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# Phytochemical investigation and evaluation of antioxidant activities of methanolic extract of *Globba marantina* leaves

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## Abstract
*Globba marantina* is a herbaceous, perennial plant with erect, unbranched stems up to 50cm tall. The plant is harvested from the wild for local use as a food and medicine. The plant used to cure earache and as eye drop to cure conjunctivitis. The small bulbils are used to stimulate the appetite. So, the present study is designed to investigate the possible chemical groups, total phenolic content and antioxidant activities of the crude methanolic extracts of *Globba marantina* leaves belonging to the family Zingiberaceae. The investigation revealed the presence of alkaloid, cardiac glycosides & steroids. Three complementary test systems, namely total phenolic content, total flavonoid content and total flavonol content were used for the analysis of quantitative determination of phenolic compounds. The presence of high phenolic, flavonoid and flavonol content in the extracts have contributed directly to the antioxidant activity. The antioxidant ability and radical scavenging properties of plants were also measured using three different assays, namely DPPH, RPA and FRAP. The leaves extract also exhibited remarkable antioxidant potentials as compared to the reference standard- ascorbic acid (AA) and butylated hydroxytoluene (BHT). Based on the results obtained in this study, the *G. marantina* leaves are the most effective antioxidants as determined through different in vitro assays which could be a potential source of natural antioxidants for use in food, cosmetics and pharmaceuticals industries.

**Keywords:** *Globba marantina*, phytochemical screening, phenolic compounds, antioxidant activity.

## 1. Introduction
Nature has been a source of medicinal agents for thousands of years and an impressive number of modern drugs have been isolated from natural sources, many of which based on their use in traditional medicine. Plant kingdom, the storehouse of thousands of unexplored compounds, possesses a great potentiality for drug search even in the day of synthetic chemistry. It has been noted that the original source of many important pharmaceuticals are plants which are currently used by indigenous people [1]. About 33% of the drugs produced in the developed countries are derived from plants [2]. Phytochemicals and Bioactive compounds can have an influence on health. It has now been established that the plants synthesize and accumulate some secondary metabolites like alkaloids, glycosides, tannins, volatile oils etc. that may possess a great potential for biological activity and can be a curative agent in therapeutic purposes. Bioactivity guided phytochemical investigation of medicinal plants may yield newer chemical constituents of remarkable therapeutic interest such as beneficial to infectious diseases, inflammation, cancer, diabetic and hypertension [3].
Root causes of numerous chronic diseases involve oxidative damage to the cellular components. The use of antioxidants to minimize the oxidative damage is one of the important approaches to the primary prevention of these health problems. Several studies have indicated that the antioxidant activities of some plants were highly correlated with their total phenolic contents and phenolic diterpenes [4-6]. Phenolic compounds are a unique category of

phytochemicals especially in terms of their vast potential health-benefiting properties including simple phenols and phenolic acids derivatives are bioactive substances occurring widely in plants. Phenolic compounds are closely associated with the sensory and nutritional quality of fresh and processed plant foods. Many phenolic compounds in plants are good sources of natural antioxidants.

Many phenolic compounds in plants are good sources of natural antioxidants. It is a great interest in recent years that many phenolic compounds in foods have inhibitory effects on mutagenesis and carcinogenesis [7]. Again, the antioxidant activities of phenolic compounds have been recognized for decades, and research and development on the use of natural substances or food ingredients containing phenolic antioxidants will continue to be of great interest to the food industry. Phenolic antioxidants not only inhibit the autoxidation of lipids, but sometimes, they also have the ability to retard lipid oxidation by inhibiting lipoxygenase activity. It is believed that the metabolism of arachidonic acid to lipid peroxides and various other oxidative products is significant in carcinogenesis [8]. It appears to play an important role in tumor promotion because inhibitors of arachidonic acid metabolism have been observed to inhibit this promotion [9].

Flavones, flavonols and their glycosides also occur widely in the plant kingdom. The most important single group of phenolics in food are flavonoids which consist mainly of catechins, proanthocyanins, anthocyanidins, flavons, flavonols and their glycosides. Some studies showed that flavonoids could protect membrane lipids from oxidation [10]. Therefore, at present study strong emphasis has been given to find natural, effective, and safe antioxidants from dietary plants.

