



Evaluation of the Anticancer Potential of a Polyherbal Vaginal Gel Against DMBA-Induced Vaginal Carcinoma in Wistar Rats

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Abstract: This study looked at how well a polyherbal vaginal gel fights vaginal cancer caused by 7,12-dimethylbenz[a]anthracene (DMBA) in Wistar rats. The gel was made from extracts of *Curcuma longa* (turmeric), *Aloe vera*, *Azadirachta indica* (neem), and several other traditional herbs. After tumors formed, the gel was applied directly to the vagina. The results showed a big drop in tumor size and a later tumor appearance in animals treated with the gel. The group that got the highest gel dose did as well as rats getting the standard cancer drug, 5-fluorouracil (5-FU). Tissue samples showed less damage to the vaginal lining and more normal-looking tissue in the treated rats. In blood tests, levels of malondialdehyde (MDA), a sign of damage from free radicals, went down. At the same time, the rats had higher levels of helpful enzymes—superoxide dismutase (SOD), catalase, and glutathione (GSH)—which shows the balance of oxidative damage improved. The inflammatory proteins TNF- α and IL-6 were also much lower in the high-dose group. Molecular tests showed that the balance of proteins that control cell death changed: less Bcl-2 and more Bax and Caspase-3 were seen, which means more cancer cells were dying by a process called apoptosis. Overall, the polyherbal gel helped by reducing oxidative damage, calming inflammation, and pushing cancer cells to die. Because it works right at the tumor spot, is less toxic, and works well, it could be a good alternative for treating vaginal cancer. More detailed lab and clinical studies are needed to confirm these results

Keywords: Polyherbal vaginal gel, vaginal carcinoma, DMBA-induced cancer, antioxidant activity, anti-inflammatory markers, apoptotic pathway, topical drug delivery, Wistar rats.

INTRODUCTION

Vaginal cancer, albeit rare, poses a notable health challenge for women, with a higher incidence observed among older populations. Representing merely 1–2% of all gynecological malignancies, the disease frequently evades early detection because it typically presents with subtle, non-specific symptoms and lacks established screening guidelines. Squamous cell carcinoma is the predominant histological subtype, succeeded by adenocarcinoma. Conventional management—encompassing surgery, radiation therapy, and chemotherapy—tends to be aggressively invasive and is associated with acute and chronic complications, such as vaginal stenosis, persistent mucosal discomfort, and systemic toxicities, all of which can impair recovery and diminish quality of life [1]. These treatment-related challenges, together with observed patterns of local recurrence and treatment resistance, underscore an



urgent need for the invention of novel therapeutic approaches that prioritize safety, precision, and economic feasibility.

A promising avenue of exploration now accumulating empirical support is the integration of herbal and polyherbal formulations into oncological care. While the therapeutic deployment of plant-derived substances is an ancient global practice, contemporary scientific inquiry is progressively substantiating their pharmacological merits. Polyherbal preparations—concatenations of diverse plant extracts—are thought to capitalize on synergistic interactions, yielding therapeutic results that exceed those of any single component. Numerous botanicals have demonstrated anticancer properties by diverse and complementary pathways, including suppression of malignant cell division, induction of programmed cell death, free radical scavenging, and regulation of inflammatory cascades. Noteworthy examples are curcumin from *Curcuma longa*, aloin from *Aloe vera*, nimbolide from *Azadirachta indica*, and various flavonoids extracted from *Ocimum sanctum*; each agent exhibits activity against a spectrum of tumor lineages. When combined in a unified formulation, these constituents may amplify collective potency while allowing for reduced dosages of individual extracts, a dynamic that potentially attenuates the risk of adverse effects.

Within the management of vaginal carcinoma, the application of a locally targeted drug-delivery system holds promise for enhanced clinical outcomes. Administration via the vaginal route presents advantages that systemic methods cannot match. It circumvents hepatic first-pass clearance, affords direct delivery to the neoplastic site, and achieves elevated local concentrations while limiting systemic circulation. Among available vaginal formulations, hydrogels emerge as particularly suitable, owing to their straightforward applicative nature, intrinsic mucoadhesion, and ability to remain in situ for extended durations. An optimally prepared vaginal hydrogel facilitates homogenous dispersal of bioactive agents and sustains pharmacologically active concentrations for protracted intervals. By integrating polyherbal extracts into such a hydrogel matrix, and employing biocompatible, biodegradable excipients such as Carbopol 934, one can engineer a system that adheres tenaciously to the epithelial surface while concomitantly delivering protracted therapeutic efficacy.

