Journal of Pharmacognosy and Phytochemistry

Available online at www.phytojournal.com

E-ISSN: 2278-4136
P-ISSN: 2349-8234
JPP 2019; 8(1): 2673-2677
Received: 01-11-2018
Accepted: 05-12-2018

P Sobiyana
PG and Research, Department of Chemistry, AVVM Sri Pushpam Arts and Science College Autonomous, Poondi, Tamil Nadu, India

G Anburaj
PG and Research, Department of Chemistry, AVVM Sri Pushpam Arts and Science College Autonomous, Poondi, Tamil Nadu, India

Dr R Manikandan
PG and Research, Department of Chemistry, AVVM Sri Pushpam Arts and Science College Autonomous, Poondi, Tamil Nadu, India

Correspondence
P Sobiyana
PG and Research, Department of Chemistry, AVVM Sri Pushpam Arts and Science College Autonomous, Poondi, Tamil Nadu, India

Comparative analysis of the *in vitro* antioxidant activity of *Tabebuia rosea* and *Tabebuia argentea*

P Sobiyana, G Anburaj and Dr. R Manikandan

Abstract
The nearness of common cancer prevention agent and antimicrobial in plants is notable. This paper reports the antioxidative and antimicrobial exercises of oil ether *Tabebuia rosea* and *Tabebuia argentea* extricates. Oil ether concentrates of *Tabebuia rosea* and *Tabebuia argentea* blossoms were assessed for their free radical searching action utilizing the DPPH radical test. Decrease of DPPH radicals can be seen by the reduction in the absorbance at λ max517nm. The tabebuia rosea blossoms remove and ascorbic corrosive indicated cell reinforcement movement with various IC50 values.

Keywords: Antioxidant activity, ascorbic acid, DPPH, free–radical assay, *Tabebuia rosea* and *Tabebuia argentea*

Introduction
Cancer prevention agents have a fundamental job in body resistances framework against Receptive Oxygen Species (ROS). Characteristic cancer prevention agents that are available in the nourishment increment the obstruction toward oxidative harms and they may essentially affect human wellbeing. In this manner, utilization of nourishment that is containing phytochemical with potential cancer prevention agent properties can diminish the peril of human infections. Chain breaking cancer prevention agents are very responsive with free radicals and structure stable aggravates that don't add to the oxidative chain reaction.[1] It is known that, during stress, the animal body often generates more free radicals, such as superoxide anion radicals (O2 −), nitric oxide (NO−), reactive oxygen species (ROS), and hydroxyl radicals (OH), than both enzymatic antioxidants (e.g., glutathione (GPx) and catalase) and synthetic antioxidants (e.g., carotenoids, flavonoids, and ascorbic acid). These free radicals which are mostly generated during biological and physiological processes are recognized as causing cell damage and, in severe cases, may result in cell death [2]. However, traditionally used medicinal plants awaits such screening. On the other hand, the medicinal properties of plants have also been investigated in the light of recent scientific developments through out the world, due to their potent pharmacological activities, low toxicity and economic viability [3]. Several medicinal plants (Rasayana) have also been extensively used in the Indian traditional (Ayurveda) system of medicine for the treatment of number of diseases [4].

Meterials and Methods
Collection of Plant Meterials
The plant were collected from the Sastar University, vallam, Thanjavur (Dt). The collected Flowers were botanically authenticated by Dr.S. John Britto Rapinat Herbarium, st.Joseph's college, Trichy.

Antioxidant activity (DPPH free radical scavenging activity) determination
The antioxidant activity of the both the flower of *Tabebuia rosea* and the *Tabebuia argentea* was examined on the basis of the scavenging effect on the stable DPPH free radical activity (Braca *et al.*, 2002) [5]. Ethanolic solution of DPPH (0.05 mM) (300 µl) was added to 40 µl of extract solution with different concentrations (100 - 1000 µg/ml). DPPH solution was freshly prepared and kept in the dark at 4 °C. Ethanol 96% (2.7 ml) was added and the mixture was shaken vigorously. The mixture was left to stand for 5 min and absorbance was measured spectrophotometrically at 517 nm. Ethanol was used to set the absorbance zero. A blank sample containing the same amount of ethanol and DPPH was also prepared. All determinations were performed in triplicate. The radical scavenging activities of the tested samples, expressed as percentage of inhibition were calculated according to the following equation (Yen and Duh, 1994) [6].