So, facilitated by the rapid development of technology of isolation and characterization process that is chromatographic and spectroscopic methods, a large number of therapeutically active plant constituents have been isolated during last two decades. Recently, investigation of natural products has gained interest due to potential therapeutic effect against infectious diseases as plant extracts and their components (alkaloids, tannins, flavonoids and phenolic compounds) have been known to exhibit health benefits against bacteria, fungi, amoebae and insects. Moreover, Phytoconstituents have gained popularity now-a-days over the synthetic medicaments due to showing little toxicity and side effects [11].

Various recent phytochemical screening and qualitative estimation show that, plants under the genus *Globba* are rich in aminoacids, cardiac glycosides, and fatty acids. Methanolic and ethanolic extracts shown some common metabolites like carbohydrates, aminoacids, cholesterol, steroids, alkaloids, cardiac glycosides, tannins, terpenoids, phlobatanins, fatty acis and phenols with health benefits [12]. So, the ultimate purpose of the present study is to examine the preliminary phytochemical screening and to evaluate the antioxidant potentials of the methanolic extract of *Globba marantina* leaves.

## 2. Methods

### 2.1. Collection of plant materials

For this present investigation, the aforementioned medicinal plant was collected from Chattogram university, Chattogram, Bangladesh and was identified by Bangladesh National Herbarium, Mirpur, Dhaka. The accession number of, *G. marantina* is DACB-43650.

### 2.2. Preparation of plant extract

The collected leaves were rinsed, cut into small pieces and dried under at room temperature for 10to12 days. The leaves were ground into a fine powder using electric blender. About 400 gm of powered material of plant was taken in a clean, flat bottomed glass container and soaked in 1600 ml of 90% methanol. The container with its contents was sealed and kept for a period of 15 days accompanying occasional shaking and stirring. The whole mixture then underwent a coarse filtration by a piece of clean, white cotton material. Then it was filtered through whatman filter paper. The filtrate (methanol extracts) obtained were concentrated under reduced pressure at 40 ºC by using a rotary evaporator. The crude extracts were then dried in a freeze dryer, weighed and stored in desiccators until furtheruse.

### 2.3. Qualitative Phytochemical screening

Testing of different chemical groups present in the extract, represent the preliminary phytochemical studies. Small quantity of partition of methanolic extract of leaves of *Globba marantina* were subjected to preliminary qualitative phytochemical investigation for detection of phytochemicals such as alkaloids, cardiac glycosides, anthraquinone, steroids, saponins, using the standard methods [13]. The detection was carried out based on visual observations of a color change or the formation of precipitates after the addition of specific reagents.

#### 2.3.1. Tests for alkaloid

Freeze-dried extract of plant material (50 mg) was dissolved in 6 ml of 1% aqueous hydrochloric acid following filtration [10]. The filtrate was divided into three test tubes and tested with different alkaloidal reagents as follows:

**A. Mayer's Test**

Filtrates were treated with Mayer's reagent (Potassium Mercuric Iodide). Formation of a white or creamy colored precipitate indicated the presence of alkaloids [14].

**B. Wagner's Test**

Wagner's reagent (Iodine in Potassium Iodide) was added by the side of the test tube. Formation of brown/reddish precipitate indicated the presence of alkaloids [15].

**C. Dragendroff's Test**

Filtrates were treated with few drops of Dragendroff's reagent (solution of Potassium Bismuth Iodide). Formation of orange-red precipitate indicated the presence of alkaloids [13].

#### 2.3.2. Tests for cardiac glycosides (Keller-Killiani test)

One hundred mg of plant extract was dissolved in 2 ml of absolute methanol and this was followed by the addition of 2 ml of glacial acetic acid. Then, one drop of 5% ferric chloride solution and 1 ml of concentrated sulphuric acid were added. The appearance of reddish brown ring at the interface and a bluish green colour in the upper (acetic acid) layer indicate the presence of a deoxysugar characteristic of cardenolides [13].

