JOURNAL OF ADVANCED MEDICAL AND PHARMACEUTICAL RESEARCH

Review Article

Evidence-Led Innovations and Experimental Approaches in Hemorrhoid Treatment

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Received 30th June 2025,
Accepted 16th September 2025
Published 8th October 2025

DOI:10.21608/jampr.2025.399256.1101

jampr.journals.ekb.eg

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ABSTRACT

Hemorrhoids, a prevalent anorectal disorder, significantly affect quality of life, yet effective treatments remain an area of ongoing research. This review provides a comprehensive analysis of preclinical studies and clinical management strategies for hemorrhoids. Preclinical studies, primarily involving animal models and *in vitro* systems, have identified various pharmacological agents with potential therapeutic benefits. Notably, compounds such as flavonoids, tannins, and certain herbal extracts have demonstrated anti-inflammatory, astringent, and venotonic effects that may help alleviate hemorrhoidal symptoms. These studies further emphasize the importance of cytokine regulation and the impact of oxidative stress on hemorrhoidal inflammation. Research on novel medication delivery methods, such as topical formulations and nanoparticles, has also shown promise in enhancing the efficacy of hemorrhoidal treatments. Meanwhile, preclinical research on surgical interventions, including laser therapy and sclerotherapy, has contributed to refining minimally invasive techniques. Clinical care often combines lifestyle changes, topical medication, and surgical procedures, but ongoing preclinical research is paving the path for more personalized and effective treatment approaches. Emerging therapies, including stem cell therapy and gene editing, are also briefly explored, pointing to future possibilities in hemorrhoid management. Overall, preclinical studies play a pivotal role in advancing both pharmacological and procedural treatments for hemorrhoidal disorders.

Keywords: Hemorrhoids, preclinical studies, herbal extract, oxidative stress, Cytokine modulation.

1. A BRIEF OVERVIEW

Hemorrhoids are anorectal disorders characterized by enlarged veins located in the lower region of the rectum and anus. They represent a common health issue that affects a large segment of the global population. Research shows that between 50% and 85% of people worldwide experience hemorrhoids at some point in their lives.

In the United States of America, approximately 10 million individuals are affected by hemorrhoid-related conditions at any given time, and nearly half of the population is likely to develop symptomatic hemorrhoids during their lifetime. The prevalence of hemorrhoids[1] varies significantly across regions. Studies indicate rates of 13.1% in Ethiopia, 16% in Israel[2], 14.4% in Korea[2], 18% in Egypt[3], and 38.9% in Austria.[4,5]

Primarily, an abnormal dilation and distortion of the vascular channel together with destructive changes in the vascular cushion within supporting connective tissues leads to hemorrhoidal conditions.

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1.1 Anatomy and pathophysiology of hemorrhoidal diseases

Hemorrhoids are common anatomical structures, both internal and external human body structures that are divided by the dentate line, which serves as an important an anatomical barrier. The three primary vascular cushions (also known as piles) that comprise the normal internal hemorrhoidal plexus are situated in the anus's left lateral (3 o'clock), right anterior (11 o'clock), and right posterior (7 o'clock) sites, as well as additional cushion-like tissues located around the anus at approximately the 1, 5, and 9 o'clock positions. These locations correspond to the major terminal branches of arteries found inside the submucosal layer.[6]

It is possible to comprehend the anal canal from both an anatomical and surgical perspective. It anatomically penetrates from the anal verge to the dentate line. However, from a surgical perspective, it is defined as stretching from the anorectal ring, which marks the transition of the rectum into the anal canal, down to the anal verge.[6]

Enlargement of the anal cushion is associated with various of pathological abnormalities and anatomical structure changes, including thrombosis, aberrant venous dilatation, thin-walled artery dilatation, connective tissue degeneration, and rupture of fibroelastic tissue.[7,8]

2. PATHOLOGICAL PROCESSES

Numerous hypotheses seek to explain the formation and progression of hemorrhoids, although the precise pathophysiology of these conditions remains not fully understood. Some of the most prominent theories include:

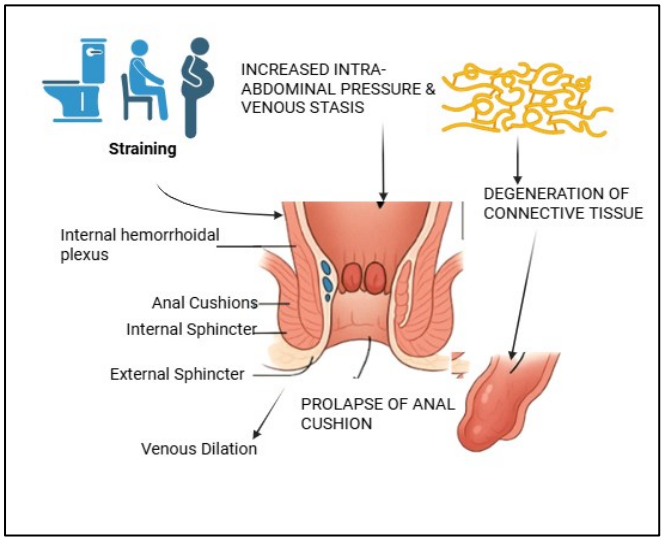

Figure 1. Pathophysiology of Hemorrhoids

2.1 Varicose vein theory

The theory posits that hemorrhoids develop due to the dilation and engorgement of veins within the anal cushions, similar to the formation of varicose veins in the legs. Increased vein compression of these veins frequently triggers venous dilatation, which can lead to hemorrhoids development.

According to this theory, elevated pressure within the anal veins causes their dilation and engorgement. Activities such as straining during bowel movements, sitting for prolonged periods, and childbearing contribute to this increase pressure. Over time, the venous walls may weaken, and the valves that prevent blood backflow may become dysfunctional, further promoting hemorrhoid formation.[9,10]

2.2 Vascular hyperplasia theory

The vascular hyperplasia theory of hemorrhoids postulates that a rise in the anal cushions' blood vessel count (vascular hyperplasia) is the root cause of hemorrhoids. The microvascular density increases in hemorrhoidal tissue, which is also known as neovascularization, due to the development of new blood vessels inside the tissue that is haemorrhage.[10]

2.3 Hyperactivity of the internal sphincter theory

In accordance with the Hyperactive of the Internal Sphincter theory, excessive constriction of the internal anal sphincter plays a crucial part in the development of hemorrhoids. This theory is based on the idea that increased sphincter tone leads to higher pressures within the anal canal, ultimately resulting in the venous congestion and hemorrhoid formation. A smooth muscle called the internal anal sphincter maintains a steady anal canal pressure. The anal cushions experience an increase in pressure when this muscle is hyperactive or contracts excessively.

These cushions, which are normally composed of blood vessels and connective tissue, become engorged with blood due to the elevated pressure. The outcome of venous congestion produces thickening and collapse of the cushions, developing hemorrhoids.

