

Available online at <http://www.harmanpublications.com>**World Journal of Science and Research****Research Article****Botany****A STUDY ON PHYTO-CONSTITUENTS AND EVALUATION OF ANTI-STRESS ACTIVITY OF *Hemidesmus indicus* (Lin.) ROOT****R. Harini and Dr. R. Sagaya Giri**PG and Research Department of Botany Kunthavai Naacchiyaar Govt. Arts College for Women (Autonomous)  
(Affiliated to Bharathidasan University), Thanjavur – 613 007, Tamil Nadu, India**ABSTRACT**

This work is aimed at evaluating the phytochemicals and the anti-stress activity of *Hemidesmus indicus* root. The qualitative phytochemical tests exhibited the presences of common phytochemicals including tannin, saponins, flavonoids, steroids, polyphenol, anthraquinones, glycosides, terpenoids, triterpenoids and coumarins in aqueous and ethanolic extracts of *Hemidesmus indicus* root. Significant amount of flavonoids ( $80.00 \pm 4.19$  mg/gm), and phenol ( $168.30 \pm 13.97$  mg/gm) were presented in *Hemidesmus indicus* root. Anti-stress activity of *Hemidesmus indicus* root extract was confirmed and the highest dose of  $400 \mu\text{g/ml}$  extract was non-significant changes as compared with normal.

**Keywords:** *Hemidesmus indicus*, qualitative, quantitative and anti-stress.**INTRODUCTION**

Stress is a feeling or condition experienced in humans when a person become frustrated and angry/nervous. Stress is actually the reaction of the body towards the demands that he faces, and a number of chemical substances are produced as a result of these reactions, collectively called stressors. Stress is known to induce alterations in various physiological responses even leading to pathological states (Sheikh *et al.*, 2013). It was demonstrated that different stress paradigms (Chrousos, 1997; Nijholt *et al.*, 2004). Recent surveys reported that psychiatric conditions especially stress and depression were among the most common mood pathologies treated with complementary and alternative therapies (Das *et al.*, 2005; Adams *et al.*, 2007). This correlates with a worldwide increasing trend to integrate traditional medicine with primary health care, because of its “green image”, its cultural significance, and its accessibility to all societal categories (Hunt *et al.*, 2010; Mander, 1998).

The medicinal value of plant lies in the phytochemical (bioactive) constituents of the plant which shows various physiological effects on human body. Therefore, through phytochemical screening one could detect the various important compounds which may be used as the bases of modern drugs for curing various diseases (Sheikh *et al.*, 2013). Chemical compounds produced as a result of metabolic reaction during plant growth are known as phytochemicals. During stressful situations, the energy requirement of an organism is increased, resulting in enhanced generation of free radicals. The generation of these free radicals induced oxidative stress. Antioxidant play a major role to overcome the oxidative stress. Keeping in view, the present study was to investigate the phytochemical and anti-stress activity of *Hemidesmus indicus* root extract.

**MATERIALS AND METHODS****Collection of plant materials**

The root powder of *Hemidesmus indicus* were purchased in January 2022 from

Siddha Medicinal shop, Thanjavur, Thanjavur district, Tamil Nadu, India.

**Preparation of plant extract:**

1 gram of the powder of *Hemidesmus indicus* root were transferred in to different conical flask (250ml). The conical flask containing 50ml of different solution (methanol and water). The conical flask *Hemidesmus indicus* root were shake it well for 30 minutes by free hand. After 24 hrs, the extracts were filtered using whatman filter paper No.1 and filtrate used for further analysis.

**Phytochemical screening**

Chemical tests were carried out on the extract using standard procedures to identify the constituents as described by Sofowara (1993), Trease and Evans (1989) and Harborne (1973 and 1984). Total phenols estimated by the method of Edeoga *et al.*, (2005). Flavonoid determine by the method of Bohm and Kocipai-Abyazan (1994).