Percent (%) inhibition of DPPH activity = [(AB – AA) / AB] x 100

Where AA and AB are the absorbance values of the test and of the blank sample, respectively. A percent inhibition versus concentration curve was plotted and the concentration of sample required for 50% inhibition was determined and represented as IC_{50} value for each of the test solutions.

Assay of nitric oxide-scavenging activity

The sodium nitroprusside (10 mM), in phosphate-buffered saline, was mixed with different concentrations of methanolic extract of flower of *Tabebuia rosea* and the flower of *Tabebuia argentea* (100-1000 µg/ml) and incubated at room temperature for 150 min. After the incubation period, 0.5 ml of Griess reagent was added. The absorbance of the chromophore formed was read at 546 nm. Vitamin C was used as positive control (Nabavi *et al.*, 2009) [7]. The mixture was recorded at 230 nm. The blank solution contains sodium phosphate buffer without H_2O_2. The percentage of nitric oxide scavenging of extract and standard compounds were calculated using the following equation

Nitric oxide scavenging effect (%) = $(1–A_s/A_c) \times 100$

Where A_C is the absorbance of the control and $A_S$is the absorbance in the presence of the sample extract or standards.

Superoxide anion scavenging (NBT reduction) assay

The superoxide anion scavenging activity was determined by spectrophotometric method (Madhumitha *et al.*, 2015) [8]. The different concentration of methanolic extract of flower of *Tabebuia rosea* and the flower of *Tabebuia argentea* (100-1000 µg/ml) was mixed with 1.4 ml of 50 mM KH_2PO4-KOH, pH 7.4 containing 1 mM EDTA, 0.5 ml of 100 µM hypoxanthine, 0.5 ml of 100 µM NBT. The reaction was started by adding 0.066 units per tube of xanthine oxidase freshly diluted in 100 µl of phosphate buffer and 0.5 ml of test extract fractions in saline. The xanthine oxidase was added last. The subsequent rate of NBT reduction was determined on the basis of spectrophotometric determinations of absorbance at 560 nm. Ascorbic acid was used as standard. The percentage inhibition of superoxide anion scavenging activity was calculated as

% Superoxide anion scavenging activity = $[(A_0–A_1)/A_0] \times 100$

Where A_0 is the absorbance of the control and $A_1$is the absorbance in the presence of the sample extract or standards.

Reducing Power Assay

1 ml of varying concentration (100-1000 µg/ml) of both the flower of *Tabebuia rosea* and the flower of *Tabebuia argentea* was mixed with 2.5 ml phosphate buffer and 2.5 ml of potassium ferricyanide. The mixture was incubated at 50 °C for 20 min. Then 2.5 ml trichloroacetic acid was added to the mixture, which was then centrifuged at 3000 rpm for 10min. The upper layer of solution (2.5 ml) was mixed with equal volume of distilled water. Then 0.5ml of freshly prepared ferric chloride solution was added and the absorbance was measured at 700 nm. Increased absorbance of the reaction mixture indicates increase in reducing power (Madhumitha *et al.*, 2015) [8].

Total antioxidant activity by phosphomolybdenum method

Different concentrations of the extracts 100-1000 µg/ml were added on the test tubes to that 1ml of reagent which contains 0.6mM of sulphuric acid, 28 mM of sodium phosphate and 4 mM of ammonium molybdenum was added and incubated for 95 °C for 90 min. The absorbance was measured at 635 nm in UV spectrometer (Madhumitha *et al.*, 2015) [8]. The total antioxidant activity was calculated by the formula

Total antioxidant =Ao –A1 / Ao× 100
Ao = Absorbance of control.
A1 =Absorbance of standard.

