Formulation and Evaluation of Anti-Varicose Cream

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Abstract

Varicose veins are a prevalent condition associated with significant variability in prevalence rates, contributing to disability and reduced quality of life. This condition arises from venous hypertension caused by the malfunction of venous valves, leading to blood reflux, elevated pressure, and the onset of symptoms. This study focuses on developing novel anti-varicose formulations utilizing the antioxidant and anti-inflammatory properties of red cabbage and Brahmi. The anthocyanin content in red cabbage is particularly effective in reducing inflammation, making it valuable for managing varicose veins. The newly developed formulations demonstrated high efficacy in combating varicose symptoms. Antioxidant activity assessed via the DPPH assay confirmed the potent antioxidant properties of red cabbage extract. All formulations exhibited good spreadability, with Formulation 2 outperforming Formulation 1 in terms of spreadability.

Keywords: *Anti-varicose cream, Red cabbage, Antioxidant activity, DPPH assay, Anti-inflammatory.*

1. Introduction

1.1 Varicose Disease:

Varicose veins are a common vascular condition that affects millions of individuals worldwide, characterized by abnormally dilated, tortuous veins, typically occurring in the lower extremities. Their distinct twisted, bulging appearance primarily affects the lower extremities but can also manifest in other areas such as the rectum (haemorrhoids) and oesophagus (oesophageal varices).[1] This condition arises due to the weakening or dysfunction of the venous valves, which leads to venous hypertension and blood pooling. Factors such as prolonged standing, aging, obesity, pregnancy, and genetic predisposition significantly contribute to the development of varicose veins. In India, the estimate suggests that between 15% to 20% of the population is affected by vein disease.[2] The condition not only has cosmetic implications but also impacts patients' physical and psychological well-being, often leading to pain, swelling, skin changes, and in severe cases, ulcers. The synthetic anti-varicose formulations currently on the market have a lot of side effects. Based on a review of the literature, it was determined that plants, which have potent antibacterial, antioxidant, and anti-inflammatory qualities, are utilized for a variety of pathological disorders.[3]

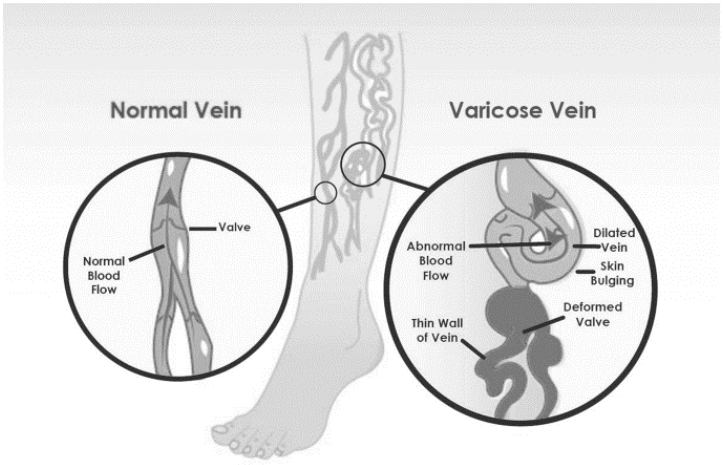


Figure 1. Varicose Veins develop from valvular incompetence, resulting in dilation of the superficial venous system

Current treatment approaches for varicose veins include compression therapy, sclerotherapy, laser treatment, and surgical interventions. While these methods are effective to varying degrees, they may involve high costs, procedural risks, or limited accessibility in resource-constrained settings. Therefore, there is growing interest in alternative and complementary therapies, particularly those utilizing the therapeutic potential of natural compounds with anti-inflammatory and antioxidant properties.

This study explores the role of plant-based bioactive compounds, such as those found in red cabbage and Brahmi, in managing varicose veins[4]. These plants are rich in anthocyanins and other phytochemicals known for their ability to reduce inflammation, neutralize free radicals, and promote vascular health. Developing effective formulations from these natural sources offers a promising, cost-effective approach to mitigating the burden of varicose veins while enhancing patient quality of life.

2. Materials and methods

2.1 Materials:

The plants used in the study were taken from the local market of Nashik, dried in an oven, powdered coarsely & used for extraction. Other excipients used for formulation are Glycerin, Beeswax, Cetyl stearyl alcohol, Propylparaben, Methylparaben, and Eucalyptus oil.

