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“ASSESSING THE ANTIOXIDANT ACTIVITIES OF METHANOLIC EXTRACT OF SARACA ASOCA LEAVES AND BARK”

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ABSTRACT: Chronic diseases such as cardiovascular, cancer, and respiratory diseases are being increased. Reactive oxygen species, free radicals, and oxidative stress play a major role in aggravating these diseases. Plants and plant-derived compounds have shown a greater potential in combating the detrimental effects of free radicals and other toxic agents. The current research work has been done on the leaf and bark of *Saraca asoca* to evaluate its free radical scavenging antioxidant properties. The antioxidant activities of *Saraca asoca* methanolic extract were assessed by employing ABTS Radical Scavenging Capacity, DPPH, Superoxide Radical, Hydroxyl Radical, Lipid Peroxidation, and Nitric Oxide Scavenging Ability. The leaf and bark extract results assessed against ARSC, DPPH, Superoxide, Hydroxyl, Lipid Peroxidation, Nitric Oxide, FRAP exhibited remarkable IC₅₀ values in a concentration-dependent manner, thus proving it as a potential antioxidant. The results of the present comprehensive analysis demonstrated that *Saraca asoca* leaf and bark extract exhibited excellent antioxidant activities and could be exploited for further nutraceutical and medicinal applications effective in treating many disorders.

INTRODUCTION: Many chronic diseases, including cancer and cardiovascular diseases are gaining prominent attention and are becoming a burning topic worldwide due to increased morbidity and mortality. Research studies have shown that herbs and plants have proven to be highly effective in treating and preventing such kind of disorders because of their excellent antioxidant properties.

The natural bioactive compounds present in herbs and plants, such as polyphenols, flavonoids, epigallocatechin gallate, quercetin, rutin *etc.*^{1, 2} have made it a pack of effective nutrients. Free radicals are unstable molecules that can cause damage to our body at the cellular level.

The damage to the cell is caused by free radicals, which have been implicated in developing several diseases and disorders, including macular degeneration, cardiovascular disease, pulmonary respiratory diseases, COPD, asthma, and impairment of the immune system^{3, 4}. Reactive Oxygen Species such as superoxide anion, hydrogen peroxide, and hydroxyl radical, also a part of particulate matter, are produced due to the

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continuous reduction of oxygen^{5,6}. ROS have been reported to play an important role as a mediator of the progression of cardiovascular disorder⁷. When the production of free radicals exceeds the threshold of the antioxidant defence system, oxidative stress occurs with successive oxidation of macromolecules such as DNA, protein, and lipids⁸. The human antioxidant defense system includes enzymes such as glutathione peroxidase, superoxide dismutase, catalase, and reduced glutathione⁹. The present study focuses on the role of herbs which can act as an immune booster and protect against the toxic effects of particulate matter and heavy metal pollutants¹⁰. Nutraceuticals in the herbal extract are significant in preventing many diseases by combating toxic external agents. *Saraca asoca* (Roxb.) De Wilde Syn., better known for its medicinal properties, belong to the family Leguminosae¹¹. All its parts, including bark, leaves, flowers, roots, seeds, and pods are highly useful and beneficial from the antioxidant properties due to the presence of potent natural bioactive compounds¹². The current study is to evaluate the free radical scavenging antioxidant abilities of this health-giving and healing plant that can justify its application as a folk therapeutic agent. The properties of the *Saraca asoca* need to be explored by assessing its free radical scavenging, antioxidant and phytochemical properties.

MATERIALS AND METHODS:

1. Plant Materials: In the present work, *Saraca asoca* plant was purchased from Samajik vaniki awadh kshetra Podhshala, Awadh Vanaprabhag, Lucknow (book no.10, serial no.036) whose bark and leaves were taken for sample preparation. The plant samples were furnished to an acknowledged taxonomist for botanical authentication. The given plant specimen for identification is *Saraca asoca* (Roxb) Willd of the family Fabaceae. The plant was identified by the Dept. of Pharmacognosy & Phytochemistry, Faculty of Pharmacy, Integral University, Lucknow, U. P. The accession no. for the specimen is IU/PHAR/HRB/22/10.

1.1 Extract Preparation: 100 g fresh leaves and bark of *Saraca asoca*, was shade dried and then grounded into fine powder. 5 gm of the fine powder was mixed with 50ml of methanol and extracted for 1 hour at 70°C by soxhlet extraction; then the

mixture was placed for continuous shaking at 25°C overnight. The next day the extract was filtered through Whatman no.1 filter paper and then concentrated at 40°C^{13,15}.

