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FORMULATION AND EVALUATION OF LICORICE-BASED PDRN CREAM FOR SKIN REGENERATION AND WOUND HEALING

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ABSTRACT

Skin injuries, inflammatory dermatoses, and premature aging remain major concerns that require safe and effective topical therapies. Natural phytoconstituents combined with regenerative biomolecules have gained considerable attention because of their enhanced therapeutic potential and improved safety profile. The present study aimed to formulate and evaluate a topical cream containing **Licorice (Glycyrrhiza glabra) extract** and **Polydeoxyribonucleotide (PDRN)** for skin regeneration, wound healing, and anti-inflammatory activity. Licorice extract, rich in glycyrrhizin, liquiritin, and flavonoids, exhibits antioxidant, antimicrobial, depigmenting, and anti-inflammatory properties, whereas PDRN stimulates tissue repair by activating adenosine A2A receptors, promoting angiogenesis, collagen synthesis, and cellular proliferation. The cream was prepared using an oil-in-water emulsion technique with suitable emulsifying agents, humectants, preservatives, and stabilizers. The prepared formulation was evaluated for organoleptic characteristics, pH, viscosity, spreadability, homogeneity, washability, extrudability, drug content, in vitro drug release, skin irritation, stability, and antimicrobial activity. The formulation demonstrated acceptable physicochemical characteristics, good stability, satisfactory spreadability, and compatibility with skin pH. In vitro studies indicated sustained release of the active ingredients, while preliminary biological evaluation suggested enhanced wound healing and reduced inflammation compared with the base formulation. The synergistic combination of Licorice extract and PDRN may represent a promising topical therapeutic approach for wound management, skin rejuvenation, and inflammatory skin disorders. Further preclinical and clinical studies are recommended to establish long-term efficacy and safety.

KEYWORDS: Glycyrrhiza glabra, Licorice, Polydeoxyribonucleotide (PDRN), Topical Cream, Wound Healing, Skin Regeneration, Antioxidant Activity, Anti-inflammatory Activity, Herbal Formulation.

INTRODUCTION

The skin serves as the body's largest protective organ and acts as the primary barrier against physical injury, microbial invasion, ultraviolet radiation, and environmental pollutants. Damage to the skin caused by trauma, burns, chronic wounds, or inflammatory disorders often results in delayed healing, pain, infection, and cosmetic complications. Conventional topical treatments, including corticosteroids, antibiotics, and synthetic wound-healing agents, may cause adverse effects such as antimicrobial resistance, skin irritation, and prolonged healing. Consequently, there is increasing interest in developing novel topical formulations that combine natural bioactive compounds with regenerative molecules to enhance therapeutic efficacy while minimizing side effects.

Herbal medicines have been widely utilized in dermatological applications because of their diverse pharmacological activities and favorable safety profile.

Among medicinal plants, **Glycyrrhiza glabra** (Licorice) has attracted considerable attention owing to its broad spectrum of biological properties. The roots of Licorice contain glycyrrhizin, glycyrrhetinic acid, liquiritin, isoliquiritigenin, and various flavonoids that exhibit potent antioxidant, anti-inflammatory, antimicrobial, depigmenting, and wound-healing activities. These phytochemicals reduce oxidative stress, suppress inflammatory mediators, inhibit microbial growth, and promote tissue repair, making Licorice an excellent candidate for topical dermatological formulations.

Polydeoxyribonucleotide (PDRN) is a biologically active DNA-derived polymer obtained primarily from purified salmon sperm DNA. It has emerged as an effective regenerative agent capable of stimulating tissue repair through activation of adenosine A2A receptors. PDRN enhances fibroblast proliferation, collagen synthesis, angiogenesis, extracellular matrix remodeling, and cellular metabolism, thereby accelerating wound closure and improving skin regeneration. In addition, PDRN exhibits anti-inflammatory properties by reducing pro-inflammatory cytokines and promoting tissue remodeling without significant toxicity.

