

Phytochemical Study, Antioxidant and Anti-inflammatory Activity of the *Carvia Callosa* (Nees) Bremek

Arun Valvi¹, Gurumeet C Wadhawa², Shubhada S. Nayak³, Vitthal S. Shivankar⁴

¹Annasaheb Awate Arts, Commerce and Hutatma Babu Genu Science College, Manchar, Pune 410503, Maharashtra, India

^{2,3}Rayat Shikshan Sanstha's, Karmaveer Bhaurao Patil College Vashi, Navi Mumbai - 400703, Maharashtra, India.,

⁴Rayat Shikshan Sanstha's, Chhatrapati Shivaji College, Satara, Maharashtra, India.

Email: ¹arunvalvi99@gmail.com, ^{2,3}wadhava.gurumeet@gmail.com

Abstract: *Carvia callosa* (Nees) Bremek is the name of a plant in the ACANTHACEAE family. This is an uncommon plant with strong activity that has been used to cure a wide range of diseases and infections. In present study, flowers and fruits of *Carvia callosa* (Nees) Bremek plant is employed for phytochemical analysis and biological activities such as antioxidants and anti-inflammatory activities. In phytochemical analysis, sterols, tannins, anthracene, saponins, flavonoids, terpenes, phenolic nucleus, terpenes, reducing sugars, alkaloids, proteins, coumarins, saponosides were present in flowers extract of *Carvia callosa* (Nees) Bremek. sterols, tannins, phenolic nucleus, terpenes, reducing sugars, coumarins, saponosides were detected in floral extract of *Carvia callosa* (Nees) Bremek. Techniques for determining antioxidant and anti-inflammatory activity that are well-known. Both the flowers and fruits extract of *Carvia callosa* (Nees) Bremek. exhibit strong antioxidant and anti-inflammatory effects. Anti-inflammatory activity is highest in ethanol extract for flowers and fruits extracts of *Carvia callosa* (Nees) Bremek.

Keywords: Antioxidant, anti-inflammatory, Fruits, flowers, *Carvia callosa* (Nees) Bremek, Phytochemicals.

1. INTRODUCTION

Family (Hindi name): ARUSA FAMILY, Family (as per The APG System III): ACANTHACEAE, Synonym(s): Strobilanthes callosa Nees, Species Name (as per The Plant List): Unresolved name, Common name: Karvi, Vernacular name: Maruadana (Hindi), Habit: Shrub, Comments / notes: Flowers once in seven to eight years, Key identifying features Include Shrubs having inflexible, rough and warted stems. Leaves elliptic-lanceolate with crenate edges and heavily lineolate on the upper surface. Flowers on sub-tetragonal spikes with rounded edges. Bracts covered with viscous secretion with a balsamic but not pleasing scent, green with pink tinge. Disc brilliant orange. Calyx segmental, leathery, gently pubescent; corolla with a white tube and a purple limb, glabrous outside with yellow hairs. Flower, Fruit: September-November, Native: India, Endemism: Central and Peninsular India [1]. Phytochemicals reveals antioxidants, anti-inflammatory, antibacterial properties which are useful to lessen the risk of chronic diseases in body. Antioxidants are substances that

protect body against damaging effect of free radicals. Inhibit oxidation and protect cell from damage caused by unstable molecule forming process of oxidation during metabolism in body such as free radicals [4]. Free radicals lead to chain reaction and start harming cells. When free radical level in body become high, it generates oxidative stress [5]. Oxidative stress plays vital part in variety of disorders like cardiovascular difficulties, malignancies, diabetes. Lipid's proteins nucleic acids are main targets of free radicals. Inflammation is critical protective response of body infection, irritation injury, swelling, heat, redness, pain are some symptoms of inflammation [6]. Cell loss, tissue injury, ischemia, cancer is also accountable for inflammation. Inflammation can be acute or persistent. Chronic inflammation last long. Anti-inflammatory chemicals are used to alleviate symptoms of inflammation in body [8]. In the present study, various phytochemicals detection analysis carried with standard methodology of flowers and fruits of *Carvia callosa* (Nees) Bremek plant. In addition, for various extract antioxidant and anti-inflammatory activity is studied of *Carvia callosa* (Nees) Bremek plant flowers and fruits (Fig. 1).