**Anti-stress activity** (Tanuj Joshi *et al.*, 2012)

**Preparation of erythrocytes suspensions**

Fresh blood sample from healthy volunteers (10–15ml) were collected and centrifuged at 3000 rpm for 15 minutes, plasma and puffy coats were removed. Red cells were washed with PBS (pH 7.00) for

three times and erythrocytes were lysed with ice-cold distilled water.

**Biochemical estimations**

Malondialdehyde was estimated by the thiobarbituric acid assay method of Beuge and Aust (1978). Reduced glutathione was estimated by method of Moron *et al.*, (1979). Copper-zinc superoxide dismutase activity was determined by the procedure of Kakkar *et al.*, (1984) in plasma. The activity of catalase was assayed by the method of Beers and Sizer, (1952). The activity of mitochondrial glutathione peroxidase was assayed by the method of Rotruck *et al.*, (1973).

**RESULTS AND DISCUSSION**

Plants are naturally gifted at the synthesis of medicinal compounds, whose characterization has led to discovery of new, cheap drugs with high therapeutic potential (Ukwuani *et al.*, 2013). The phytochemical characters of the *Hemidesmus indicus* root investigated and summarized in Table 1. The phytochemical screening *Hemidesmus indicus* root showed that the presence of tannin, saponins, flavonoids, steroids, polyphenol, anthroquinones, glycosides, terpenoids, triterpenoids and coumarins in aqueous and ethanolic extracts while alkaloids was absent in aqueous extract only of *Hemidesmus indicus* root.

**Table 1: Qualitative analysis of phytochemicals in *Hemidesmus indicus* root extract**

S. No	Phytochemicals	Extracts	
		Aqueous	Methanol
1	Tannin	+	++
2	Saponin	++	++
3	Flavonoids	++	++
4	Steroids	+	++
5	Terpenoids	++	++
6	Triterpenoids	++	++
7	Alkaloids	-	+
8	Antroquinone	+	++
9	Polyphenol	++	++
10	Glycoside	+	++
11	Coumarins	++	++

(-) Indicates Absence; (+) Indicates Presence; (++) Moderately present

Plants are reservoir for potentially useful bioactive compounds, and owing to the rising occurrences of drug. Nowadays, in several countries of the world, traditional medicines are used as a substitute to

conventional medicine (Ramawat and Mérillon, 2008). Significant amount of flavonoids (80.00±4.19mg/gm), and phenol (168.30±13.97 mg/gm) were presented (Table 2) in *Hemidesmus indicus* root.

**Table 2: Quantitative analysis of phytochemicals in *Hemidesmus indicus* root powder**

Phytochemicals	Result (mg/gm)
Flavonoids	80.00±4.19
Phenols	168.30±13.97

Value were expressed as Mean ± SD for triplicates

### Anti-stress activity of *Hemidesmus indicus* root extract

Table 3 and 4 shows the effect of *Hemidesmus indicus* root extract on MDA and antioxidants in RBC of experimental group. In this study, Malondialdehyde (MDA) was increased in stress induced rats as compared with normal rats and treatment with *Hemidesmus indicus* root decreased the MDA content. The decreased activity of antioxidant enzymes, such as superoxide dismutase (SOD) and increased activity of antioxidant enzymes, such as catalase (CAT) and glutathione

peroxidase (GPx) and non-enzymatic antioxidant, such as glutathione (GSH) in stress induced RBC blood and its retrieval towards near normalcy in *Hemidesmus indicus* root administered RBC revealed the efficacy of *Hemidesmus indicus* root in combating oxidative stress. In the present study *Hemidesmus indicus* root treated rats stress markers parameters attained an almost normal level. From these results, it was suggested that oxidative stress had been nullified due to the effect of *Hemidesmus indicus* root.

