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To evaluate the Anti-Inflammatory activity of Rhizome extract of Hedychium Coronarium by using Soxhlet Apparatus.

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ABSTRACT

Hedychium coronarium is used in traditional medicine to treat skin diseases, fever, pain and inflammation. This plant has bioactive compounds such as flavonoids, terpenoids, alkaloids, essential oils which are found in its roots and rhizomes which give it medicinal properties. The present study is aimed to evaluate experimentally the anti-inflammatory activity of Hedychium coronarium roots by phytochemical analysis and comparison with conventional anti-inflammatory drugs. Chronic inflammation is often blamed for arthritis, cardiovascular disease and many other conditions, creating a desire for safer plant-based alternatives. This research is expected to reveal significant anti-inflammatory properties in support of the traditional use of the plant and show potential of the plant as a base for herbal medicinal formulation. The study highlights the need for further studies on the active components of medicinal plants and their importance for pharmacological research. Hedychium coronarium, a medicinal plant, contains labdane diterpenes including coronarin D, which are known to have significant anti-inflammatory properties. These chemicals have been shown to inhibit inflammatory mediators such as elastase release, superoxide anion generation and COX enzymes. Coronarin D and related diterpenes are highly potent in inhibiting neutrophil mediated inflammation and oxidative stress, with a high inhibition of inflammatory markers. Their mechanisms include inhibition of reactive oxygen species, suppression of pro-inflammatory cytokines and modulation of signalling pathways such as MAPK, NF-κB.

Keywords: Inflammation, Rhizomes, Labdane Diterpenes, Coronarin D, Inflammatory Mediators, Neutrophil-Induced Inflammation, Anti-Inflammatory Mechanisms, Pharmacology.

1. Introduction -

Hedychium coronarium (white ginger lily), a member of the Zingiberaceae family, is a medicinal plant commonly grown in tropical and subtropical areas. It has been used for a long time in traditional healing systems to treat conditions such as fever, pain, skin ailments and inflammation. Rhizome is considered as the most pharmacologically active part of the plant due to its rich composition of secondary metabolites such as flavonoids, terpenoids, alkaloids, essential oils and various diterpenoid compounds which contribute to its therapeutic potential. The rhizomes contain coronarin D, a major bioactive labdane-type diterpene compound that has been shown to have significant anti-inflammatory effects in experimental studies. The compound has been reported to interfere with several inflammatory pathways by suppression of mediators such as superoxide radicals, elastase activity and cyclooxygenase (COX) enzymes. Furthermore, it has been reported that coronarin D is able

to reduce oxidative stress and downregulate the production of pro-inflammatory cytokines through modulation of signalling cascades in particular MAPK and NF-κB pathways. Inflammation is an important biological response to injury and infection, but when it persists over time, it can result in the development of chronic diseases such as arthritis, cardiovascular diseases and autoimmune diseases. The most prescribed medications are standard anti-inflammatory drugs, although side effects often limit their prolonged use.
Therefore, Hedychium coronarium is gaining attention as a potential source of natural anti-inflammatory agents. This study is designed to investigate the anti-inflammatory activity of its rhizomes, with particular focus on labdane diterpenes like coronarin D, to provide scientific support for its traditional medicinal use and explore its potential for future therapeutic development.

1. Benefits of Hedychium Coronarium Rhizome Extract: -

- Help reduce swelling and inflammation
- Fight certain bacteria and fungi
- Support skin recovery from minor wounds and irritations
- Provide mild pain-relieving effects
- Act as an antioxidant, helping protect cells from damage
- Show possible benefits for blood sugar regulation (based on early studies)
- Be explored in research for anticancer activity, evidence is limited to lab work.

2. Limitations of Hedychium Coronarium Rhizome Extract: -

- Very few studies have tested it in humans
- Safe and effective dosage has not been clearly defined
- Its chemical makeup can vary based on environment and extraction process
- Some active compounds may have poor absorption in the body
- Long-term safety information is lacking
- High concentrations may lead to unwanted effects
- Results differ depending on how the extract is prepared
- Interactions with other drugs are not well established.