This increased the anal canal's internal pressure, which not only contributes to the development of hemorrhoids but also exacerbates symptoms. An elevated sphincter tone can make defecation difficult and trigger symptoms that include bleeding from the rectal area, itchiness, and discomfort. The hyperactivity of the internal anal sphincter also hinders the normal blood flow, further contributing to venous dilation and hemorrhoid formation.[11]

2.4 Sliding theory

The sliding theory of anal cushions, proposed in 1975, posits that hemorrhoids occur when the internal anal cushions break down and slide downward due to weakened supporting structures. This weakening predisposes the anal cushions to prolapse during activities such as defecation or when there is an increase in intra-abdominal pressure. The downward displacement is linked to the inflammatory processes of the internal canal, venous dilatation, degeneration of fibro-elastic and collagen tissues, vascular thrombosis, and changes in the anal sub-epithelial muscle. This theory remains widely accepted today.[12]

2.5 Hemodynamic theory

According to the theories of hemodynamics and hypervascularization, blood is carried by the terminals of the anterior rectal artery's branches to the anal cushions, having a slightly more expansive diameter. The enlargement is associated with increased blood flow and reduced venous impairment, resulting from the collapse of arteriovenous connections. This leads to the swelling and eventual prolapse of the anal cushions.

By acting as a sphincter to reduce arterial blood intake and increase venous outflow, the smooth muscles lining the arteriovenous plexus reduced the incidence of hemorrhoids. Additionally, hemorrhoidal tissues' higher microvascular density encourages remodeling, especially following a thrombosis episode. Multiple mediators and enzymes, including zinc-dependent proteinases, thrombin-dependent matrix metalloproteinases (MMPs), and stromal vascular endothelial growth factor (VEGF), contribute to tissue remodeling and degradation, which ultimately weakens the anal cushions' structural support and triggers the development of hemorrhoids.[13,14]

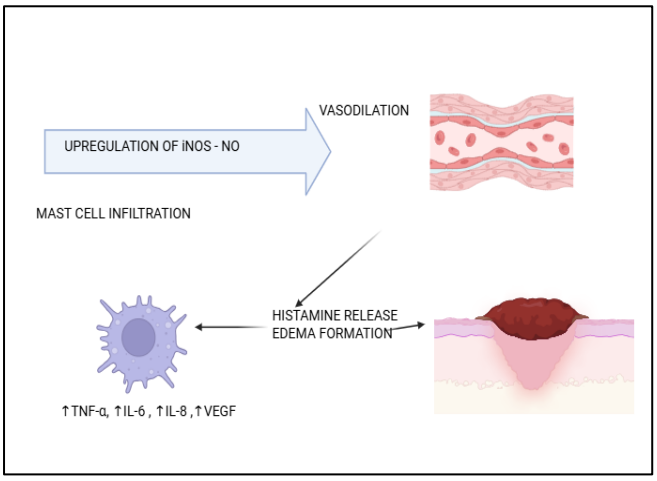

Figure 2. Vascular abnormaties and Inflammation

2.6 Inflammatory theory

The development and progression of hemorrhoids have been proposed to be significantly influenced by chronic inflammation in the anal area, according to the inflammatory theory of hemorrhoids.

Tumor necrosis factor-alpha (TNF-α), interleukin-1 (IL-1), interleukin-6 (IL-6), and other inflammatory mediators are released when the body's immune system is triggered by chronic inflammation in the anal cushions. These mediators trigger inflammatory processes, which affect tissue and allow the connective tissue supporting the anal cushions to weaken. The anal cushions' structural integrity is reduced by this tissue deterioration, resulting in the risk of prolapse and swelling. [4,9]

3. TYPES AND GRADING OF HEMORRHOIDS

The location of hemorrhoids with regard to the pectinate line, often known as the dentate line, determines their classification.

3.1 Internal hemorrhoids

Goligher's classification divides them into four stages and states that they are caused by distortion of the venous plexus over the dentate line. Depending on the severity of the anus prolapse and its proclivity to voluntarily reduce

GRADE I: There is no prolapse, but the anal cushion bleeds.

GRADE II: The anal cushions naturally diminish but collapse via the anus when there is extreme stress or an increase in intra-abdominal pressure.

GRADE III: Internal hemorrhoids that need to be manually pushed back inside the anus after prolapsing (staying out) during bowel movements.

GRADE IV: Prolapse stays out and is non-reducible.

3.2 External hemorrhoids

The swollen veins are observed below the dentate line and outside the anus. External hemorrhoids can be quite painful because of pain-sensitive nerve endings in the anoderm, in contrast to internal hemorrhoids, which are typically asymptomatic. A blood clot inside the enlarged vein can sometimes cause external hemorrhoids to thrombose, generating a hard, painful lump.[4]

4. RISK FACTORS

High body mass index [10], activities that elevate intra-abdominal pressure, such as weightlifting and straining, persistent constipation[11], pregnancy condition, which can cause constipation or reduced support of connective tissues due to increased elasticity [12], staying in the bathroom for extended periods of time might cause blood flow disruption, poor dietary habit, and hard stools or diarrhea, both of which can make bowel movements difficult [13]

5. MANAGEMENT OF HEMORRHOIDAL DISEASES

Managing hemorrhoidal diseases involves a combination of lifestyle changes, medical treatments, and sometimes surgical interventions.

5.1 Dietary and lifestyle modification

Increasing fiber intake is essential for preventing constipation and lowering the risk of hemorrhoids. By increasing intake of fruits, vegetables, whole grains, and legumes, one can lessen the need for straining by softening stools to promote regular bowel movements. Additionally, staying well-hydrated by drinking plenty of water throughout the day can aid in maintaining soft stools and preventing constipation.[10]

It is crucial to maintain a healthy weight in order to lessen the strain on the veins in the rectal region. For individuals who are overweight, weight loss can be beneficial in managing hemorrhoidal symptoms. Healthy eating habits along with regular exercise are essential for effective weight management.[10]

Hemorrhoidal disease prevention and management can be greatly aided by implementing these lifestyle modifications. By focusing on dietary modifications, proper bathroom habits, regular exercise, weight management, good hygiene, avoiding heavy lifting, and establishing regular bowel habits, individuals can reduce the risk of developing hemorrhoids and alleviate symptoms.

5.2 Medical Treatment

5.2.1 *Oral Calcium Dobesilate*

Oral calcium dobesilate is a medication primarily used to treat piles (hemorrhoids) and varicose veins. It works by improving blood flow, reducing the leakage and fragility of small blood vessels, and acting as an antioxidant to promote healing. It has been demonstrated that it primarily acts on the capillary and venous walls by decreasing their permeability and fragility. This helps in reducing edema and protecting blood vessels. It raises endothelial nitric oxide levels by increasing the activity of nitric oxide synthase. Nitric oxide acts as a vasodilator, helping to relax blood vessels and improve blood circulation, while also having anti-platelet and fibrinolytic effects. [14,15,16]

5.2.2 *Oral Flavonoids*

Oral flavonoids have been prominently proven to aid in the control of hemorrhoids because of their antioxidant, anti-inflammatory, and venotonic properties. The pharmacological actions associated with this venoactive preparation are based on the inhibition of leukocyte rolling, adhesion, and migration. It is made up of 90% micronized diosmin and 10% additional active flavonoids, including hesperidin, diosmetin, linarin, and isorhoifolin.

In a chronic venous hypertension model created by a femoral arteriovenous fistula, MPFF reduced the infiltration of granulocytes and macrophages into the venous valves, thereby preventing their degeneration.

