solution; A_{blank} is absorbance of blank

In Vitro DPPH Radial Scavenging Assay[15]: Antioxidants react with DPPH, a stable free radical and get reduced to the DPPH-H.As a consequence the absorbance's decreased from the DPPH radical to the DPPH-H form. The degree of discoloration indicate the scavenging potential of the extracts in terms of hydrogen donating ability by the method of Yohozova *et al* 1998.The free radical scavenging activity of different extracts of sample, L-ascorbic acid (vitamin c) was measured in terms of hydrogen donating or radical scavenging ability using the stable radical DPPH. About $6x10^{-5}$ M solution of DPPH (2,2diphenyl-2-picrylhydrazyl hydrate) in methanol was prepared and 1 ml of this solution was added to 3 ml of extract solution in DMSO at different concentrations (50, 100, 150µg/ml).Thirty minutes later, the absorbance was read at 517nm.Lower the discolouration of reaction mixture , higher the free radical scavenging activity. Radical scavenging activity was calculated by the following formula; % inhibition= (A_B-A_A) /A_B X 100 ; Where; A_B-absorption of blank (methanol and DPPH) sample A_A- absorption of tested extract solution

Effect of *Hemigraphis Colorata* Extracts Towards the Activity of Porcine Pancrease Elastase[16]: Various concentrations of alcoholic & aqueous extracts of the plant in DMSO were taken as test. 0.2ml of both the extract were pre incubated with 0.5ml of porcine pancreas elastase (1mg/ml in tris HCl buffer 0.2M,pH 8) at 25^0C for 15 minutes. After incubation, 0.5ml of substrate (N-Succinyl-Ala-Ala-Ala-p-nitroanilide(SANA)0.8mM in Tris HCl buffer) was added. Then the tubes were incubated for 1 hour at 37'C. Following incubation, 1ml of Soyabean Trypsin Inhibitor (0.2mg/ml) was added to arrest the enzyme reaction. Then 0.5ml of enzyme was added to the enzyme control tube. All the tubes were

vortexed and were placed in ice bath and the optical density was read at 405nm in Perkin Elmer UV/Visible Spectrophotometer. The remaining activity of elastase was calculated in comparison to the control without inhibitor, considering the influence of the buffer, substrate, solvent and test extract. Blanks contained all the components except the enzyme was also taken. IC 50 was determined.

% inhibition of elastase = [(A – B) - (C – D)]/ (A – B) X 100; Where A is the control absorbance, (Enzyme and substrate without extract); B is the control blank absorbance (Substrate without enzyme and extract); C is the sample absorbance (Enzyme and substrate with extract); D is the sample blank absorbance (Substrate and extract without enzyme))

STATISTICAL ANALYSIS

Statistical analysis was carried out and the values were reported as the average of the values from three individual experiments in each case plus Standard Error of Mean (Mean ± S.E.M). Statistical significance was estimated using One Way ANOVA, Tukey Multiple Comparison Test by SPSS 11.5 for windows Software and Regression analysis was carried out for determining IC 50 for each sample.

RESULTS

Preparation of Crude Extracts of *Hemigraphis colorata*: Aqueous & alcoholic extract of *Hemigraphis colorata* were prepared & evaporated to dryness. The yield was found to be 10g/100g plant powder for ethanolic extract & 12g/100g plant powder for aqueous extract.

Phytochemical Analysis: The alcoholic and aqueous extracts of and *Hemigraphis colorata* were analysed for the presence of various phytochemicals. Phytochemical analysis of *Hemigraphis colorata* revealed the presence of alkaloids, flavonoids, phenolic compounds and tannins in aqueous extract; carbohydrates, proteins, alkaloids, flavonoids, phenolic compounds, tanins , oils, steroids and terpenoids in ethanolic extract. Preliminary phytochemical analysis of aqueous and ethanolic extracts of *Hemigraphis colorata* are shown in table-1.

Quantitative phytochemical analysis

Total phenolic compounds: The phenol content of the plant extracts were determined and expressed in Gallic acid equivalence. The values were given in the table as mean ± standard deviation (S.D) of triplicates and expressed as mg/g plant extract in gallic acid equivalence. The content of phenol compounds in alcoholic extract and aqueous extract of *Hemigraphis colorata* were found to be 315± 0.72 and 440±0.51mg /g plant extract in gallic acid equivalents. The present study showed that ethanol extracts of the plant contained much more phenolic compounds in comparison to their aqueous extracts and is given in table 2.