2. Botanical Name Of Sontakka: - Hedychium Coronarium

GEOGRAPHICAL SOURCE: - Hedychium coronarium originates from South and Southeast Asia and now grows widely in tropical, warm, and humid wetland regions worldwide.

- **Morphology: -**

The rhizome of Hedychium coronarium is a thick, soft underground stem with a pleasant fragrance. It grows sideways beneath the soil and slowly spreads, producing new shoots and roots that help the plant grow and multiply.

- **Shape and Size: -**

The rhizome is thick and cylindrical, with a branched structure that spreads sideways under the soil.

- **Colour And Surface Texture: -**

The rhizome appears light to yellowish-brown in colour. It has a rough exterior with noticeable nodes and marks left by previous growth, and it feels firm with a slightly fibrous texture.

- **Structure And Hardness:**

The rhizome is solid and fleshy inside with a fibrous feel. It is firm but not very hard, so it can be cut easily when fresh.

3. Pharmacological Activity:

- **Anti-Imflammatory Activity:**

Inflammation is involved in many diseases including cancer, autoimmune diseases, infections and Alzheimer's. Two types of medications that help slow the progression of inflammation are corticosteroids and NSAIDs. Several essential oils, including those from Origanum, Citrus, and Pimpinella, were found to have anti-inflammatory properties in a review of forty-three plants. The major constituents, such as isoeugenol and carvacrol, showed strong anti-inflammatory activity, suggesting that these oils could be important for the treatment of inflammatory diseases.

- **Anti Aging Activity:**

Because of its potent antioxidant and enzyme-inhibiting properties, Hedychium coronarium, and especially its rhizome extracts, have demonstrated encouraging anti-aging and skin health benefits. The collagenase, elastase, and hyaluronidase enzymes that cause wrinkles and a decrease in skin elasticity are inhibited by these extracts. β-caryophyllene and eucalyptol are the primary bioactive substances.

- **Antidiabetic Activity:**

The Hedychium coronarium, which is also called the ginger lily has some good effects on diabetes. The special things in the ginger lily like the ethyl acetate extract can help with diabetes by stopping the enzymes that break down the carbohydrates. This is because the extract is very good at stopping these enzymes. The ginger lily has things like terpenes and fatty acids that're very important for its effects. The ginger lily does not just help raise the insulin levels in our body it also helps keep the pancreas safe.

- **Anti Microbial Activity:**

The essential oil of Hedychium coronarium is good for us. This oil comes from the rhizomes of Hedychium coronarium. The essential oil of Hedychium coronarium has some things in it that help fight off bad microbes. These things are called 1,8-cineole, β-pinene and α-terpineol. The essential oil of Hedychium coronarium is very good at fighting off fungus and bad bacteria. It is especially good at fighting off gram- bacteria like Candida albicans and Staphylococcus aureus. The essential oil of Hedychium coronarium also has something called coronarin D in it. This coronarin D

helps the essential oil of Hedychium coronarium fight off microbes.

- **Anti Oxidant Activity:**

Because Hedychium coronarium has stuff like flavonoids and phenols it is a strong antioxidant. The leaves of this plant usually have more of this stuff and are better at fighting antioxidants. How well the plant fights antioxidants can depend on what part of the plant is used and what liquid is used to extract it. The roots of Hedychium coronarium when using a methanol liquid are good at fighting certain types of bad molecules like DPPH, ABTS and others. The roots have a lot of stuff that helps fight these bad molecules. The leaves on the hand have better stuff and are better at fighting antioxidants especially when using an ethanol liquid.

- **Hepataprotective Activity:**

It has been found out that the plant Hedychium coronarium, which is famous for its liver-protecting qualities greatly affects liver cells. In mouse liver cells a special extract from its flowers showed protection against damage caused by a chemical. This extract, which is 80% acetone was compared to a medicine called silybin. The plants new special compounds, coronadiene and key ingredients, like coronarin C also showed liver-protecting effects. These compounds are a type of glycosides. The rhizome extracts of Hedychium species also demonstrated liver-protecting effects. The flowers and rhizome of Hedychium coronarium seem to have liver-protecting qualities.