**Table 3: Effect of *Hemidesmus indicus* root stress marker on MDA activity in experimental group in RBC**

Parameters	Group I (Control)	Group II (Negative)	Group III (100µg/ml)	Group IV (200µg/ml)	Group V (400µg/ml)
MDA (nmol of MDA formed/L)	7.33±0.03	11.80±1.29*	8.52±0.04*	8.33±0.03*	7.43±0.14 <sup>NS</sup>

**Table 4: Effect of *Hemidesmus indicus* root antioxidant status on GSH, SOD, CAT and GPx activity in experimental group in RBC**

Parameters	Group I (Control)	Group II (Negative)	Group III (100µg/ml)	Group IV (200µg/ml)	Group V (400µg/ml)
GSH (mg/dl)	6.92±0.20	2.60±0.07*	5.52±0.16*	6.17±0.19*	6.85±0.25 <sup>NS</sup>
SOD (U/ml)	8.32±0.18	4.20±0.07*	7.40±0.14*	7.77±0.13*	8.30±0.10 <sup>NS</sup>
CAT (U/ml)	7.24±0.04	5.27±0.03*	5.86±0.11*	7.18±0.01*	7.22±0.02 <sup>NS</sup>
GP <sub>x</sub> (U/ml)	3.49±0.09	1.91±0.12*	2.18±0.02*	2.91±0.05*	3.45±0.10 <sup>NS</sup>

Values were expressed as mean ± SD for triplicate in each group.

Data were analyzed by one-way ANOVA followed by post-hoc Tukey HSD test. Statistically significant variation was derived by comparing Group I versus Group II, Group III, Group IV and Group V. Significance level  $\alpha$  0.05.

\* $P < 0.05$ , statistically significant and NS= Non significant ( $P > 0.05$ ) compared with Group I (Normal).

Restraint stress is an easy and well-known method to induce chronic physical and emotional stress (Glavin *et al.*, 1994). In the present study, the potential of *Hemidesmus indicus* root extract been explored on RS-induced changes in different parameters. Oxidative stress is considered to have a critical role in changes associated with stress and it is conceivable that antioxidants are important antistress agents (Nazmun Lyle *et al.*, 2009). The intensity of oxidative stress is determined not only by the free radicals production but also by antioxidant enzymatic and non-enzymatic) defense (Beltowski *et al.*, 2000).

### CONCLUSION

Overall, it can be concluded that the anti-stress activity of *Hemidesmus indicus* root extract was dose-dependent manner against H<sub>2</sub>O<sub>2</sub>-induced oxidative stress in RBCs. Conclusively, the findings suggest the validity of the MDA assay and enzymatic and non-enzymatic antioxidants as a reliable tool in

finding out the anti-stress activity against hydrogen peroxide-induced oxidative stress. The anti-stress activity of *Hemidesmus indicus* root extract is due to the presence of phytochemicals.

### References

- Adams, M., Gmünder, F., & Hamburger, M. (2007). Plants traditionally used in age related brain disorders—A survey of ethnobotanical literature. *Journal of ethnopharmacology*, 113(3), 363-381.
- Beers, R., & Sizer I. (1952). A spectrophotometric method for measuring the breakdown of hydrogen peroxide by catalase. *Journal of Biological Chemistry*, 195, 133.
- Beltowski J., Wokcicka G., Gorny, D., & Marchiniak, A. (2000). The effect of dietary-induced obesity on lipid peroxidation, antioxidant enzymes

