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AN INVESTIGATION OF THE N-BUTANE EXTRACT OF COCOS NUCIFERA L'S ANALGESIC AND ANTI-INFLAMMATORY EFFECTS IN EXPERIMENTAL ANIMALS

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ABSTRACT: Background and Aim: Coconut sprouts, often referred to as coconut shoots or coconut hearts, have the potential to be analgesic and anti-inflammatory because of their abundance of phytochemicals, antioxidant capabilities, and traditional usage. In northeastern Brazil, herbal remedies made from coconut husk fiber are used to treat inflammatory diseases like arthritis. This study aimed to evaluate the analgesic and anti-inflammatory properties of coconut extract, *Cocos nucifera* L. **Methods:** Fresh coconut sprouts were collected, dried in the shade, grounded, and extracted using n-butanol. Phytochemical screening was carried out. The acute oral toxicity test was conducted by OECD Guidelines 423. Mice were chosen for analgesics study using Eddy's hot plate method, whereas Swiss Albino rats were used for studies on acute inflammation using the carrageen an-induced rat paw edema model. Multiple comparison tests were run after a one-way ANOVA for statistical analysis. **Results and Conclusion:** The presence of tannins, flavonoids, and alkaloids, among other compounds, which are responsible for the anti-inflammatory and analgesic properties, is revealed by the phytochemical screening. The therapeutic dosage is set between 200mg/kg and 400mg/kg in acute oral toxicity, where the animals do not exhibit toxic effects at 2000mg/kg. When compared to the control group, the test group's 400mg/kg dose significantly inhibited paw edema in acute inflammation ($P< 0.01$). In an analgesic study, the test group's 400mg/kg ($P < 0.01$) dose considerably increased the pain threshold compared to the control.

INTRODUCTION: Traditional medicine has a long history in India. Traditional medicine plays a crucial role in providing medical care. The majority of people in developing nations still rely mostly on indigenous traditional medicines to meet their basic medical needs.

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The Indian company *Materia medica* offers a wealth of knowledge about the customs and mythology around clinically significant natural products [1].

Significance of *Cocos nucifera* L., also known as the coconut palm, has been used for various purposes throughout history due to its anti-inflammatory and analgesic properties. Pain alleviation, inflammation reduction, and a natural and safe alternative to traditional painkillers and anti-inflammatory medicines are among these features. Coconut palm portions have been used in traditional medicine for millennia, and their anti-

inflammatory and analgesic characteristics have potential use in modern medicine. Inflammation is the immune system's response to damaging stimuli like pathogens, damaged cells, poisonous substances, or irradiation [2] and it functions by eradicating harmful stimuli and starting the healing process. Important micro-circulatory mechanisms during the inflammatory process include changes in vascular permeability, leukocyte recruitment and accumulation, and the release of inflammatory mediators. These signals cause leukocyte taxis from the general circulation to damaged sites. Active leukocytes create inflammatory cytokines. Your nervous system may be sending you pain as a warning. It's a bad feeling, like a prick, tingle, sting, burn, or discomfort. According to the International Society for the Study of Pain (IASP), chronic pain is any continuous or recurring pain that lasts for more than three months [3]. A bio-psychological approach should be used to assess, prevent, and manage chronic pain because of the reciprocal interaction of biological and psycho-social factors. Many biological, physical, psychological, and social consequences of chronic pain commonly accompany it, all of which add to the load placed on the patient [4].

The neurotransmitters' stimulation of the nociceptive receptor is what causes people to feel pain. For the sense of pain, three receptors mu, kappa, and delta have been discovered. They start the creation of prostaglandin I, prostaglandin II, or occasionally both. Analgesic drugs either block the COX-II receptor non-selectively or selectively. Opioids reduce pain by raising the spinal cord threshold, allowing a person to tolerate greater levels of discomfort [5].