#### 2.3.3. Tests for Anthraquinone glycosides (Borntrager's test)

One hundred mg of plant extract was dissolved in 5 ml of chloroform and filtered. The filtrate was then shaken well with equal volume of ammonia solution (10% $NH_4OH$). The appearance of pink violet or red colour in the ammoniacal layer (lower) indicates the presence of anthraquinones [14].

**2.3.4 Tests for Steroids**

**A. Liebermann–Burchard test**

50 mg of the plant extract was dissolved in 2 ml of chloroform followed by the addition of 2 ml of acetic anhydride. The mixture was then boiled and cooled. A few drops of concentrated sulphuric acid was added from the side of the test tube. The development of greenish transient colour indicates the presence of steroids [16].

**B. Salkowski Test**

50 mg of extract was dissolved in 2 ml of chloroform. Then, 2 ml of concentrated sulphuric acid was added carefully from the side of the test tube to form a layer. A color development of reddish brown at the interface indicates the presence of steroids [16].

**2.3.5. Test for saponins**

100 mg of the plant extract was shaken vigorously in a test tube with 5 ml of distilled water. The formation of characteristic honeycomb like foam which persisted for ten minutes indicates the presence of saponins [14].

**2.4. Quantitative determination of phenolic compounds of crude extract**

**2.4.1. Determination of total phenolic content (TPC)**

The TPC of the plant extract was determined using the Folin-Ciocalteu method as described by Meda *et al.*, (2005) [17], with minor modifications. The plant extract (0.5 ml of 1 mg/ml solution) was mixed with 2.5 ml of Folin-Ciocalteu reagent (10%) and 2 ml of sodium carbonate (7.5% w/v). After vortexing for 15 s, the reaction mixture was incubated at room temperature in the dark for 30 min and the absorbance was measured at 765 nm using a UV-1601 Schimadzu UV-Vis spectrophotometer. The TPC was calculated using the regression equation from the calibration curve constructed using gallic acid standards (0 to 300 ppm) treated in the same manner as the extract. The results were expressed as mg gallic acid equivalent per gram of dry weight of the extract (mg GAE/g DW).

**2.4.2. Determination of total flavonoid content**

The total flavonoid content was determined according to the method described by Lin and Tang, 2007 [18]. The test samples were dissolved in methanol and 500 µl of this sample solution was mixed with 1.5 ml methanol, 0.1 ml 10% aluminum chloride hexahydrate, 0.1 ml 1M potassium acetate and 2.8 ml distilled water. After 40 min of incubation at room temperature, the absorbance was measured at 415 nm using a UV-visible spectrophotometer. The results were expressed as mg of quercetin equivalents per gram of dry weight of the extract (mg QE/g DW). These values were calculated from a standard calibration curve of quercetin in the concentration range from 0 to 50 µg/ml.

**2.4.3. Determination of total flavonol content**

The total flavonol content was determined using the method of Kumaran and Karunakaran, 2007 [19]. An aliquot of 1 ml of the plant extract (1 mg/mL) was mixed with 1 ml of aluminium trichloride (20 mg/ml in methanol) and 3 ml of 50 g/l sodium acetate solution. After incubation for 2.5 hours at room temperature, the absorbance was measured at 440 nm. Quercetin, treated in the same manner as the sample, was used to produce a standard calibration curve in the range of 0 to 50 µg/ml. The results were expressed as mg of quercetin equivalent (QE) per gram of dry weight of extract (mg QE/g DW).

**2.5. Antioxidant activities of extracts**

**2.5.1. DPPH radical scavenging activity assay**

The DPPH radical scavenging activity of test samples was carried out according to the procedures as described by Shahid-Ud-Daula *et al.*, 2016 [20]. A solution of DPPH (0.1 mM) was prepared using methanol, and 3 ml of this solution was added to 300 µl of various concentrations (5-500 µg/ml) of methanolic extracts. After 30 min of incubation at ambient temperature, the absorbance was measured at 517 nm using a UV-Visible spectrophotometer. Ascorbic acid and BHT were used as positive controls. The inhibitory concentration (IC) was calculated according to the following equation: IC (%) = $(A_0 - A_t)/A_0 \times 100$, where $A_0$ is the absorbance value of blank sample and the absorbance value of the test samples. Percentage (%) inhibition of both standards and test samples were calculated for each concentration and graphs of% inhibition against concentration were plotted. From these graphs, the concentrations that reduce the absorption of DPPH solution by 50% ($IC_{50}$) were calculated. A lower $IC_{50}$ value indicates a greater scavenging or antioxidant activity**.**