- **Anticancer Activity:**

The phytochemical components of Hedychium coronarium, especially coronarin D, are largely responsible for the plant's anticancer effects. In cancer cells such as HeLa cervical cancer and MCF-7 breast cancer, these substances cause apoptosis, suppress cell division, and halt the cell cycle. By downregulating cyclins and CDKs that encourage cell cycle progression and upregulating tumour suppressor proteins p53 and p21, they induce G1 phase cell cycle arrest. By suppressing matrix metalloproteinases, the plant extract also prevents the migration of cancer cells.

4. Literature Review: -

Behera, Sashikanta (2023)

Hedychium coronarium is a beautiful, fragrant plant that's valued in more ways than one. People often grow it in gardens because of its striking white, butterfly-like flowers and its sweet, pleasant scent that's even used in perfumes. Beyond its looks and fragrance, it also has important medicinal value. Researchers have found that it shows antimicrobial, anti-inflammatory, antioxidant, pain-relieving, and even anticancer-related activities. These effects are mainly linked to natural compounds in the plant, especially substances like labdane diterpenes and sesquiterpenes, including coronarin D.

Kamath, Vaishali Rajesh (2024) – The study aims at large scale and sustainable propagation of healthy and disease-free plantlets of H. coronarium by developing micropropagation techniques under controlled laboratory conditions. It also discusses the pharmacological potential of the plant by identifying its bioactive compounds and supporting its traditional medicinal uses such as antimicrobial and antioxidant activities.

Bedekar, Atul N (2023) -It emphasizes the importance of linking phytochemical screening with biological activity to identify potential drug candidates. This literature shows that medicinal plant research plays an important role in drug discovery and validation of traditional medicine, although many plants still need further scientific investigation.

Prabhakar Reddy (2013) –It shows that Hedychium spicatum rhizomes are a source of new bioactive labdane diterpenes with high pharmacological potential, especially in anticancer and antidiabetic research. The work revealed that rhizomes of Hedychium spicatum are a source of new bioactive labdane diterpenes with high pharmacological potential, particularly in anticancer and antidiabetic research, making it an important plant for natural product drug discovery.

Mishra, Tripti (2018) - It emphasizes medicinal plants are rich in bioactive compounds such as alkaloids, flavonoids, and terpenoids. Literature shows their extraction, isolation, and identification using chromatographic and spectroscopic methods. These compounds exhibit antibacterial, antioxidant, anti-inflammatory, and anticancer activities. Studies highlight their pharmaceutical potential, though further research is needed for clinical validation.

Dash, Swagatika (2020)- Studies on essential oils focus on phytochemical screening to identify bioactive compounds with immunomodulatory and anti-inflammatory properties. Research shows these oils contain terpenes and phenolics that influence immune response and reduce inflammation. It has strong therapeutic potential, but further studies are needed to confirm safety, efficacy, and clinical applications.

Devi, Naorem Manglembi (2025) – The study evaluates bioactive potential of 22 Zingiberaceae rhizomes for antioxidant, anti-inflammatory, and antidiabetic activities using DPPH, ABTS, and reducing power assays. Alpinia calcarata showed highest activity. GC-MS and phytochemical analysis revealed rich phenolics and flavonoids, especially in butanol fractions. It is a promising source for diabetes treatment, with added silver nanoparticle synthesis from its extracts.

Jasmine Mary, S. (2014) This study investigates flavonoid glycosides isolated from indigenous plants for their therapeutic potential. The compounds were evaluated for anti-diabetic, anti-inflammatory, anti-ulcer, and anti-cancer activities. Results suggest that these naturally derived flavonoids exhibit strong biological effects by modulating oxidative stress and inflammatory pathways. The findings highlight indigenous plants as valuable sources of drug candidates, supporting traditional medicine while encouraging further research for safe and effective clinical applications in modern healthcare.