Estimation of flavonoid content: The content of flavonoid compound in ethanolic extract and aqueous extract of *Hemigraphis colorata* were found to be 13.6± 0.08 and 7.3± 0.13mg /g plant extract in rutin equivalents respectively, and is shown in table 3.From the table it can be concluded that the ethanol extracts of *Hemigraphis colorata* contain much more flavonoid content on compare to their aqueous extract.

Estimation of flavonol content: The flavonol content in Ethanolic extract and aqueous extract of *Hemigraphis colorata* were found to be 6.2± 0.07and 3.0± 0.15respectively mg /g plant extract in rutin equivalent, and is given in table-2. From the table we find that the ethanolic extracts contain much more flavonol compared to the aqueous extracts.

In Vitro Evaluation of Antioxidant Activities of Plant Extracts

Determination of reducing power: In the present study shows the reductive capabilities of the aqueous and ethanolic extracts of *Hemigraphis colorata* compared to Vitamin C. The reducing power of ethanolic extracts were 63.66%, 70.37%, 74.46% and of aqueous extracts are 55.62%, 59.11%, 62.63% with concentrations of 50, 100, 150 µg /ml respectively. The reducing power of standard ascorbic acid is given as 70.65%, 87.28%, 94.12% with concentrations of 50, 100, 150 µg /ml respectively. The results are shown in table-3 and are expressed in fig-1

In vitro DPPH radical scavenging assay: The percentage of DPPH radical scavenging activity of ethanolic extract was found to be 73.96%, 77.44%, 82.63% and that of

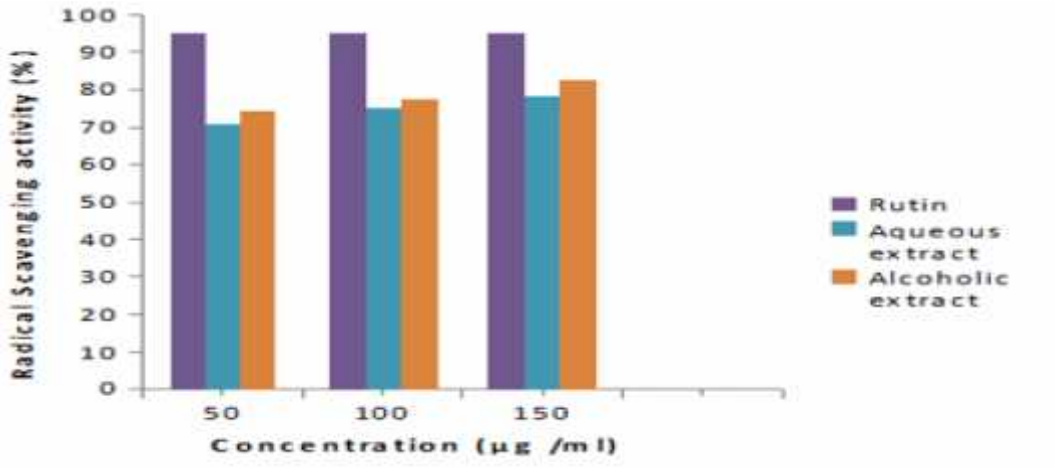

fig 2: *DPPH Radical scavenging activity.*

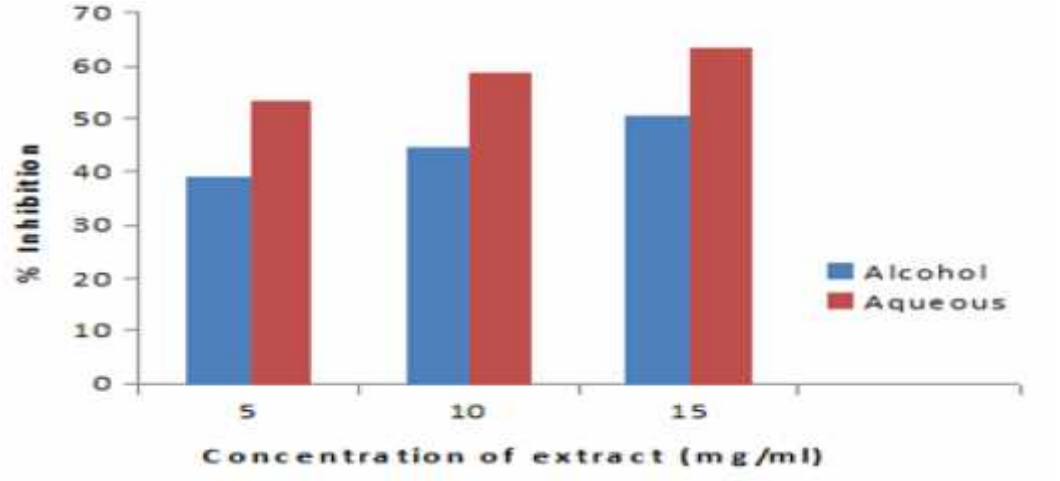

Fig 3*: Effect of aqueous and ethanolic extract of Hemigraphis colorata on % inhibition of elastase*