One way to think of germination is as a form of pre-digestion that helps break down complex molecules into their component pieces. Sprouts provide a wealth of nutrients for our bodies. Several natural products made from various plant sources are eaten straight away. Coconut sprouts derived from *Cocos nucifera L*. are used as a natural food product. They are rich in nutrients. It enhances our health while providing the nutrition we require. The pharmacological effects of *C. nucifera* L. components include Anthelmintic, anti-inflammatory, anti-nociceptive, antioxidant, antifungal, antibacterial, and anticancer properties. Depending on which portions of the plant are researched, each portion of *C. nucifera* includes a variety of phytoconstituents, and the plant's pharmacological and biological effects vary [6, 7]. Our research shows that the *Cocos nucifera* L has analgesic and anti-inflammatory properties. The Coconut sprout's analgesic and anti-inflammatory properties could be explained by its capacity to squelch free radicals, which are the main causes of inflammation. The results of the phytochemical research utilizing coconut sprouts indicated that they have analgesic and anti-inflammatory properties.

MATERIALS AND METHODS:

Plant Material: Fresh *Cocos nucifera* sprouts that I bought in Chennai, Tamil Nadu, from neighborhood markets. Authenticated by Prof. P Jayaraman, Ph. D., Retd., Professor, Presidency College, Chennai-5 from the Plant Anatomy Research Centre, and the Registration number of the certificate is PARC/2022/4686. Further research was conducted using these live sprouts. With 100 gm of coconut sprouts and the dry shade method, dried samples were analyzed for three weeks. They were then blended, kept in airtight containers for later examination, and put through a Soxhlet extraction process using n-butanol.

Preparation of the Extract: The crude extract preparation was carried out using 30gms of the fresh coconut sprouts ground with 300 ml of butanol. Then it was filtered using Whatman No.1 filter paper and was centrifuged at 5000rpm for 15 min. The supernatant was used for further phytochemical analysis. The powder (dried) was dissolved in organic solvent butanol was filtered after 48 h for the dried sprouts analysis using a hot plate. The paste that was produced was freeze-dried and stored in a refrigerator in air-tight containers for further analysis [8].

Phytochemical Screening: The procedure outlined by Lima EB et al., Chime SA *et al.*, was followed to identify glycosides, alkaloids, coumarin, terpenoids, steroids, and triterpenoids, resin, quinone, saponins, gum and mucilage, anthraquinone, phytosterol, proteins and amino acids, polyphenols, anthocyanins, and betacyanin, carbohydrates, flavonoids, cardiac glycosides, phlobatannin, phenols, and tannins [9].

Animal Requirement: The study was conducted after getting approval from the Institution Animal Ethical Committee(IAEC). CCSEA approval number from IAEC of III/ IAEC/ DrMGR /2053 /PO /ReBi /S/19/CPCSEA/07.11.2020/04. Wistar Albino rats of either sex with an average weight of 150g were used in anti-inflammatory and analgesic activity. The study was done in the Department of Pharmacology in April 2022. Animals were acclimatized to the laboratory conditions for at least 1 h before testing and were used during experiments.

Acute Oral Toxicity: Acute oral toxicity was carried out by the OECD no 423. Acute oral toxicity studies were performed on healthy adult albino rats. Animals were maintained on a 24-hour fast with unlimited access to water. The coconut sprout extract of dose 2,000 mg/kg body weight, a single dose given orally and was kept under close observation for 24hours andthe animals were monitored for any signs of mortality and signs of toxicity [10].

Anti-Inflammatory Activity:

Carrageenan-Induced Paw Edema: The most commonly used animal model for assessing the anti-inflammatory properties of drugs is Carrageenan-induced hind paw edema. The rats were divided into six groups, each with six animals. The control group animal received distilled water. The standard group animal received Indomethacin 10 mg/kg in the intra-peritoneal route. The group 3 and 4 received coconut sprout extract of 200 and 400 mg/kg. All the animals were pre-treated 30 minutes before administration of a sub-plantar injection of 100 µl of a 1% (w/v) suspension of carrageenan in the right hind paw. The paw volume was measured by a Plethysmometer at 1, 2, 3 and 4 h [11].

Analgesic Activity:

Eddy's Hot Plate Model and Procedure: This procedure described by Mohamad Ali Hijazi *et al,* was followed. Rats were divided into four groups and each group carried six animals. Group I served as control and received normal saline, whereas Group II served as standard and received the Pentazocine 10 mg/kg, and Group III and IV received coconut sprout extract 200 and 400 mg/kg, respectively.

