

In vitro Anti-inflammatory and Antioxidant Activity of *Ormocarpum cochinchinense*

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Abstract

Ormocarpum cochinchinense (OC), traditionally used for bone healing, is emerging as a plant of interest for its antioxidant and anti-inflammatory properties. This herb, from the Fabaceae family and native to Southeast Asia, India's Coromandel region, tropical Africa, and Pacific islands, shows promise in addressing oxidative stress and chronic inflammation. In the context of rising health concerns linked to these conditions, OC's bioactive compounds are being studied for their potential to scavenge free radicals and mitigate disease progression. Its effects on oxidative stress and inflammation are relevant for chronic diseases such as cardiovascular conditions, neurodegenerative disorders, and periodontitis. Despite its traditional uses and regional significance, detailed scientific research on OC's pharmacological benefits is limited. This study investigates the antioxidant and anti-inflammatory activities of the ethanolic extract of OC through in vitro analyses. The goal is to enhance understanding of OC's therapeutic potential as a natural remedy for chronic inflammatory diseases.

Keywords: Antioxidant Activity, Anti-inflammatory Activity, Bone Regeneration, *Ormocarpum cochinchinense*.

Introduction

The herb *Ormocarpum cochinchinense* (OC), known for its traditional use in bone healing, is increasingly drawing attention for its potential pharmacological benefits beyond its historical applications [1]. *Ormocarpum cochinchinense* (OC), belonging to the Fabaceae family and known by various regional names such as bone-knit or Kattu Muringai [2], is distributed across Southeast Asia, India's Coromandel region, tropical Africa, and other Pacific islands. This lesser-known plant native to Southeast Asia has garnered increasing attention in recent years due to its remarkable antioxidant and anti-inflammatory properties [3]. As society faces rising health challenges associated with oxidative stress and chronic inflammation, natural remedies like

Ormocarpum cochinchinense offer promising therapeutic potential [4]. This plant's bioactive compounds have been studied for their ability to scavenge free radicals, thereby reducing cellular damage and potentially mitigating the onset or progression of various diseases [5]. Moreover, its anti-inflammatory effects contribute to alleviating symptoms associated with inflammatory conditions, presenting a holistic approach to health maintenance and disease prevention.

Inflammation and oxidative stress are implicated in a spectrum of chronic diseases, including cardiovascular ailments, neurodegenerative disorders, and periodontitis [6]. The ability of natural compounds to combat oxidative stress and modulate inflammatory processes presents a promising avenue for

therapeutic intervention [7]. *Ormocarpum cochinchinense*, with its diverse phytochemical composition, holds promise as a source of bioactive compounds that may contribute to these therapeutic effects [8].

Despite its traditional use and regional recognition, comprehensive scientific exploration of *Ormocarpum cochinchinense*'s pharmacological activities is still in its early stages [9]. This study seeks to delve into the ethanolic extract of *Ormocarpum cochinchinense*, aiming to elucidate its antioxidant and anti-inflammatory properties through rigorous in vitro analysis. By advancing our understanding of this plant's bioactivity, we aim to uncover its potential as a natural remedy in the prevention and management of chronic inflammatory diseases.

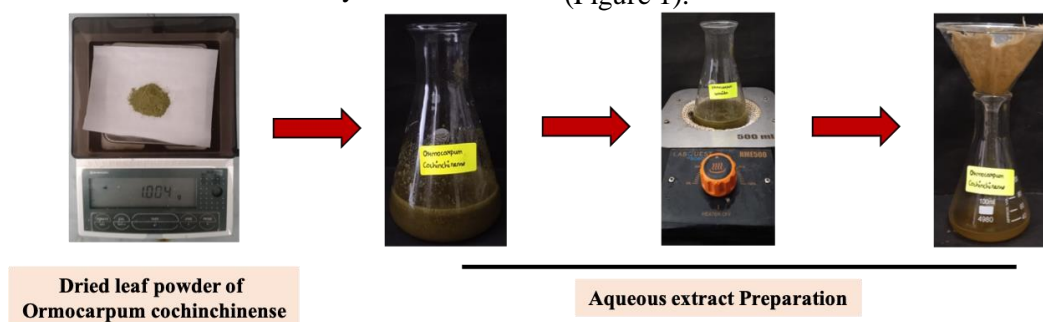


Figure 1. Schematic Representation of the Sequential Steps Involved in the Extract Preparation

Antioxidant Activity

DPPH Radical Scavenging Assay

A methanol solution of 0.1 mM 2,2-diphenyl-1-picrylhydrazyl (DPPH) was prepared as a stock solution. Before each experiment, a fresh working solution was made by diluting the stock to a final concentration of 20 μ M in methanol. *Ormocarpum*

$$\% \text{ DPPH Scavenging Activity} = [(A\text{-control} - A\text{-sample}) / A\text{-control}] \times 100$$

A control is the absorbance of the DPPH solution without the sample, and A-sample is the absorbance of the DPPH solution with the *Ormocarpum cochinchinense*. Ascorbic acid at equivalent concentrations served as the standard for comparison.