Table 4: Elastase inhibition assa

Extract	Concentration (mg/ml)	Percentage of Inhibition(%)	IC 50 (mg/ml)
Aqueous extracts of *Hemigraphis colorata*	0.5	53.7367± 0.29384[a]	0.109
	1.0	58.6867± .06960 [ab]	
	1.5	63.3900± 0.08963 [abc]	
Ethanolic extracts of *Hemigraphis colorata*	0.5	39.2667±0.4055	1.485
	1.0	44.7000± .55076 [b]	
	1.5	50.1667± 0.60093 [bc]	

The values are expressed as Mean±SEM The mean difference is significant at the .05 level ,P value<.05, a- significantly differ from alcohol extract; b- significantly differ from lowest concentration; c- significantly differ from lower concentration

aqueous extract was found to be 70.64%, 74.82%, 78.25%, and of standard rutin was 94.93%, 94.97%, and 95.03% with concentration of 50,100,150 µg /ml respectively. The results are shown in table-4 and were plotted in fig- 2. In the present study the aqueous and ethanolic extract of *Hemigraphis colorata* showed that with the increase in the concentration, percentage of Radical scavenging activity was found to be increased. Also the same concentration of ethanolic extract has high percentage of Radical scavenging activity on compared to the aqueous extracts respectively.

Inhibitory Effect of Aqueous and Alcoholic Extracts of *Hemigraphis Colorata* on Porcine Pancreas Elastase: The elastase inhibition by aqueous extraction was found to be 53.31%, 58.80%, 63.23% and that of ethanolic extract was found to be 39.20%, 44.80%, 50.50% with the concentration of 0.5, 1.0, and 1.5 mg/ml of extract in DMSO. The results was shown in table- 5 and was plotted in the fig- 4.In the present study, the aqueous and ethanolic extract of *Hemigraphis colorata* showed that with the increase in the concentration the percent inhibition towards porcine pancreatic elastase was found to be increased with IC 50 of 0.109 and 1.485 mg/ml respectively for aqueous and alcohol extracts.

DISCUSSION

Medicinal plants are immensely rich sources of new pharmaceuticals, cosmetics, agro chemicals and other economically important chemicals. Presently numerous pure compounds are isolated from plants for many therapeutic purposes. The plant *Hemigraphis colorata* is a tropical showy plant mainly used for wound healing in traditional systems of medicine. The preliminary phytochemical screening showed the presence of carbohydrates, proteins, phenolic compounds, alkaloids, flavanoids, terpenoids, steroids, and oils in alcoholic extracts and that of alkaloids, flavanoids and phenolic compounds in aqueous extracts. The phytochemical analysis indicated that ethanol can extract more active principles of the plant than the water.

Quantitative analysis of phytochemicals revealed the total amount of phenolic compounds, flavonols and flavonoids in mg/g plant extracts. The content of phenol compounds in alcoholic extract and aqueous extract of *Hemigraphis colorata* were found to be 315 and 440 mg /g plant extract in gallic acid equivalent respectively. The content of flavonoid compound in aqueous extract and ethanolic extract were found to be 7.43 and 13.68 mg /g plant extract in rutin equivalent. The flavonol content in ethanolic extract and aqueous extract of *Hemigraphis colorata* were found to be 6.27 and 3.15 mg /g plant extract in rutin equivalent. Thus from tables 4.2, 4.3 & 5.4 it is exhibited that the ethanolic extracts contain much more phytochemicals compared to the aqueous extracts.

