

Antioxidant and Antidiabetic Activities of *Alangium salvifolium* and *Bombax ceiba*

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ABSTRACT: In the present study, a methanol extract of flower of *Alangium salvifolium* (AS) and bark of *Bombax ceiba* (BC) were evaluated for its antioxidant and antidiabetic activities. Both methanol extracts exhibited DPPH free radical scavenging activity and reducing power activity in dose dependent manner. Continuous administration for of methanol extracts of AS for seven days significantly decreased blood glucose level than BC in alloxan induced diabetic rats at a dose of 200 mg/kg body weight. The IC₅₀ value of AS and BC were 17.3 µg/ml and 32.1 µg/ml, respectively. Both extract showed significant decrease of blood sugar after glucose loading points (30, 60 and 120 min). In addition, flower of AS possesses high phenol content than the bark of BC (152.73±13.60 vs 74.38±7.42 mg/g of gallic acid). This is the first report of antioxidant and reducing power evaluation and quantitative analysis of phenol content for this two plant materials. This finding suggest that the potential antioxidant activity and antidiabetic effect of flowers of AS could be due to the presence of phenolic compounds.

Key words: *Alangium salvifolium*, *Bombax ceiba*, antioxidant, antidiabetic effect.

INTRODUCTION

Alangium salvifolium Wang is a deciduous, climbing shrub or a tree belonging to the family Alangiaceae. This family consist one genus with twenty two species, out of which AS is the only species used medicinally in Bangladesh, India, China and Phillipines. Different parts of this plant are used for a wide range of diseases. Its root is used in diarrhoea, paralysis, piles and vomiting.¹ *Bombax ceiba* is also an important medicinal plant of tropical and subtropical India and has a number of traditional uses. Specially, barks are used as demulcent, diuretic, inflammation, slightly astringent and tonic. Moreover it is applied on face as freckles,

acne vulgaris and other cutenious as well as pigmentation disorders.² Literature surveys revealed³⁻⁷ that very few chemical and biological studies have been done on flowers of AS and bark of BC but there is no specific study on methanol extract of flowers and bark of AS and BC, respectively. Therefore, the present study is the first attempt to evaluate the antioxidant and antidiabetic potential of methanol extracts of the flower of AS and bark of BC and to investigate their chemical profiles in order to understand the secondary metabolites responsible for the biological activity.

MATERIALS AND METHODS

Plant collection and extraction. The barks of the plant *B. ceiba* and flowers of *A. salvifolium* were collected from Natore and Rajshahi, Bangladesh, respectively during February to March 2011. The identity of the plant was confirmed at the Botany

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Department, Rajshahi University, Bangladesh. Both plants were deposited in the same institution with a voucher specimen no. 08 (BC) and 105 (AS), respectively for future references. Shed dried flower of AS powder (750 gm) was extracted with 100% MeOH (500 ml) for 7 days and repeated the whole extraction twice. Combined extracts were filtered and evaporated to dryness with a rotary evaporator at 40°C to get a semisolid mass (approx. 5 g). Similarly, 6.7 g (approx) semisolid crude methanol extract was obtained from the bark of BC.

Evaluation of antioxidant activity. Both of these extracts were subjected to assays for total phenolic content and total antioxidant determination using the method as described involving Folin-Ciocalteu reagent as oxidizing agent, gallic acid (GAE) and ascorbic acid (AA) as standards. Free radical scavenging activity of methanol extract of flower and bark of AS and BC, respectively were performed by DPPH method. In addition, reducing power of both methanol extract were estimated by the method described by Zahan *et al.*⁸

Evaluation of antidiabetic activity. Swiss albino mice (average 25 g) of either sex was purchased from the International Centre for Diarrheal Disease Research Bangladesh (ICDDR,B) Dhaka. Throughout the experiments animal were proceeded on the basis of international ethical guidelines for the care of laboratory animals. Control, standard and experimental group contain six mice in each. Diabetes was induced in mice by a single intraperitoneal injection of 110 mg/kg body weight of Alloxan (Sigma, St Louis, MO, USA) freshly dissolved in 0.1ml of sterile normal saline water. Normal control mice were injected with physiological saline solution. In some cases, alloxan injection may trigger massive insulin release and result in fatal hypoglycemia. To prevent this, mice were fed with 30 mg/ml glucose solution for 24 h. Five days after injection, mice with fasting blood glucose levels between 10 and 26 mmol/l were used as diabetic mice for further study. After 1 week of administration an oral glucose tolerance test (OGTT) was performed on the animals after a 16 h fasting.