Materials and Methods

O. cochinchinense leaves were obtained from medicinal live herbal collections. These leaves were procured from the hills of Hosur, Tamil Nadu. The leaves were rinsed with distilled water, dried in a shaded and hot air oven to remove moisture, and then ground in an electric blender. Until further research, the substance was kept in a sealed container.

Aqueous Extract

1g of leaf powder was added to 100 ml of deionized water and then boiled at 60 C for 5 min and placed in a rotary apparatus for 3 days. The supernatant was filtered with Whatman filter paper (No.:1). The extract was further centrifuged and the pellets were collected (Figure 1).

cochinchinense was added at various concentrations (10, 20, 30, 40, 50 μ g/mL) to 200 μ L of the DPPH working solution in a 96-well plate. The plate was then incubated in darkness at room temperature for 10 minutes. Absorbance at 517 nm was measured using a spectrophotometer, with methanol used as a blank.

Hydrogen Peroxide Radical Scavenging Assay

The ability of biosynthesized *O.cochinchinensis* to scavenge H₂O₂ was evaluated in this study. A 40 mM H₂O₂ solution was prepared in a phosphate buffer (pH 7.4). Each test sample of *O.cochinchinense* and

a standard sample of ascorbic acid at various concentrations (10, 20, 30, 40, and 50 µg/mL) were separately added to 0.6 mL of the H₂O₂ solution. After incubating for 10 minutes in a dark environment, the absorbance of the

$$\% \text{ inhibition} = \frac{\text{Absorbance of control} - \text{Absorbance of sample}}{\text{Absorbance of control}} \times 100$$

A control is the absorbance of the control (H₂O₂ solution without the sample), and A-sample is the absorbance of the sample (H₂O₂ solution with either *O.cochinchinense* or ascorbic acid).

FRAP Assay

Reagents for FRAP Assay

1. **Acetate Buffer 300 mM pH 3.6:** Sodium acetate trihydrate (3.1g) was weighed and mixed with 16 mL of glacial acetic acid, then diluted to 1 L with distilled water to achieve a pH of 3.6.
2. **TPTZ (2, 4, 6-tripyridyl s-triazine):** TPTZ was prepared at a concentration of 10 mM in 40 mM HCl, with a molecular weight of 312.34.
3. **FeCl₃ · 6H₂O:** Iron chloride hexahydrate was prepared at a concentration of 20 mM, with a molecular weight of 270.30.

The working FRAP reagent was prepared by mixing components a, b, and c in a ratio of 10:1:1 immediately before use. The standard used was FeSO₄·7H₂O, prepared at concentrations ranging from 0.1 mM to 1.5 mM in methanol. All reagents were sourced from Merck (Germany).

2.3 mL of the FRAP reagent was combined with 0.7 mL of aqueous extract of *Ormocarpum cochinchinense* at various concentrations (10, 20, 30, 40, and 50 µg/mL). This mixture was then incubated in darkness at 37°C for 30 minutes. The absorbance was measured at 593 nm using a spectrophotometer against a blank containing all reagents except the sample. An increase in absorbance of the reaction mixture indicates an enhancement in reduction capability. Each sample was analyzed in

triplicate, with ascorbic acid serving as the standard. reaction mixture was measured spectrophotometrically at 230 nm. Vitamin C served as the standard reference. The percentage of H₂O₂ scavenging activity was determined using the formula:

triplicate, with ascorbic acid serving as the standard.

ABTS

ABTS radical cation (ABTS⁺) was generated by reacting 7.0 mM ABTS in 50% ethanol with 2.45 mM potassium persulfate in distilled water. This reagent was refrigerated for at least 24 hours before use. Before use, it was diluted with 50% ethanol to achieve an absorbance of 1.0 (± 0.02) at 734 nm. In 96-well microplates, 250 µL of ABTS⁺ solution and 20 µL of *Ormocarpum cochinchinense* samples (at various concentrations: 10, 20, 30, 40, and 50 µg/mL dissolved in distilled water) were added. Ascorbic acid served as the standard. The blank consisted of 20 µL of ethanol. After a 10-minute incubation in darkness, absorbance readings were taken at 734 nm using a microplate reader. Radical scavenging activity was calculated using the formula:

$I (\%) = [(Abs_0 - Abs_1) / Abs_0] \times 100$, where Abs₀ is the absorbance of the blank and Abs₁ is the absorbance in the presence of the test compound at different concentrations.

