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DPPH SCAVENGING ACTIVITY OF *DIOSCOREA BATATAS* TUBERS

R.SANDIYA[*1] AND M.MUNNIAPPAN[2]

[1, 2]Department of Pharmacology, Sree Balaji medical College and Hospital, Bharath University, Chennai, India.

ABSTRACT

Reactive oxygen species [ROS] cause oxidative damage to the tissues and protection from such damages are provided by endogenous and exogenous antioxidants. Plant based antioxidants are preferred due to the multiple mechanisms of actions and of the phytochemicals present in them. Different doses of Alcoholic Tuber extract of *Dioscorea batatas* were tested individually in equal proportion of each dose for DPPH scavenging activity and reducing power. The results indicate that the highestdoses of Alcoholic Tuber extract of *Dioscorea batatas* (100 µg) has better DPPH scavenging action and reducing power compared to the lower doses of the plant extract indicating effect of phytochemicals present in the extract indicating its antioxidant potential.

KEYWORDS: **Antioxidants, Phytochemical, DPPH scavenging activity, *Dioscorea batatas.***

R.SANDIYA
Department of Pharmacology, Sree Balaji medical College and Hospital, Bharath University,Chennai, India.

***Corresponding author**

INTRODUCTION

Reactive oxygen free radical species (ROS) are greatly reactive molecules, and includethe hydroxyl radical (•OH), the superoxide anion radical (O2•–), hydrogen peroxide (H2O2), and peroxyl (ROO•), which consequently generate metabolic products that attack lipids in cell membranes or DNA. Lipid peroxidation occurring in cell membranes or DNA which involves a series of free radical chain reaction processes is associated withseveral types of biological damage, DNA damage, carcinogenesis, and cellular degeneration related to aging.Cells are protected by their endogenous scavenging systems or by other substances [(1)]. These ROS play an important role in degenerative or pathological processes, such as aging, cancers, coronary heart diseases, Alzheimer's disease, neurodegenerative disorders, atherosclerosis, cataracts and inflammations. Living organisms have antioxidant defense systems that protects against oxidative damage by removal or repair of damaged molecules [(2)]. The term 'antioxidant' refers to the activity of numerous vitamins, minerals and phytochemicals which provide protection against the damage caused by ROS. Antioxidants interfere with the oxidative processes by scavenging free radicals, chelating free catalytic metals and by acting as electron donors. The natural antioxidant mechanisms may be insufficient in variety of conditions and hence dietary intake of antioxidant compounds are important [(3)]. The therapeutic effects of several medicinal plants are usually attributed to their antioxidant phytochemicals. It has been suggested that there is an inverse relationship between dietary intake of antioxidant rich foods and incidence of human diseases. Plant based antioxidants are preferred to the synthetic ones because of their multiple mechanisms of actions and non-toxic nature[(4)]. These facts have inspired widespread screening of plants for possible medicinal and antioxidant properties; the isolation and characterization of diverse phytochemicals and the utilization to antioxidants of natural origin prevent the diseases. In recent years, the possible toxicity of synthetic chemical antioxidants has been criticized[(5)]. Thus, recent studies have investigated the potential of plant products to serve as antioxidants to protect against various diseases induced by free radicals. Plant products including phenolics, flavonoids, tannins, proanthocyanidins, and various plant or herbal extracts have been reported to be radical scavengers and inhibitors of lipid peroxidation. Therefore, in view of the importance of these substances to health, phenolics have been proposed as health-promoting products in prophylactic medicines [(6)]. In the oriental countries and in traditional Chinese herbal medicine flour of yam tubers (*Dioscorea* species) is regarded as health food because of its antioxidant activity. Generally, oxidation is believed to be one of the primary factors for chronic degenerative processes[(7)]. *Dioscorea batatas,* also known as Huai Shan, the edible tuber of Chinese yam is used for medicinal purposes. The plant *Dioscorea batatas*was reported for its Antioxidant, Antiradicular and nootropic activity in Ayurvedic texts[(8)]. By using the scavenging activities of 1,1-diphenyl-2-picrylhydrazyl (DPPH) and hydroxyl (•OH) radicals, a systematic survey of free radical-scavenging activity of Tubers of *Dioscorea batatas* was undertaken, and results are discussed. The present study aims to spectrophotometeric quantification of DPPH radical scavenging activity in the Alcoholic Tuber Extract Of *Dioscorea batatas*.