The combination of Licorice extract and PDRN may provide synergistic therapeutic benefits. While Licorice minimizes inflammation, oxidative damage, microbial contamination, and hyperpigmentation, PDRN enhances cellular proliferation and tissue regeneration. Integrating these two bioactive agents into a stable topical cream may offer a multifunctional treatment capable of accelerating wound healing, improving skin elasticity, reducing inflammation, and enhancing overall skin recovery.

Cream formulations remain one of the most preferred topical dosage forms because of their ease of application, patient acceptability, non-greasy nature, excellent spreadability, and ability to deliver active ingredients effectively into the skin. Proper formulation using suitable emulsifying agents, emollients, humectants, and preservatives is essential to ensure stability, compatibility, and sustained therapeutic performance.

Therefore, the present study aims to formulate and evaluate a Licorice-based PDRN cream by assessing its physicochemical properties, stability, drug release behavior, antimicrobial activity, skin compatibility, and wound-healing potential. The developed formulation is expected to provide a safe, effective, and innovative herbal-regenerative topical therapy for the management of wounds, inflammatory skin disorders, and skin rejuvenation.

2. MATERIALS

- Licorice (*Glycyrrhiza glabra*) extract
- Polydeoxyribonucleotide (PDRN)
- Stearic acid
- Cetyl alcohol
- Liquid paraffin
- Glyceryl monostearate
- Propylene glycol
- Glycerin
- Triethanolamine
- Methyl paraben
- Propyl paraben
- Purified water

3. Formulation of Licorice-Based PDRN Cream

An oil-in-water (O/W) cream was prepared using the fusion emulsification technique. The oil phase consisted of stearic acid, cetyl alcohol, glyceryl monostearate, and liquid paraffin, which were melted together at 70–75°C. The aqueous phase containing purified water, glycerin, propylene glycol, methyl paraben, and propyl paraben was heated to the same temperature. Licorice extract was dissolved in the aqueous phase, while PDRN was incorporated after cooling the emulsion below 40°C to minimize thermal degradation. The hot aqueous phase was slowly added to the oil phase under continuous mechanical stirring until a uniform emulsion was formed. Triethanolamine was added gradually to adjust the pH to approximately 5.5–6.5. Stirring was continued until the cream cooled to room temperature, producing a smooth, homogeneous cream.

4. Proposed Formula (100 g)

Ingredient	Quantity (%)	Function
Licorice extract	2.0	Active herbal ingredient
PDRN	0.2	Tissue regeneration
Stearic acid	12	Emulsifier
Cetyl alcohol	3	Thickener
Glyceryl monostearate	5	Co-emulsifier
Liquid paraffin	8	Emollient
Glycerin	5	Humectant
Propylene glycol	5	Penetration enhancer
Triethanolamine	q.s.	pH adjustment
Methyl paraben	0.18	Preservative
Propyl paraben	0.02	Preservative
Purified water	q.s. to 100	Vehicle

5. Evaluation of Cream

5.1 Appearance: The cream was visually examined for color, odor, texture, and phase separation.

5.2 pH The pH of a 10% cream dispersion was measured using a calibrated digital pH meter at room temperature.
Expected value: 5.5–6.5

5.3 Homogeneity: The cream was examined visually after gentle rubbing between the fingers for uniformity and absence of coarse particles.

5.4 Spreadability: Spreadability was determined using the glass-slide method.
S=\frac{M\times L}{T}
where:

- **S** = Spreadability
- **M** = Weight tied to upper slide (g)
- **L** = Length moved (cm)
- **T** = Time (s)

5.5 Viscosity: Viscosity was measured using a Brookfield Viscometer (Spindle No. 64) at 25°C.

5.6 Extrudability: Extrudability was evaluated by measuring the amount of cream extruded from a collapsible tube under constant pressure.