Distilled water (normal control and diabetic control), AS and BC at doses of 100 and 200 mg/kg body weight and metformin at 50 mg/kg body weight were administered orally to normal and diabetic mice. After 1 h, glucose (2 g/kg body weight) was administered orally to animals as a 300 mg/ml solution. Blood glucose levels from the tail vein were analyzed using strip technique (Bioland Glucometer, Germany) at 0 (just before the oral administration of glucose), 30, 60 and 120 min after glucose loading. During the experiment, all animals were carefully monitored every day. Blood glucose levels were estimated on days 0, 3 and 7.⁹ At the end of the experiment, all rats were anaesthetized (chloral hydrate, 450 mg/kg) and blood was taken from the tail vein for biochemical analysis.

Statistical analysis. All data are presented as mean \pm standard deviation (SD). Data were evaluated by one-way analysis of variance (ANOVA) using SPSS Version 15.0 (SPSS Inc., Chicago, IL, USA), and differences between means were assessed by Dunnet's T test. The level of significance was set at $p < 0.001$ for all statistical tests.

RESULTS

Antioxidant activity. *A. salvifolium* and *B. ceiba* are used in traditional system of medicine for various diseases. However, this is the first attempt to evaluate methanol extract of flower of AS and bark of BC for antioxidant and hypoglycemic activities. In addition, total phenol content (152.73 ± 13.60 mg/g of gallic acid for AS and 74.38 ± 7.42 mg/g of gallic acid for BC) and total antioxidant (803.87 ± 12.11 mg/g of ascorbic acid for AS and 352.13 ± 10.16 mg/g of ascorbic acid for BC) were measured for both plant extracts.

Use of natural antioxidant receives a lot of attention now a days, not only for their scavenging properties but also because they are natural, non synthetic products and their appreciation by consumers are very favorable. The flowers of AS and bark of BC have shown DPPH radical scavenging activity with an IC_{50} 17.3 μ g/ml and 32.1 μ g/ml where standard ascorbic acid has shown IC_{50} 16.5

$\mu\text{g/ml}$. DPPH is commonly used as a tool to identify antioxidant molecules present in the plant extracts.⁵ Reducing power assay is another convenient and

rapid screening method for measuring the antioxidant potential. Both plant extracts showed moderate reducing power at higher concentration (Figure 1).

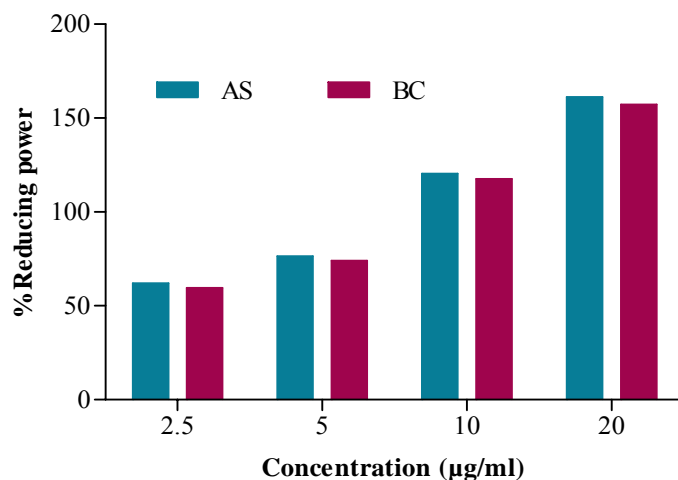


Figure 1. Reducing power assay of methanol extract of flowers of *Alangium salvifolium* and bark of *Bombax ceiba*.

Antidiabetic activity. In the OGTT, the blood glucose level measured at three time points after glucose loading showed a significant decrease after 120 min with a dose of 200 mg of AS and BC extract treatment (Table 1). In our hypoglycemic investigation, administration of methanol extract of AS and BC against alloxan induced diabetic rats revealed that both AS and BC at 200 mg displayed significant hypoglycemic effect as compared to the control group (table 2). These results suggested that both plant extracts could improve glucose intolerance in diabetic rats after glucose loading and might be effective as a hypoglycemic agent in response to glucose ingestion.

