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Antioxidant and Antibacterial Activities of *Senna tora* Roxb.

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Abstract: Methanol and aqueous extract of the dried aerial part of *Senna tora* Roxb. were subjected to the potential antioxidant and antibacterial activities. The antioxidant potential of the extract was determined on the basis of their scavenging activity of the stable 1,1-diphenyl-2-picryl hydrazyl (DPPH) free radical. IC₅₀ of the methanol extract of *Senna tora* was 109.65 µg mL⁻¹ which indicated the strong antioxidant activity of the plant extract. Methanol extract of *Senna tora* possess strong antioxidant activity. However the aqueous extract showed mild antioxidant activity. In case of antibacterial activities test, the extract was subjected for its effectiveness against both Gram-positive and Gram-negative bacteria in agar diffusion method. The zones of inhibition produced by the crude methanol and aqueous extract against few sensitive strains was measured and compared with those of standard antibiotic Gentamycin. It is evident that both extracts are active against the bacteria at low concentrations. The obtained results provide a support for the use of this plant in traditional medicine and suggest its further investigation.

Key words: Antioxidant, antibacterial, *Senna tora* Roxb., activities, extract

INTRODUCTION

Senna tora (originally described by Linné as *Cassia tora*) is a legume in the subfamily Caesalpinioideae. It grows wild in most of the tropics and is considered a weed in many places; its native range is not well known but probably South Asia. It is often confused with Chinese Senna or Sicklepod, *S. obtusifolia*. If it is given a distinct common name at all, it is called Sickle Wild Sensitive-plant (Natureserve, 2007). Bangladesh possesses rich floristic wealth and diversified genetic resources of medicinal plants. It has a widely ranging tropical and the agro climatic conditions, which are conducive for introducing and domesticating new and exotic plant varieties. The use of the plants, plant extracts and pure compounds isolated from natural sources provided the foundation to modern pharmaceutical compounds.

Senna surattensis Burm. f. is one of the important species of the Cassia group. Besides its medicinal importance, this species also has taxonomic importance (Kumar *et al.*, 2007). Methanol extract from juemingzi (*Cassia tora* L.) can be fractionated by liquid-liquid partition using ethyl acetate, n-butanol and water (Zhenbao *et al.*, 2007). An ethnobotanical search on five species of *Senna* within and around Ogbomoso, Oyo State, Nigeria showed their relevance in the local herbal medicine. These plants include *Senna tora*, *S. occidentalis*, *S. alata*, *S. podocarpa* and *S. siamea* (Ogunkunle and Ladejobi, 2006).

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Antioxidant properties (Yen and Chuang, 2000) and Inhibitory effect (Wu *et al.*, 2001) of the extract of *S. tora* have already been reported. A recent study was conducted by the scientists of the Department of Food Science and Nutrition, Catholic University of Daegu, Korea who concluded that *Cassia tora* supplements can help improve serum lipid status in type-II diabetic subjects without significant adverse effect (Cho *et al.*, 2005). In the recent study, screening for antibacterial as well as antioxidant activity of the extract of *Senna tora* was conducted to provide support for the use of this plant as traditional medicine. Phytochemical screening provides knowledge of the chemical constituents of this plant not only for the discovery of new therapeutic agents, but also for information in discovering new sources of other economic materials. Therefore, in present study an attempt was made to detect the presence of reported compounds by using various standard qualitative chemical tests and to look for possible presence of other chemical constituents in the aerial part of the extracts. In recent years one of the areas which attracted a great deal of attention is the possible therapeutic potential of antioxidants in controlling degenerative diseases associated with marked oxidative damage. Several plant extracts and different classes of phytochemicals have been found to have quite prominent antioxidant activity (Larson, 1988; Tripathi *et al.*, 1996; Rao, 1997; Vani *et al.*, 1997). The antioxidant activities can be measured using xanthine-xanthine oxidase system for generating superoxide radical ($\text{O}_2^{\cdot-}$), the DPPH (1,1-diphenyl-2-picrylhydrazyl) radical method and the Fenton system for generating hydroxyl radical (HO^{\cdot}) (Spranger *et al.*, 2008). Among the systems the DPPH methods are known as model system for determination of antioxidant activities (Jung *et al.*, 2008). We have undertaken this study to investigate the antibacterial and antioxidant activity of the crude extract of *Senna tora* Roxb.