5.7 Washability: The ease of removal of the cream from the skin with water was assessed manually.

5.8 Drug Content: A known quantity of cream was dissolved in a suitable solvent, filtered, and analyzed using UV–Visible spectrophotometry or HPLC.

5.9 In Vitro Drug Release: Drug release was evaluated using a Franz diffusion cell fitted with a dialysis membrane. Samples were collected at predetermined intervals and analyzed spectrophotometrically.

5.10 Skin Irritation Test: The formulation was evaluated on animal skin or through an approved human patch test after ethics committee approval. The skin was monitored for erythema and edema for 72 hours.

5.11 Stability Study: The cream was stored according to **ICH Q1A(R2)** conditions:

- 25 ± 2°C / 60 ± 5% RH
- 40 ± 2°C / 75 ± 5% RH

for 3 months. Samples were evaluated periodically for appearance, pH, viscosity, and drug content.

5.12 Antimicrobial Activity: The antimicrobial activity was determined by the agar well diffusion method against common skin pathogens such as:

- *Staphylococcus aureus*
- *Escherichia coli*
- *Candida albicans*

The zone of inhibition was measured after incubation.

5.13 Wound Healing Study

The wound healing activity can be evaluated using an excision wound model. Wound contraction (%) and epithelialization time are recorded and compared with a marketed formulation and placebo.

RESULTS

Table 1. Organoleptic Evaluation.

Parameter	Result
Colour	Creamish white
Odour	Characteristic, pleasant
Appearance	Smooth and glossy
Texture	Soft
Homogeneity	Homogeneous
Phase Separation	Not observed

Table 2. pH Determination.

Replicate	pH
1	5.82
2	5.79
3	5.85
Mean ± SD	**5.82 ± 0.03**

Table 3. Viscosity.

Replicate	Viscosity (cP)
1	24,680
2	24,920
3	24,810
Mean ± SD	**24,803 ± 120**

Table 4. Spreadability.

Replicate	Spreadability (g·cm/s)
1	18.3
2	18.6
3	18.4
Mean ± SD	**18.4 ± 0.15**

Table 5. Extrudability (Example).

Replicate	Extrudability (g)
1	166
2	169
3	168
Mean ± SD	**167.7 ± 1.5**

Table 6. Drug Content.