**2.5.2. Reducing power activity assay**

The reducing power of the plant extracts and standards (ascorbic acid and BHT) were measured following the procedure described by Oyaizu, 1986 [21]. An aliquot of various concentrations of plant extract (40-500 µg/ml) was mixed with 2.5 ml of sodium phosphate buffer (200 mM, pH 6.6) and 2.5 ml of 1% potassium ferricyanide. The mixture was then incubated in a water bath at 50 ºC for 20 minutes and then cooled. Reaction was then terminated by adding 2.5 ml of 10% (w/v) trichloroacetic acid. The mixture was centrifuged at 3000 rpm for 10 minutes. 2.5 ml of the supernatant was mixed with 2.5 ml distilled water and 0.5 ml freshly prepared 0.1% (w/v) anhydrous ferric chloride solution. The absorbance was measured at 700 nm against the blank. Higher absorbance values indicate stronger reducing power. The results were expressed as the effective concentration ($EC_{50}$) values, the concentration of sample which produced an absorbance value of 0.5 and obtained from a linear regression graph of absorbance against extract concentration.

**2.5.3. Ferric reducing antioxidant potential**

The ferric reducing ability of the plant extracts were determined according to the method described by Benzie & Strain, 1996 [22]. An aliquot of 0.3 ml of various concentrations of plant extract (0-500 µg/ml) was mixed with 3 ml of FRAP reagent. After vortexing for 15 seconds, the reaction mixture was incubated at room temperature in the dark for 5 minutes and the absorbance was measured at 593 nm using a UV-Vis spectrophotometer. The increase in absorbance due to the reduction of ferric to ferrous tripyridyltriazine complex (Fe3+-TPTZ → Fe2+-TPTZ) in the presence of the test sample was recorded at 593 nm. The antioxidant potential ($EC_{50}$) of the sample was calculated from a linear regression analysis and compared with those of the standards, ascorbic acid and BHT.

**2.6 Statistics and Calculations**

In this study, the results were expressed as mean ± standard deviation of at least three replicates. Statistical analyses were carried out using Statistical Package for the Social Sciences (SPSS, IBM Corp., Armonk, NY, USA). Tukey's pairwise

comparison (HSD) and least significant difference (LSD) tests were performed to analyze variance and determine significant differences among mean values (P<0.05) [10].

## 3. Result and Discussion

### 3.1. Phytochemical screening

Qualitative chemical tests were performed for the methanol extract of *G. marantina* leaves. The results of various chemical tests for the detection and identification of chemical constituents are summarized in the following table 1. These chemical tests indicated the presence (alkaloids, steroids & cardiac glycosides) and absence (Saponins & Anthraquinones) of different phytoconstituents.

**Table 1:** Phytochemical screening of methanolic extracts of *G. marantina* leaves

| Phytoconstituent | Test performed | Methanolic extract of Leaves |
|---|---|---|
| Alkaloids | Mayer's reagent | + |
| | Wagner's reagent | + |
| | Dragendoff's reagent | + |
| Cardiac Glycosides | keller-killiani Test | + |
| Anthraquinones | Borntrager's test | - |
| Steroids | Salkowski's Test | + |
| | Liberman-burchard test | + |
| Saponins | Frothing test | - |

'+' indicates presence of the constituents and '-' indicates absence.

Crude methanolic extracts of *G. marantina* leaves were used for the phytochemical screening by using appropriate reagents and proper procedure for the investigation of different chemical groups. *G. marantina* extract revealed the presence of alkaloid, cardiac glycosides, steroids and absence of saponin, anthraquinone. The various phytochemical compounds detected are known to have beneficial importance in medicinal sciences. Presence of alkaloids have been associated with medicinal uses for centuries and one of their common biological properties is their cytotoxicity [23]. Alkaloids also implicated in analgesic activities [24]. Many alkaloids derived from medicinal plants show biological activities like, anti-inflammatory, antimalarial, antimicrobial cytotoxicity, antispasmodic and other pharmacological effects. The extracts also contain cardiac glycosides. Glycosides are known to possess anti-neoplastic properties. Cardiac glycosides show cardio tonic effect and have the potential for management and control of cardiac arrest [25-26]. Steroids also have a wide variety of effects that affect the development, activity, and plasticity of the nervous system and modulate pain [27].