Results and Discussion

Determination of *in-vitro* antioxidant activity

Antioxidant activity of methanol extracts of flower of *Tabebuia rosea* and *Tabebuia argentea*

The dose-response curve of DPPH radical scavenging activity of the methanolic extracts of flower of *Tabebuia rosea* compared with *Tabebuia argentea* extracts (Table 2 and Fig. 1). It was observed that the methanol extract of *Tabebuia rosea* had the highest activity, followed by *Tabebuia argentea* respectively. At the concentration of 1000 µg/mL, the scavenging activity of methanol extract reached 89.92%, but at the same concentration, that of *Tabebuia argentea* extracts were 72.88% respectively. It was evident that the *Tabebuia rosea* extracts showed proton-donating ability and this could serve as free radical inhibitors or scavengers, acting possibly as primary antioxidants.

The reductant activity of methanolic extract of flower of *Tabebuia rosea* showed the highest activity (86.67%) when comparted to the flower of *Tabebuia argentea* (91.67%) at concentration 1000 µg/ml was shown in Fig. 2.

The nitrite oxide radical scavenging activity of flower of *Tabebuia rosea* compared with *Tabebuia argentea* extracts acts is shown in Fig. 3. Among the extracts tested, methanol extract of *Tabebuia rosea* had significant scavenging activity followed by methanolic extract of *Tabebuia argentea*. The nitrite oxide radical scavenging activity of these *Tabebuia rosea* was higher (63.33%) when compared to the *Tabebuia argentea* (43.33%) at a concentration of 1000 µg/ml.

The super oxide scavenging activity of methanolic extract of flower of *Tabebuia rosea* showed the highest activity (73.33%) when comparted to the flower of *Tabebuia argentea* (55%) at concentration 1000 µg/ml was shown in Fig. 4. Figure 5 showed the total antioxidant activity of the extracts increased with the concentration of the extract. The total antioxidant activity of flower of *Tabebuia rosea* was found to be 86% at a concentration of 1000 µg/ml, when compared with the *Tabebuia argentea* (94%).

Previous study suggested that the species *Tabebuia* ochracea and *Tabebuia rosea-alba* showed antioxidant activity (Silva *et al.*, 2017) [9]. Sobiyana *et al.*, 2015 [10] concluded that the methanolic extract of flower of *Tabebuia rosea* showed DPPH radical scavenging activity 71.63±4.6 as compared to the ascorbic acid 88.15±0.41 at a concentration 250 µg/ml. The methanol extract of endophytic fungi *Aspergillus niger* and *Penicillium* sp. of *Tabebuia argentea* was found to most effective in exhibiting *in vitro* antioxidant activity in various method (ferric thiocyanate, thiobarbituric acid, and DPPH than the chloroform extract (Govindappa *et al.*, 2013) [11]. Madhumithra *et al.*, 2015 [8]. suggested that the methanolic

extract of flower of *Tabebuia rosea* showed the antioxidant activity with high reducing potential activity and also showed larvicidal activity against *Culexquinque fasciatus* and *Anopheles subpictus.* Among the different parts of root bark, stem bark, leaf and flower of *Tabebuia pallida tested* and found that methanolic extract of *Tabebuia pallid* leaf which contains large amounts of phenolic and flavonoid compounds, exhibited the highest antioxidant and free radical scavenging and also inhibited lipid peroxidation. These *in vitro* assays indicate that *T. pallida* leaves are a significant source of natural antioxidants, which could help to prevent the progression of various diseases caused by free radicals, such as certain cancers (Rahman *et al.*, 2015) [12].

Table 1: % of Antioxidant Scavenging Activity of *Tabebuia rosea* and *Tabebuia argentea* at different concentrations

Name of samples	Concentrations of Plant Extract (µg)	% of Scavenging Activity				
		DPPH	Reductant Activity	Nitrous Oxide	Super Oxide	Total Antioxidant
Tabebuia rosea	100	21.11	36.67	31.67	13.33	21.00
	200	33.38	50.00	33.33	20.00	23.00
	300	50.69	58.33	36.67	25.00	30.00
	400	51.47	65.00	40.00	33.33	32.00
	500	59.02	68.33	41.67	40.00	44.00
	600	77.79	75.00	46.67	46.67	77.00
	700	80.43	80.00	53.33	53.33	80.00
	800	84.69	83.33	56.67	61.67	82.00
	900	87.91	86.67	60.00	70.00	84.00
	1000	89.92	86.67	63.33	73.33	86.00
Tabebuia argentea	100	17.27	55.00	25.00	1.67	15.00
	200	32.60	80.00	21.67	5.00	36.00
	300	44.94	75.00	23.33	11.67	46.50
	400	53.68	75.00	25.00	20.00	64.00
	500	59.92	80.00	25.00	30.00	78.00
	600	68.92	83.33	31.67	40.00	83.00
	700	73.69	86.67	35.00	55.00	86.00
	800	74.22	88.33	43.33	63.33	89.00
	900	73.34	90.00	50.00	71.67	91.00
	1000	72.88	91.67	53.33	78.33	94.00