The generation of prostaglandins and inflammatory substances, such as thromboxane B2, prostaglandin E2, and prostaglandin F2α, has been demonstrated to be decreased by a micronized pure flavonoid fraction in rat granulomas. It also shows antioxidant activity, reduces ischemia caused by histamines, bradykinin, and leukotriene B4, and prevents lipid peroxidation in endothelial cells. It improves lymphatic drainage and venous tone by reducing norepinephrine metabolism and altering noradrenergic signaling. It also improves microcirculation by increasing capillary resistance and decreasing capillary hyperpermeability in patients with fragile capillaries.[15]

6. CURRENT THERAPIES

6.1 Sclerotherapy

Sclerotherapy is currently recommended for the treatment of first- and second-degree hemorrhoids. The mechanism involves the injection of chemical agents (quinine hydrochloride, urea hydrochloride, and hypertonic saline) that induce fibrosis, anchoring the mucosa to the underlying muscle layer.[17,18]

Adverse effect: Potentially, incorrect injection technique may cause mucosal ulceration or localized necrosis and, in rare but notable cases, lead to serious septic complications such as retroperitoneal sepsis or prostatic abscess. In patients with immunodeficiency or severe valvular heart disease, antibiotic prophylaxis is recommended to minimize the risk of bacterial infection following sclerotherapy.[18]

6.2 Rubber Band Ligation

Rubber band ligation (RBL) is a widely preferred non-surgical treatment for second-degree hemorrhoids, offering

about 70% short-term success. It affects the tissue located above the pectinate line, leading to necrosis and separation. Radiofrequency ablation (RFA) employs a ball electrode linked to a radiofrequency generator to coagulate and vaporize hemorrhoidal tissue. This reduces vascular supply and promotes fibrosis, anchoring the hemorrhoid to underlying tissue. Studies show RBL outperforms sclerotherapy, with similar complication rates but fewer patients needing further treatment.[19,20]

Adverse effects: Some side effects of radiofrequency ablation (RFA) include perianal thrombosis, fissure infections, and significant urinary retention. Although the procedure is nearly painless, it carries a notable risk of prolapse and occasional bleeding.[21]

6.3 Hemorrhoidectomy

Hemorrhoidectomy is a surgical procedure used to remove severe or persistent hemorrhoids, typically Grade III or IV internal hemorrhoids, or complicated cases such as thrombosed or strangulated hemorrhoids.

Traditionally, third-degree hemorrhoids are treated with hemorrhoidectomy, mainly using the Ferguson (closed) or Milligan-Morgan (open) techniques. Both aims to remove internal and external hemorrhoidal tissue while preserving enough mucosa for normal function. Modern tools like harmonic scalpels, lasers, and radiofrequency devices offer reduced postoperative discomfort and faster healing.[21]

Adverse effects: Postoperative bleeding, occurring in about 2% of cases, may result from inadequate sutures, device failure, or trauma. Postoperative wound infections may result from high blood flow, bacterial exposure, and altered local immunity.[22]

6.4 Doppler-guided Hemorrhoidal Artery Ligation (DGHAL/THD)

DGHAL (also known as Transanal Hemorrhoidal Dearterialization – THD) is a minimally invasive, non-excisional technique used to treat Grade II–III internal hemorrhoids, and occasionally Grade IV. A Doppler probe inserted through a proctoscope identifies arteries that supply the hemorrhoids. These are tied with absorbable sutures to reduce blood flow. Prolapsed tissue is repositioned and secured with mucopexy.[23]

Adverse effect: Post-DGHAL may cause minor bleeding, pain, urinary retention, thrombosis, fissures, ulcers, infections, incomplete relief, recurrence, granulomas, stenosis, or rare incontinence.[23]

7. PRECLINICAL EVALUATION OF HEMORRHOIDS

7.1 Spontaneous models

Numerous dynamically occurring rat models of hemorrhoids exist; rats are commonly used in hemorrhoid research due to several scientific and practical advantages. Their anatomical and physiological responses to hemorrhoid-inducing agents closely mimic human pathophysiology, making them ideal for studying disease mechanisms and treatment options.

- Croton oil induced hemorrhoids
- Jatropha oil induced hemorrhoids
- Acetic acid induced hemorrhoids

7.1.1 *Croton oil induced hemorrhoids*

7.1.1.1 *Mechanism of induction*

Croton oil, produced from Croton tiglium L., is principally known for its unpleasant and irritant qualities due to its phorbol ester components, particularly 12-O-tetradecanoylphorbol-13-acetate. According to research, this substance activates protein kinase C, which results in an inflammatory response that releases pro-inflammatory cytokines such as tumor necrosis factor-alpha (TNF-α), interleukin-1 beta (IL-1β), and interleukin-6 (IL-6), among other inflammatory mediators.

Additionally, phorbol esters induce vasodilation and trigger the infiltration of polymorphonuclear leukocytes, thereby initiating a complex inflammatory response. Nitric oxide, cytokines, prostaglandins, leukotrienes, kinins, chemokines, and bradykinins are among the soluble pro-inflammatory mediators that have been shown to be catalyzed by these esters. The inflammatory cascade involves the manipulation of resident cells, including fibroblasts, mast cells, endothelial cells, and macrophages, and these molecular effectors work together to contribute to it. The mobilization of circulating inflammatory cells, such as neutrophils, monocytes, eosinophils, and lymphocytes, is an additional component of this response, which leads to the inflammatory process.[24,25,26]

7.1.1.2 *Induction process*

Hemorrhoids will be induced in rats by the application of prepared croton oil onto the anal portion of the animal. Hemorrhoids are induced by croton oil solution (containing deionized water, pyridine, diethyl ether, and 6% croton oil in diethyl ether in the ratio of 1:4:5:10).

Followed by overnight fasting, sterile cotton swabs of 4 mm in diameter are soaked in 100 µl of prepared croton oil and inserted into the recto-anal region of the study animals, 20 mm from the anal opening, and kept for 10 seconds. The edema will be developed linearly until 7 to 8 hours after croton oil application. [24]

Induction of croton oil is done once a day; the presence of edema would be denoted at the end of the third day after induction.

Twenty-four hours after the induction, animals of the respective group will be treated for the next 5 days. On the 9th day, 1 hour after the treatment, blood samples will be collected with retro-orbital sinus under anesthesia for the evaluation of the biochemical parameters. All animals would be euthanized by injecting an overdose of general anesthetics. Thiopental sodium (40mg/kg) was administered through the intraperitoneal route, and recto-anal tissue was taken along 20 mm, which would be separated, and the weight will be noted.

7.1.1.3 *Parameters to be evaluated* [24]

I. Hemorrhoid Grading

II. Anorectal coefficient (ARC)- Animal and isolated tissue weight to calculate the ARC would be calculated by the following formula:

$$ARC = \frac{\text{Anorectal tissue weight(mg)}}{\text{Animal body weight (g)}}$$

III. Cytokine profiling

Blood is collected from the recto-orbital sinus to measure TNFα, IL-6, and VEGF levels using an ELISA kit.

IV. Histomorphometric analysis

7.1.1.4 *Biochemical parameter*

Collection of the sample: The blood would be collected and allowed to obtain the serum; the serum would be obtained after centrifugation at 3000 rpm for 15 min and stored at -800C for further biochemical analysis.