Nitric Oxide Radical Inhibition Assay

Nitric oxide radical inhibition was assessed using the Griess Illosvoy reaction, modified with naphthyl ethylene diamine dihydrochloride (0.1% w/v) replacing 1-naphthylamine (5%). The reaction mixture (3 mL) containing sodium nitroprusside (10 mM, 2 mL), phosphate-buffered saline (0.5 mL), and *Ormocarpum cochinchinense* extract (at concentrations of 10, 20, 30, 40, and 50 µg/mL) or standard solution (ascorbic acid, 0.5 mL) was incubated at 25°C for 150 minutes. After incubation, 0.5 mL of the reaction mixture was

mixed with sulfanilic acid reagent (0.33% in 20% glacial acetic acid) and left for 5 minutes for diazotization. Then, 1 mL of naphthyl ethylenediamine dihydrochloride was added, mixed, and left for 30 minutes at 25°C to

$$\% \text{ scavenging/Reduction} = [\text{Absorbance of control} - \text{Absorbance of } O.\text{cochinchinense} \text{ sample} / \text{Absorbance of control}] \times 100$$

Anti-inflammatory Activity

Bovine Serum Albumin Denaturation Assay

The anti-inflammatory activity of *Ormacarpum cochinchinense* was evaluated using the bovine serum albumin (BSA) denaturation assay. Various concentrations (10, 20, 30, 40, 50 µg/mL) of *O.cochinchinense* were mixed with 0.45 mL of BSA, with the pH

$$\% \text{ inhibition} = \frac{\text{Absorbance of control} - \text{Absorbance of sample} \times 100}{\text{Absorbance of control}}$$

Egg Albumin Denaturation Assay

The egg albumin denaturation assay was conducted by mixing 0.2 mL of fresh egg albumin with 2.8 mL of 1X phosphate buffer. Various concentrations (10, 20, 30, 40, 50 µg/mL) of *O.cochinchinense* were introduced into the mixture, and the pH was adjusted to

$$\% \text{ inhibition} = \frac{\text{Absorbance of control} - \text{Absorbance of sample} \times 100}{\text{Absorbance of control}}$$

Membrane Stabilization Assay

The *in vitro* membrane stabilization assay evaluates how well compounds stabilize cell membranes and prevent the release of intracellular contents (10). The assay uses human red blood cells (RBCs), phosphate-buffered saline (PBS), Tris-HCl buffer (50 mM, pH 7.4) *Ormacarpum* extract at various concentrations (10, 20, 30, 40, 50 µg/mL) were evaluated.

To prepare the RBC suspension, fresh blood with anticoagulant is centrifuged at 3000 RPM for 10 minutes. The supernatant is discarded,

develop a pink chromophore under diffused light. The absorbance of these solutions was measured at 540 nm against corresponding blank solutions.

adjusted to 6.3. The mixture was then left at room temperature for 10 minutes and incubated at 55°C for 30 minutes. Diclofenac sodium served as the positive control, while dimethyl sulfoxide was used as the negative control. Absorbance was measured at 660 nm using a spectrophotometer.

The percentage of protein denaturation:

6.3. The mixture was then incubated at room temperature for 10 minutes before being heated in a water bath at 55°C for 30 minutes. Diclofenac sodium was used as a standard reference, while dimethyl sulfoxide served as the control. Absorbance was recorded at 660 nm using a spectrophotometer.

and the RBCs are washed three times with PBS before being resuspended in Tris-HCl buffer to create a 10% (v/v) suspension.

For the assay, 1 mL of the RBC suspension is added to each centrifuge tube, followed by different concentrations of silver nanoparticles. The tubes are mixed and incubated at 37°C for 30 minutes, then centrifuged at 2500 RPM for 5 minutes. The absorbance of the supernatant is measured at 560 nm using a UV-Vis spectrophotometer to assess membrane stabilization.

The percentage inhibition of haemolysis using the following formula:

$$\% \text{ inhibition} = [(\text{OD control} - \text{OD sample}) / \text{OD control}] \times 100$$

Where OD control is the absorbance of the RBC suspension without the test compound(s) and OD sample is the absorbance of the RBC suspension with the test.