Table 1: DPPH Scavenging Activity of Plant Extracts

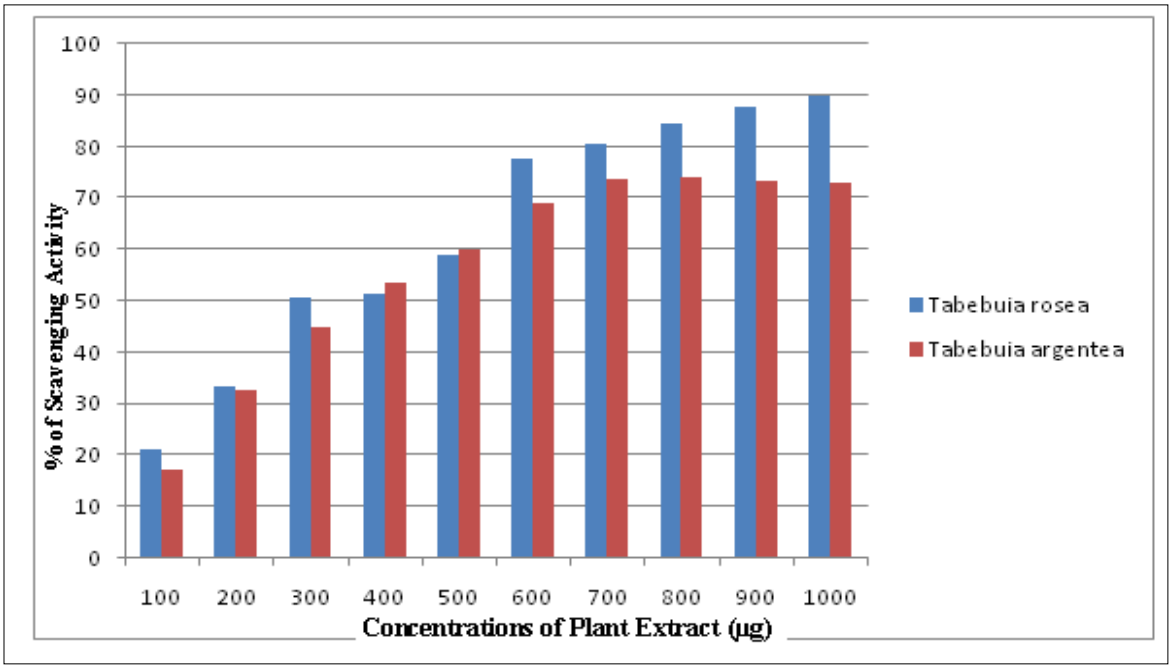

Fig 1: Reductant Activity of Plant Extracts *Tabebuia rosea* and *Tabebuia*

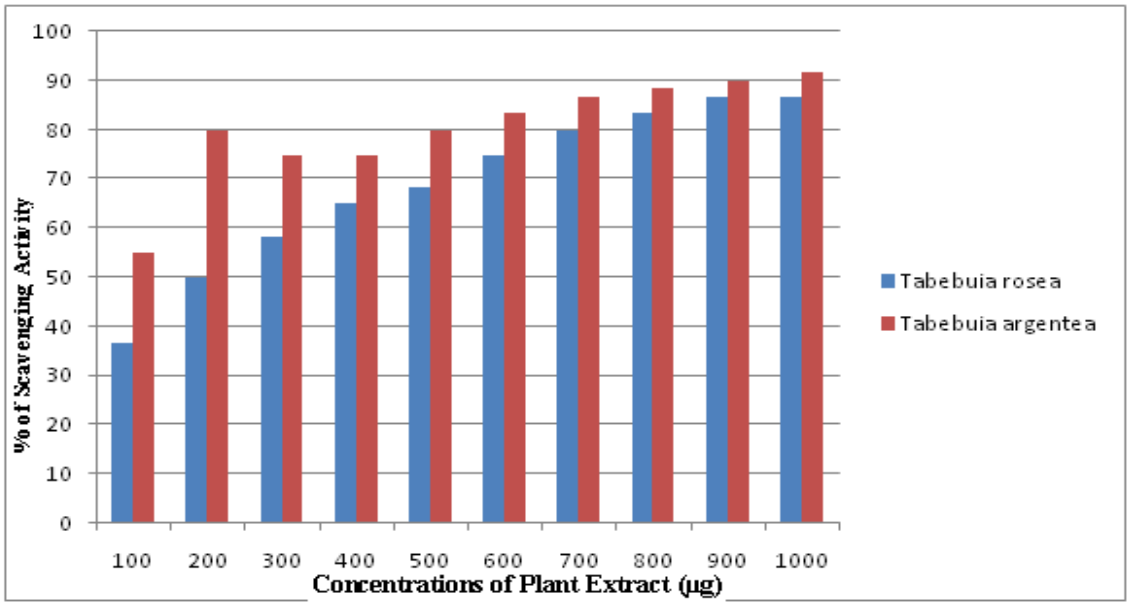

