

**NOTE*****In vitro* Antioxidant Activity of Leaves of *Ficus rumphii* Blume**

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The *in vitro* antioxidant activity of aerial parts of *Ficus rumphii* Blume has been investigated by estimating degree of non-enzymatic haemoglobin glycosylation measured colorimetrically at 520 nm. The chloroform and aqueous extract of aerial parts of *F. rumphii* showed higher antioxidant activity than other extracts of it. The antioxidant activity of the extracts is close and comparable to that of standard antioxidant compounds used.

**Key Words:** Antioxidant activity, *Ficus rumphii*, Non-enzymatic haemoglobin glycosylation.

*Ficus rumphii* Blume (Pakar in Hindi, Gajaswat in Begali, Pakri in Assam; family: Moraceae) is an interesting group of trees in Nepal and India, not only of their useful value but also of their growth habits and religious significance. It is a moderate deciduous usually epiphytic tree, found in Punjab, Assam, West Bengal, Madhyabharat, West Peninsula, Northern and Southern India. The juice of the plant is used to kill worms and given internally with turmeric, pepper and ghee for the relief of asthma. Bark is used in snake bite. It is also useful in burning sensations, leucoderma, ulcers, leprosy, itching, biliousness and diseases of blood. The leaves boiled in oil form good applications for wounds and bruises<sup>1-4</sup>. *F. rumphii* on preliminary chemical analysis are found to contain flavonoids, tannins and phenolic compounds<sup>5</sup>. Recently, a great deal of interest has been directed towards the bioactivity of these compounds as sources of antioxidant<sup>6-8</sup>. Hence, the present communication deals with the evaluation of the antioxidant activity of leaves of *F. rumphii* Blume.

Evaluation of the antioxidant activity of any drug sample or herbal extract can be carried out either by *in vitro* or *in vivo* models. Various procedures are available in each model to determine the antioxidant capacity. Here, the evaluation is carried out by *in vitro* non-enzymatic glycosylation of haemoglobin method. Since non-enzymatic glycosylation of haemoglobin is an oxidation reaction, an antioxidant is expected to inhibit the reaction. The degree of haemoglycosylation *in vitro* in the presence of different concentration of extracts can be measured colorimetrically.

Haemoglobin was purchased from Nice Chemicals Pvt. Ltd., Cochin. Glucose, phosphate buffer and D- $\alpha$ -tocopherol were procured from Merck, Mumbai. Ascorbic acid and gentamycin were obtained from Biokem International Pvt. Ltd., Bangalore and Nicholas Piramol India Ltd., Pithampur, respectively. All other reagents and solvents used were of analytical grade.

**Preparation of extracts:** The leaves of *F. rumphii* were collected from Tamluk, East Midnapur District, West Bengal in the month of December and were authenticated by Mondal, Additional Director, Central National Herbarium, Botanical Survey of India, Howrah and West Bengal. A voucher specimen has been preserved in our laboratory for future reference (DPK1). Shade-dried, powdered, sieved (40 mesh size) plant materials were exhaustively extracted successively with petroleum ether (40-60 °C), chloroform, ethyl acetate, ethanol and distilled water using a soxhlet extractor. The extracts were concentrated to dryness in vacuum. The yields of petroleum ether, chloroform, ethyl acetate, ethanol and water extracts were 2.27, 1.45, 1.95, 4.27 and 9.65 % w/w, respectively. The extracts were subjected to antioxidant studies.

### Antioxidant studies

**Non-enzymatic haemoglycosylation method:** The antioxidant activities of different extracts were investigated by estimating degree of non-enzymatic haemoglobin glycosylation measured colorimetrically. Haemoglobin, 60 mg/100 mL in 0.01 M phosphate buffer (pH 7.4) was incubated in presence of 2 g/100 mL concentration of glucose for 72 h in order to find out the best condition for haemoglobin glycosylation. The assay was performed by adding 1 mL of glucose solution, 1 mL of haemoglobin solution and 1 mL of gentamycin (20 mg/100 mL) in 0.01 M phosphate buffer (pH 7.4). The mixture was incubated in dark at room temperature for 72 h. The degree of glycosylation of hemoglobin in the presence of different concentration of extracts and their absence were measured colorimetrically at 520 nm<sup>9-13</sup>.

Results of antioxidant activity of leaves of *F. rumphii* extracts are summarized in Table-1. The results obtained indicate that chloroform and aqueous extract of leaves of *F. rumphii* have better antioxidant activity than petroleum ether, ethyl acetate and ethanol extract. The activities were compared with D- $\alpha$ -tocopherol (vitamin E) and ascorbic acid (vitamin C) that were used as standard antioxidant compounds.

TABLE-1  
ANTIOXIDANT ACTIVITY OF DIFFERENT EXTRACTS OF *F. rumphii*

Samples	Final concentration of the tested compound (mg/mL)	
	0.5	1.0
Petroleum ether extract	3.0 ± 0.15	7.5 ± 0.19
Chloroform extract	13.5 ± 0.21	25.3 ± 0.50
Ethyl acetate extract	9.0 ± 0.23	17.5 ± 0.30
Ethanol extract	6.1 ± 0.15	11.3 ± 0.24
Aqueous extract	10.3 ± 0.18	20.2 ± 0.38
D- $\alpha$ -tocopherol	11.3 ± 0.16	21.4 ± 0.45
Ascorbic acid	5.4 ± 0.10	15.4 ± 0.40

Percent inhibition of haemoglobin glycosylation was measured at two concentrations of petroleum ether, chloroform, ethyl acetate, ethanol and aqueous extract. The activities were compared with those of D- $\alpha$ -tocopherol and ascorbic acid. Values are mean ± SEM of three replicates.

The detailed chemical nature of the active principle(s) responsible for antioxidant activity is not known. However, preliminary phytochemical screening has confirmed

the presence of flavonoids, tannins and phenolic compounds, which might be responsible for this activity<sup>8</sup>.

### ACKNOWLEDGEMENTS

The authors are grateful to the Principal and President, S.I.P.S., Jharpokharia, Orissa for providing necessary facilities.

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(Received: 31 July 2009;

Accepted: 7 August 2010)

AJC-8963