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FORMULATION AND EVALUATION OF ANTI ARTHRITIC HERBAL CHOCOLATE

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ABSTRACT: Chocolate is the most loving food among the people. It is the easiest form to chew and absorb for every individual. This research deals into the Innovative application of herbal chocolates, a delightful fusion of traditional enjoyment and holistic well-being. The blending of herbal ingredients with the luxurious texture of chocolate has given rise to a new category of confectionery that not only satisfies sweet cravings but also offers potential health advantages. The essential target of this study was to formulate and evaluate nutritious chocolate and nutritional supplement containing anti arthritis property. Chocolate is a range of products derived from cocoa (cocoa) mixed with fat and finely powered coconut sugar to produce a solid confectionery. Sesame is the herbal drug which has several medicinal properties, like anti arthritis property, antioxidant property, from the prevention of rheumatoid arthritis. Sesame was used to improve blood pressure and act as anti aging agent. Thus, we have to formulate the chocolate with aqueous extract of sesame. Chocolate is a sophisticated and infinitely adaptable food that can be mixed and matched to generate a variety of taste and texture sensation. Hence worth, in the present examination, endeavor was to make to get ready chocolate plan of sesame which enhances the patient's compliances and worthiness.

INTRODUCTION: Sesame (*Sesamum indicum* L.) is one of the earliest human production and consumption oil crops in the family of Pedaliaceae¹, rape, soybean, and peanuts, known as China's four major oil crops. First discovered in ancient sites in Pakistan, sesame is a long-established cultivated crop². It is distributed in countries such as India, China, and Malaysia. Chinese people have used sesame seeds for more than 5000 years^{1, 3}. Globally, India, Sudan, Myanmar, China, and Tanzania are the major producers of sesame.

In recent years, the production of sesame seeds in African countries has increased, and Tanzania has replaced India as the leading producer of sesame seeds. According to the Food and Agriculture Organization of the United Nations, the global production of sesame in 2017 was 5.899 million tons, of which 806,000 tons were produced in Tanzania and 733,000 tons in China⁴.

Sesame is widely grown and popular because of its highly aromatic odor and mellow flavor. In people's lives, sesame seeds are often used to make a variety of foods, such as sesame oil, sesame paste, or to decorate other foods. The scientific nutritional value status of sesame seeds was established on a legal level in 2002 when they were included in the list of medicinal and food ingredients published by the former Chinese



Ministry of Health. Among the reviews on sesame, only one detailed review on the phytochemistry and ethno-pharmacology of sesame has been published in recent years ⁵. Other reviews are either about a certain chemical composition in the sesame seeds ⁶, on some of the pharmacological effects of sesame ⁷, or the technical aspects of its production ⁸. This review will focus on not only the phytochemical and pharmacological properties of sesame but also the economic-phytological and nutritional value of sesame.

MATERIAL AND METHODS:

Sesame:

Botanical Name: *Sesamum indium* L

Synonyms: Sesame, benne, beni, beni seed, benne seed, Til

Morphological Characteristics ⁹: Sesame is an erect annual herb that grows 60–150 cm tall. The stem is hollow or has white pith. The sesame leaves are 3–10 cm long, 2.5–4 cm wide, and rectangular or ovate in shape with a slightly hairy surface. They are borne singly or 2–3 together in the leaf axils. The calyx lobes of sesame are 5–8 mm long and 1.6–3.5 mm wide, lanceolate in shape, and have a pilose appearance. The corolla of sesame is 2.5–3 cm long in a tube shape about 1–1.5 cm in diameter. It is white, often with a purplish-red or yellow halo. The four stamens are hidden inside the flower, the ovary is superior, 4-loculed and pilose outside, and flowering occurs in late summer and early autumn. The sesame capsule is rectangular in

shape, 2–3 cm in length, and 6–12 mm in diameter, with longitudinal ribs on the surface and microscopic hairs on the epidermis.

Extraction of Sesame Seed ¹⁰: The essential oils were extracted by hydro-distillation method using Clevenger equipment, by placing a quantity of 30 g of dry sesame powder sample in a 500 ml glass flask, to which a quantity of distilled water (400 ml) was added until the entire sample was immersed.