Fig 2: Nitrous Oxide Scavenging Activity of Plant Extracts *Tabebuia rosea* and *Tabebuia*

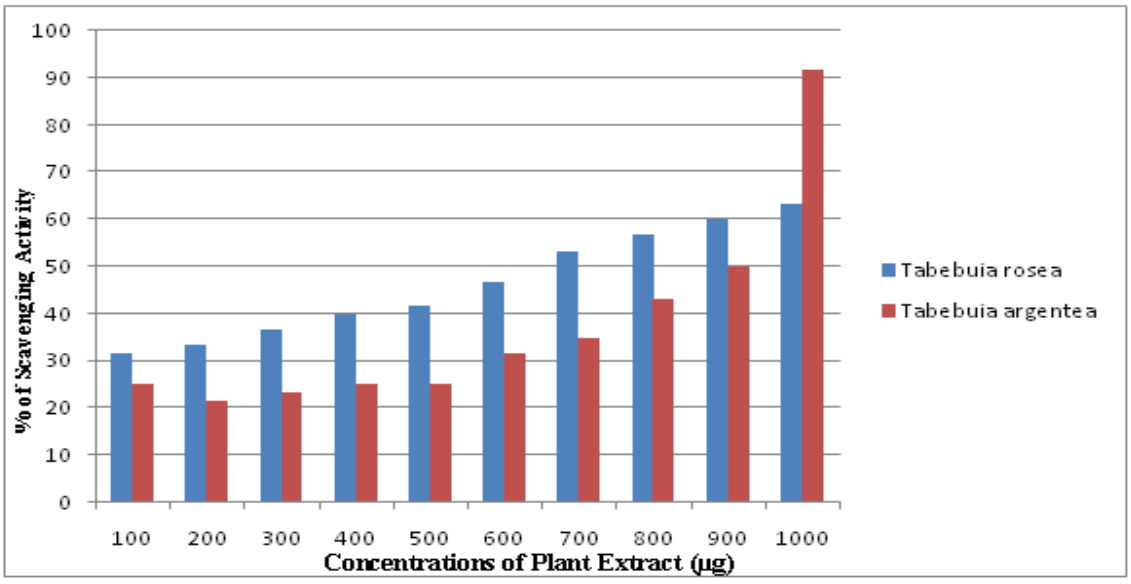

Fig 3: Super Oxide Scavenging Activity of Plant Extracts *Tabebuia rosea* and *Tabebuia*