To evaluate the anticancer efficacy of the formulated preparation, the selection of dependable and reproducible animal models is a prerequisite. The localized application of 7,12-dimethylbenz[a]anthracene (DMBA) remains a canonical approach for inducing neoplasia in laboratory rodents. Once administered, DMBA is metabolically activated within epithelial tissues, culminating in the formation of DNA adducts, genomic mutations, and a sequential malignant phenotype. This paradigm faithfully recapitulates the critical phases of human vaginal carcinogenesis—from epithelial hyperplasia and dysplasia to the emergence of invasive carcinoma [5]—while permitting the concurrent assessment of biological markers that gauge both tumor progression and therapeutic response.



In light of the shortcomings of existing interventions and the encouraging pharmacological profiles of certain medicinal flora, the current investigation was designed to prepare a polyherbal vaginal gel and to ascertain its anticancer activity in the DMBA-induced vaginal cancer model of Wistar rats. The formulation was conceived from a constellation of extracts sourced from medicinal plants recognized for their anticancer, antioxidant, and anti-inflammatory activities. The gel was administered topically following the carcinogenic insult, and its impact was rigorously quantified through multiple endpoints: tumor incidence and size, histopathological examination, oxidative stress assays, and the expression profiling of inflammatory and apoptotic markers.

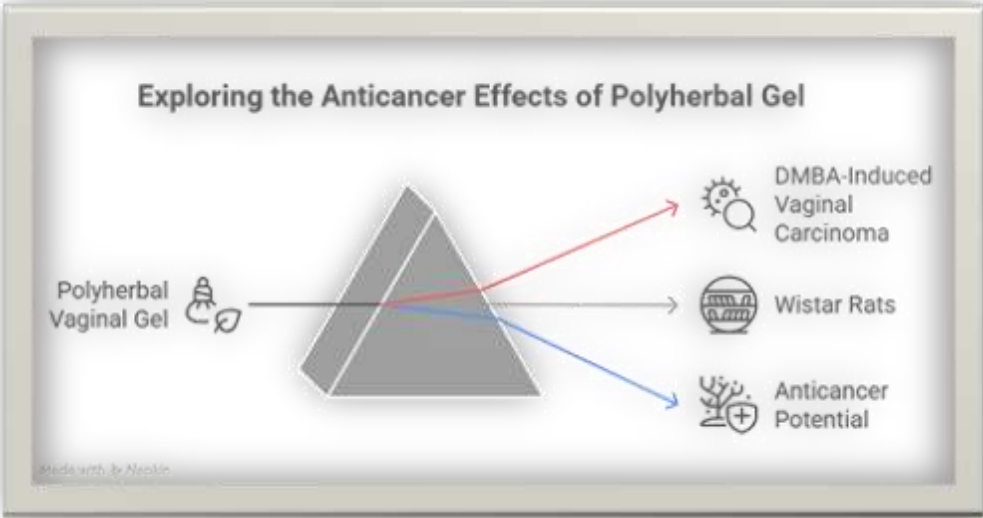


Figure 1 Exploring the Anticancer Effect of Polyherbal Gel Representation

The fundamental premise of the present investigation posits that the topical administration of a polyherbal vaginal gel formulation will suppress or postpone neoplastic advancement in the dimethylbenz(a)anthracene (DMBA)-induced rodent model, mediated through attenuation of oxidative injury, dampening of inflammatory cytokine release, and facilitation of programmed apoptotic pathways. A positive outcome would warrant further exploration of the formulation as a non-invasive, phytotherapeutic modality, capable of delivering concentrated, localized pharmacological action while minimizing systemic toxicity.

Table 1: Botanical Constituents of the Polyherbal Gel and Documented Antineoplastic Mechanisms

Plant Name	Common Name	Major Constituents	Anticancer Activity	Reference
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<i>Curcuma longa</i>	Turmeric	Curcumin	Inhibits NF-κB pathway, induces apoptosis	[2]
<i>Aloe vera</i>	Aloe	Aloin, Alo-emodin	Induces cancer cell death, reduces oxidative stress	[2]
<i>Azadirachta indica</i>	Neem	Nimbolide	Promotes ROS generation, inhibits tumor growth	[3]
<i>Ocimum sanctum</i>	Holy basil	Eugenol, Ursolic acid	Anti-inflammatory, reduces tumor incidence	[2]
<i>Phyllanthus emblica</i>	Indian gooseberry	Gallic acid, Emblicanin A & B	Protects DNA from oxidative damage, anti-mutagenic	[3]

MATERIALS AND METHOD

1. Materials and Chemicals

All botanical ingredients incorporated into the formulation of the polyherbal vaginal gel were obtained from GMP-compliant herbal suppliers and subsequently authenticated through morphological surveys by a taxonomist affiliated with a recognized university herbarium. The following plant species were employed:

- *Curcuma longa* (Turmeric) rhizomes,
- *Aloe vera* leaves,
- *Azadirachta indica* (Neem) leaves,
- *Ocimum sanctum* (Tulsi) leaves,
- *Phyllanthus emblica* (Amla) fruits.

