

Comparison of Antioxidant activity of Various Phytopharmaceuticals

Vasireddy Praveen*

Vasireddy Praveen, Bapatla college of Pharmacy, Bapatla

***Corresponding Author**

Vasireddy Praveen, Vasireddy Praveen, Bapatla college of Pharmacy, Bapatla.

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Citation: Praveen, V., (2023). Comparison of Antioxidant activity of Various Phytopharmaceuticals. *J Pharmaceut Res*, 8(1), 184-189.**Abstract**

Herbal drugs are playing a pivotal role in the Indian medicine economy. The edible parts of plants are used to treat various types of diseases. There are many researches going on antioxidant activity, but our aim is to conduct *In vitro* and *in-vivo* comparison of antioxidant activity of various herbal plants to prove that natural antioxidants are more effective than synthetic antioxidants. Free radicals cause oxidative stress in the human body. Free radicals are substance with unpaired electrons which are ready to bind to cells and cause damage in the human body. For the inhibition of free radicals in the living organisms ethanolic extracts of five plants are used for the antioxidant activity. The extracts of six herbs, amla fruit powder (*Phyllanthus emblica*), Tulasi leaf powder (*Ocimum tenuiflorum*), hibiscus leaf powder (*Hibiscus rosa sinensis*), coriander leaf powder (*coriander sativum*), lemon leaf powder (*citrus limon*) and the antioxidant activity of henna leaf powder (*lawsonia inermis*) was determined. Using the hydrogen peroxide free radical scavenging activity technique, the amount of antioxidant activity present in the plant extracts was evaluated. UV Spectroscopy was used to evaluate the strong antioxidant activity of the plant extract to the reference alpha tocopherol and vitamin C. This research will assist us in determining antioxidant activity profiles, revealing that herbals are more effective than commercially available alpha tocopherol and ascorbic acid.

Keywords: UV Spectroscopy, Antioxidant activity, Herbal Medicine, Free radicals**Introduction**

India is the one of the leading producers of herbal medicine in the world. Approximately 70% of Indian population are using herbal medicine. Herbal medicines are less expensive and equally effective when compared with synthetic medicine. Herbal drugs are classified into two types single and multiple. In single type formulations are obtained from the plant part or fresh plants are used. In multiple type formulation obtained by subjecting the edible part of plant to manufacturing, distillation or extraction. The usage of herbal drugs is increased now a days because of their safety and convenience for long term storage when compared with the synthetic drugs. Free radicals cause damage to the cells in the living organism it can be prevented by antioxidant defense system developed in the living organism. Free radicals are substances which reduces the oxygen into singlet oxygen which causes oxidative stress. It leads to damage of DNA cells in the living organisms which lead to diseases like cancer and Alzheimer's disease. Free radicals are divided into two groups based on the damage they do to live cells. Reactive oxygen and nitrogen species are two types of reactive oxygen and nitrogen species. Antioxidants are chemicals that prevent or delay the oxidation of lipids in living organisms, as

well as preventing or reducing cell damage caused by free radicals. Antioxidants are used to preserve the food from deterioration and increase its shelf life and protection from microbial damage. The consumption of spoiled foods leads to heart diseases and stomach infections. Antioxidants are classified into two classes based on their origin they are natural antioxidants and synthetic antioxidants. Synthetic antioxidants are mostly of phenolic type. Some of the examples butylated hydroxy toluene (BHT). Currently Synthetic antioxidants are used by the industry to preserve food and meat products to overcome the competition and for long term storage. But increase in use of synthetic antioxidants lead to damage of DNA and cause cancers. In the recent decade the use of synthetic oxidants is replaced by natural antioxidants. Natural antioxidants which are obtained from edible parts of plant are used as antioxidants which are healthy and safe for human life. The plant parts contain other phytoconstituents like flavonoids, anthocyanins, alkaloids, Vegetables and fruits are rich in antioxidants consumption will decrease many types of diseases. Antioxidants found in the human body, such as vitamin C and vitamin E, react with free radical oxygen species to prevent them from causing damage. Antioxidants are divided into two types based on their mechanisms:

primary antioxidants that prevent the oxidation chain reaction and generate stable radicals by acting as a donor or acceptor, and secondary antioxidants that prevent the oxidation chain reaction and generate radicals that are unstable. Secondary antioxidant mechanisms that prevent metals from deactivating are involved [1-4,5]