Biochemical parameters to be estimated are: Superoxide Dismutase SOD, Lipid peroxidation activity, Catalase activity CAT, Reduced Glutathione (GSH), Malondialdehyde, Myeloperoxidase, and Total Protein

7.1.2 *Jatropha oil induced hemorrhoids*

7.1.2.1 *Mechanism of jatropha oil*

Studies reported [27] that Jatropha oil contains phorbol esters, which may inhibit the production of prostaglandins. Prostaglandins help regulate blood flow and inflammation. Reduced prostaglandin synthesis might lead to increased blood vessel constriction, potentially exacerbating hemorrhoids. Phorbol esters in Jatropha oil can activate protein kinase C (PKC), triggering the activation of several inflammatory pathways. This might result in increased inflammation and swelling in the rectal veins, contributing to hemorrhoid development, and it also may increase vascular permeability, allowing more fluid to leak into the tissues and contributing to swelling and inflammation in the rectal area.

Additionally, Jatropha oil showed that it altered gut motility by altering the balance of gut hormones, such as gastrin and cholecystokinin. Altered gut motility can lead to constipation or diarrhea, both of which can contribute to hemorrhoid development.

7.1.2.2 *Process of induction*

The induction of hemorrhoids was carried out with jatropha with slight modification. The application of jatropha is done by soaking a sterile cotton swab inserted into the anus about 20 mm from the anal opening of a rat and keeping it there for 20 seconds in all groups (except the normal control). Administration of jatropha oil is done once a day for 5 consecutive days.

A day after induction, all groups will be subjected to their relevant treatment for the next five days. On the last day, one hour after the relevant treatment, record the body weight of all rats. Then, euthanize the rats using an overdose of CO2 for histopathological examination and measurement of the recto-anal coefficient (RAC) determination, and collect the blood samples from the retro-orbital route for biochemical evaluation.[27]

7.1.2.3 *Parameters to be evaluated*

I. Hemorrhoid Grading Severity Score
II. Cytokines profiling i.e., PEG-2, TNF-α, IL-6

III. Histopathological analysis
IV. Anorectal coefficient (ARC)- Animal and isolated tissue weight to calculate the ARC would be calculated by the following formula:

$$ARC = \frac{\text{Anorectal tissue weight(mg)}}{\text{Animal body weight (g)}}$$

7.1.3 *Acetic acid induced hemorrhoids*

7.1.3.1 *Mechanism of induction of acetic acid*

Acetic acid is a weak organic acid that has been reported to induce hemorrhoids in individuals by disrupting the mucosal barrier, leading to increased permeability and allowing toxins and irritants to penetrate the tissue. According to research, acetic acid can cause oxidative stress and inflammation in the rectal mucosa, which results in the generation of reactive oxygen species (ROS) and pro-inflammatory cytokines.

The transcription factor nuclear factor kappa B (NF-κB), which controls the production of genes that promote inflammation, can be activated by acetic acid. Furthermore, it has the ability to enhance the production of adhesion molecules, including intercellular adhesion molecule-1 (ICAM-1), which can promote leukocyte recruitment and inflammation.[28]

7.1.3.2 *Induction of hemorrhoids*

Hemorrhoids were induced by rectally administering 2 mL of a 3% (v/v) acetic acid solution using a 2 mm diameter polyurethane tube, which was inserted 4.5 cm into the rectum. To prevent solution extravasation, the rats were maintained in a Trendelenburg position throughout the rectal instillation procedure and for an additional minute post-instillation, ensuring optimal retention of the irritant solution.

7.1.3.3 *Parameters*

Biochemical analyses, including measurements of caspase-3 and myeloperoxidase (MPO) levels, were performed on tissue samples taken from the proximal region of the anus (2–3 cm).[29]

7.2 Stable internal hemorrhoid model

Establishing a stable internal hemorrhoid model in rats is crucial for researching the pathophysiology and potential treatments of this condition. A notable method involves the induction of a croton oil mixture to the recto anal region, combined with specific physical activities.[30,31]

7.2.1 *Procedure*

Inhalation isoflurane anesthesia- Rats were anesthetized using inhaled isoflurane (induction: 3–3.5%, maintenance: 1.5–2.5%) and placed supine on a custom fixation frame with the anus fully exposed. Following sterilization and positioning in the lithotomy posture, a longitudinal incision of approximately 0.5 cm was made in the canal sulcus at the 12 o'clock position using tissue scissors. The incision included the inner and 12-point dentate lines along with the outer sulcus margin to facilitate effective drainage. After the procedure, the rats were allowed to eat normally for three days. A pale-yellow ulcer developed at the site of the 12-point incision, without notable swelling. The experiment proceeded once the absence of bleeding was confirmed.[32]

Croton oil stimulation: A croton oil mixture was created by combining distilled water, pyridine, ether, and 6% croton oil in a ratio of 1:4:5:10. After immobilizing the rat for 10 seconds, 0.05 mL of the croton oil combination was pipetted into the mucosa on the rat's tooth line. Fingers were then used to gently massage the perianal region for 10 seconds, and the rat was then put back in its cage.

A plastic cylinder of 5 cm in diameter and 35 cm in length roughly three times the rat's body length was chosen for the standing experiment at a temperature between 18 and 20°C. The cylinder was made to fit snugly around the rat's abdominal circumference. The rat was positioned inside the cylinder with its head facing upward and feet downward. Once the feet were exposed, the cylinder was secured in a vertical position, keeping the rat upright and stationary for 6 hours.[33]

Swimming experiment: For an hour, the standing position rats were submerged in water with a temperature between 20 and 25 °C. After 1 hour, the rats exhibited signs of distress, including sparse and disheveled fur, lethargy, The rats exhibited reduced responsiveness to external stimuli, hind limb weakness, trembling, and signs of overall exhaustion and fatigue. They were then placed in a room maintained at 25°C to allow their fur to dry for 1 hour.[34]

8. TRADITIONAL HERBAL EXTRACT FOR ANTI-HEMORRHOIDAL ACTIVITY

Traditional herbal extracts have been used for centuries to treat hemorrhoids due to their anti-inflammatory, astringent, and soothing properties. These herbs are believed to alleviate symptoms like swelling, pain, and itching associated with hemorrhoids. Some commonly used herbs are listed in **Table 1**.

9. NANOPARTICLE-BASED HERBAL FORMULATIONS FOR HEMORRHOIDS

Nanoparticle-based herbal formulations (**Table 2**) present a propitious therapeutic strategy for hemorrhoid management. By augmenting bioavailability, facilitating site-specific targeting, and enhancing therapeutic efficacy, these innovative formulations may afford symptomatic relief and significantly improve patients' quality of life.