Data Analysis

Data analysis was conducted using IBM SPSS version 21, with statistical significance set at the 0.05 level. Descriptive statistics, including means and standard deviations, were calculated for continuous variables to summarize the data. To assess the distribution of the data, the Shapiro-Wilk test was performed to evaluate normality. For analyzing the effects of the aqueous extract of *Ormocarpum cochinchinense* on antioxidant and anti-inflammatory activities, one-way Analysis of Variance (ANOVA) was employed for normally distributed data, while the Kruskal-Wallis test was utilized for non-normally distributed data. These tests were used to determine significant differences in antioxidant and anti-inflammatory effects across different concentrations of the extract. This approach allowed for a robust assessment of the extract's efficacy and ensured an accurate interpretation of its biological activities.

Results and Discussion

The study evaluated the antioxidant and anti-inflammatory effects of the ethanolic extract of *O. cochinchinense*. Antioxidants are commonly used alongside traditional treatments to mitigate oxidative stress in inflammatory diseases. Plants are a key source of natural antioxidants, though their effectiveness can vary depending on the timing of their collection.

Antioxidant Activity

Research into *Ormocarpum cochinchinense* (OC) has revealed a variety of bioactive compounds that contribute to its notable antioxidant activity. The plant is rich in flavonoids, such as quercetin and kaempferol, along with their glycosides, which play a crucial role in scavenging free radicals by

donating electrons to neutralize them. This action helps inhibit lipid peroxidation, thus protecting biological membranes from oxidative damage. Additionally, phenolic acids like gallic and caffeic acid enhance the plant's antioxidant capacity by serving as hydrogen donors and metal chelators, preventing oxidative chain reactions. The antioxidant activity of OC operates through multiple mechanisms, including strong free radical scavenging, particularly against reactive oxygen species (ROS) like superoxide anions, hydroxyl radicals, and peroxy radicals. These effects are largely due to the plant's phenolic content, which stabilizes free radicals by donating hydrogen atoms or electrons. Moreover, OC demonstrates metal-chelating abilities that reduce the availability of transition metals like iron and copper, which can catalyze free radical production [10,11].

Its antioxidant compounds are also effective in inhibiting lipid peroxidation, a key process in oxidative damage to cell membranes, and may further enhance the body's natural antioxidant defences by upregulating endogenous enzymes such as superoxide dismutase (SOD), catalase, and glutathione peroxidase. Comparative studies have shown that OC exhibits antioxidant activity comparable to, or even surpassing, standard antioxidants like ascorbic acid and butylated hydroxytoluene (BHT) in various in vitro assays. Given this, the plant holds significant potential for therapeutic applications, particularly in the prevention and management of oxidative stress-related diseases like cancer, diabetes, and cardiovascular conditions. Natural antioxidants, like those found in OC, are often less toxic and have fewer side effects compared to synthetic alternatives, making this plant a valuable addition to nutraceutical formulations designed to boost antioxidant defences. Overall, the rich phytochemical composition of OC underscores its potential as a natural source of antioxidants with promising applications in medicine, food preservation, and other

industries, highlighting the importance of its conservation and further research [12].

The antioxidant activity of *Ormocarpum cochinchinense* was evaluated utilizing a variety of assays, including DPPH radical scavenging, H₂O₂ scavenging, FRAP, ABTS, and nitric oxide.

DPPH

Table 1. Antioxidant Activity of *Ormocarpum cochinchinense* using DPPH Radical Scavenging Assay

DPPH	Standard	<i>Ormocarpum cochinchinense</i>
10	66.25	62.16
20	78.52	74.37
30	85.63	83.39
40	88.68	86.58
50	93.15	91.45

P value: 0.6934 - 69.34%

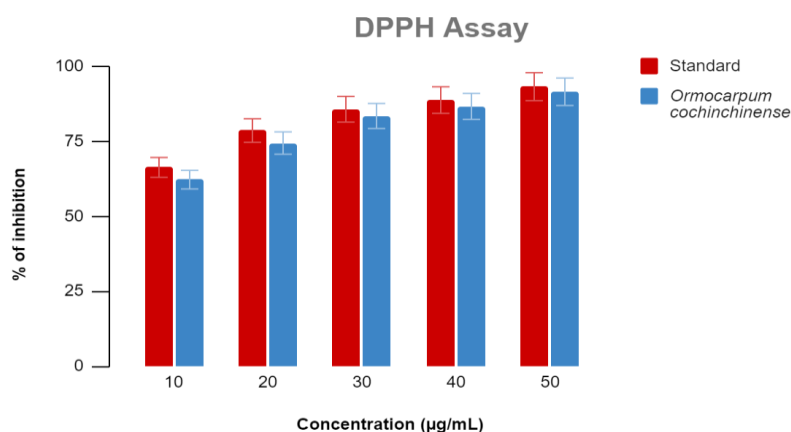


Figure 2. Antioxidant Activity of *Ormocarpum cochinchinense* - DPPH ASSAY

H₂O₂

Table 2 and Figure 3 demonstrate that the scavenging activity of *Ormocarpum cochinchinense* against hydrogen peroxide increased with concentration, reaching 87.9%