Fig. 1. *Carvia callosa* (Nees) Bremek plant.

2. RESEARCH ELABORATIONS

The blooms and fruits of *Carvia callosa* (Nees) Bremek plant were taken from Lonavala, Maharashtra. The specimen of fruits and flowers cleansed with distilled water then specimens are dried. After completion of drying, then crushed the specimens and powered of fruits and flowers *Carvia callosa* (Nees) Bremek plant kept in air tight bottles. The flower and fruits extract of the *Carvia callosa* (Nees) Bremek plant are CC-F and CC-L extract.

Phytochemical Analysis

Phytochemical testing carried using estimated protocol for CC-F and CC-L extract [9].

Sterols

An equal volume of acetic anhydride was added to test tube and gently swirled. After that, 1 ml of concentrated H₂SO₄ was poured down the tube's side [10]. The presence of sterols is indicated by the creation of a brownish-red ring at the contact zone of the two liquids and a greenish tint in the separation layer. Test is Positive for both CC-F and CC-L.

Tannins

Ferric chloride test was done for detection of tannins [11]. In this test, look of a blue changed to olive green as additional ferric chloride was applied. Ferric chloride test is positive for CC-F and CC-L.

Anthracene

5 mL chloroform was added to the powder of flowers or fruits in a test tube and stirred for 5 minutes. The mixture was filtered, and the filtrate was agitated with a 10 percent ammonia solution in an equal amount. When the aqueous layer is stirred, it turns pink, crimson, or violet, signifying the existence of free anthraquinone [12]. Anthracene detection test was positive for the CC-F and negative for CC-L.

Saponins

The powdered flowers or fruits were put to a test tube with 10 mL distilled water and vigorously shaken for 30 seconds. Afterwards, it was left to stand for 30 minutes. The presence of saponins is indicates by the production of honeycomb foam [6]. Saponins's detection test was positive for CC-F as well as negative for CC-L.

Flavonoids

Acetone was employed to entirely retain two grammes of powder of flowers or fruits. After evaporating the acetone over a water bath, the residue was removed with warm water. After sifting the mixture while it was still hot, the filtrate was allowed to cool before being used for the next test: Shinoda's experiment in 3 ml of an aqueous solution, a few magnesium chips were added, and 2 drops of mild HCl were added and warmed. The presence of flavonoids is suggested by a pink or red tint [13]. Flavonoid's detection test was Positive for CC-F as well as negative for CC-L.

Phenolic nucleus

Sodium hydroxide test is utilised for identification of phenolic nucleus [14]. Phenolic nucleus's detection test was positive for CC-F as well as negative for CC-L.

Terpenes

The Liebermann reagent test aids in the identification of terpene, resulting in the development of a blue green colour that demonstrates the presence of a terpene, whilst no pink colour shows the lack of terpenes [15]. Terpene's detection test was Positive for CC-F as well as negative for CC-L.

Reducing Sugars

The Fehling reagent was used to identify reducing sugars, which was subsequently verified by the Tollens reagent test [13]. Reducing Sugar's detection test was Positive for CC-F and CC-L.

Alkaloids

Bouchardat reagent and (reagent iodo-iodized) were used to characterize alkaloids [16]. Alkaloid's detection test was Positive for CC-F as well as negative for CC-L.

Proteins

The biuret reaction was used to detection the proteins. Add 2 -3 drops of an aqueous portion of CuSO₄ to 2% to a small volume of extract diluted in 2 mL of 20% aqueous NaOH in a test

tube. Purple colour formations indicate the presence of protein [17]. Proteins' detection test was positive for CC-F as well as negative for CC-L.