MATERIALS AND METHODS

Plant Materials

The plant was collected from Bagerhat Municipal Park, Bangladesh during mid July, 2006 on the daytime. The plant (Accession No. DACB-30.216) was identified by the expertise of Bangladesh National Herbarium, Mirpur, Dhaka by using standard taxonomical method. The identified aerial parts of the plant were cut into small pieces separately and then dried by shed drying for about 1 week. The plant parts were ground into coarse powder with the help of a suitable mechanical grinder and the powder was stored in a suitable container for extraction.

Preparation of the Extract

The powdered plant material (about 150 g) was taken in a clean, flat-bottled glass container and soaked in 650 mL of Methanol up to 2 inch. height above the sample surface as solvent can sufficiently cover the sample surface. The container with its contents was sealed and kept for a period of 2 weeks accompanying occasional shaking and stirring. The whole mixture was then underwent a coarse filtration by a piece of clean, white cotton material. Then it was filtered through filter paper and after filtration the remaining portion of the plant extract was given for re-extraction for 7 days with another 150 mL of Methanol. The mixture was again filtered by the same way as previous. The filtrate (Methanol extract) obtained was evaporated by air supplied from a continuously moving electric fan until dried. It rendered a greenish black type of residue of 3.2 g (yield: 2.13%). The greenish black type concentrate residue was designated as crude Methanolic extract of the aerial part of *Senna tora*. One gram of re-extracted residue was found followed by the evaporation of remaining part. The crude extract was then stored in a cool and dry place prepared for studies.

Determination of Antioxidant Activities

The anti-oxidant potential of the Methanolic extract was determined on the basis of their scavenging activity of the stable 1,1-diphenyl-2-picryl hydrazyl (DPPH) free radical. DPPH is a stable

free radical containing an odd electron in its structure and usually utilized for detection of the radical scavenging activity in chemical analysis. The aliquots of the different concentrations (1-500 $\mu\text{g mL}^{-1}$) of the extract was added to 3 mL of a 0.004% w/v solution of DPPH. Absorbance at 517 nm was determined after 30 min and IC_{50} (Inhibitory concentration 50%) was determined. IC_{50} value denotes the concentration of sample required to scavenge 50% of the DPPH free radicals (Gupta *et al.*, 2003). After making the desired concentrations 3 mL of 0.004% DPPH solution was applied on each test tube by pipette. The room temperature was recorded and kept the test tubes for 30 min in light to complete the reactions. DPPH was also applied on the blank test tubes at the same time where only Methanol was taken as blank. After 30 min, absorbances of each test tube were determined by UV spectrophotometer. IC_{50} was determined from % inhibition vs concentration graph.

Determination Antibacterial Activities

Nutrient agar media was prepared by adding water to a dehydrated product that contains all the ingredients. Practically all media are available commercially in powdered form (Pelczar *et al.*, 1993). Three types of discs were prepared for antibacterial screening: One gram sample extracts was dissolved in 10 mL of Methanol to prepare sample solution, 0.03 g/10 mL gentamicin standard disc used as positive control to ensure the activity of standard antibiotic against the test organisms as well as for comparison of the response produced by known antibacterial agent with that produced by test samples and third one was a blank sample (only Methanol) which was used as negative control to ensure that the residual solvents was not active. Specific organisms were inoculated into previously sterilized nutrient agar media, mixed thoroughly and transferred immediately to the sterile petri dish in an aseptic condition. It was stored in an incubator for about 24 h to allow the proper growth of microbes. Prepared sample solutions were applied to the corresponding cups or holes with the help of a micropipette. The plates were then allowed to stand to diffuse the sample solution into the antibiotic medium at room temperature for 2 h. The plates were then incubated at 37°C overnight. After proper incubation, clear zones of inhibition around the point of application of sample solution were formed. These inhibition zones were measured by slide calipers and expressed in millimeter.