### 3.2. Total phenolic, flavonoid, and flavonol contents

The crude extracts of *Globba marantina* leaves were tested for total phenolic content. Folin-Ciocalteu reagent was used for the test. Based on the absorbance values of the extract solutions the colorimetric analysis of the total phenolic content of extracts were determined and compared with the standard solutions of gallic acid and the total flavonoid content and the total flavonol content of the extract is expressed as quercetin equivalent. The results (Table 2) revealed that the extract of *G. marantina* leaves contained considerable amount of phenolic compounds (129.44 mg GAE/g DW). However, measured total flavonoid content was (97.958mg QE/g DW) and total flavonol content was (120.639mg QE/g DW) in the leaves extract of *Globba marantina*.

**Table 2:** Total phenolic, flavonoid, and flavonol contents of methanolic extracts of leaves of Globba marantina

| Plant extract | Total phenolic content (mg GAE/g DW)a | Total flavonoid content (mg QE/g DW)b | Total flavonol content (mg QE/g DW)b |
|---|---|---|---|
| Leaves | 129.44 ± 0.24 | 97.958± 0.033 | 120.639± 0.047 |

Values are means ± SD (n=3). Within rows, mean values were significantly different (P< 0.01; Tukey's HSD and LSD test) a Total phenolic content is expressed as mg gallic acid equivalents per g dry weight (mg GAE/g DW).

B Total flavonoid and flavonol contents are expressed as mg quercetin equivalents per g DW (mg QE/g DW). Phenolic compounds have multiple biological effects and also act as antioxidants by preventing the oxidation of Low-Density Lipoproteins (LDL), platelet aggregation and damage of red blood cells [28]. These chemical constituents (secondary metabolites) present in plant vary according to their age and maturity. The antioxidant activity of phenolic compounds is mainly due to their redox properties, which can play an important role in absorbing and neutralizing free radicals, quenching singlet and triplet oxygen or decomposing peroxides [29]. Further, phenolic compounds are effective hydrogen donors, which make them antioxidant [30]. Many important medicinal tree species possess antioxidant properties due to the presence of phenolic compounds which may be associated with lower incidence and lower mortality rates of cancer in several human populations [31]. The obtained result is expressed as the number equivalents to gallic acid per gram of the plant extract. The plant extract was found to contain large amount of phenolic content. Thus, the plant extract demonstrated moderate total antioxidant capacity.

Flavonoids are a group of polyphenolic compounds, which exhibit several biological effects such as anti- inflammatory, anti-hepatotoxic, anti-ulcer, anti-allergic, anti-viral, anti-cancer activities. They also inhibit enzymes such as aldose reducatse and xanthine oxidase. Flavonoids and flavonols are capable of effectively scavenging the reactive oxygen species because of their phenolic hydroxyl groups and are potent antioxidants [32]. Flavonoid and flavonol content is increased when concentration of the extract is increased.

So, in view of their wide pharmacological and biological actions, *G. marantina* leaves have a greater therapeutic potential. The presence of high phenolic, flavonoid and flavonol content in the extracts have contributed directly to the antioxidant activity by neutralizing the free radicals and have been shown to be effective in the treatment of various health problems, including neurodegenerative, systemic and infectious diseases [33].

### 3.3. Antioxidant activities

The antioxidant ability and radical scavenging properties of plants are associated with its medicinal values. In this study, the antioxidant activity of *G. marantina* leaves were measured using three different assays, namely DPPH, RPA and FRAP. Performing a single assay to evaluate the antioxidant properties would not give the correct result because antioxidant activity of plant extract is influenced by many factors, for example the test system and composition of extract. Therefore, it is important to carry out more than one type of antioxidant capacity measurement to cover the various mechanisms of antioxidant action.