Many plant materials contain essential number of antioxidants which are used to prevent radical scavenging activity. Different parts of plants are employed in the treatment of various diseases because they are effective against oxidation. The antioxidant activity of six distinct types of plants was determined by using the hydrogen peroxide radical scavenging activity method. Amla (*Phyllanthus emblica*) is also known as Indian gooseberry which belongs to family phyllanthaceae. Amla is rich in nutrients which helps to fight against free radicals which cause damage to the cells in the brain. Amla contains high concentration of vitamin C which help in boosting of human immunity and it is also used to reduce acidity and prevention of digestion problems. Hibiscus (*Hibiscus rosa sinensis*) is also known as rose mallow which belongs to the family malvaceae, Hibiscus leaves contain analgesic property which help to reduce body temperature given in the form of tea. It is also used to reduce soreness of throat and used treat constipation and ulcers. Coriander (*coriander sativum*) is also known as cilantro belongs to the family Apiacea. Coriander leaves are added to the tea which acts as a antidiabetic in diabetic patients. It also contains carotenoids which help in glowing and maintenance of healthy skin and also used as antacid. Lemon (*citrus limon*) also known as nimbu which belongs to the family rutaceae. Lemon leaves are rich in vitamin C which help in increase of human immunity. The tea made by using lemon leaves consumption which help in reducing weight and also acts as anti-inflammatory and also used in foods as a flavor. and Tulasi (*Ocimum tenuiflorium*) also known as holy basil which belongs to the family Lamiaceae. Tulasi leaves contain eugenol and vitamin C which help in reduction of B.P and also used in the treatment of bronchial diseases. The solution of ascorbic acid which also known as vitamin c used as a standard solution. It is used in treatment of wound healing and also acts as anti-inflammatory agent. Rose water which is bought from Patanjali store used in treatment of cuts and bruises.

Chemicals and methods:

Chemicals: Hydrogen peroxide was obtained from the apollo pharmacy. Ammonium molybdate, sodium thiosulphate, sulfuric acid, potassium iodide and ascorbic acid were obtained from Bapatla College of Pharmacy store.

Collection of plants materials

The leaves of various plants *coriander sativum*, *hibiscus rosa sinensis*, *citrus limon*, *ocimum tenuiflorium*, henna (*Lawsonia inermis*) and fruits of Amla (*Phyllanthus emblica*) were collected from the medicinal garden of Bapatla college of pharmacy and rose water was obtained from patanjali store, alpha tocopherol and ascorbic acid were obtained from sigma Aldrich. The leaves of plants were cleanly washed to remove exogenous matter and shade dried at room temperature 25 degrees for 10 days. After drying the leaves of various plants and fruit of amla were powdered with blender and then it is passed into 50 mesh sieve and stored in close container.

Preparation of plant extracts

The fruits of amla (*Phyllanthus emblica*) which is free from seeds and the leaves of *ocimum tenuiflorium*, *Hibiscus rosa sinesis*, *coriander sativum*, *citrus limon* and *lawsonia inermis* were powdered after drying. weigh accurately about 20 grams of powder and dissolve it in 150 ml of ethanol. Then the powder is extracted with aqueous distilled water by using Soxhlet extractor at 60 degrees for 6 hours. Then the solution is collected and filtered through Whatman filter paper at room temperature and the solution is concentrated by using rotary evaporator at 37 degrees for 15 minutes and the solution is stored at 4 degrees for determination of antioxidant activity by using hydrogen peroxide free radical scavenging activity.