Table 1 and Figure 2 show that *Ormocarpum cochinchinense* has significant antioxidant activity in the DPPH assay. The greatest concentration of 50 µg/mL resulted in 91.45% radical scavenging activity. This implies that *Ormocarpum cochinchinense* can effectively neutralize DPPH radicals, indicating a robust free-radical scavenging potential.

at 50 µg/mL. This assay assesses the plant's ability to neutralize hydrogen peroxide, a reactive oxygen species that can cause oxidative damage. The reported p-value of 79.52% suggests significant antioxidant activity over H₂O₂.

Table 2. Antioxidant Activity of *Ormocarpum cochinchinense* - H₂O₂ ASSAY

H ₂ O ₂	Standard	<i>Ormocarpum cochinchinense</i>
10	51.1	49.4
20	56.9	52.1
30	66.1	65.6
40	78.8	74.3
50	89.9	87.9

p-79.52%

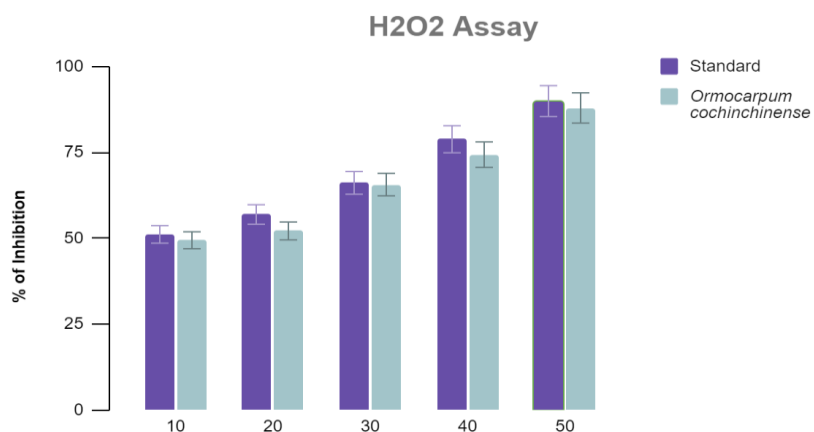


Figure 3. Antioxidant Activity of *Ormocarpum cochinchinense* - H₂O₂ ASSAY

FRAP

Table 3 and Figure 4 show that *Ormocarpum cochinchinense* has a progressive rise in antioxidant activity in the FRAP experiment,

reaching 87.63% at 50 µg/mL. The FRAP assay detects the reduction in ferric ions to ferrous ions and indicates the plant's reducing power. This corroborates the plant's considerable antioxidant capability.

Table 3. Antioxidant Activity of *Ormocarpum cochinchinense* - FRAP ASSAY

FRAP		
Conc	Standard	<i>Ormocarpum cochinchinense</i>
10	72.98	70.27
20	76.84	74.52
30	81.31	78.38
40	85.84	81.64
50	90.89	87.63

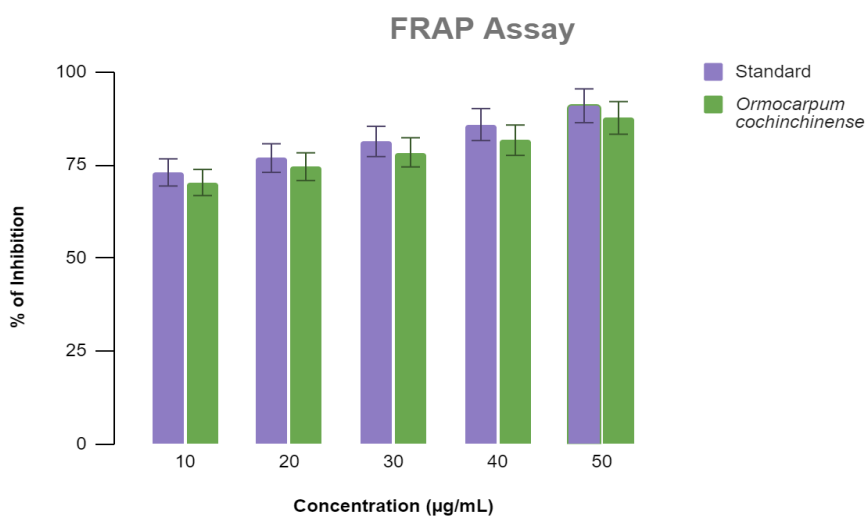


Figure 4. Antioxidant Activity of *Ormocarpum cochinchinense* - FRAP ASSAY

ABTS

Table 4 and Figure 5 show that *Ormocarpum cochinchinense* has strong ABTS radical scavenging activity, with a maximal activity of

88.35% at 50 $\mu\text{g/mL}$. The ABTS assay evaluates the capacity of the plant to neutralize the ABTS radical cation, providing additional proof of its antioxidant activity.