1.2 Chemicals and Standard: Ascorbic Acid, 2, 2 –Azino-bis (4-Ethylbenzthiazoline-6-Sulfonic acid (ABTS), ammonium persulphate, methanol, 2,2-diphenyl-1-picrylhydrazyl (DPPH), Nitroblue tetrazolium (NBT), Nicotinamide adenine dinucleotide (NaDH), Tris HCl Buffer and Phenazine methosulphate (PMS), EDTA, Ferric chloride (FeCl₃), Ascorbic Acid), hydrogen peroxide (H₂O₂) and Deoxyribose, Trichloroacetic acid (TCA), thiobarbituric acid (TBA), KCl–Tris-HCl buffer, liver homogenate, distilled water, *n*-butanol, HCl, sodium dodecyl sulfate, sodium nitroprusside, phosphate buffer saline, Griess reagent, acetate buffer, 2, 4, 6-tripyridyl-*s*-triazine (TPTZ), ferrous sulphate.

1.3 Evaluation of Antioxidant Activity: Following antioxidant assays were performed on both leaves and bark extract of *Saraca asoca*.

1.4 ABTS Radical Scavenging Activity: The ABTS (2, 2 -Azinobis (4-Ethylbenzthiazoline-6-Sulfonate) radical scavenging activity of the extracts was measured by first preparing the ABTS radicalcation (ABTS+). It was produced by reacting ABTS solution (7mM) with 2.45 mM of ammonium persulphate, and the mixture was allowed to stand in the dark at room temperature for 12-16 hour before use¹⁶.

Different concentrations (50-250µg/ml) of the bark and leaves extract prepared in methanol were taken in the volume of 0.5ml and mixed with 0.4 ml of ABTS solution, and the final volume was made up to 1ml with methanol¹⁸. For control, the absorbance of ABTS solution was measured. The absorbance was read at 745 nm, and the % inhibition was calculated. The optical density value for control at 745nm was 0.245. This value was used in the formula for calculating the % scavenging activity for each extract concentration.

The % inhibition was calculated using the formula;

$$\% \text{ ABTS radical scavenging activity} = \frac{\text{Abs. of control} - \text{Abs. of test sample}}{\text{Abs. of control}} \times 100$$

The IC₅₀ values for the extracts were calculated by plotting the curve of % inhibition and concentration of extract.

1.5 DPPH Radical Scavenging Activity: The DPPH radical scavenging activity was performed using 1, 1-diphenyl-2-picrylhydrazyl (DPPH) according to the technique outlined by W. Brand-Williams *et al.* (1995). For this activity, 0.3 Mm DPPH solution was prepared in methanol; from this, 0.5 mL of DPPH solution was transferred to a tube. Then, 100 µL of bark and leaf extracts prepared at variable concentrations ranging from 50-250 µg/mL was added to the same tube. The mixture was kept in the dark at 37°C for 30 min. The absorbance was measured spectrophotometrically at 517 nm. For control, the absorbance of freshly prepared DPPH solution was measured immediately against methanol as blank¹⁸.¹⁹ The optical density value for control at 517 nm was 0.195. This value was used in the formula for calculating the % scavenging activity for each extract concentration. The ability to scavenge DPPH radical, represented by a decrease in absorbance, was calculated using following formula, described by Brand-Williams *et al.*¹⁹

$$\% \text{ DPPH radical scavenging activity} = \frac{\text{Abs. of control} - \text{Abs. of test sample}}{\text{Abs. of control}} \times 100$$

1.6 Superoxide (SO) Radical Scavenging Activity: For this activity, 1 ml of the extract at variable concentration (50-250 µg/mL) was mixed with 0.5 ml (0.3 mM) Nitroblue tetrazolium (NBT), Nicotinamide adenine dinucleotide (NADH) (0.936 mM), Tris HCl Buffer (16 mM) (pH-8.0) and Phenazine methosulphate (PMS) (0.12 mM). The reaction mixture was left for incubation at 25°C for 5 min. A control, replacing plant extract with methanol, was prepared and treated the same way as sample extract tubes. Next measurement of absorbance at 560 nm spectrophotometrically was done¹³. The superoxide scavenging activity was calculated using the formula,

$$\% \text{ SO scavenging activity} = \frac{\text{Abs. of control} - \text{Abs. of test sample}}{\text{Abs. of control}} \times 100$$

1.7 Hydroxyl Radical Scavenging Activity: The hydroxyl radical scavenging activity was executed as per the method described by Klein, S M, *et al.* 1981 with minor modifications¹⁴. Stock solutions

of EDTA (1 mM), FeCl₃ (10 mM), Ascorbic Acid (1 mM), H₂O₂ (10 mM) and Deoxyribose (10 mM) were prepared in distilled deionized water. The process was initiated by preparing a reaction mix containing, 0.1 mL EDTA, 0.01 mL of FeCl₃, 0.1 mL H₂O₂, 0.36 mL of deoxyribose, 1.0 mL of the extract of unlike concentration (50-250 µg/mL) dissolved in methanol, 0.33 mL of phosphate buffer (50 mM, pH 7.9), 0.1 mL of ascorbic acid in succession. A control with all the components and plant extract replaced with methanol was prepared alongside. The mixture was then incubated at 37°C for 1 hour, then 1.0 mL portion of the incubated mixture was mixed with 1.0 mL of 10% TCA and 1.0 mL of 0.5% TBA to develop the chromogen, and its OD was measured at 532 nm¹⁶. The percentage inhibition was computed by comparing the results of the test with those of the control using the formula;

$$\% \text{ Hydroxyl scavenging activity} = \frac{\text{Abs. of control} - \text{Abs. of test sample}}{\text{Abs. of control}} \times 100$$