Table 1. Traditional herbal extract for anti-hemorrhoidal activity

No.	Botanical name	Family	Extraction	Formulation	Model	Activity /MOA	Reference
1	***Allium iranicum***	Amaryllidaceae	Leaves extraction	Cream	A double blind randomized controlled clinical trial	Leaf shows significant better outcomes in the leek group compared to the routine ant hemorrhoid and placebo groups in the field of bleeding severity and overall subjective improvement.	35
2	***Cissus quadrangularis* L.**	Vitaceae	Leaves ethanolic extract	Enteric film coated tablets	• Anti-inflammatory • Vasoconstrictive effects • Acute oral toxicity	↑ Healing rate ↓ Reoccurrence rate	36
3	***Teucrium Orientale L.***	Lamiaceae	Aqueous extract	Extract	Randomized clinical trail	↑ Healing rate ↓ Reoccurrence rate	37
4	***Aesculus hippocastanum* Seed** ***V. Vinifera* leaves**	Sapindaceae Vitaceae	Ethanolic extract	Hydrogel	Writhing test Plethysmometric method	Antinociception effect (pain) Anti inflammatory effect	38
5	***Pluchea indica* Leaves**	Asteraceae	Aqueous extract	Extract	• Whole gut transit time in mice • Intestinal transit rate in mice • Croton oil-induced hemorrhoids in rats	No effect on the gastrointestinal. Significant decrease in the inflammation of hemorrhoid .	39
6	***Acacia ferruginea* DC**	Fabaceae	Hydroalcoholic extract	Extract	Croton oil-induced hemorrhoids in rats	Reduced the inflammatory cytokines TNF-α, IL-6 ,PGE2.	40
7	***Terminalia chebula Retz.***	Combretaceae	Powder	Capsule	A double – blind randomized placebo – controlled clinical trial	Repaired hemorrhoid damaged tissues.	41
8	***Phlomis grandiflora***	Lamiaceae	Methanolic extract	Cream	Croton oil-induced hemorrhoids in rats	VEGF ↓ Capillary permiability .	42
9	***Solanum melongena . L***	Solanaceae	Methanolic extract	Extract Tropical cream	Croton oil-induced hemorrhoids in rats	Reducing the capillary permeability.	43
10	***Annona muricata L.***	Annonaceae	Ethanolic extract	Extract	Croton oil-induced hemorrhoids in rats	Inhibition of TNF-α,IL-β,IL-6, Nitric oxide .	44

No.	Botanical name	Family	Extraction	Formulation	Model	Activity /MOA	Reference
11	***Capsella bursa-pastoris***	Brassicaceae	Ethanolic extract Aqueous extract	Extract	Croton oil-induced hemorrhoids in rats	Ameliorated the levels of the cytokines, LPO, MPO, and the antioxidants.	45
12	***Dolichandrone falcata***	Bignoniaceae	Ethanolic extract	Extract	Croton oil-induced hemorrhoids in rats	Restore the alterations in LPO and CAT levels.	46
13	***Lawsonia inermis***	Lythraceae	Ethanolic extract	Extract	Croton oil-induced hemorrhoids in rats	Anti inflammatory activity	47
14	***Graptophyllum pictum (L.)***	Acanthaceae	Ethanolic extract	Oral and tropical formulation	Croton oil-induced hemorrhoids in rats	Repaired hemorrhoid damaged tissues.	48
15	***Launea procunbence (Linn.)***	Asteraceae	Aqueous extract	Suspension	Inflammatory induction of hemorrhoidal disease by injection of kaolin suspension	Anti-inflammatory effect is maxima.	49
16	***Aloe vera***	Asphodelaceae (Liliaceae)	Scoped gel	Oral and tropical formulation	Croton oil-induced hemorrhoids in rats	Oral formulation is not much effective as compared to standard.	50
17	***Newbouldia laevis***	Bignoniaceae	Ethanolic extract	Emulgels	Croton oil-induced hemorrhoids in rats	Novel formulation has better anti inflamatory property.	51
18	***Actiniopteris radiata***	Pteridaceae	Ethanolic extract	Extract	Croton oil-induced hemorrhoids in rats	Showed reduction inflammatory extent, severity, edema, granulation tissue, congestive and thickened blood vessels, suggestive of anti-inflammatory and anti-oxidant properties.	52
19	***Cistus laurifolius***	Cistaceae	Methanolic and aqueous extracts	Extract	Croton oil-induced hemorrhoids in rats	Improved blood markers of disease progression and histomorphological scores.	53
20	***Lantana Camara***	Verbenaceae	Extract	Candy	Croton oil-induced hemorrhoids in rats	Lantana Camara leaves have anti-inflammatory, analgesic action.	54
21	***Achillea millefolium***	Asteraceae	Hydroalcoholic extract	Ointment	Randomized double-blind placebo-controlled clinical trial	Positive effect on improving pain, discomfort during defecation, intensity and frequency of bleeding in patients with grade 1 and 2 internal hemorrhoids.	55

No.	Botanical name	Family	Extraction	Formulation	Model	Activity /MOA	Reference
22	***Ocimum gratissimum***	Labiateae	Ethanolic extract	Extract	Croton oil-induced hemorrhoids in rats	Reparation of anorectal tissue .	56
23	***Boswellia serrata***	Burseraceae	Hexane, chloroform, and methanol	Extract	Croton oil-induced hemorrhoids in rats In silico methods	The identified phytoconstituent(s) that could potentially interact with the target protein cyclooxygenase-2 (COX-2) (PDB: 4RRW) using molecular dynamics simulation and in silico docking.	57
24	***Pithecellobium Dulce***	Fabaceae	Aqueous	Extract	Jatropha oil-induced hemorrhoidal disease in rats.	Anti-hemorrhoidal properties via mechanism involving vasoconstriction, alongside enhancement of anti-inflammatory and antioxidant status.	58
25	***Laurus nobilis***	Lauraceae	Ethanolic	Extract	Croton oil-induced hemorrhoids in rats	Higher dose posses anti hemorrhoidal acivity.	59
26	***Scaphium Affine***	Malvaceae	Ethanolic	Extract	Croton oil-induced hemorrhoids in rats Jatropha oil-induced hemorrhoidal disease in rats.	Histopathological analysis confirmed the tissue recovery as it revealed minimal inflammation and decreased dilated blood vessels in treated animals.	60
27	***Moringa Oleifera***	Moringaceae	Ethanolic	Extract	Croton oil-induced hemorrhoids in rats	Moringa oleifera extract reduced inflammation, cytokines, extravasation, and tissue damage in croton oil-induced hemorrhoidal rats, proving anti-hemorrhoidal potential.	61
28	***Tridax procumbens***	Asteraceae	Ethanolic whole plant extract	Extract	Croton oil-induced hemorrhoids in rats	Reduced inflammation, neutrophils, TNF-α, and IL-6 in hemorrhoidal rats.	62
29	***Lawsonia inermis***	Lythraceae	Ethanolic leaf extract	Extract	Croton Oil Induced Hemorrhoidal Rats	Reduced inflammation, TNF-α, IL-6, and neutrophils in hemorrhoidal rats.	63
30	***Capsella bursa-pastoris***	Brassicaceae	Ethanolic leaf extract	Extract	Croton Oil Induced Hemorrhoidal Rats	Confirmed its anti-hemorrhoidal, antioxidant effects, recommending its organic acid-rich extracts for treatment.	64