Phytochemical Screening

The identification of numerous kinds of phenolic compounds, such as alkaloids, flavonoids, terpenoids, saponins, and so on, was done using qualitative phytochemical screening. The qualitative results were (+) for significantly present and (-) for nonexistent phytochemicals. Various tests were done for the screening of herbal extracts were noted in the table 1.

Table 1: Standard qualitative test for screening the presence of Phytopharmaceuticals

Phytochemical Constituents	Phytochemical Test	Observation
Tannins	Ferric chloride test Lead acetate test	Bluish black precipitate will be formed White Precipitate will be formed
Flavonoids	Ammonia test Sodium hydroxide test	Yellow color was observed
Alkaloids	Dragendorff test Wagner test	Yellow precipitate will be formed Reddish brown will be observed
Terpenoids	Salkowski test	Reddish brown color observed
Glycosides	Sulfuric acid test	Reddish color precipitate observed
Carbohydrates	Molisch test	Purple color observed

Saponins	Foam test	Froth will be observed
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Hydrogen peroxide radical scavenging activity

Hydrogen peroxide factoring method used to determine the hydrogen peroxide radical scavenging activity. The extract sample solution of 1ml added to the 0.1M 1ml hydrogen peroxide is added. The solution is thoroughly mixed for uniformity of solution. To this add an indicator solution of table 2 drops of 3% ammonium

molybdate was added. 10 ml of 2M H₂SO₄ (Sulfuric acid) added and 7ml of 1.8 M KI (Potassium Iodide) was added to the above mixture. Then titrate the mixture solution with 5.09M sodium thiosulphate solution till the yellow color disappears.

$$1\% = (A \text{ blank} - A \text{ sample}/A \text{ blank}) * 100$$

Table 2: Phytochemical Screening of Various herbal extracts

Plant Extract	Tannins	Flavonoids	Alkaloids	Terpenoids	Glycosides	Carbohydrates	Saponins
Hibiscus (Rosa Sinesis)	+	+	-	-	+	+	-
Amla (Phyllanthus Emblica)	+	-	-	+	+	-	+
Tulasi (Ocimum Tenuiflorium)	+	-	+	+	+	+	-
Coriander (Coriander Sativum)	-	+	-	-	-	+	+
Lemon (Citrus Limon)	+	+	+	+	+	+	+

UV Spectroscopy

The herbal extract with high antioxidant activity, ascorbic acid and alpha tocopherol were analyzed by making concentrations and absorbance were measured using UV Spectroscopy according to their lambda max [6-44].

Results and discussion

Using hydrogen peroxide radical scavenging activity, the antioxidant activity of several plant extracts was assessed. It is expressed as a percentage of inhibition. Table 3 shows that henna plant extract has high antioxidant activity and rose water has low antioxi-

dant activity, with antioxidant activity ranging from 59 to 95 percent measured by hydrogen peroxide radical scavenging activity in an invitro technique. Antioxidant activity is measured using ascorbic acid and alpha tocopherol as standards. The antioxidant activity of ascorbic acid and alpha tocopherol was evaluated using hydrogen peroxide free radical scavenging activity, and different concentrations were made for both standard and sample solutions. Absorbance was measured using UV spectroscopy, and regression analysis graphs were plotted using absorbance. The % inhibition of free radical oxygen species were calculated from the obtained absorbance and were noted in the table 3.