DISCUSSION

Antioxidant activity had a linear relationship with the total phenolic content in some plants which can be validated by a number of previous reports.¹⁰⁻¹² Methanol extract had better reducing capacity of DPPH radical. A strong positive correlation has been reported between total polyphenol content and DPPH free radical scavenging activity. Therefore, the higher phenolic content in methanol extract might account

for the better results found in their reducing power and scavenging effect on DPPH. Also, phenolic compounds which originate from plant resources have multiple biological effects such as antioxidant activity and mainly attribute antioxidant activity to their redox properties.¹³ Many plant exhibits efficient antioxidant properties owing to their phenolic constituents.¹⁴ Therefore, it can be suggested that the obtained antioxidant activity might be due to the presence of phenolic compound in these tested plant extract.

These antioxidant properties further established the efficiency of both plant extracts in providing protection against oxidative stress mediated diseases such as diabetics and heart diseases. Antioxidants are substances or nutrients in our foods which can prevent or slow the oxidative damage to our body. When our body cells use oxygen, they naturally produce free radicals (by-products) which can cause damage. Antioxidants act as "free radical scavengers" and hence prevent and repair damage done by these free radicals. Health problems such as heart disease, muscular degeneration, diabetes mellitus, cancer etc are all contributed by oxidative damage. Hence the antioxidant levels were measured in diabetic rats.¹⁵

The phytochemical^{9,16} analysis of AS and BC showed the presence of phenols, tannins, flavanoids, saponins and sterols. The antidiabetic ability of these classes of

compounds to regenerate the pancreatic β -cell has already been justified.¹⁷ Sterols can decrease blood sugar in experimental animal models.¹⁸

Table 1. Glucose level (mmol/l) after treatment with plant extract (*A. salvifolium* and *B. ceiba*) from 0 to 120 min.

Group	Initial 0 min	30 min	60 min	120 min
Control	5.01 ± 0.22	14.5 ± 0.37	15.3 ± 0.39	16.2 ± 0.28
Met. HCl 50 mg	5.11 ± 0.20	7.9 ± 0.11	7.4 ± 0.15	6.5 ± 0.19
AS 100 mg	5.43 ± 0.19	6.96 ± 0.35**	6.28 ± 0.30**	5.55 ± 0.12**
BC 100 mg	5.40 ± 0.21	7.21 ± 0.56**	6.31 ± 0.41**	5.76 ± 0.39**
AS200 mg	5.25 ± 0.12	6.15 ± 0.21**	5.15 ± 0.12**	4.48 ± 0.14**
BC 200 mg	5.09 ± 0.11	6.48 ± 0.20**	5.98 ± 0.10**	4.51 ± 0.11**

Values are mean ± SEM, (n = 6) **p < 0.001 Dunnet's T test, compared to control.

Table 2. Fasting blood glucose level (mmol/L) after treatment with plant extract (*A. salvifolium* and *B. ceiba*) for one week.

Group	Control (non diabetic)	Control (Diabetic)	1st day	3rd Day	7th Day
Metformin HCl	5.10 ± 0.17	18.16 ± 0.49	12.46 ± 0.67	8.75 ± 0.31	5.53 ± 0.27
AS 100 mg	5.30 ± 0.21	17.06 ± 0.19	7.93 ± 0.14**	4.85 ± 0.24**	4.18 ± 0.17**
BC 100 mg	5.12 ± 0.30	16.99 ± 0.42	9.46 ± 0.44**	8.00 ± 0.24**	6.21 ± 0.28**
AS 200 mg	5.43 ± 0.13	17.5 ± 0.38	7.13 ± 0.15**	4.43 ± 0.18**	3.53 ± 0.13**
BC 200 mg	5.22 ± 0.19	18.12 ± 0.31	8.20 ± 0.30**	6.31 ± 0.48**	5.46 ± 0.36**

Values are mean ± SEM, (n=6) **p < 0.001 Dunnet's T test, compared to control. An experimental group at the doses of 100 and 200 mg/kg b. wt. of both plant extract were compared with diabetic control group on corresponding day using Dunnet's T test.

Moreover, in the present study both plant extracts possess high amount of total phenolic compounds which speculate that the hypoglycemic activity might be mainly due to their phenol content. But this is the first report demonstrating the hypoglycemic activity of flower of AS and bark of BC. Currently more work is going on in our lab to isolate active compounds which are responsible for the antioxidant and hypoglycemic effects.

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