#### 3.3.1. DPPH radical scavenging activity

The DPPH radical has been considered as a model system to analyze the scavenging activities of many natural compounds

[34]. The DPPH shows their antioxidant activity based on the principle of decolorizing in the presence of free radical scavenger. The color turns purple to yellow is an indication of the reaction in which DPPH radical serves as the oxidizing radical to be reduced by the antioxidant. The change of color is due to the presence of odd electron in DPPH and is also responsible for the absorbance at 517 nm. The methanolic extract of *Globba marantina* showed the antioxidant activity by reacting with DPPH which was reduced to DPPH-H. The degree of antioxidant activity was determined by the discoloration tendency. In the DPPH assay, radical scavenging potential of the methanolic extract of leaves of *Globba marantina* and two reference standards: ascorbic acid (AA) and BHT are presented in Figure 1A demonstrating a concentration dependent radical scavenging effect. The activity of ascorbic acid (95.64%) was the highest, followed by BHT (92.788%) and leaves (88.62%) (Fig 1A).

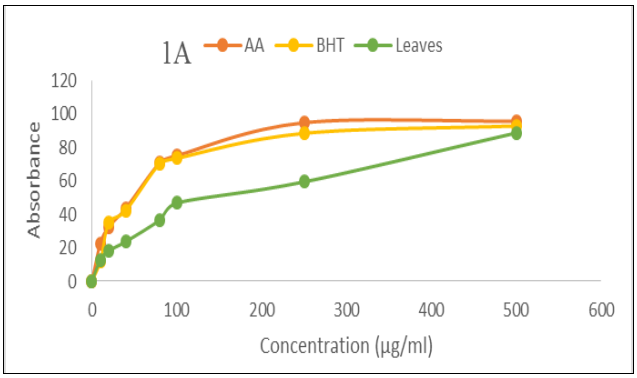


**Fig 1:** DPPH free radical scavenging activity assay of methanolic extract of *Globba marantina* (L.) in comparison with ascorbic acid and butylated hydroxy toluene as standards.

The $IC_{50}$ values of leaves extract, AA and BHT are shown in the leaf extract of *G. marantina* ($IC_{50}$ =145.80µg/ml) showed moderate antioxidant activity as compared with those of the standards, ascorbic acid ($IC_{50}$ = 100.27µg/ml) and BHT ($IC_{50}$ = 113.89 µg/ml). A lower $IC_{50}$ value indicates stronger DPPH scavenging activity, while a higher $IC_{50}$ value indicates a lower scavenging activity

### 3.3.2. Reducing power activity assay

The reducing capacity of a compound may serve as a significant indicator of its potential antioxidant activity [35]. The reduction of Fe3+ is often used as an indicator of electron donating activity where the yellow colour of the solution changes to different shades of blue and green depending on the power of a compound to reduce the free radicals. The basic principle is based on the conversion of ferric–ferricyanide complex to prussian blue ferrocyanide complex by the phenols. Figure 1B shows the dose–response curves for the reducing power of methanolic extracts of *Globba marantina* and reference standards. Both the plant extracts and standards were able to reduce the ferric ions (Fe3+) to ferrous ions (Fe2+) and the activities of the extracts varied significantly. Reducing power of the plant extracts increased with increasing concentration of the extract which is evident from the absorbance of the reaction mixture at 700 nm. The extract showed some degree of electron donation but lower than that of reference standard, ascorbic acid and BHT (Fig 1B). In this assay, the standards and plant extract could be ranked based on their $EC_{50}$ values, from strongest reducing power to weakest, as follows: BHT ($EC_{50}$ =60.76 µg/ml) > ascorbic acid ($EC_{50}$ =68.67 µg/ml) > leaves ($EC_{50}$ = 142.85µg/ml). (Table-3).