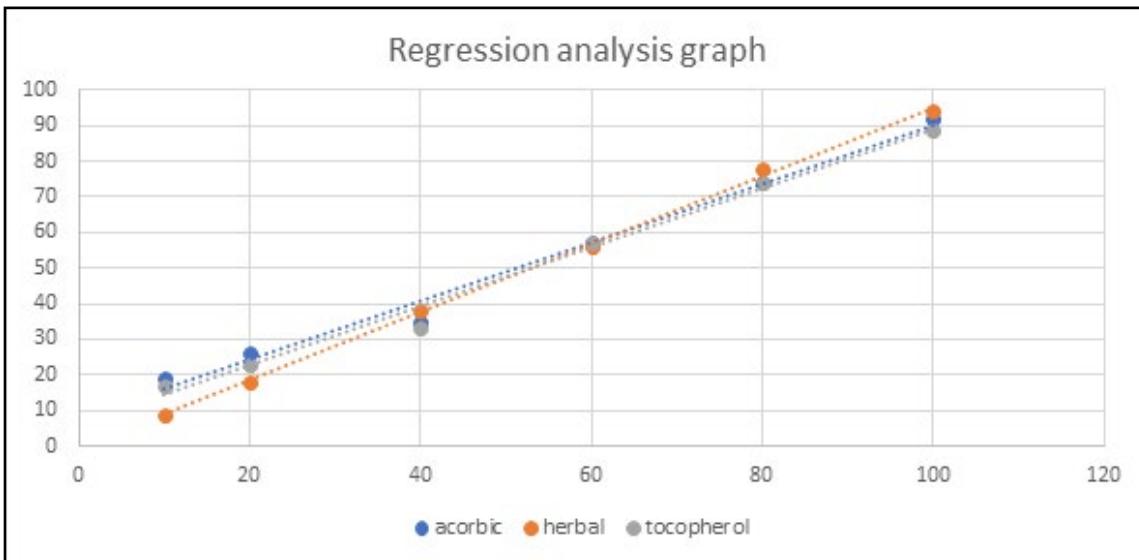
Table 3: Hydrogen Peroxide radical scavenging activity of various herbal extracts

S. No	Plant Extract	Hydrogen Peroxide radical Scavenging Activity (%)
1	Phyllanthus Emblica	85.96
2	Rose Water	58.9
3	Ocimum Tenuiflorium	72.88
4	Hibiscus rosa sinesis	78.9
5	Coriander Sativum	74.57
6	Citrus Limon	87.71
7	Henna Lawsonia inermis	91.22
8	Alpha Tocopherol	87.71
9	Ascorbic acid	89.47

Table 4: % Percentage scavenging activity according to their concentration

Concentration	Absorbance of Ascorbic Acid	Absorbance of Alpha tocopherol	Absorbance of Herbal extract	% inhibition (Ascorbic Acid)	% inhibition (Alpha Tocopherol)	% inhibition (Lawsonia Inermis)
10	0.09	0.08	0.09	19	17	9
20	0.026	0.023	0.018	26	23	18

40	0.035	0.033	0.038	35	33	38
60	0.057	0.052	0.056	57	57	56
80	0.074	0.070	0.078	74	74	78
100	0.092	0.089	0.094	92	89	94



Conclusion

We have concluded that the antioxidant activity of natural antioxidants are more effective than synthetic antioxidants.