Coumarins

2 mL ethanolic solution created from each residue during extraction in two test tubes, heats both test tubes in a water bath until boiling, then add 0.5 mL of 10 percent NaOH to one of the test tubes. 4 mL distilled water in each test tube to cool it down. If the liquid from the test tube in which the alkaline solution was added is transparent or more translucent than the liquid from the control test tube (without the alkaline solution), a faint yellow solution indicates the presence of coumarin [3]. Coumarin's detection test was positive for CC-F as well as negative for CC-L.

Saponosides

8-10 mL aqueous full extract in a test tube to find saponosides. The tube was shaking for 10-15 seconds before being left alone for 12-15 minutes. saponosides Saponins are identified when the height of persistent foam was larger than 1 to 2 cm [14]. Saponoside's test was positive for CC-F as well as negative for CC-L.

Antioxidant Activity Determination

DPPH Scavenging Test: The proportion of the antioxidant contained in the sample was measured using the usual technique of the DPPH scavenging test. This test was carried out following the specific procedure. This test was carried out by making the various extracts of the plant material [18].

Study of anti-inflammatory activity (In-vitro models)

The anti-inflammatory activity of the different extracts was carried out using a slight modification of Mizushima and Kobayashi protocol with doses. The albumin test method was used [19].

3. RESULTS

Phytochemical Analysis

Based on the present investigation it can be stated that the ethanolic extract from fruits and flowers *Carvia callosa* (Nees) Bremek indicated the presence of several phytochemicals. In phytochemical analysis, sterols, tannins, anthracene, saponins, flavonoids, terpenes, phenolic nucleus, terpenes, reducing sugars, alkaloids, proteins, coumarins, saponosides were present in flowers extract of *Carvia callosa* (Nees) Bremek sterols, tannins, phenolic nucleus, terpenes, reducing sugars, coumarins, saponosides were present in flowers extract of *Carvia callosa* (Nees) Bremek.

Antioxidant Activity Determination

The antioxidant activities of the organic solvents of flowers and fruits extract of *Carvia callosa* (Nees) Bremek reported in table 1 and table 2 for BHT, C₂H₅OH, CHCl₃ and CCl₄ extract. The graphical performance of BHT, C₂H₅OH, CHCl₃ and CCl₄ for EM-F presented in Tab. 1. It shows that C₂H₅OH and CHCl₃ have stronger antioxidants performance than CCl₄. Tab. 2 revealed that DPPH radical activity of CC-L and CHCl₃ and CCl₄ exhibited greater performance than C₂H₅OH extract.

Table 1: Antioxidant activity of CC-F

Extract Conc. Mg/ml	BHT	Ethanol	CHCl ₃	CCl ₄
0.05	48.1	22.30	24.90	34.82
0.1	49.91	32.77	29.50	46.79
0.2	52.24	39.66	34.54	48.34
0.3	60.57	42.12	39.50	55.60

Table 2: Antioxidant activity of CC-L

Extract Conc. Mg/ml	BHT	Ethanol	CHCl ₃	CCl ₄
0.05	48.1	41.34	28.44	24.88
0.1	49.91	44.34	34.44	32.45
0.2	52.24	48.63	36.50	34.40
0.3	60.57	54.66	38.67	34.50

Determination of Anti-inflammatory Activity

Anti-inflammatory activity (In-vitro models) tested for *Carvia callosa* (Nees) Bremek plant's flowers and fruits extract tabulated in Tab. 3 and Tab. 4 correspondingly for standard (Ibuprofen), petroleum ether, chloroform, ethyl acetate, n-butanol and ethanol. Tab. 3 and 4 demonstrated that anti-inflammatory effectiveness is greatest in ethanol extract for both CC-F and CC-L than other extract. Mostly Ethanol extract of the flowers and fruits of *Carvia callosa* (Nees) Bremek possess in-vitro anti-inflammatory action which might be attributed to the presence of different phytochem. The chloroform extract found less performance in CC-L while ethyl acetate extract found less performance in CC-F.