MATERIALS AND METHODS

(i) Plant materials

Chinese yam (*Dioscorea batatas)* was Purchased from the local market in Chennai and identified by The Director, National institute of herbal science, West Tambaram, Chennai, India.

(ii) Reagents required

1)1,1Diphenyl 2-picryl hydrazide (DPPH)
2)Ethanol
Reagent preparation: DPPH= 200µM

(iii) Preparation of Alcoholic Tuber Extract Of Dioscorea batatas

Tubers of *Dioscorea batatas* were obtained from the local market were shade dried and powdered using mechanical mixer. The plant extract was prepared using soxhlet apparatus at 60-80^{o}C, by maceration of 50 gm of the chopped, dried Tubers of *Dioscorea batatas*in a mixture of 200 ml ethanol and 200 ml distilled water by shaking them for 48 hours and pressing the solution out of the material using a filter press. The extract was then transferred into the previously weighed empty beaker and was subjected to evaporation on a water bath at 50^{0}C till a thick paste of extract remained in the beaker [9].

(iv) DPPH Radical Scavenging Assay

This assay is based on reduction of absorbance of methanol solution of DPPH by free radical scavengers.DPPH radical scavenging activity was done using the method of Yohozowa et al [10]. The reaction mixture containing 1.9ml of DPPH solution (200µM in ethanol) with different concentrations of the substance was shaken and incubated in dark for 20min at room temperature. The resultant absorbance was recorded at 517nm (Table 1). The percentage inhibition was calculated using the formula

$$\text{Percentage inhibition} = \frac{\text{Abs}_{\text{control}} - \text{Abs}_{\text{sample}}}{\text{Abs}_{\text{control}}} \times 100$$

Table 1
Antioxidant activity of Dioscorea batatas

S.No	Sample taken(ml)	Conc (µg)	Reagent taken(ml)		Absorbance at 517nm		
1	0.01	100	1.990	Incubation In dark for 30min	0.1772	0.1776	0.2116
2	0.02	200	1.980		0.1650	0.1706	0.2001
3	0.04	400	1.960		0.1602	0.1630	0.1709
4	0.08	800	1.920		0.0935	0.1158	0.089
5	0.10	1000	1.900		0.0862	0.086	0.080

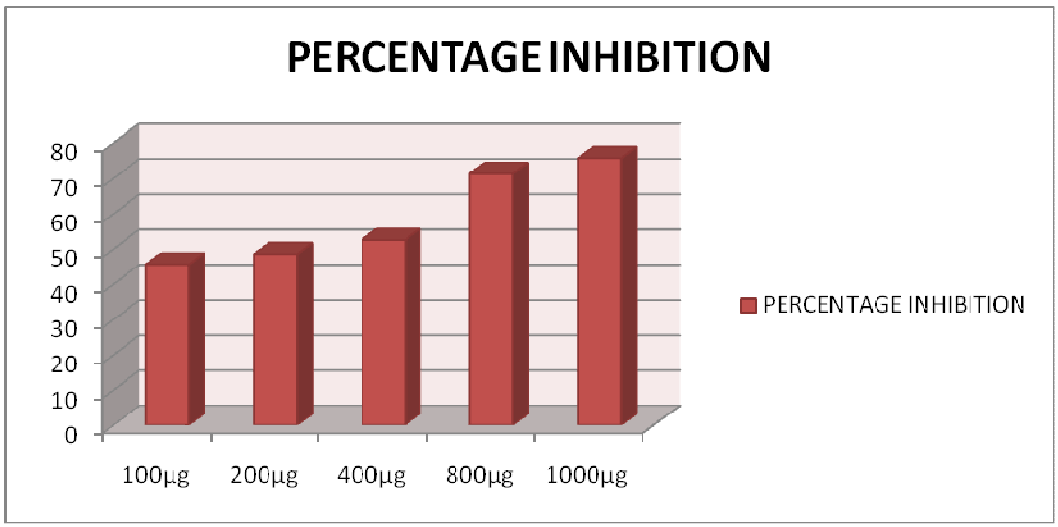
Control: 1.9ml of DPPH+ 0.1ml of DMSO =0.3470 Blank: 1.9ml of ethanol +0.1ml of DMSO % Scavenging Activity= Control- Test/Test X 100