RESULTS AND DISCUSSION

Antioxidant Activities Test

DPPH is one of the free radicals widely used for testing preliminary radical scavenging activity of a compound or a plant extract. Zhenbao *et al.* (2007) found ethyl acetate fraction exhibited more antioxidant potency than other fractions. They found the function of ethyl acetate to be more effective in protecting LDL against oxidation in a concentration-dependent manner in *Cassia tora* L. The data suggest that juemingzi especially ethyl acetate-soluble fraction may have a preventive effect against atherosclerosis by inhibiting LDL oxidation. In the present study, methanol extracts of the leaves of *Senna tora* showed potential free-radical scavenging activity but aqueous extract showed very little free-radical scavenging activity. The free radical scavenging property may be one of the mechanisms by which this drug is effective in traditional medicine. Most of the tannins and flavonoids are phenolic compounds and may be responsible for antioxidant properties of many plants (Larson, 1988). IC_{50} of the methanol extract of *Senna tora* was 109.65 $\mu\text{g mL}^{-1}$ which indicated the strong antioxidant activity of the plant extract. However the aqueous extract showed mild antioxidant activity (Table 1).

Antibacterial Activity Test

An antimicrobial activity of the methanolic extract of the aerial part of the plant was compared with the standard antibiotic gentamicin (control) by measuring the zone of inhibition diameter. The methanolic and aqueous extract of *Senna tora* was tested for antibacterial activity against a number

Table 1: Antioxidant activity of *Senna tora* (DPPH Scavenging Assay)

Sample	Concentration ($\mu\text{g mL}^{-1}$)	Average absorbance at 517 nm	Inhibition (%)	IC ₅₀ ($\mu\text{g mL}^{-1}$)
Methanol extract of <i>Senna tora</i>	0	0.978	0.00	109.65
	1	0.964	1.32	
	5	0.875	10.53	
	10	0.811	17.08	
	50	0.690	29.44	
	100	0.487	48.18	
	500	0.186	82.18	
Aqueous extract of <i>Senna tora</i>	0	0.978	0.00	Na
	1	0.971	0.71	
	5	0.955	2.35	
	10	0.939	3.98	
	50	0.923	5.62	
	100	0.888	9.20	
	500	0.827	15.43	
Ascorbic acid	0	0.978	0.00	7.08
	1	0.872	10.83	
	5	0.631	35.48	
	10	0.286	70.75	
	50	0.110	88.87	
	100	0.084	91.41	
	500	0.048	95.19	

Na = Not applicable

Table 2: Antibacterial activity of the methanolic and aqueous extract of *Senna tora*

Name of bacteria	Diameter zone of inhibition in mm		
	Gentamycin (30 $\mu\text{g/well}$)	Methanol extract (500 $\mu\text{g/well}$)	Aqueous extract (500 $\mu\text{g/well}$)
Gram positive bacteria			
<i>Staphylococcus aureus</i>	13	9	11
<i>Staphylococcus epidermidis</i>	11	9	11
<i>Staphylococcus saprophyticus</i>	12	-	13
<i>Streptococcus pyogenes</i>	11	-	10
Gram negative bacteria			
<i>Plesiomonas shigelloides</i>	14	8	9
<i>Shigella dysenteriae</i>	14	9	10
<i>Vibrio cholerae</i>	18	9	9
<i>Salmonella typhi</i>	11	-	-
<i>Shigella flexneri</i>	19	-	10
<i>Shigella boydii</i>	13	-	-
<i>Shigella sonnei</i>	14	-	9
<i>Pseudomonas aeruginosa</i>	17	-	10

-: No inhibition

of both gram negative and gram positive bacteria. The highest zone of inhibition (19.0 mm) was recorded against *Shigella flexneri* (Table 2). The result can be compared with Chukeatirote *et al.* (2007) where they found both crude ethanol and aqueous extracts of *S. tora* were active against *E. coli* growth (6-10 mm) and only crude water extracts of *S. tora* were able to inhibit the *S. cerevisiae* growth.

CONCLUSION

Methanol extract of *Senna tora* possess strong antioxidant activity. However the aqueous extract showed mild antioxidant activity. This activity may be due to the presence of phenolic compounds (tannins and flavonoids) present in the extract. Both methanol and aqueous extract of *Senna tora* show significant antibacterial activity against few gram positive and gram negative bacterial strains. Bangladesh imports a large quantity of pharmaceutical raw materials including medicinal plants and

semi processed plant products to produce drugs and medicines. This huge foreign exchange can be saved if the indigenous medicinal plants or their semi-processed products are utilized by the manufacturers to satisfy their needs. So, further pharmacological and toxicological study is required to establish the therapeutic uses of the plant and particularly with its active principles.

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