No.	Botanical name	Family	Extraction	Formulation	Model	Activity /MOA	Reference
31	***Amorphophallus paeoniifolius***	Araceae	Methanolic extract Aqueous extract	Extract	Croton Oil Induced Hemorrhoidal Rats	Reduced inflammation, oxidative damage, vasodilation, and hemorrhagic severity.	65
32	***Tephrosia linearis (Wild) Pers***	Fabaceae	Aqueous extract	Extract	Jatropha Oil Induced Hemorrhoidal Rats	*T. Linearis* extract showed stronger anti-hemorrhoid effects than Daflon, supporting traditional use.	66
33	***Blumea lacera (Burm.f.) DC.***	Asteraceae	Ethanolic leaf extract	Extract	Croton Oil Induced Hemorrhoidal Rats	Significantly reduced inflammation, cytokines, RAC, and oxidative stress, supporting *Blumea lacera*'s traditional hemorrhoid treatment, likely due to quercetin.	67
34	***Pithecellobium dulce***	Fabaceae	Aqueous Extract of the seeds	Extract	Induction of hemorrhoidal disease by acetic acid 5 %	Seed extract reduced acetic acid-induced hemorrhoids by decreasing inflammation, oxidative stress, and tissue damage, enhancing antioxidant defense and healing.	68
35	***Moringa oleifera***	Moringaceae	Seed oil	Suppositories	Croton Oil Induced Hemorrhoidal Rats	Moringa seed oil suppositories with macrogol and dika fat demonstrated stability, anti-inflammatory effects, and potential for hemorrhoid treatment via optimized delivery.	69
36	***Ageratum conyzoides L***	Asteraceae	Ethanolic leaf extract	Tropical and oral	Croton Oil Induced Hemorrhoidal Rats	Treatment with the test extract dose-dependently decreased inflammation, rectal bleeding and hemorrhoid size, when compared to the control group, histopathological analysis of the anal area of treated rats revealed a reduction in congestion, edema, and vascular dilatation.	70

Table 2. Nanoparticle based herbal formulation for hemorrhoid

No.	Plant scientific name & common name	Part used	Metal/metal oxide nanoparticles	Method used for characterization	Anti hemorrhoidal activity	Results	Signaling pathway		References
1	Blumea lacera	Seeds	Silver nanoparticles (PLSNPs) (spherical)	HPLC, TEM, SEM	Induced hemorrhoids	Restoration of altered antioxidant status Reduced inflammation zone with minimal dilation of blood vessels	AMPK pathway	Activates AMPK, Reduction of inflammation Endothelin B receptor modification.	[71]
2	GraptophylluSm pictum	leaves	Lipid nanoparticles (liposomal and ethosomal)	DSC, SEM, EE, TLC, FTIR	Croton oil induced hemorrhoids Skin penetration study	GE revealed better anti-hemorrhoid activity as it showed a lesser number of inflammatory cells, goblet cells, and necrosis cells. Additionally, it exhibited a smaller hemorrhage area and muscular mucose cell thickness compared to GL	Unspecified	It revealed better anti-hemorrhoid activity as it showed a lesser number of inflammatory cells, goblet cells, and necrosis cells.	[72]

10. SUMMARY AND PROSPECTS

Although numerous animal models claim to replicate hemorrhoids, only a few truly meet key criteria: clinical and pathological resemblance to hemorrhoids, involvement of relevant pathogenic mechanisms, and validation through response to established hemorrhoid therapies.

Preclinical studies have significantly advanced our understanding of hemorrhoidal disease, elucidating its complex pathophysiology involving vascular hyperplasia, inflammation, and connective tissue deterioration. These insights have established the groundwork for the development of targeted therapeutic strategies beyond conventional symptomatic relief. Key preclinical findings emphasize the role of oxidative stress, vascular dysfunction, and inflammatory mediators in hemorrhoid progression.

Animal models have been instrumental in testing pharmacological agents such as flavonoids, anti-inflammatory compounds, and novel sclerosing agents, which demonstrate promising results in reducing hemorrhoidal swelling, bleeding, and discomfort.

Evidence-based approaches integrate these findings into clinical practice, promoting minimally invasive and conservative treatments as first-line options. Techniques such as rubber band ligation, infrared coagulation, and doppler-guided hemorrhoidal artery ligation are supported by robust clinical data. Additionally, phototherapeutics like micronized purified flavonoid fractions (MPFF) have shown efficacy in both acute and chronic hemorrhoidal disease management.

Looking forward, further integration of molecular and genetic research could enable the development of personalized treatment strategies. Nanotechnology-based drug delivery systems and bioengineered tissue scaffolds represent promising future avenues. Continued collaboration between preclinical research and clinical trials will be crucial in optimizing therapeutic outcomes and improving patient quality of life in hemorrhoidal disease management.

ACKNOWLEDGMENT

We sincerely appreciate all who shared their insights and experiences, and we especially thank Mrs. Samhitha J of Visveswarapura College of Pharmaceutical Sciences, Bangalore, for the opportunity to contribute to this work.

AUTHOR CONTRIBUTIONS

Lekhashree. M. G. was responsible for the overall development of the article, including conceptualization, conducting the literature review, and drafting the manuscript. Samhitha J contributed to data analysis, provided critical revisions, and assisted in the final editing of the manuscript. The final draft of the manuscript was reviewed and approved by both authors.

CONFLICT OF INTEREST

The authors declare that there are no conflicts of interest or competing interests related to this publication.

REFERENCES

1. A A Kibret, Oumer M, Moges A M .(2021). Prevalence and associated factors of hemorrhoids among adult patients visiting the surgical outpatient department in the University of Gondar Comprehensive Specialized Hospital, Northwest Ethiopia. *PLOS ONE,*16(4): e0249736. https://doi.org/10.1371/journal.pone.0249736
2. Carter D, Beer Gabel M, Zbar A, Segev S, Kopylov U. Prevalence and Clinical Associations of Hemorrhoids at Screening Colonoscopy. *World Journal of Colorectal Surgery*. 2013;3(2):10
3. Lee J-H, Kim H-E, Kang J-H, Shin J-Y, Song Y-M. Factors associated with hemorrhoids in korean adults: korean national health and nutrition examination survey. *Korean journal of family medicine.* 2014;35(5):227.
4. Lohsiriwat V. (2022). Hemorrhoids: from basic pathophysiology to clinical management. *World J Gastroenterol*,18(17):2009-17
5. F.Johanson,A Sonnenberg. (1990). The prevalence of hemorrhoids and chronic constipation: An epidemiologic study.*Gastroenterology.* 98(2):380-86
6. Mahadevan, V. Anatomy of the rectum and anal canal surgery (2019). *Surg.J,*35(3):1-5
7. Peter,J,bazira . Anatomy of the rectum and anal canal surgery (2022). *Surg.J*,41(1)
8. Lee J M, Kim NK. Essential Anatomy of the Anorectum for Colorectal Surgeons Focused on the Gross Anatomy and Histologic Findings (2019). *Ann Coloproctol*,34(2):59-71
9. Aigner F, Gruber H, Conrad F et al. (2009) Revised morphology and hemodynamics of the anorectal vascular plexus: impact on the course of hemorrhoidal disease. *Int J Colorectal Dis,*24(1):105-13
10. Lalisang TJ. (2022). Hemorrhoid: Pathophysiology and Surgical Management Literature Review. New Ropanasuri J Surg,1(9):31-6
11. De Marco S, Tiso D. (2021). Lifestyle and Risk Factors in Hemorrhoidal Disease. *Front. Surg,*1-5
12. Sandler, Robert S.(2019). Rethinking What We Know About Hemorrhoids. *Clin Gastroenterol Hepatol,*17;8-15