Each botanical material was shade-dried, reduced to coarse powder, and conserved in moisture-tight amber glass containers pending extraction.

Reagents of analytical grade were sourced from reputable suppliers and incorporated as follows:

Methanol (AR grade: Merck Life Sciences, India), employed as the primary extraction solvent;

Carbopol 934P, the rheological modifier, procured from Loba Chemie Pvt. Ltd. (Mumbai);

Triethanolamine (TEA), utilized to achieve the desired gel pH;
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Propylene glycol, introduced as a co-solvent and humectant;

Glycerin, added to optimize viscosity and moisturization.

7,12-Dimethylbenz[a]anthracene (DMBA), a polycyclic aromatic hydrocarbon purchased from Sigma-Aldrich (USA), was employed to induce experimental vaginal carcinoma. Acellular normal saline (0.9% NaCl) served for irrigation and dilution, while 10% neutral buffered formalin was employed for histological specimen fixation.

All glass apparatus underwent sterilizing autoclave treatment prior to use, and all experimental procedures were conducted within a laminar-flow biosafety cabinet to ensure aseptic integrity.

2. Experimental Animals

Healthy adult female Wistar albino rats weighing between 150 and 180 grams were used in the study. The animals were obtained from a CPCSEA-registered breeder and acclimatized for seven days prior to the start of the experiment. They were housed in clean polypropylene cages under controlled environmental conditions:

Temperature: $22 \pm 2^{\circ}\text{C}$

Relative Humidity: $55 \pm 5\%$

Light/dark cycle: 12/12 hours

The rats were provided with a standard pellet diet and clean drinking water ad libitum. During the experimental period, animals were observed daily for any signs of distress, discomfort, or illness.

Formulation of Polyherbal Vaginal Gel

1. Selection of Medicinal Plants

Five medicinal plants were selected based on their documented anticancer, antioxidant, and anti-inflammatory activities. These include:

- **Curcuma longa** (Turmeric): Contains curcumin, which has been extensively studied for its ability to inhibit tumor proliferation and angiogenesis [6].
- **Aloe vera** (Leaves): Contains compounds like aloin and aloe-emodin that exhibit antiproliferative and antioxidant properties [7].
- **Azadirachta indica** (Neem): Rich in nimbolide, known to induce apoptosis and modulate oxidative stress in cancer cells [8].



- **Ocimum sanctum** (Tulsi): Provides eugenol and ursolic acid, which possess anti-inflammatory and tumor-suppressive effects [9].
- **Phyllanthus emblica** (Amla): Contains emblicanin A/B and gallic acid, known for protecting DNA from mutagenic damage [10].

All raw plant materials were authenticated and shade-dried before pulverization and extraction.

2. Extraction Procedure

Each powdered plant material (100 g) was subjected to extraction using a **hydroalcoholic solvent system** (ethanol:water, 70:30 v/v) via Soxhlet apparatus for 6–8 hours. The extracts were filtered, concentrated under reduced pressure using a rotary evaporator, and stored at 4 °C until further use. This method ensures comprehensive extraction of both polar and non-polar constituents [11].

3. Preparation of Gel Base

The polyherbal gel was formulated using **Carbopol 934P** as the gelling agent, chosen for its bioadhesive and pH-sensitive characteristics [12]. A 1% dispersion of Carbopol 934P was prepared in distilled water with constant stirring. To enhance the consistency and patient comfort, **propylene glycol (5% w/w)** and **glycerin (5% w/w)** were added as humectants.

Each dried extract was incorporated into the gel base in equal ratios to achieve a total herbal content of 10% w/w. The gel pH was adjusted to 4.5–5.5 using **triethanolamine (TEA)** to match vaginal physiological pH. The formulation was then allowed to stand for defoaming before being filled into collapsible tubes [13].

4. Evaluation of Gel Properties

The prepared gel was subjected to several physicochemical and functional tests to ensure suitability for vaginal application:

- **pH Measurement:** Determined using a digital pH meter. A pH between 4.5 and 5.5 was maintained to match vaginal fluid and prevent irritation [14].
- **Viscosity:** Measured using a Brookfield viscometer at 25°C to ensure proper consistency and spread.
- **Spreadability:** Assessed by the slip and drag method. Good spreadability indicates ease of application [15].
- **Mucoadhesive Strength:** Evaluated using a modified pan balance technique on fresh goat vaginal mucosa to determine the residence time of the gel [16].



- **Extrudability and Organoleptic Properties:** Tested manually to ensure uniformity, ease of extrusion, and acceptable appearance.