The extraction process continued for 4-5 hours at 80°C temperature, after which the essential oil was separated from the aqueous extract by a 500 ml separating funnel. A separator funnel is used to separate immiscible liquids.

Chocolate Formulation ¹¹: All of the materials were precisely weighed. Sugar was placed in one of the beakers. Dark chocolate cocoa butter was heated in a separate beaker, then added to a powder combination and thoroughly combined to achieve a fine consistency.

After that, an emulsifier, honey, was added and blended. Finally, the herbal medication extract i.e. crude extract of sesame seed oil was precisely measured and put to the previously made chocolate. Before entering into the moulds, vanilla was added as a flavoring ingredient. The prepared chocolate containing herbal medication extract was then put into moulds and allowed to set overnight in the freezer.

TABLE 1: CHOCOLATE FORMULATION

Sr. no.	Ingredients	Quantity	Role
1.	Sesame oil	5 mg	Anti arthritis, Antioxidant
2.	Cardamom	q. s.	Antioxidant
3.	Honey	q. s.	Emulsifier
4.	Cocoa Butter	3 g	Solidifying agent
5.	Sugar	5g	Sweetening agent
6.	Milk	q. s.	Making consistency
7.	Vanilla	q. s.	Flavoring agent

Evaluation Test:

Chemical Test ^{12, 15}:

Test for Carbohydrate (Fehling's Test): To 1 ml of the solution, equal quantities of Fehling's solution A and B was added and heated.

The formation of brick red precipitate indicates the presence of carbohydrates.

Test for Protein (Biuret Test/General Test):

Take 3ml of chocolate formulation; add 4% NaOH and few drops of 1% copper sulphate solution, violet colour indicate presence of protein.

Test for Amino acids (Ninhydrin Test):

3ml of test solution was heated and 3 drops of 5% Ninhydrin Solution was added in boiling water and

was boiled for 10 min. purple and bluish color indicates presence of amino acid.

Test for Saponins (Foam Formation): Place 2ml of chocolate formulation in water and this was added in test tube, shake well and stable foam is form indicate the presence of saponins.

pH: 2gm of prepared chocolate was dissolved in 100ml of phosphate buffer solution and pH of the resulted solution was studied by digital pH meter with glass electrode.

Blooming Test ¹³:

Fat Bloom: When a thin layer of fat crystals forms on the surface of the chocolate formulation. This will cause the chocolate to lose its gloss and a soft white layer will appear, giving the finished article an unappetizing look.

Fat bloom is caused by the recrystallization of the fats and/or a migration of a filling fat to the chocolate layer. Storage at a constant temperature will delay the appearance of fat bloom.

Sugar Bloom: This is a rough and irregular layer on top of the chocolate formulation. Sugar bloom is caused by condensation (when the chocolate is taken out of the refrigerator). This moisture will dissolve the sugar in the chocolate. When the water evaporates afterwards, the sugar recrystallizes into rough, irregular crystals of the surface.

This gives the chocolate an unpleasant look Each sample was subjected to treatment cycles comprised (1) 30°C for 11 hours, (2) temperature shifting for 1 hour, (3) 18°C for 11 hours, and (4) temperature shifting for I hour. At least chocolate formulation observed, after the step at 18°C for 11 hours, whether or not blooming has taken place.

In-vitro Anti-arthritis Activity ¹⁴:

1. Protein denaturation testing was used to assess this activity. Plant extracts with varying concentrations (100–500 µg /ml) were produced.
2. One ml of the test substance and one ml of solutions containing 1% bovine serum albumin were combined to create reaction mixtures of each concentration.

3. For 15 minutes, these produced solutions were incubated at 27°C.
4. The reaction mixture was then maintained in a water bath at 70°C to cause denaturation. Turbidity was determined spectrophotometrically at 660 nm after cooling these solutions.
5. Diclofenac sodium was utilized as a test extract and employed as a standard medication at concentrations of 50–1000 g/ml.
6. Utilizing a control with no medication administered, the percentage of inhibition of denaturation was assessed.
7. Averaging was done after each experiment was carried out in triplicate.
8. Equation was used to determine the percentage inhibition of protein denaturation.

$$\% \text{ Inhibition of protein denaturation} = 100 \times [A_1 - A_2] / A$$

RESULT:

Phytochemical Test: The extract of *Sesamum indium* L seed oil shows presence of phytochemical constituents like Carbohydrates, Proteins and Amino acids, absence of saponins.