RESULTS

The Percentage inhibitions were

For 100µg concentration = 0.3470-0.1888/0.3470*100= **45.59%**
For 200µg concentration = 0.3470-0.17857/0.3470*100= **48.53%**
For 400µg concentration = 0.3470-0.1647/0.3470*100= **52.53%**
For 800µg concentration = 0.3470-0.09943/0.3470*100= **71.35%**
For 1000µg concentration = 0.3470-0.0841/0.3470*100= **75.76%**

Graph 1
Percentage Inhibition of Dioscorea batatas



Standard Curcumin[11] The DPPH radical scavenging activity of the standard Curcumin is about 90.63% The DPPH radical scavenging activity was recorded in terms of percentage Inhibition. It was observed from that *Dioscorea batatas* (100 µg /ml) has minimum DPPH scavenging activity (45.59%) and *Dioscorea batatas* (1000 µg /ml) has maximum DPPH scavenging activity (75.76%) (Graph 1). The Results obtained were comparative to Curcumin standard. Higher Percentage Inhibition indicates better scavenging activity or antioxidant potential. The given sample showed the dose dependent activity in scavenging the free radicals compared to that of the Curcumin standard.

DISCUSSION

Flavonoids are the most widespread group of natural compounds and probably the most important natural phenolics. Total phenolics and flavonoids possess a broad spectrum of chemical and biological activities including radical scavenging properties. Such property is especially distinct for flavonols .The medicinal effects of plants are often attributed to the antioxidant activity of phytochemical constituents, mainly phenolics, flavonoids and flavonols[3].It is claimed that phenolic compounds are powerful chain breaking antioxidants. The scavenging activity of phenolic group is due to its hydroxyl group [12]. The antioxidant activity has been reported to be concomitant with the development of reducing power [13]. Herbal preparation revealed synergistic effects both in DPPH scavenging and reducing power in comparison with the individual plant extracts selected for the study. The crude extracts of plants are pharmacologically more active than their isolated active principles due to the synergistic effects of various components present in the whole extract [14]. Traditional Chinese herbal medicine, literature reports that *Dioscorea batatas* (yam) possesses anti-aging activity. It is generally believed that oxidation is one of the primary factors for developing many chronic degenerative processes, including aging [15]. The antioxidant is defined as a molecule that has the ability to slow or prevent the oxidation of other molecules. The oxidation damages on physiological substances are highly related to many diseases, such as atherosclerosis, aging, neurodegenerative diseases and cancers showed that dioscorin isolated from *D. batatas* Decne had both dehydroascorbate reductase and monodehydroascorbate reductase activities *in vitro* at the pH close to neutral, by these, dioscorin could reduce dehydroascorbate and monodehydroascorbate to generate ascorbate in turn to reduce the ROS and increase the scavenger concentration

at the same time. Dioscorin exhibited the scavenging activity against both 1, 1-diphenyl-2-picrylhydrazyl (DPPH) and hydroxyl radical in a dose-dependent manner *in vitro* [16] However, it is also possible to study the in-vitro antioxidant potential of *Dioscorea batatas* in terms of various other experimental models such as 2, 2' azino-bis (3-ethylbenzothiazoline-6- sulphonate) radical cation assay, superoxide scavenging assay, hydroxyl scavenging activity, nitric oxide scavenging assay, in-vitro antioxidant activity and total antioxidant activity in order to confirm the synergistic effect of the combination of individual extracts compared to single plant extract.

CONCLUSION

The Alcoholic Tuber Extract Of *Dioscorea batatas* exhibited significant antioxidant activity compared to Curcumin standard and the activity may be related to theflavonoids and phenolic compounds in this plantextract. Since reactive oxygen species are importantcontributors to several serious ailments, in the presentstudy, the observed DPPH scavenging activity of the Tuber Extract of *Dioscorea batatas* might be useful for thedevelopment of newer and more potent natural antioxidants.

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