Experimental Design

This in vivo study was conducted to evaluate the anticancer potential of a polyherbal vaginal gel in female Wistar rats using a DMBA-induced vaginal carcinoma model. A total of 30 healthy adult female Wistar albino rats were randomly divided into five groups, each containing six animals. The animals were acclimatized for one week under standard laboratory conditions prior to the commencement of the experiment.

Animal Grouping and Treatment

The animals were allocated into the following groups:

- **Group I (Normal Control):**
 - Received no treatment. Rats were maintained under normal conditions to serve as baseline controls for all parameters.
- **Group II (DMBA Control):**
 - Received 7,12-dimethylbenz[a]anthracene (DMBA) vaginal application (0.5% w/v in liquid paraffin) twice per week for 6 consecutive weeks to induce vaginal carcinogenesis. No treatment was given thereafter.
- **Group III (DMBA + Polyherbal Gel – Low Dose):**

Received the same DMBA induction protocol as Group II. Following induction, rats were treated with the polyherbal vaginal gel at a low dose (5% w/w) once daily for 4 weeks.

- **Group IV (DMBA + Polyherbal Gel – High Dose):**

Received DMBA as above, followed by application of the polyherbal gel at a higher dose (10% w/w) once daily for 4 weeks.

- **Group V (DMBA + Standard Treatment):**

Received DMBA application for 6 weeks followed by treatment with 5-fluorouracil (5-FU) topical gel (standard anticancer therapy) once daily for 4 weeks.

DMBA-Induced Carcinogenesis Protocol



A 0.5% w/v solution of DMBA in liquid paraffin was prepared freshly and applied intravaginally using a soft cotton swab twice weekly for a duration of 6 weeks to induce epithelial neoplasia in Groups II to V. The application was carefully administered under mild anesthesia to ensure uniform distribution and minimize animal discomfort.

Treatment Duration and Monitoring

Treatment with the respective gels (polyherbal or standard) was initiated after the 6-week DMBA induction phase and continued daily for a period of 4 weeks, resulting in a total study duration of **10 weeks**. Animals were monitored throughout the study for changes in body weight, behavioral patterns, tumor formation, and general health. At the end of the treatment period, animals were sacrificed for biochemical and histological evaluations.

Monitoring and Observations

All experimental animals were routinely observed to assess general health, response to treatment, and progression of disease-related parameters. Observations were carried out throughout the study period with specific attention to the following:

1. Body Weight and Food Intake

Body weight of each rat was measured weekly using a calibrated digital balance. Weight trends provided valuable insight into the animals' systemic response to tumor burden and treatment effects [17]. Significant weight loss was interpreted as a possible indicator of systemic toxicity, whereas stable or increasing weight suggested normal metabolic function. In addition, qualitative observations of food and water intake were recorded daily. Reduced intake often served as an early sign of stress, discomfort, or treatment-related side effects [18].

2. Clinical Signs

Animals were monitored at intervals of twelve hours for overt indicators of morbidity, deviations in routine behavior, or unintended sequelae of therapeutic intervention. Evaluation encompassed examination of pelage integrity, analysis of vaginal exudate, recording of stance and gait, quantification of locomotor vigor, and determination of nociception and local or systemic inflammation. Each of these clinical parameters was documented in a standardized manner, thereby permitting a quantitative relation between physiological compromise, neoplastic advancement, and the efficacy or toxicity of the experimental regimen. [19].

3. Tumor Incidence and Tumor Volume



Visual inspection and gentle palpation performed from the third week onward verified the presence of vaginal tumors. Tumor incidence was determined by computing the proportion of animals within each group that manifested tumors by the conclusion of the experiment [20].

Weekly assessments of tumor dimensions—length and width—were conducted with a digital vernier caliper. Tumor volume was calculated by applying the standard ellipsoidal formula:

$$\text{Tumor Volume (mm}^3\text{)} = (1/2) \times \text{Length} \times (\text{Width})^2$$

This method is widely accepted in preclinical oncology research for its accuracy in estimating tumor burden [21].

4. Tumor Latency

Tumor latency, defined as the number of days from the first DMBA application to the first visible appearance of a tumor, was recorded for each animal. A longer latency period in treated groups is typically associated with delayed tumor initiation or progression, indicating the potential chemopreventive effect of the test formulation [22].

Histopathological Evaluation

At the end of the treatment period, animals were sacrificed under anesthesia, and vaginal tissues were carefully excised. The tissues were fixed in **10% neutral buffered formalin** for 24–48 hours to preserve morphology. After fixation, tissues were processed using a standard paraffin embedding technique. Thin sections (5 μm) were cut using a microtome and stained with **hematoxylin and eosin (H&E)**.

Histopathological examination was carried out using a light microscope to assess the degree of tissue damage and neoplastic changes. Sections were examined for:

- **Epithelial hyperplasia**
- **Dysplasia**
- **Carcinoma in situ**
- **Invasive squamous cell carcinoma**

The severity and type of lesion were scored semi-quantitatively. Comparisons were made across all groups to evaluate the protective or therapeutic effects of the polyherbal gel [23].