1.8. Lipid Peroxidation Inhibition Activity: For this activity determination, 2 gm of rat liver tissue was sliced and homogenized in 10 mL 15 mM KCl-Tris-HCl buffer (pH 7.2). The reaction mixture was prepared as 0.25 mL liver homogenate, 0.1 mL Tris-HCl buffer (pH 7.2), 0.05 mL 1 mM ascorbic acid, 0.05 mL 4 mM FeCl₂ and 0.05 mL of plant extract of variable concentration ranging from 50-250 µg/ml prepared in methanol was taken in tube. The reaction tubes were incubated at 37°C for 1 h.

After incubation 0.5 mL 0.1 N HCl, 0.2 mL 9.8% sodium dodecyl sulfate, 0.9 mL distilled water, and 2 mL 0.6% Thiobarbituric acid were added to each tube and vigorously shaken. Then, the tubes were placed in a boiling water bath at 100°C for 30 min. After cooling, the flocculent precipitate was removed by adding 5 mL *n*-butanol, mixed well, and centrifuged at 9,000 rpm for 10 min²⁰. The absorbance (Abs) of the supernatant was measured at 532 nm with a UV-visible double-beam spectrophotometer (Systronic, India). A control was prepared with an equal amount of all the solutions and reagents and without the plant extract, while methanol was used as the blank. The IC₅₀ values for the extracts were calculated by plotting the curve of % inhibition and concentration of extract.

After OD measurement, the % inhibition was calculated using the formula;

$$\% \text{ Lipid Peroxidation inhibition} = \frac{\text{Abs. of control} - \text{Abs. of test sample}}{\text{Abs. of control}} \times 100$$

1.9 Nitric Oxide Scavenging Activity: To measure the nitric oxide free radical scavenging activity, 50 μL of plant extract of different concentrations ranging from 50-250 $\mu\text{g/ml}$ dissolved in DMSO was taken in tube and then to the same tube methanol was added to make the volume 150 μL . After this all the tubes were loaded with 2.0mL of sodium nitroprusside (10mM) in phosphate buffer saline was added and the mixture was incubated at room temperature for 150 min. After the incubation period, the reaction mixture was loaded with 5mL of Griess reagent and the absorbance of chromophore formed was measured at 546 nm with UV-visible double beam spectrophotometer (Systronic, India). A control was prepared with equal amount of all the solutions and reagents and without the plant extract while methanol was used as the blank ²⁰. After OD measurement, the % inhibition was calculated using the formula;

$$\% \text{ Hydroxyl scavenging activity} = \frac{\text{Abs. of control} - \text{Abs. of test sample}}{\text{Abs. of control}} \times 100$$

The IC₅₀ values for the extracts was calculated by plotting the curve of % inhibition and concentration of extract.

1.10 Ferric Reducing Antioxidant Power: The reducing power of the extracts was determined according to the method described by Oyaizu, M. (1986) ¹⁷. FRAP assay measures the amount of antioxidants based on their ability to reduce Fe³⁺ to Fe²⁺. The method started with the preparation of a reaction mix containing 100 μl of extracts (50-250 $\mu\text{g/ml}$), 300 μl of distilled water, and 3ml of FRAP reagent. Freshly prepared FRAP reagent was used, which was a mixture of TPTZ (10mmol/L) dissolved in HCl (40 mmol/L), Ferric chloride (20 mmol/L), and acetate buffer (300 mmol/L, pH 3.6) in the ratio of 31:1:10. The sample mixture was then incubated for some time of 30 mins at 37°C, and later the absorbance was recorded at 593 nm. Ferrous sulfate was chosen as the standard, and a standard curve was plotted using absorbance against concentration (5–25 μM) ^{18, 21}.

The concentration of ferrous sulphate was calculated using the equation obtained from the standard curve. The amount of ferrous produced was used as the parameter to represent the antioxidant potential of the extracts.

2. Statistical Analysis: All antioxidant activities like DPPH radical scavenging activity, hydroxyl radical scavenging activity, superoxide radical activity, ABTS radical action scavenging activity were carried out in triplicate and results were expressed as mean \pm standard deviation (SD) using one-way ANOVA. $P \leq 0.05$ has been considered statistically significant for this research work. IC₅₀ values were calculated using Biod at a fit online software.

RESULTS AND DISCUSSION:

3. Antioxidant Property Determination in Leaf and Bark Methanolic Extract of *Saraca asoca*:

3.1 ABTS Radical Scavenging Activity: ABTS (2, 2'-Azino-Bis-3-Ethylbenzothiazoline-6-Sulfonic acid) is a stable free radical used for assessing the total antioxidant capacity (TAC) of natural herbal products. The optical density value for control at 745nm was 0.245. This value was used in the formula for calculating the % scavenging activity for each extract concentration. The ABTS radical scavenging activity of *Saraca asoca* leaves and bark exhibited the radical scavenging activities of 21.2% and 19.1% at the concentration of 50 $\mu\text{g/ml}$ with the O.D. of 0.193 and 0.198 at 745nm **Fig. 1**.