All animals received treatment by intra-peritoneal injection 30 min before the experiment. The pain was induced by placing it on the hot plate. The latency time for responses was measured at different time intervals as 0, 30, 60, 90, and 120 min. To conduct our experiment, the hot plate's temperature was between 50°C and 55°C. To prevent any thermal harm to the paws, a 15-second cut-off duration was used.

The duration of the animal's reflexive pain behavior was used to determine the reaction time. At 0, 30, 60, and 90 minutes following the corresponding treatment, the reaction time in seconds was recorded [13].

Statistical Analysis: Statistical analyses were performed using one-way ANOVA and data were presented as mean ± standard deviation by using Graph Pad Prism Software.

RESULTS:

Phytochemical Screening: This study revealed that fresh and dried *Cocos nucifera* L extracts contained alkaloids, saponins, Glycosides, Terpenoids, steroids, resins, Quinone, Gum and Mucilage, Coumarin, Anthraquinone, Protein and Amino acids, Anthocyanin and Betacyanin, Carbohydrates, Phlobatannin, Flavonoids, Cardiac glycosides, Phenols, Tannins, Phytosterols, Polyphenols, Fixed oils and fats, Fatty acids. However, the cardiac glycosides and Anthocyanin and Betacyanin were found only in fresh extract. The phytoconstituents present (+) and absence (-) were presented in the **Table 1.**

TABLE 1: PHYTOCHEMICAL EVALUATION OF BUTANOL SOLVENT EXTRACTS OF FRESH AND DRIED COCONUT SPROUT

S. no.	Phytochemical Constituents	Butanol	
		(F)	(D)
1.	Alkaloids	-	-
2.	Saponins	+	+
3.	Terpenoids	+	+
4.	Glycosides	-	-

5.	Steroids and Triterpenoids	+	+
6.	Resin	+	+
7.	Quinone	+	+
8.	Gum and Mucilage	-	-
9.	Coumarin	-	-
10.	Anthraquinone	-	-
11.	Protein and Amino acids	+	+
12.	Anthocyanin And Betacyanin	+	-
13.	Carbohydrates	+	+
14.	Phlobatannin	-	-
15.	Flavonoids	-	-
16.	Cardiac glycosides	+	-
17.	Phenols	-	-
18.	Tannins	-	-
19.	Phytosterols	-	-
20.	Polyphenols	-	-
21.	Fixed oils and fats	-	-
22.	Fatty acids	-	-

(F)-Fresh; (D)-dried; (+)-Presence; (-)-absence

Analgesic Activity: Analgesic activity of coconut sprouts in thermal pain method-Eddy's hot plate, the effects of butanol extract of coconut sprout at doses of 200 mg/kg (low) and 400 mg/kg (high) and Pentazocine 10mg/kg were compared to the control group at various hours. In comparison to the control, the test of 400mg/kg significantly increased the Pain threshold. The data is expressed as the mean standard deviation of n=6 **P<0.05 Compared with the control

TABLE 2: ANALGESIC ACTIVITY OF COCONUT SPROUT EXTRACT

Groups	0 min	30 min	60 min	90 min	120 min
Control	1.83±0.75	1.8±0.75	1.66±0.51	1.5±0.54	1.6±0.54
Standard (Pentazocine 10mg/kg)	1.5±0.54****	2.6±0.81****	4.2±0.82****	6.8±1.03****	8.7±0.83****
Coconut sprout extract 200mg/kg	1.2±0.7*	1.5±0.5*	1.8±0.5*	2.7 ±1.1*	3.2±0.75**
Coconut sprout extract 400mg/kg	1.4 ± 0.12****	2.4±0.2****	3.8±0.21****	4.5±0.1****	6.8±1.23****

Value expressed as mean ±SD symbols represents statistical significance ***P<0.001, ** P <0.01,*P<0.05.

Anti-Inflammatory Activity: In a Carrageenan-induced paw edema model, the effects of butanol extract of coconut sprout at doses of 200 mg/kg (low) and 400 mg/kg (high) and Indomethacin 10 mg/kgwere compared to the control group at various hours. In comparison to the control, the test of 400mg/kg significantly decreased the edema in the Carrageenan-induced paw. Paw edema considerably (P < 0.001) decreased during the second, third, and fourth hours with the test 400mg/kg compared to the control.