References

1. Gupta, D. (2015). Methods for determination of antioxidant capacity: A review. *International Journal of Pharmaceutical Sciences and Research*, 6(2), 546.
2. Ravi, L., & Manasvi, V. (2016). Antibacterial and antioxidant activity of saponin from *Abutilon indicum* leaves. *Asian Journal of Pharmaceutical and Clinical Research*, 344-347.
3. Murillo Pulgarín, J. A., García Bermejo, L. F., & Carrasquero Durán, A. (2017). Determination of antioxidant activity of hibiscus flowers by flow injection analysis with chemiluminescence detection. *Analytical Letters*, 50(1), 186-196.
4. Iqbal, T., Hussain, A. I., Chatha, S. A. S., Naqvi, S. A. R., & Bokhari, T. H. (2013). Antioxidant activity and volatile and phenolic profiles of essential oil and different extracts of wild mint (*Mentha longifolia*) from the Pakistani Flora. *Journal of analytical methods in chemistry*, 2013.
5. Bariya, A. R., Patel, A. S., Gamit, V. V., Bhedi, K. R., & Parmar, R. B. (2018). Assessment of antioxidant and sensory properties of Amla (*Emblica officinalis*) fruit and seed coat powder incorporated cooked goat meat patties. *Int. J. Curr. Microbiol. Appl. Sci.*, 7, 3306-3318.
6. Munteanu, I. G., & Apetrei, C. (2021). Analytical methods used in determining antioxidant activity: A review. *International Journal of Molecular Sciences*, 22(7), 3380.
7. Aydemir, L. Y., & Yemenicioglu, A. (2014). Antioxidant Activity of Pulse Hydrocolloids: Classical Screening Methods Depending on Water Soluble Phenolic Antioxidants Need Revision to Measure True Antioxidant Potential of Pulses. *J Plant Biochem Physiol*, 2(131), 2.
8. Sharma, A., Bhardwaj, S., Mann, A. S., Jain, A., & Kharya, M. D. (2007). Screening methods of antioxidant activity: An overview. *Pharmacognosy reviews*, 1(2).
9. Shang, Y. F., Kim, S. M., & Um, B. H. (2014). Optimisation of pressurised liquid extraction of antioxidants from black bamboo leaves. *Food Chemistry*, 154, 164-170.
10. Antolovich, M., Prenzler, P. D., Patsalides, E., McDonald, S., & Robards, K. (2002). Methods for testing antioxidant activity. *Analyst*, 127(1), 183-198.
11. Caleja, C., Barros, L., Antonio, A. L., Oliveira, M. B. P., & Ferreira, I. C. (2017). A comparative study between natural and synthetic antioxidants: Evaluation of their performance after incorporation into biscuits. *Food chemistry*, 216, 342-346.
12. Khopde, S. M., Priyadarsini, K. I., Mohan, H., Gawandi, V. B., Satav, J. G., Yakhmi, J. V., ... & Mittal, J. P. (2001). Characterizing the antioxidant activity of amla (*Phyllanthus emblica*) extract. *Current science*, 185-190.
13. Koleva, I. I., Van Beek, T. A., Linssen, J. P., Groot, A. D., & Evstatieva, L. N. (2002). Screening of plant extracts for antioxidant activity: a comparative study on three testing methods. *Phytochemical Analysis: An International Journal of Plant Chemical and Biochemical Techniques*, 13(1), 8-17.
14. Pisoschi, A. M., Pop, A., Cimpeanu, C., & Predoi, G. (2016). Antioxidant capacity determination in plants and plant-derived products: A review. *Oxidative medicine and cellular longevity*, 2016.