13. Octavi ZN, Suryaningtyas W. (2024). Factors Affecting the Incidence of Hemorrhoids: A Literature Review. *IJRP,*140(1):658-61
14. Cintron, J.R., Abcarian, H. (2007). Benign Anorectal: Hemorrhoids. In: Wolff, B.G., *et al.* The *ASCRS Textbook of Colon and Rectal Surgery. Springer*,156-177
15. Das Gracas C, de Souza M, Cyrino FZ,et,al.(2018). Protective Effects of Micronized Purified Flavonoid Fraction (MPFF) on a Novel Experimental Model of Chronic Venous Hypertension. *Eur J Vasc Endovasc Surg*,55(5):694-702.
16. Bulent Mentes, B., Gorgul, A., Tatlicioglu, E. *et al.* (2001).Efficacy of calcium dobesilate in treating acute attacks of hemorrhoidal disease. *Dis Colon Rectum* ,1489–95
17. He A, Chen M. (2023). Sclerotherapy in Hemorrhoids. *Indian J Surg*,85(2):228-232.
18. Juan C G, Gonzalo L G. (2024).Effective non-surgical treatment of hemorrhoids with sclerosing foam and novel injection device.*Gastroenterology & Endoscopy*.2(4)176-180
19. Albuquerque A.(2016). Rubber band ligation of hemorrhoids: A guide for complications. *World J Gastrointest Surg,*8(9):614-620
20. Chew SS, Marshall L, Kalish J,et al. (2003). Short-term and long-term results of combined sclerotherapy and rubber band ligation of hemorrhoids and mucosal prolapse. *Dis Colon Rectum*, 46:1232–1237.
21. Cocorullo G, Tutino R, Falco N, et al. (2017) The non-surgical management for hemorrhoidal disease. A systematic review. *G Chir*,38(1):5-14.
22. Guttadauro A, Maternini M, Chiarelli M, et al. (2018) Evolution in the surgical management of hemorrhoidal disease. *Ann Ital Chir*,89:101-106
23. Yilmaz I , Sucullu I, Akin ML. (2012). Doppler-guided hemorrhoidal artery ligation: experience with 2 years follow-up. *Am Surg*, 78(3):344-48.
24. Azeemuddin, Mohammed & G.L.et al.(2001). An Improved Experimental Model of Hemorrhoids in Rats: Evaluation of Antihemorrhoidal Activity of an Herbal Formulation. *ISRN pharmacology,*1-7.
25. [18] Kusumo M H B, Prayitno A, . S, Laqif A. (2004).Synergistic therapeutic approach for hemorrhoids: integrating mesenchymal stem cells with diosmin-hesperidin to target tissue edema and inflammation. *Arch.Med, Sci*,20(5):1556-66.
26. [19] Nallajerla SK, Ganta S. (2022). Evaluation of Anti-inflammatory Mediated Anti-hemorrhoidal Activity of Lawsonia inermis on Croton Oil Induced Hemorrhoidal Rats. *Antiinflamm Antiallergy Agents Med Chem*,21(1):62-73
27. Ubana, Muhammad, B, Zaruwa, et al. (2022).Studies on the Effect of Tephrosia linearis (Wild) Pers. Aqueous Extract on Jatropha curcas Oil Induced Hemorrhoids in Rats.*SJBAS*, 4(1): 54-57
28. Gisele, A Francis, Takvou ,et al .(2023). Aqueous Extract of the seeds of Pithecellobium dulce (Fabaceae) Demonstrates Anti-inflammatory Effect Through Anti-oxidant Activity Enhancement in a Rodent Model of on Hemorrhoidal disease induced by Acetic Acid. *TJP*, 12(3):135-142
29. M yang, Liang H ,Sun C et al .(2024).Puji Zhichuang Suppositories Alleviate Acetic Acid-Induced Hemorrhoids in Rats by Inhibiting the Pi3k-Akt Signaling Pathway and Complement and Coagulation Cascades.*SSRN,*1-44
30. Minhui Ke, Shuyan Huang , Haorong Lin ,et al. (2023).Establishment and study of a rat internal haemorrhoid model.*Sci.Rep*,13:1-10
31. Professional Committee of Chinese Medicine Experimental Pharmacology, China Association of Chinese Medicine. (2017). Specifications for the preparation of animal models of hemorrhoids in Chinese medicine (draft). *Pharmacol. Clin. Chin. Med*,33(5), 211–212.
32. Zhou, J. et al. (2019).Research on the method of establishing a new animal model of hemorrhoids. *Chin. Mod. Appl. Med*,36(5), 526–31.
33. Wu, L. L. et al.(2021). Evaluation of the effect of establishing a hemorrhoid model by acetic acid-induced anorectal mucosal injury in rats. *Colorectal Anal. Surg*, 27(1), 19–23
34. Veskoukis, A. S. et al. (2018).A novel swimming performance test in rats. *Chin. J. Physiol*, 61(3), 144–151 .
35. Seyed Hamdollah Mosavat, Leila Ghahramani, et al.(2015).The effect of leek (Allium iranicum (Wendelbo)) leaves extract cream on hemorrhoid patients: A double blind randomized controlled clinical trial. *Eur J Integr Med*,7(6):669-73
36. Ahmadi Pirshahid, P., Thisayakorn, C.et al. Anti-hemorrhoid activities from *Cissus quadrangularis* L. 2016.*Acta Hortic*: 161-72
37. Emin N, Nural K, Guzel A. (2018). Investigation of The Effectiveness of Teucrium Orientale L. Plant in Hemorrhoid Treatment. *TURJAF*,6(3);326-32
38. Mihai D P, Seremet O C, Nitulesci G et al .(2019). Evaluation of Natural Extracts in Animal Models of Pain and Inflammation for a Potential Therapy of Hemorrhoidal Disease .*Sci Pharm*,87(2);1-14
39. Senvorasinh K, Phunikhom K, Sattayasai J et al .(2019). Anti-Hemorrhoidal Activity of Pluchea indica Leaves Aqueous Extract in Croton Oil-Induced Hemorrhoids in Experimental Animals.*SRIMEDJ*,34(6):1-9
40. Faujdar, Samriti , Sati ,et al. (2018).Phytochemical evaluation and anti-hemorrhoidal activity of bark of Acacia ferruginea DC. *JTCM*,9(2):85-9
41. Andarkhor, Pouran et al.(2019). Effects of Terminalia chebula Retz. in treatment of hemorrhoids: A double – blind randomized placebo – controlled clinical trial. *Eur J Integr Med* ,30:1-35
42. Ceylan, Koca U.(2019). Ethnopharmacological Survey on Phlomis grandiflora: In vivo Ant hemorrhoidal Model.*CUPMAP,*2(1):40-46
43. Donmez, Ceylan et al.(2020). From Nutrition to Medicine: Assessing hemorrhoid healing activity of Solanum melongena L. via in vivo experimental models