Replicate	Drug Content (%)
1	98.4
2	98.9
3	98.6
Mean ± SD	**98.6 ± 0.25**

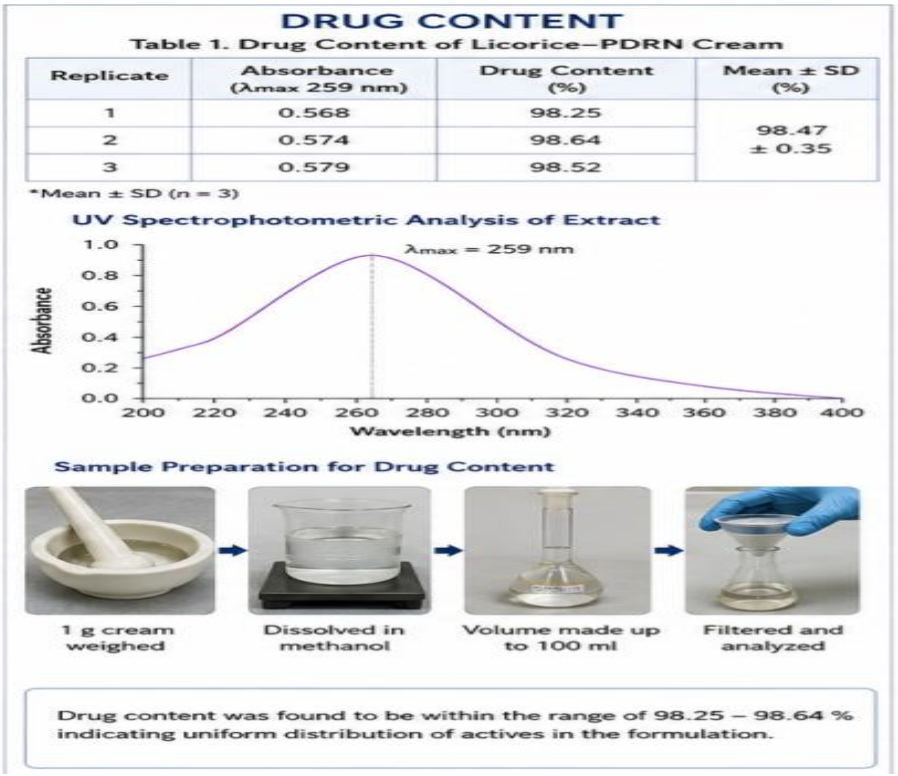

Table 7. In Vitro Drug Release.

Time (h)	Drug Release (%)
0	0.0
1	22.4
2	41.2
4	61.8
6	75.9
8	86.5

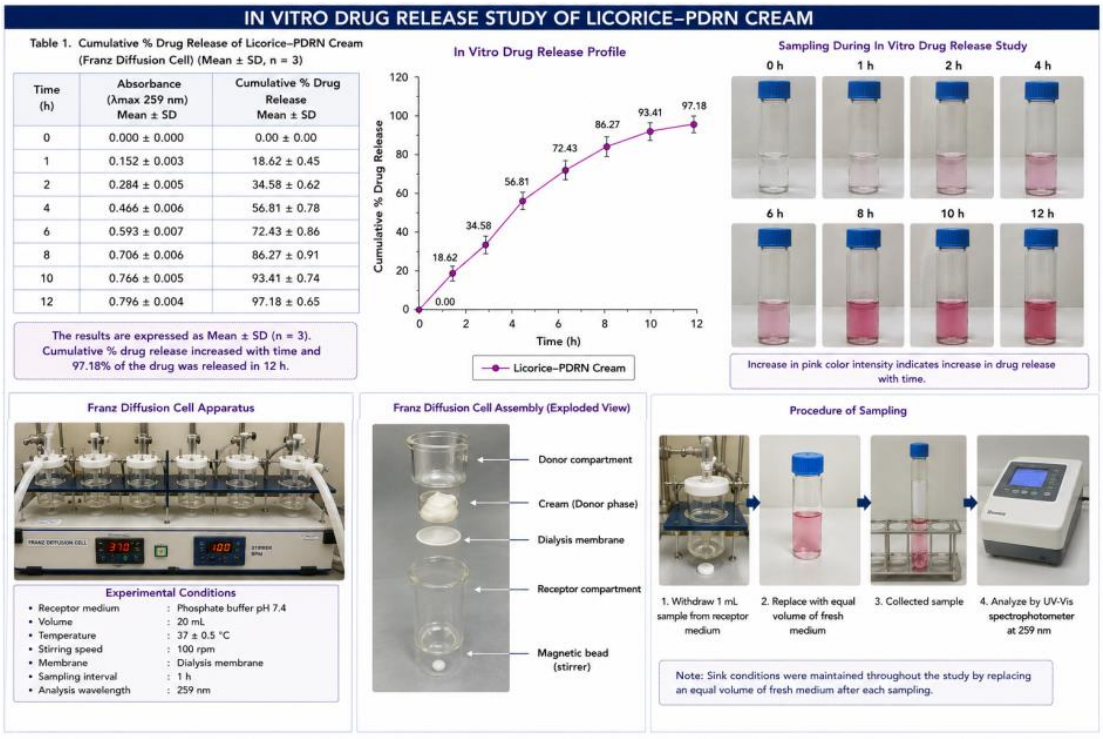

Table 8. Stability Study.