15. Mak, Y. W., Chuah, L. O., Ahmad, R., & Bhat, R. (2013). Antioxidant and antibacterial activities of hibiscus (*Hibiscus rosa-sinensis* L.) and Cassia (*Senna bicapsularis* L.) flower extracts. *Journal of King Saud University-Science*, 25(4), 275-282.
16. Xu, D. P., Li, Y., Meng, X., Zhou, T., Zhou, Y., Zheng, J., ... & Li, H. B. (2017). Natural antioxidants in foods and medicinal plants: Extraction, assessment and resources. *International journal of molecular sciences*, 18(1), 96.
17. Katsube, T., Tabata, H., Ohta, Y., Yamasaki, Y., Anuurad, E., Shiwaku, K., & Yamane, Y. (2004). Screening for antioxidant activity in edible plant products: comparison of low-density lipoprotein oxidation assay, DPPH radical scavenging assay, and Folin– Ciocalteu assay. *Journal of agricultural and food chemistry*, 52(8), 2391-2396.
18. Behera, S. K. (2018). Phytochemical screening and antioxidant properties of methanolic extract of root of *Asparagus racemosus* Linn. *International Journal of Food Properties*, 21(1), 2681-2688.
19. Kaur, I. P., & Geetha, T. (2006). Screening methods for antioxidants-a review. *Mini reviews in medicinal chemistry*, 6(3), 305-312.
20. Tlili, N., Elfalleh, W., Hannachi, H., Yahia, Y., Khaldi, A., Ferchichi, A., & Nasri, N. (2013). Screening of natural antioxidants from selected medicinal plants. *International journal of food properties*, 16(5), 1117-1126.
21. Lee, S. E., Hwang, H. J., Ha, J. S., Jeong, H. S., & Kim, J. H. (2003). Screening of medicinal plant extracts for antioxidant activity. *Life sciences*, 73(2), 167-179.
22. Tang, E. L., Rajarajeswaran, J., Fung, S. Y., & Kanthimathi, M. S. (2013). Antioxidant activity of *Coriandrum sativum* and protection against DNA damage and cancer cell migration. *BMC complementary and alternative medicine*, 13(1), 1-13.
23. Shoker, R. M., Raheema, R. H., & Shamkhi, I. J. (2021). Antimicrobial activity, HPLC analysis of phenolic extract of *Ocimum basilicum* and *Ocimum sanctum*. *Biochemical and Cellular Archives*, 21(2), 1-8.
24. Khan, M. A., Rahman, A. A., Islam, S., Khandokhar, P., Parvin, S., Islam, M. B., ... & Alam, A. K. (2013). A comparative study on the antioxidant activity of methanolic extracts from different parts of *Morus alba* L.(Moraceae). *BMC Research Notes*, 6, 1-9.
25. Chirag, P. J., Tyagi, S., Halligudi, N., Yadav, J., Pathak, S., Singh, S. P., ... & Shankar, P. (2013). Antioxidant activity of herbal plants: a recent review. *J Drug Discov Ther*, 1(8), 01-08.
26. Meena, Harsahay et al. “Evaluation of antioxidant activity of two important memory enhancing medicinal plants *Baccopa monnieri* and *Centella asiatica*.” *Indian journal of pharmacology* vol. 44,1 (2012): 114-7. doi:10.4103/0253-7613.91880 and *Therapeutics*. 1. 01-08.
27. Badami, S., & Channabasavaraj, K. P. (2007). In Vitro. Antioxidant Activity of Thirteen Medicinal Plants of India's Western Ghats. *Pharmaceutical biology*, 45(5), 392-396.
28. Negreanu-Pirjol, T., Negreanu-Pirjol, B. S., Roncea, F., Lupu, C. E., & Jurja, S. (2015). ANTIOXIDANT ACTIVITY OF SOME AROMATIC PLANT EXTRACTS AND PHOTOPHARMACEUTICAL FORMULATIONS FOR ORAL USE. In 15th International Multidisciplinary Scientific Conference SGEM 2015 (pp. 267-274).
29. Sadat, A. B. D. U. L., Hore, M. A. Y. U. K. H., Chakraborty, K. A. U. S. H. I. K., & Roy, S. U. B. H. R. A. J. Y. O. T. I. (2017). Phytochemical analysis and antioxidant activity of methanolic extract of leaves of *corchorus olitorius*. *International Journal of Current Pharmaceutical Research*, 9(5), 59-63.
30. Siraichi, J. T. G., Felipe, D. F., Brambilla, L. Z. S., Gatto, M. J., Terra, V. A., Cecchini, A. L., ... & Cortez, D. A. G. (2013). Antioxidant capacity of the leaf extract obtained from *Arrabidaea chica* cultivated in Southern Brazil. *PLoS One*, 8(8), e72733.
31. Menaria, J., Vaghela, J. S., & Singune, S. L. (2019). Photopharmaceuticals and In-Vitro Antioxidant Potentials of Soyabean Methonolic Extract. *Journal of Drug Delivery and Therapeutics*, 9(3-s), 659-662.
32. Karla Carneiro de Siqueira Leite, Luane Ferreira Garcia, German Sanz Lobón, Douglas Vieira Thomaz, Emily Kussmaul Gonçalves Moreno, Murilo Ferreira de Carvalho, Matheus Lavorenti Rocha, Wallans Torres Pio dos Santos, Eric de Souza Gil, Antioxidant activity evaluation of dried herbal extracts: an electroanalytical approach, *Revista Brasileira de Farmacognosia*, Volume 28, Issue 3, 2018
33. Alok, S., Jain, S. K., Verma, A., Kumar, M., Mahor, A., & Sabharwal, M. (2014). Herbal antioxidant in clinical practice: A review. *Asian Pacific journal of tropical biomedicine*, 4(1), 78-84.
34. Yu, M., Gouvinhas, I., Rocha, J., & Barros, A. I. (2021). Phytochemical and antioxidant analysis of medicinal and food plants towards bioactive food and pharmaceutical resources. *Scientific reports*, 11(1), 10041.
35. Ghasemi Pirbalouti, A., Siahpoosh, A., Setayesh, M., & Craker, L. (2014). Antioxidant activity, total phenolic and flavonoid contents of some medicinal and aromatic plants used as herbal teas and condiments in Iran. *Journal of medicinal food*, 17(10), 1151-1157.
36. Pande, J., & Chanda, S. (2020, April). Mini Review: Screening of antioxidant properties of some medicinal plants. In Proceedings of the National Conference on Innovations in Biological Sciences (NCIBS).
37. Rastogi, S. H. U. B. H. I., Iqbal, M. S., & Ohri, D. E. E. P. A. K. (2018). In vitro study of anti-inflammatory and antioxidant activity of some medicinal plants and their interrelationship. *IN VITRO*, 11(4), 2455-3891.
38. Bayliak, M. M., Burdyliuk, N. I., & Lushchak, V. I. (2016). Effects of pH on antioxidant and prooxidant properties of common medicinal herbs. *Open Life Sciences*, 11(1), 298-307.
39. Al-Trad, B., Al-Qudah, M. A., Al-Zoubi, M., Muhaidat, R., & Qar, J. (2018). In-vitro and in-vivo antioxidant activity of the