Desu Brahma Srinivasa Rao (2021)- This study looks at two traditional medicinal plants, Strobilanthes kunthianus and Strobilanthes cuspidatus, to understand their potential in treating inflammation and joint-related problems. Based on traditional knowledge, their ethanolic leaf extracts were tested in animal models and laboratory assays to evaluate pain relief, anti-inflammatory, and anti-osteoarthritic effects. The results showed strong biological activity, supporting their traditional use.

Manem Satya Prasad (2023)- This study evaluates the anti-inflammatory and anti-arthritic potential of Polyalthia korinti, a medicinal plant used in traditional systems of medicine. The research involved pharmacological testing of plant extracts to assess their ability to reduce inflammation and arthritis symptoms in experimental models.

Jasmine Mary, S. (2018)-This study explores flavonoid glycosides isolated from indigenous plants for their therapeutic potential. The compounds were tested for anti-diabetic, anti-inflammatory, anti-ulcer, and anti-cancer activities using experimental models. Results showed strong biological effects, suggesting these natural compounds help regulate oxidative stress and inflammation, which are linked to many diseases.

5. Material and Methods: -

- **Material** –

Collect the Rhizomes and check the activity of Hedychium Coronarium

- **Powder Form –**
  - BIOLOGICAL NAME – Hedychium coronarium
  - COMMAN NAME – Butterfly Ginger, Sontakka
  - FAMILY – Zingiberaceae

- **Chemical Constituents –**

Diterpenes: Coronarin: series (e.g., coronarin A, D, E, F, G, H, I), Hedyforrestin C, Labdane-type diterpenes,

Sterols: daucosterol, stigmasterol, and β-sitosterol.

Flavonoids :5-hydroxy-3,7,4'-trimethoxyflavone and Chrysin.

Monoterpenes: 1,8-cineole, linalool, α-pinene, β-pinene, α-terpineol, erpinene-4-ol, eucalyptol, p-cymene

Sesquiterpenes: Z-caryophyllene, caryophyllene oxide.

6. USES-

1. Traditionally used to ease respiratory problems such as cough, asthma, bronchitis, and tonsillitis.
2. Helps in reducing pain and inflammation, including headaches and joint stiffness.
3. Used for digestive issues, especially indigestion and stomach discomfort.
4. Applied in treating fevers, infections, bleeding piles, and even snakebites.
5. Shows strong anti-inflammatory and pain-relieving effects in studies.
6. Demonstrates antimicrobial and antifungal activity against harmful microbes.
7. Contains antioxidant properties that help protect cells from damage.
8. Shows promising anti-diabetic potential for blood sugar control.

7. Method –

- **Extraction:**

1. Collect Hedychium coronarium plant parts and wash them clean.
2. Dry them completely in shade until they become brittle.
3. Grind the dried material into a fine powder.
4. Put the powder into a Soxhlet thimble and close it with cotton.
5. Set up the Soxhlet apparatus with a round-bottom flask, extractor, and condenser.
6. Add a suitable solvent (like hexane) into the flask.
7. Heat the setup so the solvent keeps boiling and cycling through the plant material.
8. Let it run for several hours until the plant is fully extracted.
9. Stop heating and allow it to cool.
10. Collect the liquid extract.
11. Remove the solvent using evaporation to get the crude extract containing coronarin D.

Fig.Extract

- **Activity Checked on Hedychium Coronarium:**
- **Activity Checked:** In Vitro Anti-Inflammatory Activity by Protein Denaturation Method
- **Objective:**

To evaluate the anti-inflammatory potential of a test sample by assessing its ability to inhibit protein denaturation, an in vitro indicator of anti-inflammatory activity.

- **Principle:**

Inflammation is associated with the denaturation of proteins. Certain anti-inflammatory agents prevent protein denaturation. This method measures the ability of a test sample to inhibit the denaturation of proteins (commonly bovine serum albumin (BSA) or egg albumin) induced by heat.