2.2 Preparation of extracts:

2.2.1 Cabbage extract: Take 50 gm of red cabbage from the market, separate the leaves, and cut it into small pieces. These small pieces are placed on a petri plate and kept in a hot air oven for 30 min at 70 ℃. Take out the dried leaves and grind them with 10ml of water in a grinder. Filter the material and concentrate it on the hot plate for 15 minutes at 80°C. [5]

2.2.2 Brahmi extract: Take the 15gm of Brahmi stem powder in a volumetric flask by weighing it accurately and refluxing it in 130 ml of water for 3 hrs. After refluxing, filter the solution and concentrate it on a hot plate by keeping it for 15 minutes at 80°C. [6]

2.3 Preparation of formulation:

Initially, lanolin was used as a thickener in our formulation but the batches formed do not have proper consistency. After that, rose water was used in formulation for its anti-inflammatory properties. However, batches failed because of phase separation as shown in Figure 2.

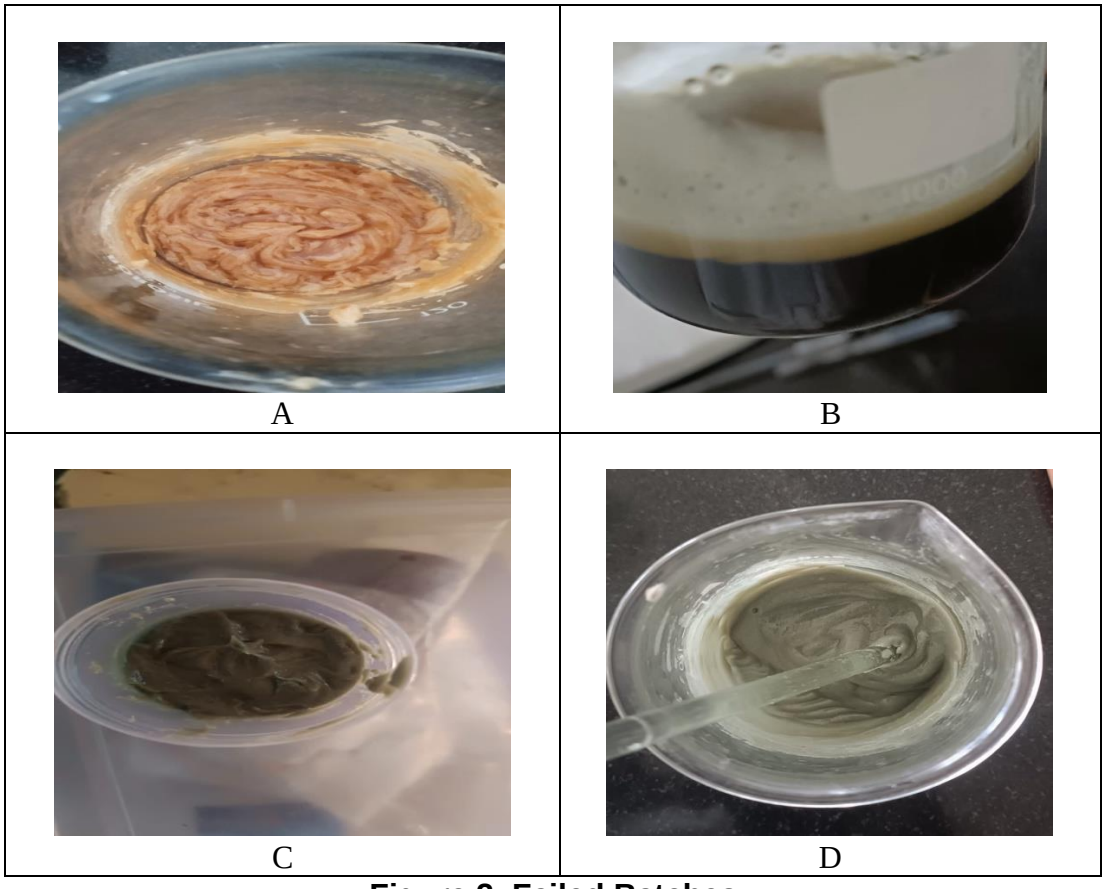


Figure 2. Failed Batches

The ingredients taken for Formulation 1 (denoted by F1) such as beeswax and ceto stearyl alcohol which are oil soluble in a beaker and ingredients for Formulation 2 (denoted by F2) are water soluble ingredients such as Red cabbage extract, Brahmi extract, Red rose oil, glycerin in another beaker. The constituents are shown in Table 1. Both parts are heated separately on a heating mantle at 70°C. After heating, the aqueous soluble components are added slowly in oil soluble components with continuous stirring.[7,8]