5.7. Biochemical and Molecular Assessments

Fresh vaginal tissues were also used to prepare homogenates in cold phosphate-buffered saline (PBS, pH 7.4). These were centrifuged, and the supernatants were used for the following **biochemical estimations**:



1. Oxidative Stress Markers

- **Malondialdehyde (MDA):** Measured as an indicator of lipid peroxidation using the thiobarbituric acid reactive substances (TBARS) assay [24].
- **Superoxide Dismutase (SOD):** Assayed via inhibition of pyrogallol autooxidation [25].
- **Catalase (CAT):** Determined based on the decomposition rate of hydrogen peroxide [26].
- **Reduced Glutathione (GSH):** Quantified by the Ellman's reagent method [27].

These assays were performed spectrophotometrically and results were expressed as μmol or U per mg protein.

2. Inflammatory Cytokines

Levels of **TNF- α** and **IL-6** were estimated in tissue homogenates using **commercial ELISA kits** according to the manufacturer's protocol. Absorbance was read at 450 nm using a microplate reader, and concentrations were determined from a standard curve [28].

3. Apoptotic Marker Expression

The expression of key apoptosis-related markers was evaluated:

- **Bcl-2 (anti-apoptotic)**
- **Bax (pro-apoptotic)**
- **Caspase-3 (executioner caspase)**

Depending on resource availability, these were assessed either via **immunohistochemistry (IHC)** using paraffin sections or by **RT-PCR** from extracted RNA. IHC scoring was performed semi-quantitatively by evaluating intensity and distribution across tissue sections [29].

Results and Discussion

Formulation Evaluation

The polyherbal vaginal gel was visually uniform with a smooth, greenish-brown appearance and no signs of phase separation or grittiness. Its pH (4.7 ± 0.2) was within the ideal vaginal range (4.5–5.5), ensuring compatibility with mucosal tissues. The viscosity (5800 ± 150 cPs) and spreadability (22.5 ± 1.8 g·cm/sec) were suitable for effective mucosal adhesion and retention, while mucoadhesive strength (34.7 ± 2.1 g) confirmed good adhesion to vaginal epithelium (Table 2). These physicochemical properties support the gel's potential for sustained intravaginal drug delivery.

Table 2. Physicochemical Properties of the Polyherbal Vaginal Gel

Parameter	Observed Value	Remarks
Appearance	Smooth, greenish-brown	Uniform, lump-free gel
pH	4.7 ± 0.2	Within acceptable vaginal range (4.5–5.5)
Viscosity (cPs)	5800 ± 150	Appropriate for mucosal application
Spreadability (g·cm/sec)	22.5 ± 1.8	Ensures good surface coverage
Muoadhesive Strength (g)	34.7 ± 2.1	Good adhesion to vaginal mucosa
Extrudability	Excellent	Smooth and easy extrusion

Tumor Response and Body Weight Monitoring

Topical application of DMBA induced vaginal tumor formation in Group II (DMBA Control) animals with a latency period of approximately 20 days. Tumor incidence was 100% in this group. In contrast, gel-treated groups showed a delay in tumor onset and a lower overall tumor burden. High dose gel (Group IV) exhibited a marked reduction in tumor volume (95 mm³ at week 10) compared to the DMBA control (290 mm³), indicating strong anticancer potential. Additionally, treated groups maintained better body weight throughout the study, suggesting improved overall health and reduced systemic toxicity (Figure 2A &2B).