Table 4. Antioxidant Activity *Ormocarpum cochinchinense* - ABTS ASSAY

ABTS		
Conc	Standard	<i>Ormocarpum cochinchinense</i>
10	70.56	67.28
20	75.68	72.91
30	82.43	78.49
40	86.57	84.72
50	91.39	88.35

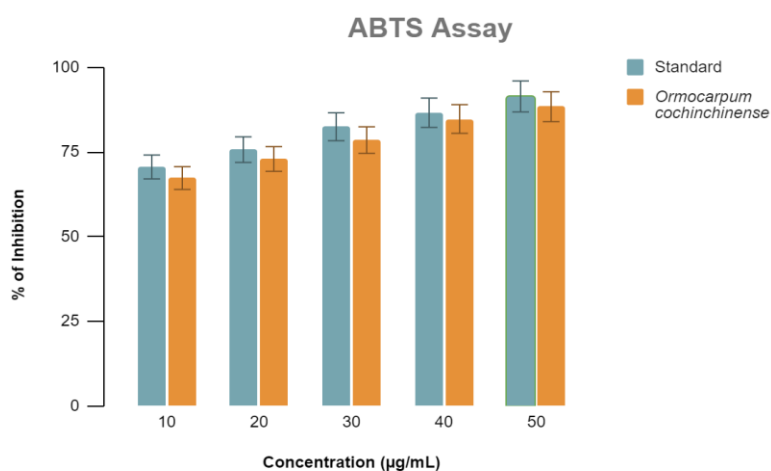


Figure 5. Antioxidant Activity of *Ormocarpum cochinchinense* - ABTS Assay

Nitric Oxide

As shown in Table 5 and Figure 6, *Ormocarpum cochinchinense* effectively

scavenged nitric oxide radicals, with 86.15% activity at the maximum concentration.

Table 5. Antioxidant Activity of *Ormocarpum cochinchinense* - Nitric Oxide Assay

Nitric oxide		
Conc	Standard	<i>Ormocarpum cochinchinense</i>
10	72.43	68.62
20	77.94	73.34
30	80.37	77.46
40	84.28	82.79
50	88.67	86.15

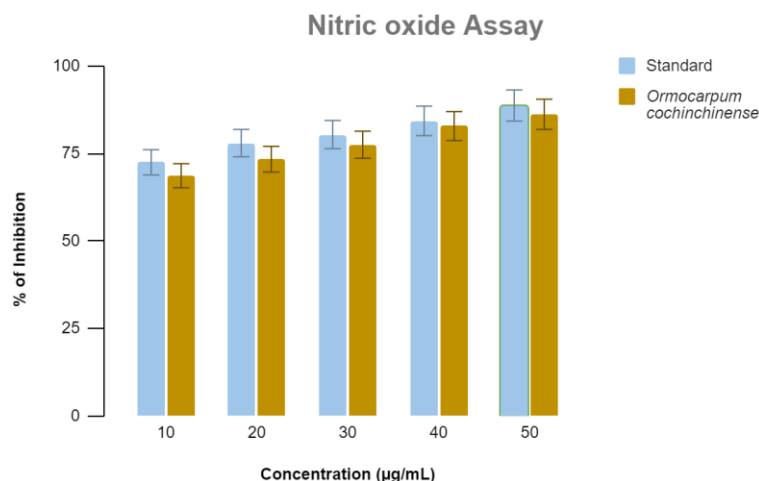


Figure 6. Antioxidant Activity of *Ormocarpum cochinchinense* - Nitric Oxide Assay

Anti-inflammatory Activity

The anti-inflammatory activity of OC has garnered interest due to its potential in managing inflammatory conditions, which are often precursors to various chronic diseases, such as arthritis, cardiovascular disease, and certain cancers. The anti-inflammatory properties of this plant are largely attributed to its diverse phytochemical composition, which includes flavonoids, alkaloids, tannins, and terpenoids. These bioactive compounds interact with inflammatory pathways to inhibit the production of pro-inflammatory mediators and modulate immune responses [3].