Table 1. Formulation Table

Sr.No.	Ingredients	F1	F2	Uses
1	Red cabbage ext.	3.5gm	3.5gm	Anti-inflammatory
2	Brahmi extract	4gm	4gm	Anti-inflammatory
3	Ceto stearyl alcohol	1gm	1.5gm	Thickener, emulsifier
4	Bees wax	5gm	4gm	Thickener
5	Glycerin	6gm	7.5gm	Moisturizer
6	Methyl paraben	0.5gm	0.5gm	Preservative
7	Propyl paraben	0.5gm	0.5gm	Preservative
8	Water	q.s	q. s	Solvent
9	Eucalyptus oil	q.s	q. s	Additives

3. Evaluation:

3.1 Evaluation of extracts:

3.1.1 Physical evaluation: Colour was checked against the white background and odour was identified by mixing the gel in water and taking the smell. The evaluation of the extracts is shown in following Table 2.

Table 2: Evaluation of the extract

Parameters	Observation	
	Red cabbage extract	Brahmi extract
A] Physical Evaluation:		
Color	Dark brown	Dark green
Odour	Aromatic	Aromatic
B] Phytochemical evaluation:		
Carbohydrate	+	+
Protein	+	+
Anthraquinone	-	+
Alkaloids	+	+
Tannins	-	+

3.1.2 Phytochemical evaluation:

a) Carbohydrates (Benedict test -) 5 ml of Benedict's solution and a few drops of diluted HCl were added to ml of the sample aqueous extract, and the mixture was heated for minutes. Precipitation turned the solution red, indicating the presence of glycosides.

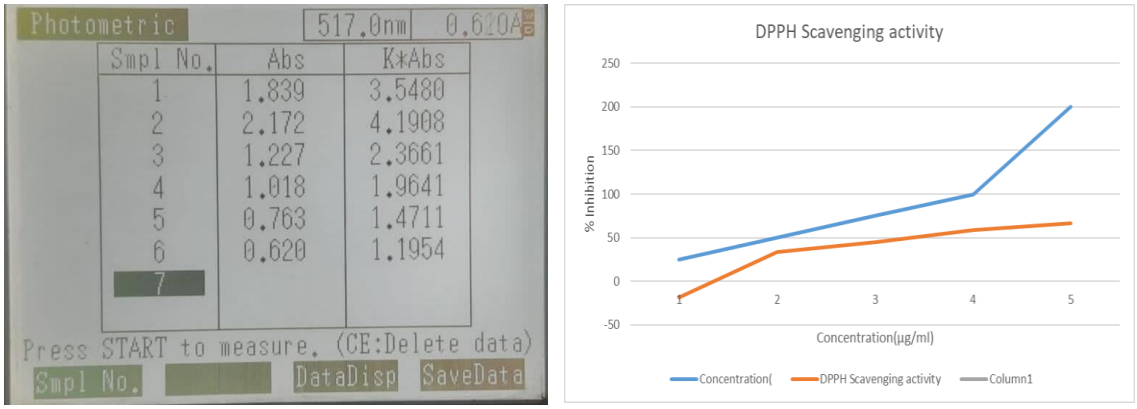
b) Proteins (Millions test) -The test solution is then gradually heated after a few drops of the reagent are added. A precipitate or reddish-brown hue denotes the presence of tyrosine residues, which are found in almost all proteins.

c) Anthraquinone (Brontrager's Test)- After adding 3 ml of chloroform and shaking it, separating the chloroform layer, and adding 10% ammonia to 2 ml of filtered hydrolysate solution was included with it. Glycosides were denoted by the pink hue.