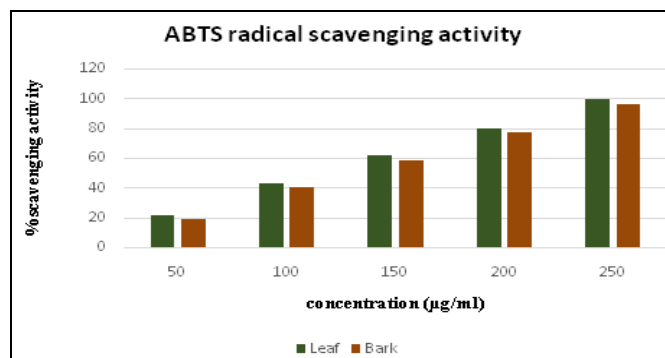


FIG. 1: ANTIOXIDANT PROPERTY OF SARACA ASOCA LEAF AND BARK METHANOLIC EXTRACTS ESTIMATED BY ABTS RADICAL SCAVENGING ACTIVITY

As the concentration of the reaction mixture increases, the absorbance decreases, and the radical scavenging activity increases.

Thus leaves exhibit slightly higher potential of ABTS radical scavenging activity than bark. In another study on the flowers of *Saraca asoca*, the IC₅₀ values were 26.18µg/mL, which shows a considerable difference from the present research work¹⁶.

3.2 DPPH Radical Scavenging Activity: The DPPH free radical scavenging activity is a broadly used method for assessing various compounds' free radical scavenging ability. DPPH assay is the most commonly used method to assess the antioxidant potential in herbal extracts to assess the free radical scavenging ability. The optical density value for control at 517nm was 0.195. This value was used in the formula for calculating the % scavenging activity for each extract concentration. A lower value of IC₅₀ indicates higher antioxidant activity. The absorbance decreased, resulting in a color change from purple to yellow, as antioxidants scavenged radicals by donating hydrogen to form the stable DPPH molecule. The result is represented in **Fig. 2** given below. In a *Saraca asoca* extract study, the DPPH radical scavenging activity exhibited 38.5 IC₅₀ (µg/mL), contrasting with the current research work²⁰.

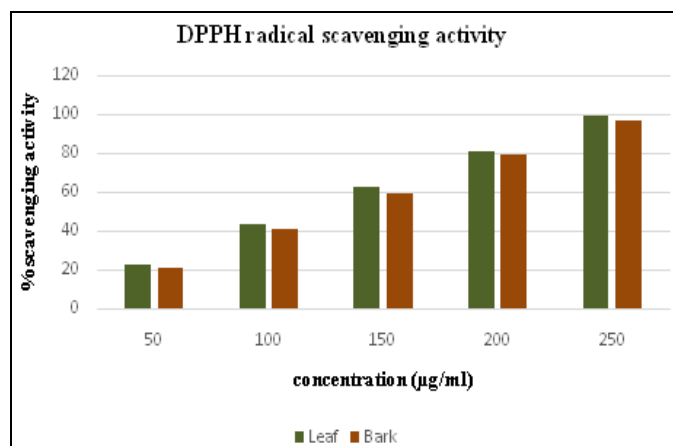


FIG. 2: ANTIOXIDANT PROPERTIES OF SARACA ASOCA LEAF AND BARK METHANOLIC EXTRACTS BY DPPH RADICAL SCAVENGING ACTIVITY

3.3 Superoxide (SO) Radical Scavenging Activity: **Fig. 3** shows the O₂⁻ scavenging activity. Superoxide anions are reactive oxygen species. It forms toxic hydroxyl radicals and singlet oxygen, leading to oxidative stress as it is a weak oxidant. The optical density value for control at 560nm was 0.257. This value was used in the formula for calculating the % scavenging activity for each concentration of extract. In the PMS/NADH-NBT

system, the O₂⁻ that has been obtained from the dissolved oxygen reduces NBT by the PMS/NADH coupling reaction, which absorbs at 560 nm. In the presence of antioxidants, the absorbance gets reduced, which exhibits superoxide scavenging in the reaction mixture. It can be concluded that as the concentration increases, the superoxide scavenging activity increases. The methanolic *Saraca asoca* leaf extract displayed a greater degree of inhibition at 75.8%, while the bark extract at 74.3% was displayed in **Fig. 3**. The result is represented in the figure below.