TABLE 3: ANTI-INFLAMMATORY ACTIVITY USING COCONUT SPROUT

Group	Mercury displacement volume				
	0hr	1hr	2hr	3hr	4hr
Control	3.65±0.18	4.73±0.19	4.78±0.18	4.75±0.37	4.83±0.08
Standard	3.61±0.18***	3.9±0.12***	3.71±0.04***	3.61±0.13***	3.45±0.05***
Test 200mg/kg	3.65±0.16**	4.33±0.10**	3.98±0.11**	3.75±0.05**	3.7±0.10**
Test 400mg/kg	3.61±0.13***	4.16±0.08***	3.83±0.15***	3.66±0.08***	3.56±0.12***

The value expressed as mean ±SD symbols represents statistical significance ***P<0.001, ** P <0.01,*P<0.05.

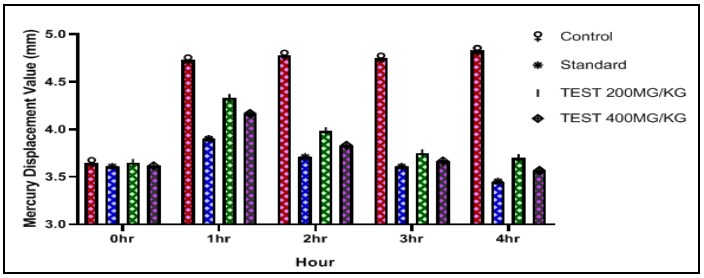

FIG. 1: ANTI-INFLAMMATORY ACTIVITY OF MERCURY DISPLACEMENT VOLUME USING COCONUT SPROUT

DISCUSSION: Throughout the ages, a variety of cultures have used medicinal plant extracts. The hot plate test, which is a common method to evaluate analgesics that act on the central and peripheral nerve systems, was used to examine the analgesic activity of coconut sprouts. Reduced discomfort from the hot plate after administration of coconut sprout extract suggests that it has some analgesic impact on both the central and peripheral nervous systems [13].

The efficacy of non steroidal anti-inflammatory drugs, which largely block the cyclooxygenase implicated in prostaglandin generation, has been assessed using the carrageenan-induced rat paw edema model [14]. The phytochemical components of CA flower extracts, which include phenolic terpenoids, tannins, and flavonoids, are thought to be responsible for their anti-inflammatory properties. These findings support earlier research and are in line with numerous studies in the literature that claim that many plants containing these chemical classes of compounds have powerful anti-inflammatory properties that act by inhibiting prostacyclin synthesis [15, 16].

The results of the phytochemical research utilizing coconut sprouts indicated that they have analgesic and anti-inflammatory properties. Using the hot plate method and a Plethysmometer, an *in-vivo* study was conducted to assess the anti-inflammatory and analgesic properties of coconut sprouts. Positive findings from the trial in terms of analgesic and anti-inflammatory efficacy were found. In an animal investigation, the extract exhibits substantial results at higher doses as compared to lower doses [17].

CONCLUSION: Our research indicates the analgesic and anti-inflammatory properties of *Cocos nucifera* L. Coconut sprout's analgesic and anti-inflammatory properties could be explained by its capacity to combat free radicals, which are the primary causes of inflammation.

Additionally, it might have prevented the induction of pro inflammatory cytokines like NF-dB, TNF, IL-1, and IFN as well as the activity of the inflammatory cyclooxygenase enzymes. Coconut sprouts may include flavonoids and other polyphenols, which have anti-inflammatory and analgesic properties.