d) Tannins -Add a few drops of 5% $FeCl_3$ solution to 1-2 ml of plant extract. A green tint denoting the existence of Brown-coloured gallotannins was indicative of tannins.

e) Alkaloids (Mayer test) A drop or two of Mayer's reagent were added to a few milliliters of filtrate by the test tube's edge. a creamy or white Precipitate showed that the test was successful. [9,10,11]

3.1.3 DPPH Assay: A stock solution of DPPH reagent was prepared by taking 12mg of 2,2-diphenyl-1-picrylhydrazyl (DPPH) in 50 mL of methyl alcohol and this solution was carefully kept in the freezer. 10 mL of stock solution was taken, diluted with methanol and the absorbance was set in the given range as shown in Figure. 3.



A B

Figure 3. A-DPPH assay observations and B-Graph

The antioxidant activity of cabbage extract was checked with a spectrophotometer by taking a reading of dilution at the wavelength of 517nm. [12] Antioxidant capacity was measured by;

%inhibition = (Absorbance of control) -(absorbance of sample) / (absorbance of control) ×100

The DPPH radical scavenging activity was calculated using the following formula[13]-

Radical Scavenging (%) = Absorbance of Control – Absorbance of Sample / Absorbance of Control × 100

The percentage inhibition is shown in Table 3.

Table 3. % inhibition in DPPH assay

Concentration (µg/mL)	DPPH radical scavenging activity (% inhibition)
25	-18.10
50	33.27
75	44.64
100	58.51
200	66.28

3.2 Evaluation of formulation:

3.2.1 Physical evaluation: Colour was checked against a white background and odour was identified by mixing the gel in water and taking the smell. [14]

a) pH testing- pH paper was used to measure the pH. The color shift indicated the pH when pH paper was dipped immediately into the mixture.
b) Washability- After applying the formulation to the skin, the degree and ease of washing with water were manually assessed. [14]
c) Spreadability- The amount of space that a topical formulation (cream) easily distributes when applied to the skin or the affected area is known as spreadability. Carefully, a 20 g weight was fastened to the upper slide. It was recorded how long it took the higher slide, acting as a force of gravity, to move 6.0 cm and split off from the bottom slide. Three times the experiment was conducted, and the average time required for computation.[8] Spreadability was calculated by using the following formula:

$$S = (M \times L) / T$$

Where, S= Spreadability,
M = Weight in the pan (tied to the upper slide),
L = Length glass. [13]
d) Dye test- Sudan red III, an oil-soluble dye, was employed to conduct this test. A sample of cream is placed on a glass slide, and two to three drops of Sudan red III dye are added. The slide was examined under a microscope.[15] The observations for the evaluation of the final product are shown as follows in Figure 4.