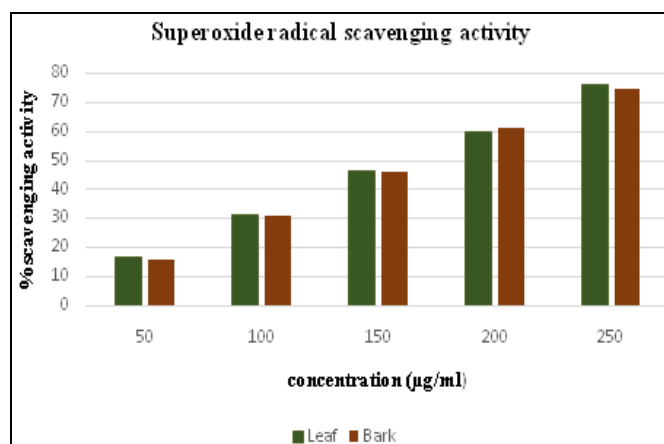


FIG. 3: ANTIOXIDANT PROPERTIES OF SARACA ASOCA LEAF AND BARK METHANOLIC EXTRACTS ESTIMATED BY SUPEROXIDE RADICAL SCAVENGING ACTIVITY

3.4 Hydroxyl Radical Scavenging Activity: The hydroxyl radical is a powerful oxidant and is one of the highly toxic reactive oxygen species as it leads to the formation of hydrogen peroxide and singlet oxygen. Hydroxyl radical attacks a broad range of molecules, including carbohydrates, lipids, and nucleic acids in living cells. So, eliminating the hydroxyl radical is of utmost importance for protecting living organisms. The optical density value for control at 532nm was 0.236. This value was used in the formula for calculating the % scavenging activity for each extract concentration. The hydroxyl radical scavenging activity increased with the concentration of both leaf and bark extracts. In the current investigational study, the hydroxyl radical scavenging activity observed was in the range of 16.1%-80.0% in *Saraca asoca* leaf extracts and 18.2%-83.4% in bark extracts at the different concentrations of 50, 100, 150, 200, and 250 (µg/ml) as represented in **Fig. 4**.

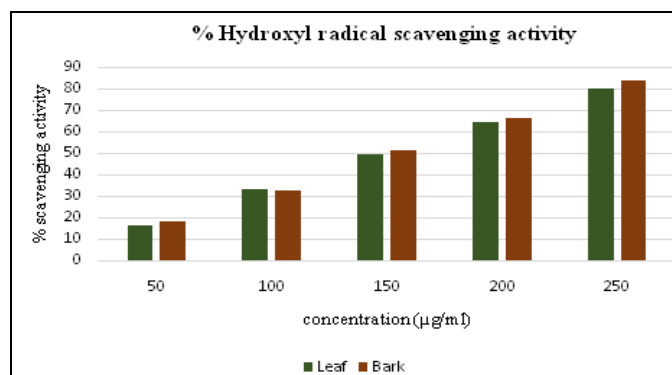


FIG. 4: ANTIOXIDANT PROPERTIES OF SARACA ASOCA LEAF AND BARK METHANOLIC EXTRACTS ESTIMATED BY HYDROXYL RADICAL SCAVENGING ACTIVITY

3.5 Lipid Peroxidation Inhibition Activity: Lipid peroxidation is the oxidative degradation of lipids where peroxides and aldehydes are generated. High levels of free radicals damage the cells by altering the protein and DNA. The effectiveness of Lipid peroxidation inhibition activity of both the *Saraca asoca* leaf and bark extracts is evident in the figure. In the current research work, the optical density value for control at 532nm was 0.252. This value was used in the formula for calculating the % inhibition activity for each extract concentration. As the concentration increases from 50 µg/ml -250 µg/ml, the absorbance decreases and % inhibition increases. So, the methanolic *Saraca asoca* leaf extract exhibits slightly greater lipid peroxidation inhibition activity with 20.2% inhibition at 50 µg/ml than *Saraca asoca* bark extract with 19.4% of inhibition at the same concentration range **Fig 5**. This is because antioxidants in the extract scavenge free radicals by resisting oxidative stress, thus inhibiting lipid peroxidation²².

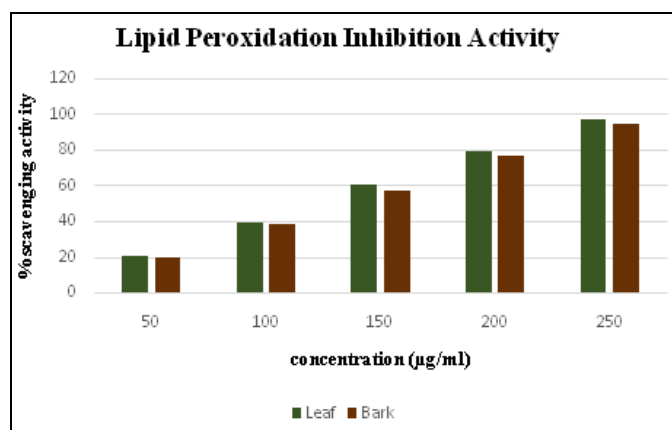


FIG. 5: ANTIOXIDANT PROPERTY OF SARACA ASOCA LEAF AND BARK METHANOLIC EXTRACT ESTIMATED BY LIPID PEROXIDASE INHIBITION ACTIVITY