- and total plasma antioxidant capacity. *Journal of Physiology and Pharmacology*. 51, 883-896.
- Beuge, J. A., & Aust, S. D. (1978). Estimation of serum malondialdehyde level. *Methods in enzymology Hoffee Jones ed. By Hoffee PA and Jone ME. Academic Press, a Subsidiary of Harcoart Brace Jovanovich Publisher, New York*.
- Bohm, B. A., & Kocipai-Abyazan, R. (1994). Flavonoids and condensed tannins from leaves of *Hawaiian vaccinium* and *V calycinium*. *Pacific Sci*, 48, 458-463.
- Chrousos, G. P. (1997). Stress as a medical and scientific idea and its implications. In *Advances in Pharmacology* (Vol. 42, pp. 552-556). Academic Press.
- Das, A., Rai, D., Dikshit, M., Palit, G., & Nath, C. (2005). Nature of stress: differential effects on brain acetylcholinesterase activity and memory in rats. *Life Sciences*, 77(18), 2299-2311.
- Edeoga, H. O., Okwu, D. E., & Mbaebie, B. O. (2005). Phytochemical constituents of some Nigerian medicinal plants. *African journal of biotechnology*, 4(7), 685-688.
- Glavin, G. B., Paré, W. P., Sandbak, T., Bakke, H. K., & Murison, R. (1994). Restraint stress in biomedical research: an update. *Neuroscience & Biobehavioral Reviews*, 18(2), 223-249.
- Harborne, J. B. (1973). *Phytochemical methods*, London. Chapman and Hall, Ltd. pp. 49-188.
- Harborne. J. B. (1984). *Phytochemical Methods. A Guide to Modern Technique of Plant analysis*. London: *Chapman and Hall*, 78-210.
- Hunt, K. J., Coelho, H. F., Wider, B., Perry, R., Hung, S. K., Terry, R., & Ernst, E. (2010). Complementary and alternative medicine use in England: results from a national survey. *International journal of clinical practice*, 64(11), 1496-1502.
- Kakkar, P., Das, B., & Viswanathan, P. N. (1984). A modified spectrophotometric assay of SOD. *Ind J Biochem Biophy*, 21: 130-132.
- Mander M., (1998). Marketing of Indigenous Medicinal Plants in South Africa, A Case Study in KwaZulu-Natal, *FAO, Rome, Italy*.
- Moron M.S., DsePierre J. W., & Manerwik K.B. (1979). Levels of glutathione, glutathione reductase and glutathione-s-transferase activities in rat lung and *liver Acta of Biochemica ,biophysica* 582:67-68.
- Nazmun Lyle., Gomes, A., Sur, T., Munshi, S., Paul, S., Chatterjee, S., & Bhattacharyya, D. (2009). The role of antioxidant properties of *Nardostachys jatamansi* in alleviation of the symptoms of the chronic fatigue syndrome. *Behavioural Brain Research*, 202(2), 285-290.
- Nijholt, I., Farchi, N., Kye, M., Sklan, E. H., Shoham, S., Verbeure, B., & Blank, T. (2004). Stress-induced alternative splicing of acetylcholinesterase results in enhanced fear memory and long-term potentiation. *Molecular psychiatry*, 9(2), 174-183.
- Ramawat K. G. & Méridon, J. M. (2008). *Bioactive Molecules and Medicinal Plants*, Springer, *Berlin, Germany*.
- Rotruck, J. T., Pope, A. L., Ganther, H. E., Swanson, A. B., Hafeman, D. G., & Hoekstra, W. G. (1973). Selenium: biochemical roles as component of glutathione peroxidase. *Sci*, 179, 588-590.
- Sheikh N., Y. Kumar, A. K. Misra, and L. Pfoze, "Phytochemical screening to validate the ethnobotanical importance of root tubers of *Dioscorea* species of Meghalaya, North East India," *Journal of Medicinal Plants Studies*, vol. 1, no. 6, 69, 2013.
- Sofowara, A. (1993). *Medicinal plants and Traditional medicine in Africa*. *Spectrum Books Ltd, Ibadan, Nigeria*. 191-289.
- Tanuj Joshi, T., Sah, S. P., & Singh, A. (2012). Antistress activity of ethanolic extract of *Asparagus racemosus* Willd roots in mice.
- Trease G. E. E & vans W. C (1989). *Pharmacognsy*. 11<sup>th</sup> edn. Brailliar Tiridel can. Macmillian Publishers.U.S. (1984). Environmental protection Agency, Draft Criteria document for carbon tetrachloride, criteria and standards Division, office of Drinking, Washington, DC.
- Ukwuani A.N., Abubakar M.G., Hassan S.W., & Agaie B.M., 2013. Antinociceptive Activity of Hydromethanolic Extract of Some Medicinal Plants in Mice. *International Journal of Pharmacy*. Photon 104, 120-125.