Table 3: Anti-inflammatory activity of CC-F

In-Vitro Anti – inflammatory activity	Dose (mg / kg)	Absorbance value (Mean + SE)	Inhibition of denaturation (%)
Control	5ml / kg	0.098	----
Standard (Ibuprofen)	100mg/kg	0.18	90.32
Petroleum ether extract	200mg/kg	0.15	47.40
Chloroform extract	200mg/kg	0.14	36.25
Ethyl acetate extract	200mg/kg	0.12	33.26
n-Butanol	200mg/kg	0.16	54.30
Ethanol	200mg/kg	0.17	82.40

Table 4: Anti-inflammatory activity of CC-L

In-Vitro Anti – inflammatory activity	Dose (mg / kg)	Absorbance value (Mean + SE)	Inhibition of denaturation (%)
Control	5ml / kg	0.098	----
Standard (Ibuprofen)	100mg/kg	0.18	90.32
Petroleum ether extract	200mg/kg	0.12	28.19
Chloroform extract	200mg/kg	0.13	34.20
Ethyl acetate extract	200mg/kg	0.12	64.10
n-Butanol	200mg/kg	0.14	56.70
Ethanol	200mg/kg	0.12	88.19

4. CONCLUSIONS

In phytochemical examination, sterols, tannins, anthracene, saponins, flavonoids, terpenes, phenolic nucleus, terpenes, reducing sugars, alkaloids, proteins, coumarins, saponosides were detected in floral extract of *Carvia callosa* (Nees) Bremek. sterols, tannins, phenol nucleus, terpenes, reducing sugars, coumarins, saponosides were present in flowers extract of *Carvia callosa* (Nees) Bremek anti-inflammatory activity is maximal in ethanol extract for both fruits and flower extract. Fruits and flowers of *Carvia callosa* (Nees) (Nees) Bremek strong antioxidant activity as evidenced by the free radical scavenging property, can be a very effective antioxidant and can protect against the oxidative stress that is found to be an important path physiological event in a variety of diseases including aging, diabetes, cancer, cardiovascular disorders, and rheumatoid arthritis. Overall, it is a source of natural antioxidant that can be essential in illness prevention and health maintenance. Therefore, its ethnomedical claims was true according to the aforementioned testing results. This offers support to the argument for the traditional usage of the herb in the treatment of inflammation. The result of the study has seen to provide support for the use fruits and flowers of *Carvia callosa* (Nees) Bremek to promote proper conservation sustainable use of such plant resources, awareness of local communities should be enhanced incorporating the traditional knowledge with scientific findings.

ACKNOWLEDGMENT

We are very much thankful to our parent institute Rayat Shikshan Sanstha, Satara for providing the facilities for research.

5. REFERENCES

- [1] K Sankara Rao, R.K. Swamy, D. Kumar, A. Singh and K. G. Bhat. *Flora of Peninsular India*, 2019. <http://peninsula.ces.iisc.ac.in/plants.php?name=Carvia callosa>.
- [2] S. S. Nayak, N. A. Mirgane, K. B. Pathade, V. S. Shivankar and G. C. Wadhawa. "Phytochemical analysis, antioxidant and anti-inflammatory activity of leaves and bark of *Ceropegia rollae* Hemadri," *Plant Sci. Today*, 8(3), 2021, 425–428.