and its major chemicals. *J Ethnopharmacol*,261(6)113-20.
44. Ayun N Q,Nurhuda K et al .(2020). Anti-inflammation of soursop leaves (annona muricata l.) against hemorrhoids in mice induced by croton oil. *Pharmacogn J* ,12(4):784-92
45. Apaydin Yildirim B, et al .(2020). Antihemorrhoidal activity of organic acids of Capsella bursa-pastoris on croton oil-induced hemorrhoid in rats. *J Food Biochem*,44(9):1-9
46. Dhaswadikar, S.R., Parmar, et al.(2022). Anti-hemorrhoidal potential of standardized leaf extract of Dolichandrone falcata. *Phytomedicine Plus*,2(1):1-8
47. Nallajerla, Sai Krishna, and Suhasin Ganta.(2022).Evaluation Of Anti-inflammatory Mediated Anti-hemorrhoidal Activity of Lawsonia inermis on Croton Oil Induced Hemorrhoidal Rats. *Antiinflamm Antiallergy Agents Med Chem*,21(1):62-73
48. Kusumawati I, Rullyansyah S, Rohmania, et al . Histomorphometric study of ethanolic extract of Graptophyllum pictum (L.) Griff. leaves on croton oil-induced hemorrhoid mice: A Javanese traditional anti-hemorrhoid herb. *J Ethnopharmacol*. 2022; 284:114765
49. Pant R, Irchhaiya R, Mishra K .(2022). Anti-Hemorrhoidal Activity of Leaf Extractof Launea procunbence (Linn.). *NeuroQuantology* ,20(13):1504-10
50. Swathi S,Singh A,Vellapandian C.(2022). Therapeutic Effect of Aloe vera on Croton Oil Induced Hemorrhoids.*J nat remedies*,22(3):472-81
51. Olayeme O J , lucy Binda et al . (2023).Effect of emulgel containing Newbouldia laevis stem bark extract on croton oil-induced hemorrhoids in mice.*JRP*,27(4):1488-98
52. Anna M K, Vimala K S ,priyalatha et al. (2023).PRESENTATION OF THESIS IN SCIENTIFIC FORUM Anti-haemorrhoidal activity of Actiniopteris radiata (Sw.) Link (Mayurasikha) paste mixed with milk internally in Wistar Albino Rats.*Eur.Chem Bull*,12(4):20043-66
53. Pekacar, Sultan et al.(2024). Identification of bioactive components on antihemorrhoidal activity of Cistus laurifolius L. using RP-HPLC and LC-QTOF-MS. *J. Ethnopharmacol*,319(1):117122
54. Khandagale. S, Salve Dvitiya S.et al. (2024). Formulation Development and Evaluation of Herbal Candy Containing Lantana Camara Leaves Extract for Treatment of Hemorrhoid. *Int. J. of Pharm. Sci*,2(2), 509-19.
55. Mahmoudi A, Seyedsadeghi M, Miran M, et al .(2023). Therapeutic Effect of Achillea millefolium on the Hemorrhoids; A Randomized Double-Blind Placebo-Controlled Clinical Trial. *J. Herb. Med*,39(1):100657
56. Moneme E C, Nwaka A C,Oladejo A, et al . (2023). Histopathological and Anti-hemorrhoid Effect of the Leaf Extract of Ocimum gratissimum in Croton Oil-induced Hemorrhoid.*J Altern Complement Med*,25(12):1-10
57. Rao H, Maurya A, Kumar Raidas H, et al. (2024). In Silico Exploration of Potential Phytoconstituents from the Bark Extract of Boswellia serrata for Hemorrhoidal Disease: Molecular Docking and Molecular Dynamics Analysis. *Chem Biodivers*,21(2):1-5
58. Atsang A, Francis T, Finsia E, Mathieu, et al. (2023).Aqueou Extract of the seeds of Pithecellobium (Fabaceae) Demontrates Anti-inflammatory Effect Through Anti-oxydant Activity Enhancement in a Rodent Model of on Hemorrhoidal disease induced by Acetic Acid. *TJP*,12(3);135-42
59. Verma V,Ghai Roma , Nagaranjan et al .(2024). Pharmacological Investigation of Laurus nobilis Extract in Wistar Rats towards Anti-Hemorrhoidal Potential.*Indian J Pharm sci*,86(4)1324-30.
60. Abbas, Syed A et al. (2024). Gas Chromatography-Mass Spectrometry (GC-MS) Analysis of Scaphium Affine Seed Extract and Assessment of Its Anti-hemorrhoidal Efficacy. *Anti-inflammatory & anti-allergy agents in medicinal chemistry*,23(2): 118-128.
61. Nallajerla S K, Ganta Suhasim et al. (2022).Ethanolic Leaf Extract of Moringa Oleifera A Potential Agent for Treatment of Hemorrhoids: And Its Evaluation in Croton Oil Induced Hemorrhoidal Rats. *IJADR,*13(8):1-7
62. Nallajerla S K, Ganta Suhasim et al. (2023). Croton Oil-induced Hemorrhoidal Rats: Evaluation of Anti-inflammatory Mediated Anti-hemorrhoidal Activity of Tridax procumbens Whole Plant. *Curr. Bioact. Compd*,19(7):1-8
63. Nallajerla, Sai Krishna, and Suhasin Ganta. (2022). Evaluation Of Anti-inflammatory Mediated Anti-hemorrhoidal Activity of Lawsonia inermis on Croton Oil Induced Hemorrhoidal Rats.*Anti-inflammatory & anti-allergy agents in medicinal chemistry*,21(1): 62-73.
64. Apaydin Yildirim, Betul et al. (2020).Antihemorrhoidal activity of organic acids of Capsella bursa-pastoris on croton oil-induced hemorrhoid in rats. *Journal of food biochemistry*,44(9);1-10
65. Dey YN, Wanjari MM, Kumar D, et al. (2016).Curative effect of Amorphophallus paeoniifolius tuber on experimental hemorrhoids in rats. *J Ethnopharmacol*,192:183-191.
66. Zaruwa , Ubana M A , Muhammad et al. (2022).Studies on the Effect of Tephrosia linearis (Wild) Pers. Aqueous Extract on Jatropha curcas Oil Induced Hemorrhoids in Rats.*SJBAS,*4(1):54-7
67. Dubey T, Bhanukiran K, Prasad K S et al.(2023). Optimization of Extraction Process and Anti-Hemorrhoidal Activity of Blumea lacera (Burm.f.) DC. Leaves in Croton Oil-induced Hemorrhoid Model.Pharmacogn.Mag,19(1):709-19
68. Atsang,Francis T egre Finsia et al.(2023). Aqueou Extract of the seeds of Pithecellobium dulce (Fabaceae) Demontrates Anti-inflammatory Effect Through Anti-oxydant Activity Enhancement in a Rodent Model of on Hemorrhoidal disease induced by Acetic Acid.*TJP*,12(3):135-42
69. Lucy B, Ekere E ,Olayeme et al. Formulation, Evaluation and Anti-Hemorroidal Activity of Suppositories Containing Moringa Oleifera Lam. Seed Oil. *ACTA Pharmaceutics Sciencia*59(1):150-74

70. Nitisha , Virender Kaur, Tirath Kumar .(2025). Pharmacological Evaluation of Anti-hemorrhoidal activity in ethanolic leaf extract of Ageratum conyzoides L. S*EEJPH*,16:3384-3405
71. Dubey, Tarkeshwar ,Bhanukiran et al.(2024) Development of phytosterol-loaded silver nanoparticles for ameliorating haemorrhoidal complications via the AMPK pathway a mechanistic approach. *Biomed.Mater,*19(3):1-10
72. Kusumawathi I, Rullyansyah S, Warsito M F et al.(2024).Formulation and evaluation of encapsulated Graptophyllum pictum (L.) Griff. ethanolic extract in lipid vesicles for hemorrhoid treatment.*J Pharm Pharmacogn res*,12(1)27-38.