3.6 Nitric Oxide Scavenging Activity: Like other reactive oxygen species, nitric oxide is another free radical responsible for inflammation, stroke, cancer, blood disorders, and other serious health diseases. Nitric oxide is a free radical and is very unstable²³. The herbs and plant products can curtail the disastrous effects of nitric oxide, thus protecting the human body against the toxic effects of nitric oxide generation. The chain of reactions generated by excessive nitric oxide that is destructive to human health is reduced by herbal extracts' antioxidant properties. The figure shows the scavenging activity of *Saraca asoca* leaf extract against nitric oxide radical released by sodium nitroprusside in a concentration-dependent manner. The figure shows the scavenging activity of *Saraca asoca* leaf extract by sodium nitroprusside against nitric oxide radical in a concentration-dependent manner. A comparable proportional scavenging activity was observed between the *Saraca asoca* leaf and bark extract. At 250 µg/mL, the percentage inhibitions of the *Saraca asoca* leaf extract and *Saraca asoca* bark extract were 94.3% and 92.7%, respectively **Fig. 6**. The optical density value for control at 546nm was 0.249. This value was used in the formula for calculating the % scavenging activity for each extract concentration. With the increase in the concentration of the extracts, the percentage of inhibition increases^{23, 24, 25}. In this study, as compared to the nitric oxide scavenging activity of 20.0-92.7%, in *Saraca asoca* bark extract, the *Saraca asoca* leaf extract exhibits 21.2-94.3% of nitric oxide scavenging activity at a concentration range of 50-250 µg/ml which indicates that leaves are better in their nitric oxide scavenging power.

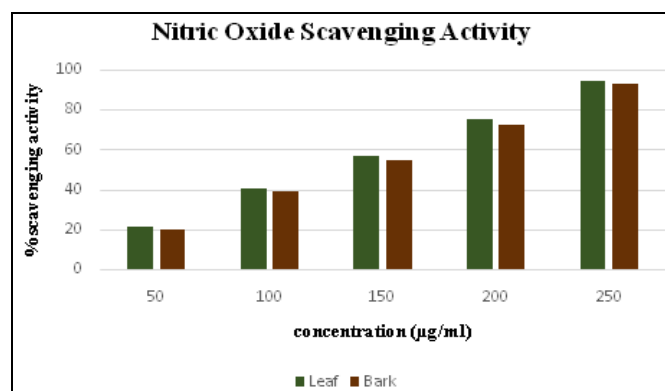


FIG. 6: ANTIOXIDANT PROPERTIES OF SARACA ASOCA LEAF AND BARK METHANOLIC EXTRACTS ESTIMATED BY NITRIC OXIDE RADICAL SCAVENGING ACTIVITY

3.7 Ferric Reducing Antioxidant Power: The result for antioxidant potential in this assay is represented as the amount of FeSO_4 generated by the action of extracts on the reaction mixture. The amount of FeSO_4 liberated at different concentrations of leaf and bark extract of *Saraca asoca* was calculated from the equation generated on the standard curve of FeSO_4 .

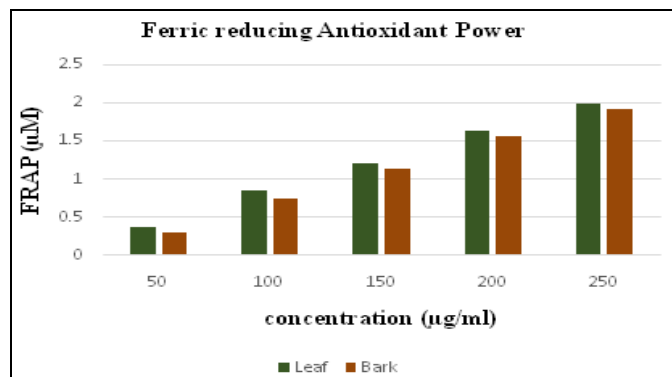


FIG. 7: ANTIOXIDANT PROPERTY OF SARACA ASOCA LEAF AND BARK EXTRACTS ESTIMATED BY FERRIC REDUCING ANTIOXIDANT POWER ASSAY

FRAP (Ferric Reducing Antioxidant Power Assay) assesses the antioxidant potential directly through the transformation of ferric ions (Fe^{3+}) to ferrous ions (Fe^{2+}) by antioxidants in the sample, unlike other antioxidant assays. The other methods indirectly assess the inhibition of free radicals in the reaction mixture, and the outcome strongly relies on the type of free radicals. For the standard preparation, a solution of FeSO_4 is prepared at different concentrations and then the optical density of this solution is measured at 593nm using a spectrophotometer. The concentration of FeSO_4 solution is in micromolar^{26, 27, 28}. For the bark sample at the lowest concentration of 50 µg/ml the optical density obtained is 0.021, so the concentration of FeSO_4 in it will be 0.31, while in the leaves sample, the optical density obtained is 0.023, so the concentration of FeSO_4 in it will be 0.38 **Fig. 7**. The ferric ion (Fe^{3+}) get reduced to ferrous ion (Fe^{2+}) by antioxidants through donating a hydrogen electron of phenolic compounds. The color observed in the test was changed to green and blue shades from yellow.