- [3] S. Velavan “Phytochemical Techniques-A Review,” *World J. Sci. Res.*, 1(2), 2015, 80–91.
- [4] N. A. Sasane, P. S. Gaikar, S. L. Gaikwad, A. K. Valvi, and G. C. Wadhawa, “Phytochemical analysis, anti-oxidant and anti-inflammatory of *Belosynopsis vivipara* leaves and roots,” *International Journal of Botany Studies*, 6(4), 2021, 138–142.
- [5] A. Phaniendra, D. B. Jestadi, and L. Periyasamy. “Free Radicals: Properties, Sources, Targets, and Their Implication in Various Diseases,” *Indian Journal of Clinical Biochemistry*, 30(1), 2015, 11–26.
- [6] M. S. Auwal, S. Saka, I. A. Mairiga, K. A. Sanda, A. Shuaibu, and A. Ibrahim, “Preliminary phytochemical and elemental analysis of aqueous and fractionated pod extracts of *Acacia nilotica* (Thorn mimosa).,” *Vet. Res. forum an Int. Q. J.*, 5(2), 2014, 95–100.
- [7] F. Ayertey et al., “Anti-inflammatory activity and mechanism of action of ethanolic leaf extract of *Morinda lucida* Benth,” *J. Tradit. Complement. Med.*, 11(3), 2020, 249–258.
- [8] D. Schottenfeld and J. Beebe-Dimmer, “Chronic Inflammation: A Common and Important Factor in the Pathogenesis of Neoplasia,” *CA. Cancer J. Clin.*, 56(2), 2006, 69–83.
- [9] J. Senguttuvan, S. Paulsamy, and K. Karthika, “Phytochemical analysis and evaluation of leaf and root parts of the medicinal herb, *Hypochoeris radicata* L. for in vitro antioxidant activities,” *Asian Pac. J. Trop. Biomed.*, 4, 2014, Suppl 1, S359–S367.
- [10] J. B. Harborne, *Phytochemical Methods*. Springer Netherlands, 1984.
- [11] A. Z. Yusuf, A. Zakir, M. Abdullahi and Halima, “Phytochemical analysis of the methanol leaves extract of *Paullinia pinnata* linn” *Journal of Pharmacognosy and Phytotherapy*, 6(2), 2014, 10–16.
- [12] D. K. Mhaske, D. D. Patil and G. C. Wadhawa, Antimicrobial activity of methanolic extract from rhizome and roots of *Valeriana wallichii*, *Int J Pharm Biomed Res.* 2(4), 2011, 107.
- [13] R. Gul, S. U. Jan, S. Faridullah, S. Sherani and N. Jahan, “Preliminary Phytochemical Screening, Quantitative Analysis of Alkaloids, and Antioxidant Activity of Crude Plant Extracts from *Ephedra intermedia* Indigenous to Balochistan,” *Sci. World J.*, 2017, 1.
- [14] F. Mujeeb, P. Bajpai and N. Pathak, “Phytochemical evaluation, antimicrobial activity, and determination of bioactive components from leaves of *aegle marmelos*,” *Biomed Res. Int.*, 2014, 1.
- [15] S. S. Nayak, G. C. Wadhawa, V. S. Shivankar, R. Inamadar, and M. C. Sonawale, “Phytochemical Analysis and Dpph Antioxidant Activity of Root And Bark of *Syzygium Stocksii* (Duthie) Plant,” *Eur. J. Mol. Clin. Med.*, 7(10), 2021, 2655–2668.
- [16] G. C. Wadhawa, M. A. Patare, D. D. Patil and D. K. Mhaske, Antibacterial, antioxidant and anti-inflammatory studies of leaves and roots of *Anthocephalus kadamba*. *Universal Journal of Pharmacy*, 2013.
- [17] N. A. Mirgane, A. Chandore, V. Shivankar, Y. Gaikwad and G. C. Wadhawa, “Phytochemical Study and Screening of Antioxidant, Anti-inflammatory Typhonium *Flagelliforme*,” *Res. J. Pharm. Technol.*, 14(5), 2021, 2686–2690.
- [18] S. B. Kedare and R. P. Singh, “Genesis and development of DPPH method of antioxidant assay,” *Journal of Food Science and Technology*, 48(4), 2011, 412–422.
- [19] D. D. Patil and G. C. Wadhawa, Antibacterial, Antioxidant and Anti-Inflammatory Studies of Leaves and Roots of *Solanum Xanthocarpum*, *Unique Journal of Ayurvedic and Herbal Medicines.* 1(3), 2013, 59-63.