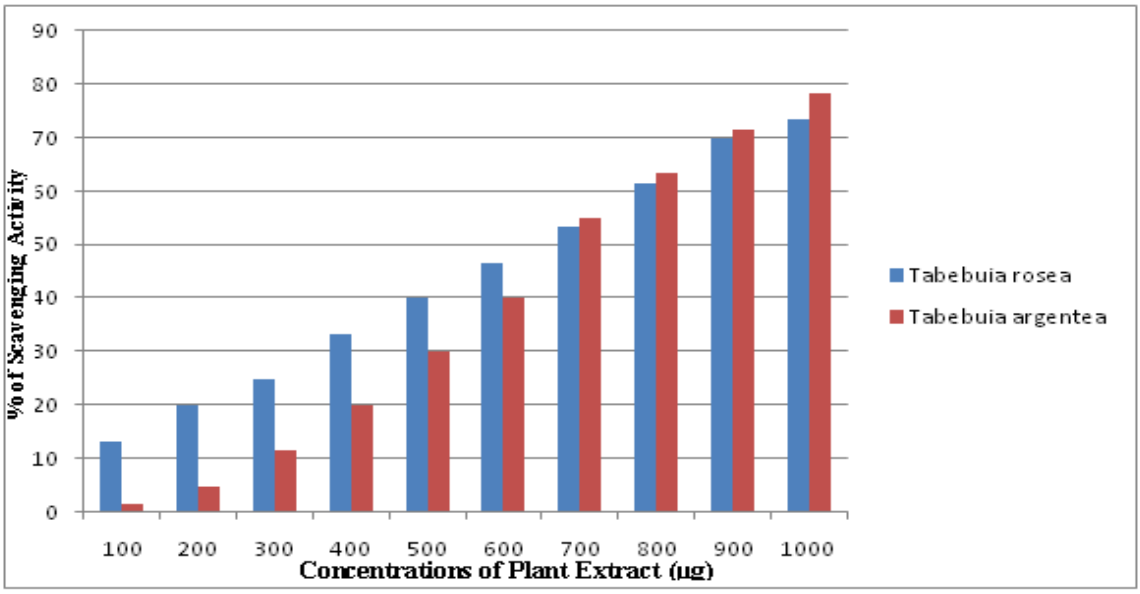

Fig 4: Total Antioxidant Capacity of Plant Extracts *Tabebuia rosea* and *Tabebuia*

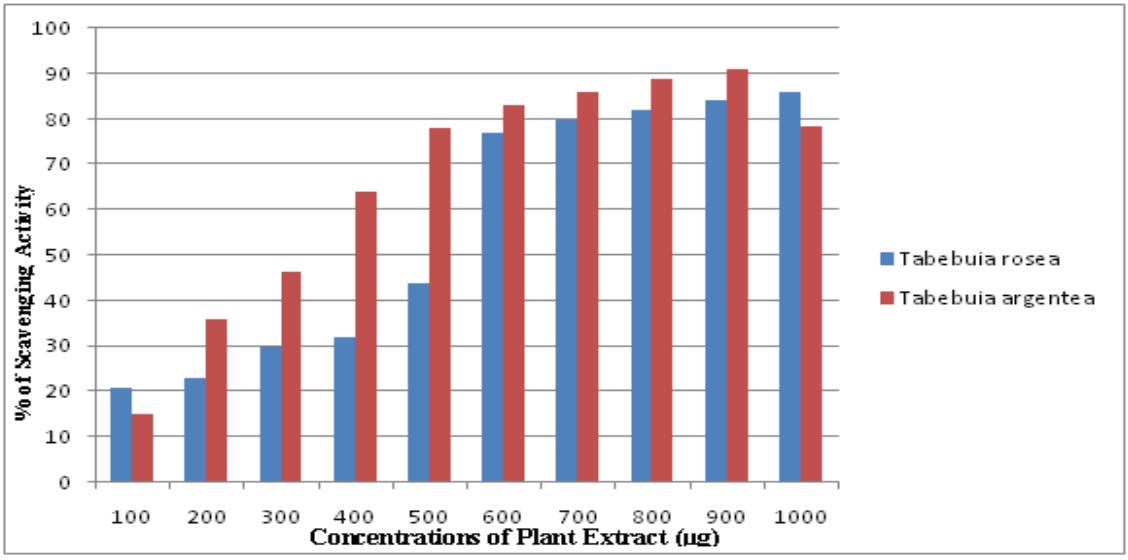

Fig 5: Superoxide Anion Radical Scavenging Assay (%) *Tabebuia rosea* and *Tabebuia*