8. Materials Required:

- Test sample (extract, formulation, or compound)
- Standard anti-inflammatory drug (e.g., Diclofenac sodium, Ibuprofen)
- Protein solution: Bovine Serum Albumin (BSA) 1% w/v in phosphate buffer (pH 6.4)
- Phosphate buffer saline (PBS) pH 6.4
- Distilled water
- Test tubes
- Water bath
- Spectrophotometer (UV-Vis)
- Pipettes and micropipettes
- Incubator

- **Preparation Of Solutions:**

1. BSA Solution (1% w/v) :/ egg albumin

- Dissolve 1 g of BSA in 100 mL phosphate buffer (pH 6.4).

2. Standard Solution:

- Prepare a stock solution of Diclofenac sodium/Ibuprofen (e.g.,250,500,1000 µg/mL) in distilled water or PBS.

3. Test Sample:

- Prepare various concentrations (e.g.,250,500,1000 µg/mL) in suitable solvent.

- **Procedure**:

1. Reaction Setup: separate test tubes, mix:

- 0.5 mL of test sample or standard solution
- 0.5 mL of 1% BSA solution
- for control, mix:

0.5 mL phosphate buffer + 0.5 mL BSA solution

2. **Incubation:** Incubate all tubes at 37°C for 15–20 minutes to allow protein-sample interaction.

4. Denaturation:

- Heat the samples at 70°C for 5–10 minutes in a water bath to induce protein denaturation.
- Cool the samples to room temperature.

5. Measurement:

- Measure the absorbance at 660 nm using a UV-Vis spectrophotometer.

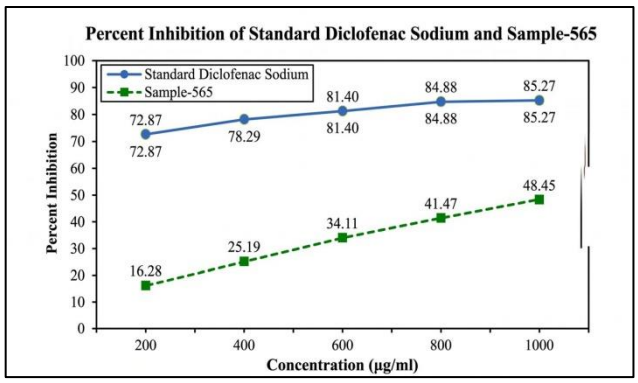
- **Calculations:**

Sr. no.	Sample	Conc. µg/ml	O. D.			Mean	Percent inhibition
1	Control		0.82	0.89	0.87	0.86	
2	Standard Diclofenac sodium	200	0.25	0.24	0.21	0.23	72.87
		400	0.19	0.18	0.19	0.19	78.29
		600	0.15	0.17	0.16	0.16	81.40
		800	0.13	0.14	0.12	0.13	84.88
		1000	0.11	0.18	0.09	0.13	**85.27**
3	Sample – 565	200	0.73	0.72	0.71	0.72	16.28
		400	0.65	0.66	0.62	0.64	25.19
		600	0.58	0.55	0.57	0.57	34.11
		800	0.52	0.51	0.48	0.50	41.47
		1000	0.47	0.45	0.41	0.44	**48.45**

% inhibition = absorbance of control - absorbance of test / absorbance of control x 100

In vitro anti-inflammatory activity by Protein denaturation method

- **Evaluation Parameters: -**



1. Liebermann–Burchard Test

- **Reagent:** Acetic anhydride + concentrated H_2SO_4
- **Procedure:** The sample dissolved in chloroform is treated with acetic anhydride followed by addition of concentrated H_2SO_4.
- **Result:** Development of blue-green or yellow coloration indicates the presence of terpenoid skeleton (supporting Coronarin D).

2. Salkowski Test:

- Reagent: Chloroform + concentrated H_2SO_4
- Procedure: The sample dissolved in chloroform is carefully layered with concentrated H_2SO_4 along the side of the test tube.

- Result: Reddish-brown coloration at the interface confirms the presence of terpenoid compounds

3. Copper Acetate Test

- Reagent: Copper acetate solution
- Procedure: The sample extract is mixed with copper acetate solution and observed for colour change.
- Result: Emerald, green coloration suggests the presence of diterpenes.