TABLE 2: PHYTOCHEMICAL TEST OF HERBAL CHOCLATE

Name of test	Test group	Control group
Carbohydrate	+	+
Protein	+	+
Amino acid	+	-
Saponin	-	-

In-vitro Anti-arthritis Activity: The anti-arthritis potential of *Sesamum indium* L hydroalcoholic extract utilizing the Inhibition of protein denaturation assay has not previously been investigated.

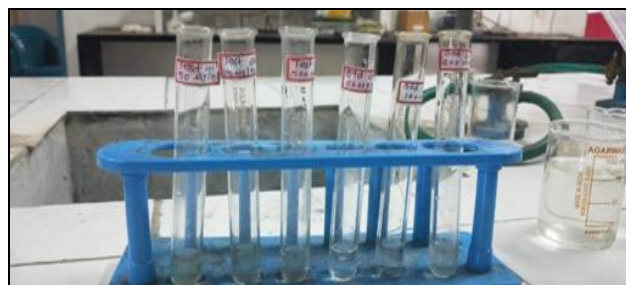
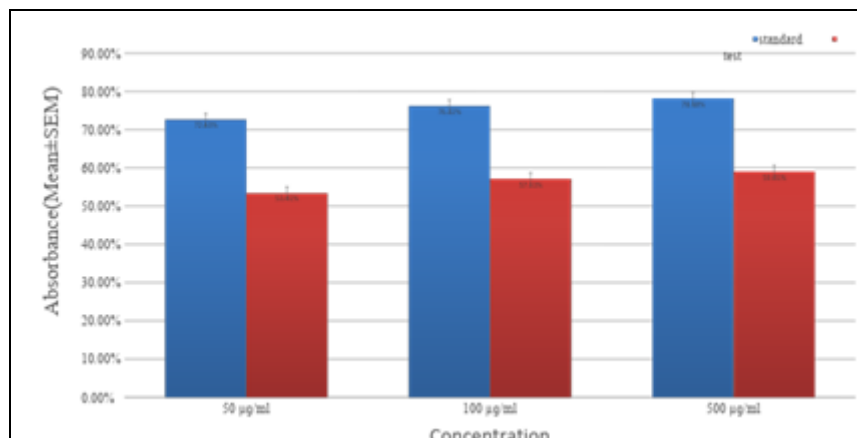


FIG. 1: IN-VITRO ANTI-ARTHRITIC ACTIVITY

TABLE 3: PERCENTAGE INHIBITION FOR OF PROTEIN DENATURATION INHIBITORY ASSAY OF LEAVES EXTRACT OF *SESAMUM INDIUM* L

Sample	Concentration	Absorbance (Mean±SEM)	% Inhibition
Standard	50 µg/ml	0.0384±0.01369***	72.63 %
	100 µg/ml	0.0790±0.030631	76.22 %
	500 µg/ml	0.0584±0.038891***	78.18%
Test	50 µg/ml	0.0681± 0.015782**	53.41 %
	100 µg/ml	0.0382±0.019964	57.11 %
	500 µg/ml	0.2308±0.078234**	59.01 %

**FIG. 2: PERCENTAGE INHIBITION BY PROTEIN DENATURATION INHIBITORY ASSAY OF HYDRO ALCOHOLIC EXTRACT OF *SESAMUM INDIUM* L.**

CONCLUSION: *Sesamum indicum* L. is a significant medicinal plant that grows throughout India. In the present study, development of Herbal Chocolate having anti arthritic activity was carried out using sesame seed oil was prepared and phytochemical analysis was carried out to check the presence of desired compounds that shows acceptable results.

The experiments were also carried out for *in-vitro* anti-arthritis activity on seed oil of *Sesamum indicum* L. The assay uses Diclofinac sodium as a standard drug. The maximal percentage of inhibition was discovered to be 59.01% at 500 µg/ml. Further studies are ongoing to isolate and characterize the compounds responsible for anti-arthritis activity.

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CONFLICT OF INTEREST: The author declare that there are no commercial or financial

relationships that could be construed as a potential conflict of interest in the conduct and publication of this research.

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