Conclusion

The results of the present study indicate that ethanol extract of *Tabebuia rosea* and *Tabebuia argentea* exhibit strong antioxidant activities. The scavenging activities observed against DPPH, hydroxyl radicals, metal chelating, ferric thio cyanate as well as the thiobarbituric acid assay, lead us to propose *Tabebuia rosea* and *Tabebuia argentea* as promising natural sources of antioxidants suitable for application in nutritional/pharmaceutical fields, in the prevention of free radical- mediated diseases. Further studies are needed to explore the potential phenolics compound(s) from *Tabebuia rosea* and *Tabebuia* and *in vivo* studies is needed for a better understanding their mechanism of action.

Reference

1. Akira Yamauchi, Akitoyo Ichinose, Chikage Kawai and Futoshikuribayashi, Thephyagocyte NADPH oxidase and bacterial infections, Kawasaki Medical Journal, 2012, 11-18
2. Kataria AK, Kataria N. Evaluation of Oxidative Stress in Sheep Affected with Peste des petits ruminants. J Stress Physiol. Biochem. 2012, 8:72-77.
3. Auddy B, Ferreira M, Blasina F, Lafon L, Arredondo F, Dajas F *et al.* Screening of antioxidant activity of three Indian medicinal plants traditionally used for the management of neurodegenerative diseases. Journal of Ethnopharmacology. 2003; 84:131-138.
4. Chopra RN, Nayer SL, Chopra IC. Glossary of Indian medicinal plants. 3rd ed. Council of Scientific and Industrial Research, New-Delhi, India, 1992.
5. Braca A, Sortino C, Politi M. Antioxidant activity of flavonoids from Licania licaniaeflora. J Ethnopharmacol. 2002; 79:379-381
6. Yen GC, Duh PD. Scavenging effect of methanolic extracts of peanut hulls on free-radical and active oxygen species. J Agric. Food Chem. 1994; 42:629-632.
7. Nabavi SM, Ebrahimzadeh MA, Nabavi SF, Fazelian M, Eslami B. *In vitro* antioxidant and free radical scavenging activity of Diospyros lotus and Pyrus boissieriana growing in Iran. Pharmacogn Mag. 2009; 4(18):123-7.
8. Madhumitha G, Divya K, Fowsiya J. A study on phytochemical analysis, antioxidant and larvicidal activity of dried flowers of Tabebuia rosea. Journal of Chemical and Pharmaceutical Research. 2015; 7(10):693-698.
9. Sand Jeferson Caetano da Silva1, Wanderlei Barbosa dos Santos1, Maria Gabriella Silva Araujo1, Joice Fragoso da Silva Oliveira1, Regina Célia Sales Santos Veríssimo1, Thaís Honório Lins Bernardo1 *et al.* Evaluation of the Cytotoxic, Antimicrobial and Antioxidant Activity of the Plant Especies Tabebuia roseo-alba (Ridl) Journal of Chemical and Pharmaceutical Research. JC da Silva *et al.* 2017, 9(4):148-15.
10. Sobiyana P, Anburaj G, Manikandan R. Antioxidant and Antimicrobial Activities of Tabebuia Rosea. Int J Nano Corr Sci and Engg. 2015; 2(6):24-30.
11. Govindappa M, Channabasava R, Sunil Kumar KR, Pushpalatha KC. Antioxidant Activity and Phytochemical Screening of Crude Endophytes Extracts of Tabebuia argentea Bur. & K. Sch. American Journal of Plant Sciences. 2013; 4:1641-1652
12. Md. Mahbubur Rahman, Md. Badrul Islam, Mohitosh Biswas, Khurshid Alam AHM. *In vitro* antioxidant and free radical scavenging activity of different parts of *Tabebuia pallida* growing in Bangladesh. BMC Res Notes. Rahman *et al.* 2015; 8:621 DOI 10.1186/s13104-015-1618-6