One of the primary mechanisms of OC anti-inflammatory activity is the inhibition of pro-inflammatory cytokines such as interleukin-1 (IL-1), interleukin-6 (IL-6), and tumour necrosis factor-alpha (TNF- α), which are key players in the inflammatory process. By reducing the levels of these cytokines, the plant helps to alleviate inflammation and prevent tissue damage. Additionally, OC has been found to inhibit the expression of cyclooxygenase-2 (COX-2) and lipoxygenase (LOX) enzymes, which are responsible for the production of inflammatory mediators like prostaglandins and leukotrienes. This inhibition reduces the synthesis of these pro-inflammatory compounds, providing relief from inflammation and associated pain [13-15].

Flavonoids, such as quercetin and kaempferol, present in OC are particularly noted for their ability to inhibit the activation of nuclear factor-kappa B (NF- κ B), a transcription factor that regulates the expression of genes involved in inflammation. By suppressing NF- κ B activity, the plant further downregulates the inflammatory response at the molecular level. Moreover, the plant's phenolic compounds exert antioxidant effects, which indirectly contribute to its anti-inflammatory action by reducing oxidative stress that often exacerbates inflammation [16-20].

Studies have demonstrated that extracts of *Ormocarpum cochinchinense* (OC) exhibit significant anti-inflammatory effects in both in vitro and in vivo models. In animal studies, the administration of OC extracts has led to a marked reduction in inflammation-induced oedema, supporting its potential as a natural anti-inflammatory agent. These effects are believed to stem from the plant's ability to modulate the immune system and inhibit the migration of inflammatory cells to the site of inflammation [13].

Given these promising results, OC holds considerable potential for therapeutic applications in the management of inflammatory conditions. Its anti-inflammatory activity could be beneficial in treating conditions such as rheumatoid arthritis,

inflammatory bowel disease, and other chronic inflammatory disorders. Additionally, the plant's natural origin may offer a safer alternative to synthetic anti-inflammatory drugs, which are often associated with side effects such as gastrointestinal irritation and cardiovascular risks.

The anti-inflammatory activity of *Ormocarpum cochinchinense* is driven by its rich phytochemical profile, particularly its flavonoids and phenolic acids. These compounds act on multiple inflammatory pathways, reducing the production of pro-inflammatory cytokines, inhibiting key enzymes involved in inflammation, and modulating immune responses. The plant shows potential for use in both pharmaceutical and nutraceutical formulations aimed at

managing inflammatory conditions, highlighting the need for further research to explore its clinical efficacy and safety in human populations.

Ormocarpum cochinchinense's anti-inflammatory properties were evaluated using three different assays: BSA, EA, and MSA. Tables 6–8 detail the results, which are represented in the related graphs.

BSA

Table 6 and Figure 7 show that *Ormocarpum cochinchinense* increased anti-inflammatory activity with concentration, reaching 81% at 50 µg/mL. This assay assesses the plant's potential to reduce protein denaturation, a process linked to inflammation.

Table 6. Anti-Inflammatory Activity of *Ormocarpum cochinchinense* - BSA Assay

BSA	10	20	30	40	50
<i>Ormocarpum cochinchinense</i>	45	57	69	76	81
Standard	47	60	72	78	84

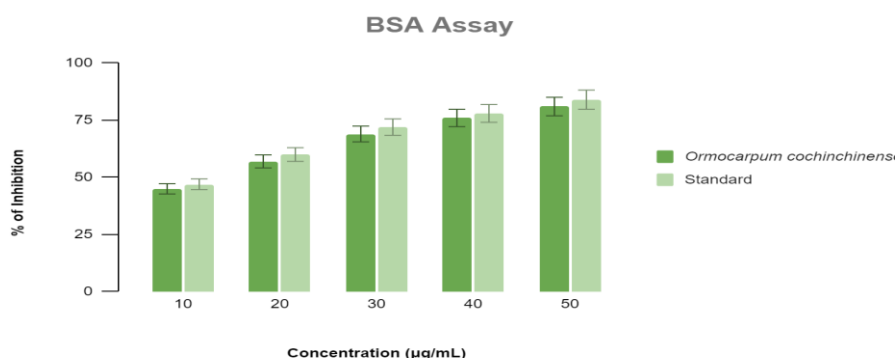


Figure 7. Anti-Inflammatory Activity of *Ormocarpum cochinchinense* - BSA Assay

EA

Table 7 and Figure 8 reveal that *Ormocarpum cochinchinense* suppressed

inflammation in the EA assay, with an activity of 78% at 50 µg/mL. The EA assay assesses the plant's ability to alleviate inflammation-related oedema.