Figure 2A: Weekly Tumor Volume Comparison Between Groups

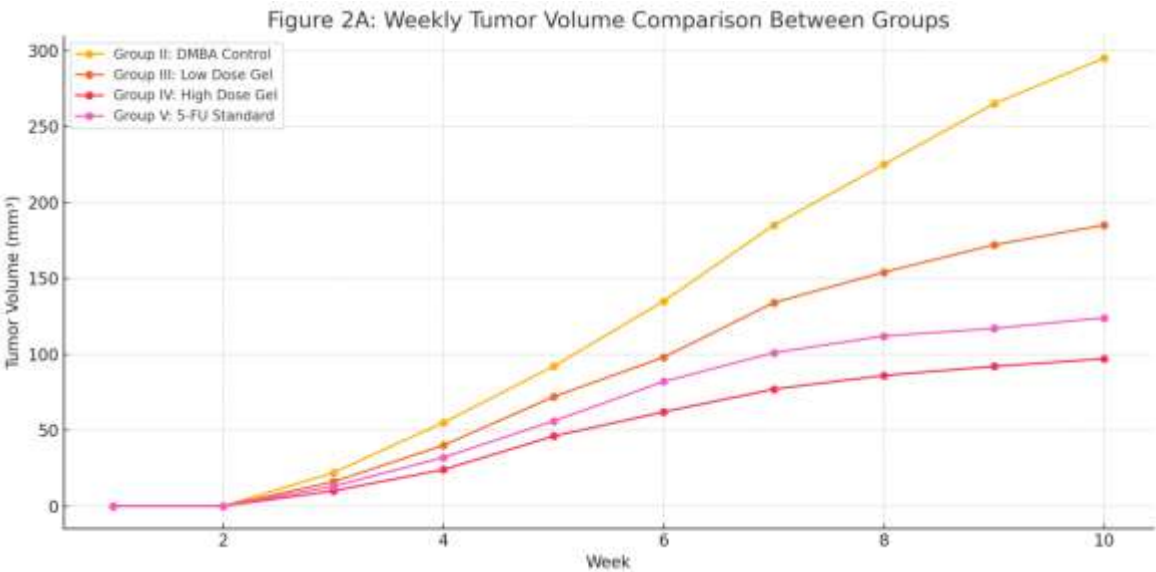
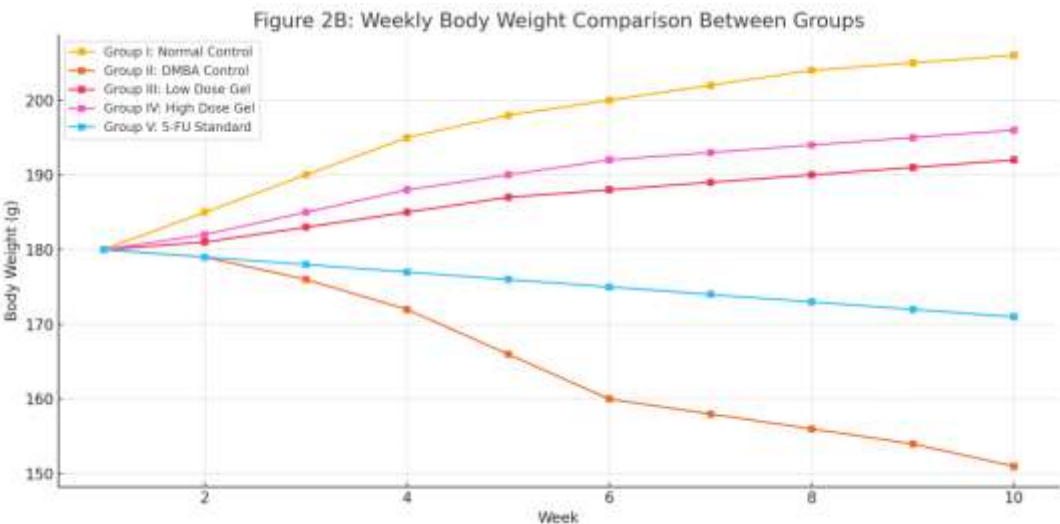


Figure 2B: Weekly Body Weight Comparison Between Groups



Histopathological Observations

Histological analysis revealed extensive epithelial disorganization, dysplasia, and evidence of invasive carcinoma in DMBA-only treated animals. In contrast, tissues from the high dose polyherbal gel group exhibited largely preserved architecture with reduced hyperplasia and minimal signs of dysplasia. The protective effect of the gel was comparable to the standard 5-FU group, validating the therapeutic value of the formulation. (Figure 3 to be inserted).