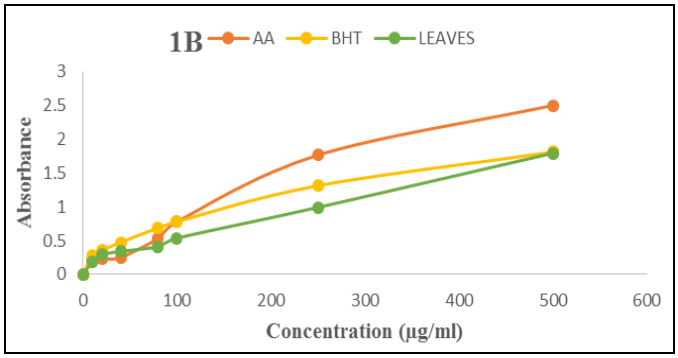


**Fig 2:** Reducing power activity assay of methanolic extract of *Globba marantina* (L.) in comparison with ascorbic acid and butylated hydroxy toluene as standards.

### 3.3.3. Ferric reducing antioxidant power

The FRAP assay is one of most widely accepted method for evaluating the antioxidant capacity of plant extract. The principle of this method based on the conversion of ferric tripyridyltriazine (Fe3+-TPTZ) complex to coloured ferrous tripyridyltriazine (Fe2+-TPTZ). The results of FRAP assay for methanolic extracts of leaves of G. *marantina* and standards are shown in Figure 1C. It was observed that the FRAP activity of extracts of leaves and standard antioxidants (AA and BHT) increased with increasing concentration. However, they were significantly different from each other. In this assay, BHT ($EC_{50}$ =20.49 µg/mL) showed the highest activity followed by leaves extract of *G. marantina* ($EC_{50}$ = 34.95 µg/ml) and ascorbic acid ($EC_{50}$ = 32.90 µg/ml). (Table 3)

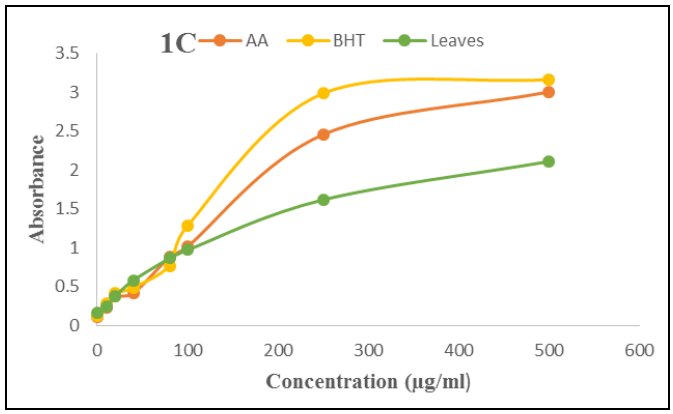


**Fig 3:** Ferric reducing power activity assay of methanolic extract of *Globba marantina* (L.) in comparison with ascorbic acid and butylated hydroxy toluene as standards.

**Table 3:** Antioxidant activities of methanolic extract of leaves of *Globba marantina* in DPPH, RPA, and FRAP assays.

| Plant part | IC50 in DPPH radical scavenging assay (µg/ml) | EC50 in reducing power assay (µg/ml) | EC50 in ferric reducing assay (µg/ml) |
|---|---|---|---|
| Leaves | 145.80±1.28 | 142. 85 [a] ±5.09 | 34.95 [a] ±0.09 |
| Ascorbic Acid | 100.27[a]±2.50 | 68.67 [a] ±2.44 | 32.90 [a] ±0.90 |
| BHT | 113.89[a] ±0.63 | 60.76 [a] ±2.05 | 20.49 [a] ±1.70 |

Values are mean ± SD (n=3). Within each column and row, means followed by the same superscript letter and number of asterisks are not statistically different (P< 0.05; Tukey's HSD).

## 4. Conclusion

The result of this study shows the presence of potent phytochemicals such as alkaloids, steroids, and cardiac glycosides in methanolic leaves extracts of *G. marantina*. The leaves extract of *G. marantina* also possesses high content of phenolic compounds. The present study also revealed that the leaves extracts have moderate antioxidant activity in comparison with reference standard. Thus, it can be concluded that its antioxidant properties might be due to the presence of high content of phenolic compounds and other phytochemicals present in this plant. So, the isolation of bioactive constituents from this plant might be a potential alternative for synthetic antioxidant.

## 5. Acknowledgements

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