Table 7. Anti Inflammatory Activity of *Ormocarpum cochinchinense* - EA ASSAY

EA	10	20	30	40	50
<i>Ormocarpum cochinchinense</i>	5	61	66	70	78
Standard	55	64	69	72	81

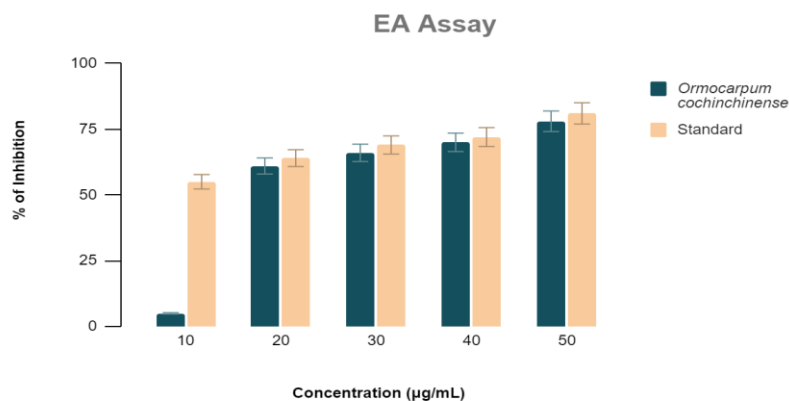


Figure 8. Anti-Inflammatory Activity of *Ormocarpum cochinchinense* – EA Assay

MSA

Table 8 and Figure 9 show that *Ormocarpum cochinchinense* has a significant anti-inflammatory action in the MSA assay, with 85% inhibition at the maximum concentration. This assay measures the plant's ability to minimize inflammation by reducing the synthesis of inflammatory mediators. Overall, *Ormocarpum cochinchinense* has substantial

antioxidant and anti-inflammatory properties across a variety of assays. The constant rise in activity with increasing concentrations indicates a dose-dependent responsiveness in both antioxidant and anti-inflammatory actions. The plant's capacity to scavenge various reactive species and reduce inflammation indicates its promise as a source of alternative chemicals for therapeutic purposes.

Table 8. Anti-Inflammatory Activity of *Ormocarpum cochinchinense* - MSA Assay

MSA	10	20	30	40	50
<i>Ormocarpum cochinchinense</i>	55	68	75	78	85
Standard	58	70	77	82	89

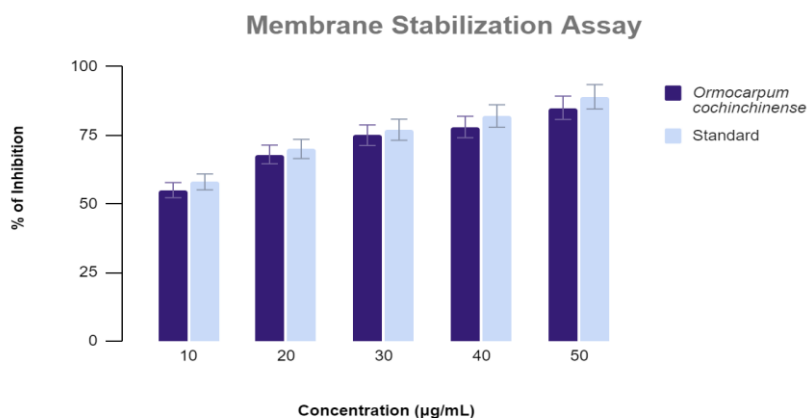


Figure 9. Anti-Inflammatory Activity of *Ormocarpum cochinchinense* – MSA Assay

Further research could look into the exact chemicals responsible for these effects, as well as their methods of action, to better understand and use *Ormocarpum cochinchinense* in medicinal and nutritional settings.

Conclusion

In analyzing the data from the present study, a clear trend emerges regarding the antioxidant and anti-inflammatory activities of the ethanolic extract of *Ormocarpum*

cochinchinense. The antioxidant capacity, assessed using Nitric Oxide and DPPH assays, demonstrated that the OC extract exhibited significant inhibition of oxidative stress compared to the standard ascorbic acid. Specifically, the extract showed increasing antioxidant activity with higher concentrations, achieving up to 79.52% inhibition in the H₂O₂ assay and 85.17% in the DPPH assay at the highest concentration tested. Similarly, the anti-inflammatory effects, measured through protein denaturation and HRBC membrane stabilization assays, revealed that the OC extract provided significant inhibition of protein denaturation and stabilization of the erythrocyte membrane, with up to 66.09% and 79.51% inhibition, respectively. The data

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indicate that the ethanolic extract of OC is effective in reducing oxidative stress and inflammation, with its activities comparable to or exceeding those of the standard drugs used. These results underscore the potential of OC extract as a natural therapeutic agent for managing oxidative stress and inflammatory conditions.

Conflict of Interest

The authors declare no conflicts of interest

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