4. Vanillin–Sulfuric Acid Test

- Reagent: Vanillin–sulfuric acid reagent
- Procedure: The TLC plate is sprayed with vanillin–sulfuric acid reagent and gently heated.
- Result: Coloured spots indicate the presence of diterpenes

5. Anisaldehyde–Sulfuric Acid Test

- Reagent: Anisaldehyde–sulfuric acid reagent
- Procedure: The developed TLC plate is sprayed with anisaldehyde–sulfuric acid reagent and heated at 100–110°C.
- Result: Appearance of purple, blue, or green coloured spots confirms diterpenoid compounds.

6. Evaluation of Test:

1. Liebermann–Burchard Test

Evaluation

This test helps find terpenoid compounds. It works like this: A sample is mixed with anhydride and strong sulfuric acid. If there's a terpenoid nucleus the mixture turns blue-green or yellow. This colour change is a sign. It means diterpenoids like Coronarin D are probably present. The test is useful, for identifying these types of compounds. Coronarin D is one example that can be detected with this test.

2. Salkowski Test

Evaluation:

The Salkowski test is a simple preliminary test for terpenoids. The formation of a reddish-brown colour at the junction of chloroform and sulfuric acid layers indicates the presence of terpenoid compounds in the sample.

3. Copper Acetate Test

Evaluation:

In this test, diterpenes react with copper acetate solution to produce an emerald, green colour. This colour change provides evidence for the presence of diterpene compounds in the extract.

4. Vanillin–Sulfuric Acid Test

Evaluation:

This test is mainly used during TLC analysis to visualize diterpenoid compounds. After spraying and heating, coloured spots appear on the TLC plate, indicating the presence of diterpenes. The colour intensity may also help in comparing different compounds.

5. Anisaldehyde–Sulfuric Acid Test

Evaluation:

Anisaldehyde–sulfuric acid reagent is a sensitive detecting reagent for terpenoids. On heating, diterpenoid compounds produce purple, blue, or green spots on the TLC plate, confirming their presence and helping in compound identification.

9. Authentification Certificate: -

- **Discussion: -**

The current study looked at how Sample–565 works to reduce inflammation in a lab setting. It used a method that checks how well a substance can stop proteins from breaking down when they get too hot which is something that happens when there is inflammation. The standard drug Diclofenac sodium was very good at reducing

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Date: 25/04/2026

Authentication Certificate

This is to certify that, the plant specimen submitted by **Harshada Vikas Patil** of Nootan Pharmacy College, Landagewadi has been authenticated and identified as follows:

Specimen:

- Scientific/ Botanical Name: ***Hedychium coronarium*** J.Koenig
- Family: Zingiberaceae
- Common Name: Sontakka
- Collection Location: Kavathe Mahankal

The authentication has been carried out based on morphological characteristics of specimen and comparison with existing literature and Botanical Flora's.

Authenticated by,
Mr. S. M. Sabale

Acting Principal
Padmabhushan Vasantraodada Patil Mahavidyalaya, Kavathe Mahankal, Dist-Sangli.

inflammation it worked well with 72.87 percent less inflammation at a low dose of 200 µg/ml and 85.27 percent less inflammation at a higher dose of 1000 µg/ml. This shows that the test they used was a one and it helps us compare Sample–565 to something that we know works.

Sample–565 was pretty good at reducing inflammation it just worked a little slower. At a dose of 200 µg/ml it reduced inflammation by 16.28 percent and, at a higher dose of 1000 µg/ml it reduced inflammation by 48.45 percent. This means that Sample–565 has things in it that can help keep proteins from breaking down and reduce inflammation.

1. Result: -

The study was conducted to evaluate the Anti - Inflammatory activity was successful.

10. Conclusion: -

The results of the protein denaturation assay show that Sample-565 has anti-inflammatory activity when tested in a lab. This means it can help reduce inflammation. The activity of Sample-565 depends on the dose used. However, Sample-565 is not as strong as the drug Diclofenac sodium. The fact that Sample-565 has some activity suggests that it contains compounds that're biologically active and may have anti-inflammatory properties. These compounds in Sample-565 could be useful, for reducing inflammation.

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