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CONFLICTS OF INTEREST: Nil

REFERENCES:

1. Rathod R and Chandak RR: Review on “standardization an imp tool for herbal drug development”. Journal of Drug Delivery and Therapeutics 2019; 9(6): 253-7.
2. Fernandes SM, Watanabe M and Vattimo MD: Inflammation: improving understanding to prevent or ameliorate kidney diseases. Journal of Venomous Animals and Toxins including Tropical Diseases 2021; 27.
3. Treede RD, Rief W, Barke A, Aziz Q, Bennett MI and Benoliel R: A classification of chronic pain for ICD-11. Pain [Internet]. 2015 [cited on 05 Feb 2020]; 156 (6): 1003–7.
4. Kaundal SP, Sharma A, Kumar R, Kumar V and Kumar R: Exploration of medicinal importance of an underutilized legume crop, *Macrotyloma uniflorum* (Lam.) Verdc.(Horse gram): a review. Int J Pharm Sci Res 2019; 10(7): 3178-86.
5. Ally A, Powell I, Ally MM, Chaitoff K and Nauli SM: Role of neuronal nitric oxide synthase on cardiovascular functions in physiological and pathophysiological states. Nitric Oxide 2020; 102: 52-73.
6. Alam MM, Emon NU, Alam S, Rudra S, Akhter N, Mamun MM and Ganguly A: Assessment of pharmacological activities of *Lygodium microphyllum* Cav. leaves in the management of pain, inflammation, pyrexia, diarrhea, and helminths: *In-vivo*, *in-vitro* and *in-silico* approaches. Biomedicine & Pharmacotherapy 2021; 139: 111644.
7. Venkataramaiah C: Chemical constituents of plants as promising drug candidates: nature’s (god) benefaction to mankind. Chief Editor 2020; 53.
8. Belem-Kabr&e WL, Kabor&e BE, Compaor&e-Coulibaly A, Traor&e TK, Thiombiano EA, Nebi&e-Traor&e M, Compaor&e M, Kini FE, Ou&edraogo S, Kiendrebeogo M and Ou&edraogo N: Phytochemical and biological investigations of extracts from the roots of *Cocos nucifera* L.(Arecaceae) and *Carica papaya* L.(Caricaceae), two plants used in traditional medicine. African Journal of Biochemistry Research 2021; 15(2): 28-35.
9. Omitola OJ: Molecular and Biological Standardisation of Alstonia Boonei De Wild. And *Alstonia congensis* Engl. Leaves and Stem-Barks (Doctoral dissertation).
10. Ormazabal P, Cifuentes M, Varì R, Scazzocchio B, Masella R, Pacheco I, Vega W, Paredes A and Morales G: Hydroethanolic Extract of *Lampaya medicinalis* Phil. (Verbenaceae) decreases proinflammatory marker expression in palmitic acid-exposed macrophages. Endocrine, Metabolic & Immune Disorders-Drug Targets (Formerly Current Drug Targets-Immune, Endocrine & Metabolic Disorders) 2020; 20(8): 1309-20.
11. Rafiee L, Hajhashemi V and Javanmard SH: Maprotiline inhibits COX2 and iNOS gene expression in lipopolysaccharide-stimulated U937 macrophages and carrageenan-induced paw edema in rats. Central European Journal of Immunology 2019; 44(1): 15-22.
12. Shorinwa OA and Obioha CG: Evaluation of the analgesic activity of ethanol extract of the stem and leaf of *Cissus gracillis* Gull and Perr (Vitaceae).

13. Komakech R, Kim YG, Matsabisa GM and Kang Y: Anti-inflammatory and analgesic potential of *Tamarindus indica* Linn. (Fabaceae): a narrative review. Integrative Medicine Research 2019; 8(3): 181-6.
14. Modi AD, Parekh A and Pancholi YN: Evaluating pain behaviours: widely used mechanical and thermal methods in rodents. Behavioural Brain Research 2023; 114417.
15. Onyango IG, Jauregui GV, Čarná M, Bennett JP and Stokin GB: Neuroinflammation in Alzheimer's disease. Biomedicines 2021; 9(5): 524.
16. Semis HS, Gur C, Ileriturk M, Kaynar O and Kandemir FM: Investigation of the anti-inflammatory effects of caffeic acid phenethyl ester in a model of λ-Carrageenan–induced paw edema in rats. Human & Experimental Toxicology 2021; 40(12_suppl): 721-38.
17. Servi H, Vatansever C, Doğan A and Majeed V: Antibacterial activity and essential oil composition of *Calendula arvensis* L. IJSM 2020; 7(4): 229-36.

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