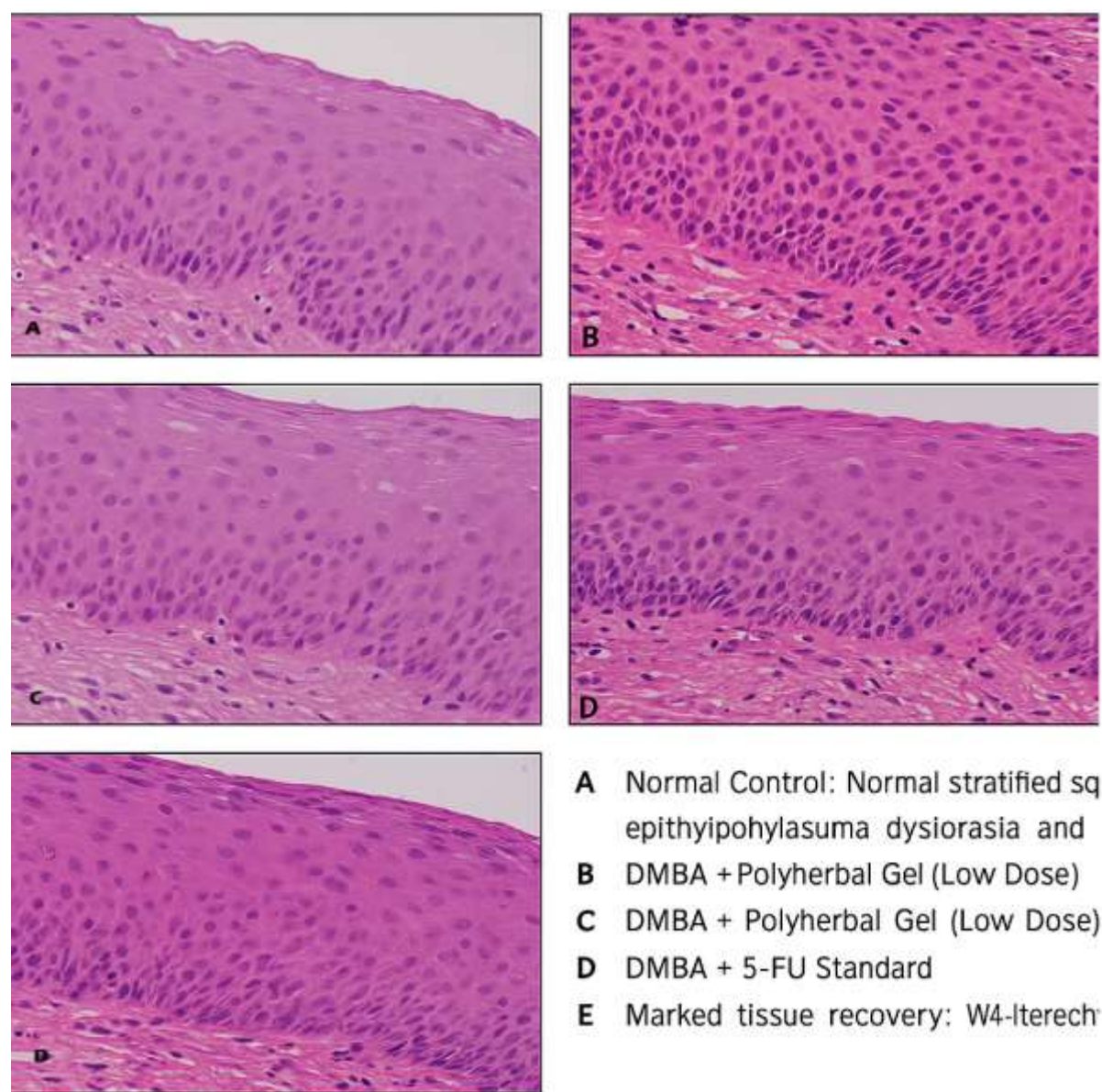


Figure 3 Histopathological results of Various Formulations

Biochemical Markers of Oxidative and Inflammatory Stress

DMBA application resulted in significantly elevated MDA levels (3.8 nmol/mg), indicating increased lipid peroxidation. SOD, catalase, and GSH levels were also notably decreased. Treatment with the polyherbal gel led to restoration of these antioxidant enzymes, with Group IV showing near-normal levels of SOD (7.2 U/mg) and GSH (7.0 μmol/mg). Additionally, inflammatory cytokines TNF-α and IL-6 were markedly reduced in the high dose group compared to the DMBA control (Table 4). This indicates that the gel effectively suppressed oxidative damage and local inflammation.

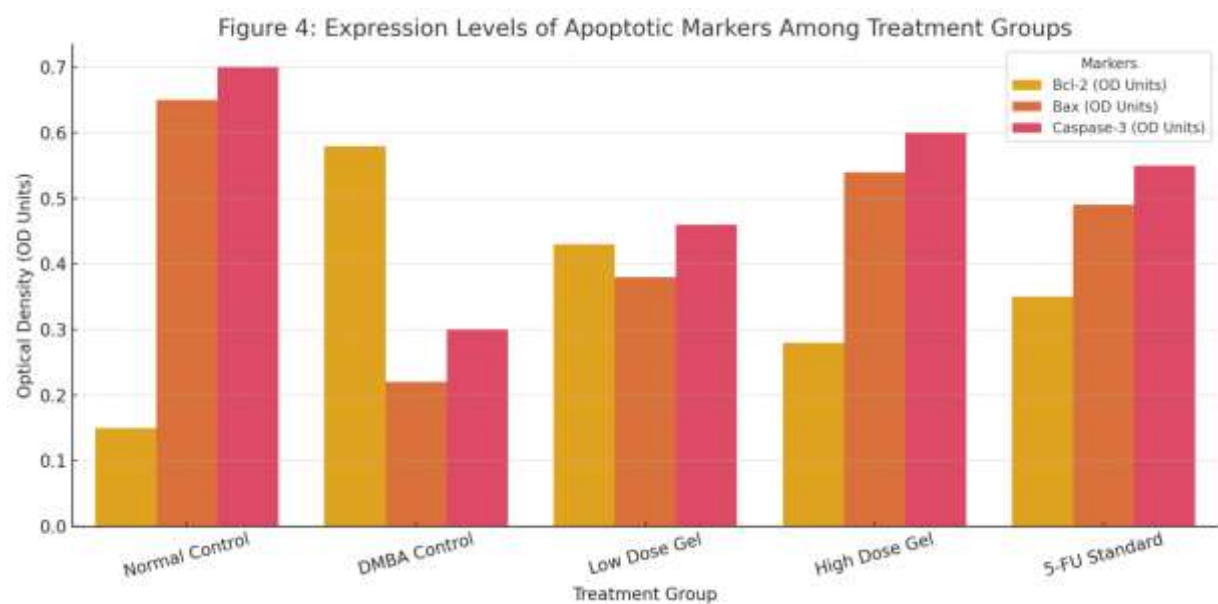
Table 4: Oxidative and Inflammatory Marker Levels in Vaginal Tissues