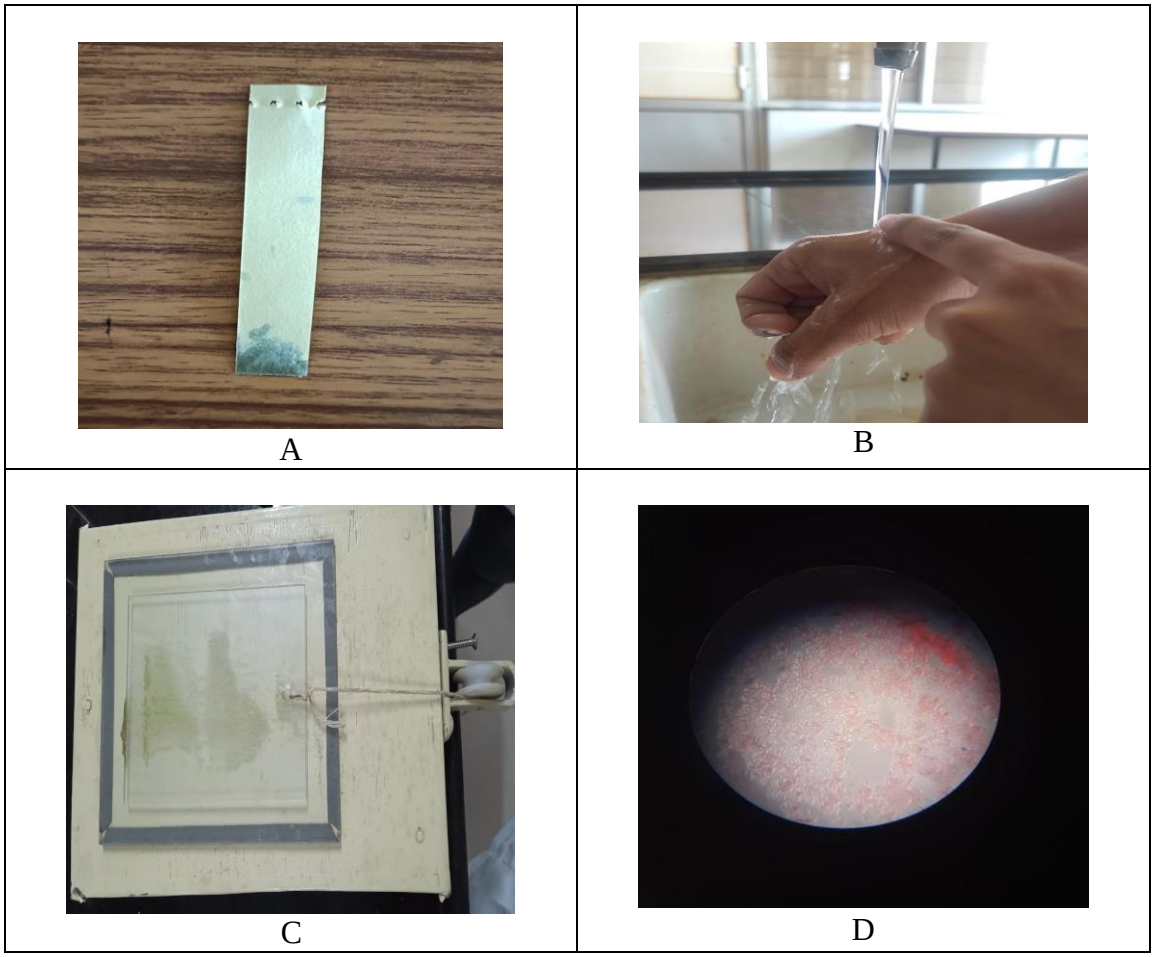


Figure 4. Evaluation results of formulation: A- pH testing, B- Washability test, C- Spreadability test, D- Dye test.

The evaluation of formulations, F1 and F2, provides a comprehensive overview of the key parameters assessed to determine the quality, stability, and effectiveness of a formulated

product. It typically includes metrics such as physical appearance, pH, viscosity, drug content, stability under various conditions, and any observed incompatibilities. These evaluations ensure that the formulation meets the desired specifications and regulatory requirements, offering consistent performance. The data aids in identifying any potential issues and optimizing the formulation process for enhanced reliability and efficacy. The evaluation of formulations is shown in Table 4.

Table 4. Evaluation of formulations

Characteristics	F1	F2
Color	Pale brown	Brown
Odour	Aromatic	Aromatic
Appearance	Smooth	Smooth
Physical state	Semi solid	Semi solid
pH	7-8	8-9
Homogeneity	Homogenous	Homogenous
Washability	Good	Good
Spreadability (g-cm/sec)	20.61	21.20
Dye test	O/W	O/W

The final formulations as shown in Figure 5, represent the optimized and thoroughly evaluated versions of a product, developed to meet desired quality, stability, and efficacy standards. These formulations are typically selected after extensive research, testing, and refinement to ensure compliance with regulatory requirements and the intended purpose of the product. Key factors such as composition, physical properties, and manufacturing feasibility are carefully balanced to achieve consistent performance. Final formulations undergo rigorous quality checks, including stability studies, compatibility assessments, and performance evaluations, to ensure their safety, effectiveness, and market readiness.

A B

Figure 5. Final Formulations: A- After Preparation B- Final Product

Conclusion

Synthetic anti-varicose formulations currently available on the market are associated with numerous adverse effects. A literature review highlights that plants, known for their potent antibacterial, antioxidant, and anti-inflammatory properties, are widely utilized not only for treating various pathological conditions but also as food preservatives and flavoring agents. Leveraging these plant-based properties, the newly developed formulations

demonstrated significant effectiveness in addressing varicose veins, offering a promising and natural alternative to synthetic options.

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