It is increasingly being realized that a majority of the present day diseases are due to the shift in the balance of the pro-oxidant and the antioxidant homeostatic phenomenon in the body. Pro-oxidant conditions dominate either due to the increased generation of the free radicals caused by excessive oxidative stress of the current life, or due to the poor scavenging/ quenching in

the body caused by depletion of the dietary antioxidants.[17,18]

The present study aims to assess the *in vitro* antioxidant potential of leaf extract of *Hemigraphis colorata* by using reducing power and DPPH radical scavenging activity. The *Hemigraphis* species possess very good antioxidant activities. The phenolic compounds are effective hydrogen donors, which makes them good antioxidants. This study demonstrated that the aqueous and ethanolic extracts of *Hemigraphis colorata* possess a high DPPH radical scavenging activities compared to standard Vitamin C. The reducing power of aqueous and ethanolic extracts of *Hemigraphis colorata* were increased with quantity of samples. The plant extracts could reduce most Fe^{3+} ions, which had a lesser reductive activity than the standard Vitamin C showed that with the increase in the concentration of extracts, percentage of radical scavenging activity was found to be significantly increased.

Our preliminary data shows that the plant has efficient antioxidant activity and also, with the increase in the concentration, the percentage of Radical scavenging activity and reductive capabilities found to be increased. Also the same concentration of ethanolic extract has high percentage of antioxidant potential as compared to the aqueous extracts. It may be due to two reasons :- 1) the nature of biological active components like alkaloids, flavanoids etc, which may be enhanced in the presence of alcohol 2) the stronger extraction capacity of alcohol may have produced a greater number of active principles responsible for the activity[19].

Elastase are a group of serine proteases that possess the ability to cleave the important connective tissue protein elastin. Elastin is a key component in keeping parts of the body flexible, such as the elasticity in the skin and in the lungs. Elastase inhibitors have been used in the treatment of HNE associated diseases. From table 5:7 & fig.5:3 , the aqueous & alcoholic extracts of *Hemigraphis colorata* showed a considerable inhibition towards porcine pancreatic elastase with IC 50 of 0.109 and 1.485 mg/ml respectively and the inhibitory effect increases significantly with increase in concentration of both extracts. The effect is more for aqueous extract .This suggest that the polar compounds in the plant are responsible for the inhibitory effect on elastase and this result is same as in the reports of Tanaka T *et al* 1993 [20].This is also proved by the high amount of phenol compounds in the aqueous extract (440mg /g Gallic acid equivalents) compared to alcoholic extract (315 mg/g Gallic acid equivalents).

As elastase may be used as a nonspecific indicator to screen inflammation, infection, Cardiovascular complications and obesity elastase inhibitors can be used to treat these complications. Elastase inhibitors are also used as anti ageing and anti wrinkling agent .Thus it can be used in skin and cosmetic industry. Wound healing also require the inhibition of elastase (Subhramaniam *et al*) .The traditional use of *Hemigraphis colorata* as a wound healing agent can be confirmed by this study.

CONCLUSION

One of the important functions of skin is protection from harmful environments. Many studies have explored how to prevent skin from wrinkling and the occurrence of pigmentation changes. Skin wrinkling and pigmentation changes could be caused by unusual disruption of connective tissue, formation of free radicals and ultraviolet radiation. Natural products are interesting, intriguing and extremely popular agents among the human population In this study, extracts obtained from *Hemigraphis colorata* were screened for inhibitory effects on elastase, and for free radical scavenging effects. Investigations were carried out to evaluate the phytochemicals of aqueous & alcoholic extracts of *Hemigraphis colorata.* The result indicated that the crude extract at various concentration showed significant amount of phytochemicals such as phenolic compounds,flavonols and flavonoids. However the amount of phytochemicals in alcoholic extract was considerably more compared to its aqueous extracts[21]. From the study it can be concluded that both aqueous and alcoholic extracts of *Hemigraphis colorata* showed remarkable antioxidant activities like DPPH radical scavenging activity and reducing power. The study also showed that both aqueous & alcoholic extracts of *Hemigraphis colorata* exhibited an inhibition against porcine pancreatic elastase and the effect is more for aqueous extract. From these results we can conclude that the traditional use of this plant for infectious diseases is promising. The result also supports the traditional use of *Hemigraphis* as an effective anti ageing[22], wound healing[24], antiwrinkling[23]and anti cancer agent[22].

FUTURE PROSPECTS

Our study supports the medicinal value of *Hemigraphis colorata* considering its anti oxidant potential and anti elastase activity. Although crude extracts of the plant contain various secondary metabolites, a systematic screening and characterization of these active principles is needed for finding out the specific constituent in the *Hemigraphis colorata*, which is responsible for the anti elastase effect and pharmacological validation in terms of modern medicine will be of great medicinal importance in future. Ancient knowledge coupled with scientific principle, can come to the forefront and provide us with powerful remedies to eradicate diseases and give betterment of mankind.

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