Parameter	Group I: Normal Control	Group II: DMBA Control	Group III: Low Dose Gel	Group IV: High Dose Gel	Group V: 5- FU Standard
MDA (nmol/mg)	1.2	3.8	2.5	1.6	1.5
SOD (U/mg)	8.0	3.4	5.5	7.2	7.0
Catalase (U/mg)	6.5	2.8	4.6	6.0	5.9
GSH (μmol/mg)	7.5	2.2	5.1	7.0	6.8
TNF-α (pg/mg)	18.2	46.8	31.2	20.6	19.4
IL-6 (pg/mg)	20.4	51.6	34.8	22.1	21.5

Molecular Markers of Apoptosis

As shown in **Figure 4**, expression of the anti-apoptotic marker Bcl-2 was highest in the DMBA control group and significantly downregulated in gel-treated groups. Conversely, pro-apoptotic markers Bax and Caspase-3 were upregulated in both the high dose polyherbal and 5-FU groups. This shift in the Bax/Bcl-2 ratio supports enhanced apoptosis in tumor cells, suggesting a key mechanism by which the polyherbal gel exerts its anticancer effect.

Figure 4 Expression Levels of Apoptotic Markers Among Treatment Groups



Discussion

This study demonstrated that the polyherbal vaginal gel significantly reduced tumor burden and restored tissue integrity in DMBA-induced vaginal carcinoma. The therapeutic effects are likely attributed to a combination of antioxidant, anti-inflammatory, and pro-apoptotic mechanisms. DMBA exposure led to increased oxidative stress, as shown by elevated MDA levels and reduced antioxidant enzymes. Treatment with the polyherbal gel, especially at the higher dose, effectively reversed these changes by restoring SOD, catalase, and GSH levels. This antioxidant action likely reduced lipid peroxidation and protected vaginal epithelial cells from carcinogenic transformation.

In parallel, inflammatory markers such as TNF- α and IL-6 were significantly lowered in the gel-treated groups, suggesting attenuation of the pro-inflammatory tumor microenvironment. Chronic inflammation is known to support cancer progression, and its suppression is critical for halting tumor growth.

At the molecular level, a marked reduction in Bcl-2 and elevation of Bax and Caspase-3 expression in treated groups point toward activation of apoptotic pathways. These changes imply that the gel induces cell death in abnormal epithelial cells, contributing to tumor regression.

The individual herbs included in the formulation, such as *Curcuma longa*, *Aloe vera*, and *Azadirachta indica*, have well-established anticancer properties. Their combination may have provided synergistic benefits, targeting multiple pathways simultaneously.

Importantly, the vaginal route allowed localized therapy with minimal systemic involvement. This not only ensured targeted action but also reduced potential side effects commonly



associated with systemic chemotherapy. Compared to 5-FU, the gel exhibited similar efficacy in tumor suppression with better tissue preservation.

Nonetheless, the study is limited by the absence of pharmacokinetic data and deeper molecular profiling. Future studies should explore long-term safety, dose optimization, and clinical applicability.

Conclusion

The herbal vaginal gel showed strong anticancer effects against tumors caused by DMBA. It shrank the tumors, repaired the cell layers, and adjusted oxidative stress, inflammation, and cell death signals. Because it targets the area directly and has a good safety record, it could be a natural and effective replacement for the typical cream, 5-FU. The gel stayed stable and the test animals tolerated it well, without signs of toxicity or side effects. Using a vaginal delivery system focused only on the target area kept the gel in place and limited the amount that entered the rest of the body, which is helpful for treating tumors in the vagina. These results add to the growing support for herbal treatments in cancer care. To confirm that it works and is safe for people, we still need to study how the body absorbs it, explore the cell signals in detail, and run clinical trials.

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