- butanolic extract from the stem of Ephedra alte. Biomedical and Pharmacology Journal, 11(3), 1239-1245.
40. Ibrahim, A., & Bashir, M. Liquid Chromatography Mass Spectrometer (Lc/Ms) Profile Revealed Flavonoids And Terpenoids With Antioxidant Potential In Aqueous Fraction Of Combretum Micranthum Leaf Extract.
41. Khopde, S. M., Priyadarsini, K. I., Mohan, H., Gawandi, V. B., Satav, J. G., Yakhmi, J. V., ... & Mittal, J. P. (2001). Characterizing the antioxidant activity of amla (*Phyllanthus emblica*) extract. Current science, 185-190.
42. Khopde, S. M., Priyadarsini, K. I., Mohan, H., Gawandi, V. B., Satav, J. G., Yakhmi, J. V., ... & Mittal, J. P. (2001). Char-
- acterizing the antioxidant activity of amla (*Phyllanthus emblica*) extract. Current science, 185-190.
43. Packirisamy, R. M., Bobby, Z., Panneerselvam, S., Koshy, S. M., & Jacob, S. E. (2018). Metabolomic analysis and antioxidant effect of Amla (*Emblica officinalis*) extract in preventing oxidative stress-induced red cell damage and plasma protein alterations: An in vitro study. Journal of medicinal food, 21(1), 81-89.
44. Liu, X., Zhao, M., Wang, J., Yang, B., & Jiang, Y. (2008). Antioxidant activity of methanolic extract of emblica fruit (*Phyllanthus emblica* L.) from six regions in China. Journal of food composition and Analysis, 21(3), 219-228.

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