3.8 IC₅₀ Values of Leaf and Bark Extract: The activities of bark extract against ARSC, DPPH, Superoxide, Hydroxyl, Lipid Peroxidation, Nitric Oxide, FRAP was concentration dependent with

IC₅₀ value of 128.8, 126.3, 165.6, 149.2, 131.3, 134.7, 48.9 µg/ml respectively **Fig. 8**. On the other hand, the activities of leaf extract assessed against ARSC, DPPH, Superoxide, Hydroxyl, Lipid Peroxidation, Nitric Oxide, FRAP was concentration dependent with IC₅₀ value of 122.5, 121.3, 163.7, 154.7, 126.5, 130.4, 60.7 µg/ml respectively **Fig. 8**. So, this shows that both leaves and bark of *Saraca asoca* offer better free radical scavenging activities.

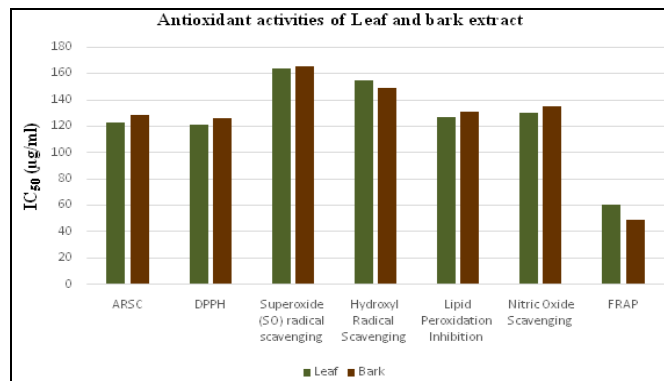


FIG. 8: IC₅₀ VALUES OF LEAF AND BARK EXTRACT

CONCLUSION: Based on the research work on antioxidant activity done on *Saraca asoca*, it can be concluded that the methanolic extract of *Saraca asoca* leaves and bark possesses excellent antioxidant and free radical scavenging properties. This indicates its potential to act as an anti-carcinogenic, cardioprotective, anti-inflammatory, anti-diabetic, hypolipidemic, and immune booster agent. Moreover, the results of the antioxidant activity show that *Saraca asocac* can prevent and treat the body from the toxic effects of free radicals and particulate matter pollutants, which are ubiquitous in the atmosphere and are highly hazardous for humans. The bioactive compounds present in it could be used in the formation of a dietary supplement or as a herbal extract which can help boost immunity and combat many disorders. So, it could be an innovative study to explore the role of *Saraca asoca* as a wonder herb using it for the development of medicinal drugs and nutraceuticals as well.

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CONFLICTS OF INTEREST: Authors declare no Conflict of Interest.

REFERENCES:

- Panche AN, Diwan AD and Chandra SR: Flavonoids: an overview. *J Nutr Sci* 2016; 29(5): 47.
- Moriassi GA, Ireri AM and Ngugi MP: *In-vivo* cognitive-enhancing, *ex-vivo* malondialdehyde-lowering activities and phytochemical profiles of aqueous and methanolic stem bark extracts of *Piliostig mathonningii* (Schum.). *Int J Alzheimers Dis* 2020; 24: 1367075.
- Chakraborty S and Roychoudhury S: Pathological Roles of Reactive Oxygen Species in Male Reproduction. *Adv Exp Med Biol* 2022; 1358: 41-62.
- Oshi M, Gandhi S, Yan L, Tokumaru Y, Wu R, Yamada A, Matsuyama R, Endo I and Takabe K: Abundance of reactive oxygen species (ROS) is associated with tumor aggressiveness, immune response, and worse survival in breast cancer. *Breast Cancer Res Treat* 2022.
- Agapov A, Olina A and Kulbachinskiy A: RNA polymerase pausing, stalling and bypass during transcription of damaged DNA: from molecular basis to functional consequences. *Nucleic Acids Res* 2022; 50(6): 3018-3041.
- Nabavi SF, Nabavi SM, Ebrahimzadeh MA, Eslami S, Jafari N & Moghaddam AH: The protective effect of curcumin against sodium fluoride-induced oxidative stress in rat heart. *Archives of Biological Sciences* 2011; 63: 563-569.
- Adegbola P, Aderibigbe I, Hammed W and Omotayo T: Antioxidant and anti-inflammatory medicinal plants have potential role in the treatment of cardiovascular disease: a review. *Am J Cardiovasc Dis* 2017; 7(2):19-32.
- Singh A, Kukreti R, Saso L and Kukreti S: Oxidative Stress: A Key Modulator in Neurodegenerative Diseases. *Molecules* 2019; 24(8): 1583.
- Sheweita SA, Abd El-Gabar M & Bastawy M: Carbon tetrachloride-induced changes in the activity of phase II drug-metabolizing enzyme in the liver of male rats: role of antioxidants. *Toxicology* 2001; 165(2-3): 217-24.
- Newman DJ and Cragg GM: Natural Products as Sources of New Drugs over the Nearly Four Decades from 01/1981 to 09/2019. *J Nat Prod* 2020; 83(3): 770-803.
- Smitha GR and Thondaiman V: Reproductive biology and breeding system of *Saraca asoca* (Roxb.) De Wilde: a vulnerable medicinal plant. *Springer Plus* 2016; 5(1): 2025.
- Ahmad SR & Ghosh PA: systematic investigation on flavonoids, catechin, β -sitosterol and lignin glycosides from *Saracaasoca* (ashoka) having anti-cancer & antioxidant properties with no side effect. *Journal of the Indian Chemical Society* 2022; 99(1): 100293.
- Vignesh A, Selvakumar S & Vasanth K: Comparative LC-MS analysis of bioactive compounds, antioxidants and antibacterial activity from leaf and callus extracts of *Saracaasoca*. *Phytomedicine Plus* 2022; 2(1): 100167.
- Klein SM, Cohen G and Cederbaum AI: Production of formaldehyde during metabolism of dimethyl sulfoxide by hydroxyl radical generating systems. *Biochemistry*. 1981; 20(21): 6006-12.
- Mittal A, Kadyan P, Gahlaut A and Dabur R: Nontargeted identification of the phenolic and other compounds of *Saracaasoca* by high performance liquid chromatography-positive electrospray ionization and quadrupole time-of-flight mass spectrometry. *ISRN Pharm* 2013; 293935.
- Mohan VR, Tresina PS, Paulpriya K and Sornalakshmi V: Antioxidant Activity of *Saraca asoca* (Roxb.) Wilde Flower: An *in-vitro* Evaluation. *International J of Pharma and Phytochemical Research* 2019; 10: 139-145.
- Oyaizu M: Studies on Products of Browning Reactions: Antioxidative Activities of Product of Browning Reaction Prepared from Glucosamine. *Japan Journal of Nutrition* 1986; 44: 307-315.
- Asokan A, Thangavel M and Nisha P: *In-vitro* antioxidant activity of *Saracaindicamethanolic* bark extract. *Int J Curr Microbiol App Sci* 2015; 4(3): 515-520.
- W Brand-Williams, ME Cuvelier and C. Berset: "Use of a free radical method to evaluate antioxidant activity," *LWT Food Science and Technology* 1995; 28(1): 25-30.
- Yadav NK, Saini KS, Hossain Z, Omer A, Sharma C, Gayen JR, Singh P, Arya KR and Singh RK: *Saracaindica* bark extract shows *in-vitro* antioxidant, antibreast cancer activity and does not exhibit toxicological effects. *Oxidative Medicine and Cellular Longevity* 2015; 205360.
- ShahinTaj RA, Sharath J and Bhagya M: Effect of Extraction Solvents on Total Phenol, Flavonoid Content and Free Radical Scavenging Potential of *Saraca asoca*, an Indian Medicinal Plant. *J Appl Pharm* 2021; 13: 287.
- Forni C, Facchiano F, Bartoli M, Pieretti S, Facchiano A, D'Arcangelo D, Norelli S, Valle G, Nisini R, Beninati S, Tabolacci C & Jadeja RN: Beneficial Role of Phytochemicals on Oxidative Stress and Age-Related Diseases. *BioMed Research International* 2019; 8748253.
- Ahmad A, Dempsey SK, Daneva Z, Azam M, Li N, Li PL & Ritter JK: Role of Nitric Oxide in the Cardiovascular and Renal Systems. *Inter J of Mole Sci* 2018; 19(9): 2605.
- Moncada A, Palmer RM and Higgs EA: Nitric oxide: physiology, pathophysiology and pharmacology. *Pharmacol Rev* 1991; 43(2): 109-42.
- Ebrahimzadeh MA, Pourmorad F and Hafezi S: Antioxidant Activities of Iranian Corn Silk. *Turkish Journal of Biology* 2008; 32: 43-49.
- Gulcin I, Oktay M, Kirecci E and Kufrevioglu I: Screening of antioxidant and antimicrobial activities of anise (*Pimpinella anisum* L.) seed extracts. *Food Chem* 2003; 83(3): 371-382.
- Chou ST, Chao WW and Chung YC: Antioxidative activity and safety of 50% ethanolic red bean extract (*Phaseolus radiatus* L. var. Aurea). *J Food Sci* 2003; 68(1): 21-25.
- Suriyaprom S, Mosoni P, Leroy S, Kaewkod T, Desvaux M and Tragoolpua Y: Antioxidants of Fruit Extracts as Antimicrobial Agents against Pathogenic Bacteria. *Antioxidants (Basel)* 2022; 11(3): 602.

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