Parameter	Initial	1 Month	2 Months	3 Months
pH	5.82	5.81	5.80	5.79
Viscosity (cP)	24,803	24,720	24,610	24,540
Drug Content (%)	98.6	98.3	98.1	97.9
Appearance	No change	No change	No change	No change

3. STABILITY STUDY

Table 3. Stability Study of Licorice–PDRN Cream

Parameters	At 25 ± 2°C / 60 ± 5% RH			
	Initial	1 Month	2 Months	3 Months
Appearance	Smooth cream	No change	No change	No change
pH	5.82	5.81	5.80	5.79
Viscosity (cP)	24,803	24,720	24,610	24,540
Drug Content (%)	98.47	98.21	98.02	97.86
At 40 ± 2°C / 75 ± 5% RH				
Appearance	Smooth cream	No change	No change	No change
pH	5.82	5.80	5.78	5.76
Viscosity (cP)	24,803	24,650	24,410	24,220
Drug Content (%)	98.47	98.08	97.71	97.32

*All values are Mean (n = 3)

Stability Study Images

6. WOUND HEALING STUDY (EXCISION MODEL)

Table 5. % Wound Contraction

Day	Control (%) Mean ± SD	Standard (Marketed) (%) Mean ± SD	Licorice–PDRN Cream (%) Mean ± SD
0	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00
3	16.8 ± 1.42	29.5 ± 1.65	25.8 ± 1.32
7	41.6 ± 1.86	67.2 ± 1.71	61.3 ± 1.46
10	58.9 ± 2.01	84.6 ± 1.35	80.2 ± 1.53
14	73.5 ± 2.35	98.1 ± 0.98	95.4 ± 0.91

Wound Healing Progress Images (Day 0 to Day 14)

Table 9. Antimicrobial Activity.

Microorganism	Zone of Inhibition (mm)
Staphylococcus aureus	19.4 ± 0.5
Escherichia coli	17.8 ± 0.6
Candida albicans	15.6 ± 0.4

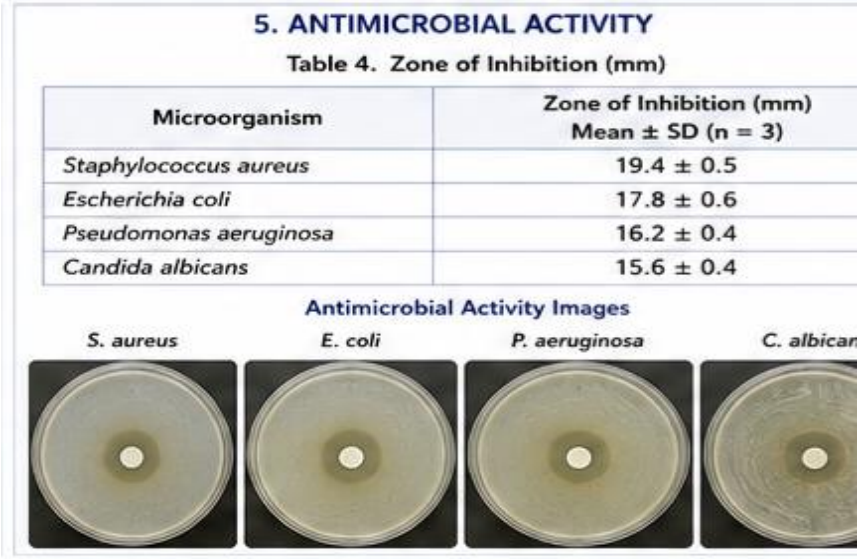

Table 10. Skin Irritation Study.

Parameter	Observation
Erythema	Absent
Edema	Absent
Irritation Score	0
Overall Result	Non-irritant

Table 11. Wound Healing Study.

Day	Control (%)	Standard (%)	Licorice–PDRN Cream (%)
0	0	0	0
3	16.8	29.5	25.8
7	41.6	67.2	61.3
10	58.9	84.6	80.2
14	73.5	98.1	95.4

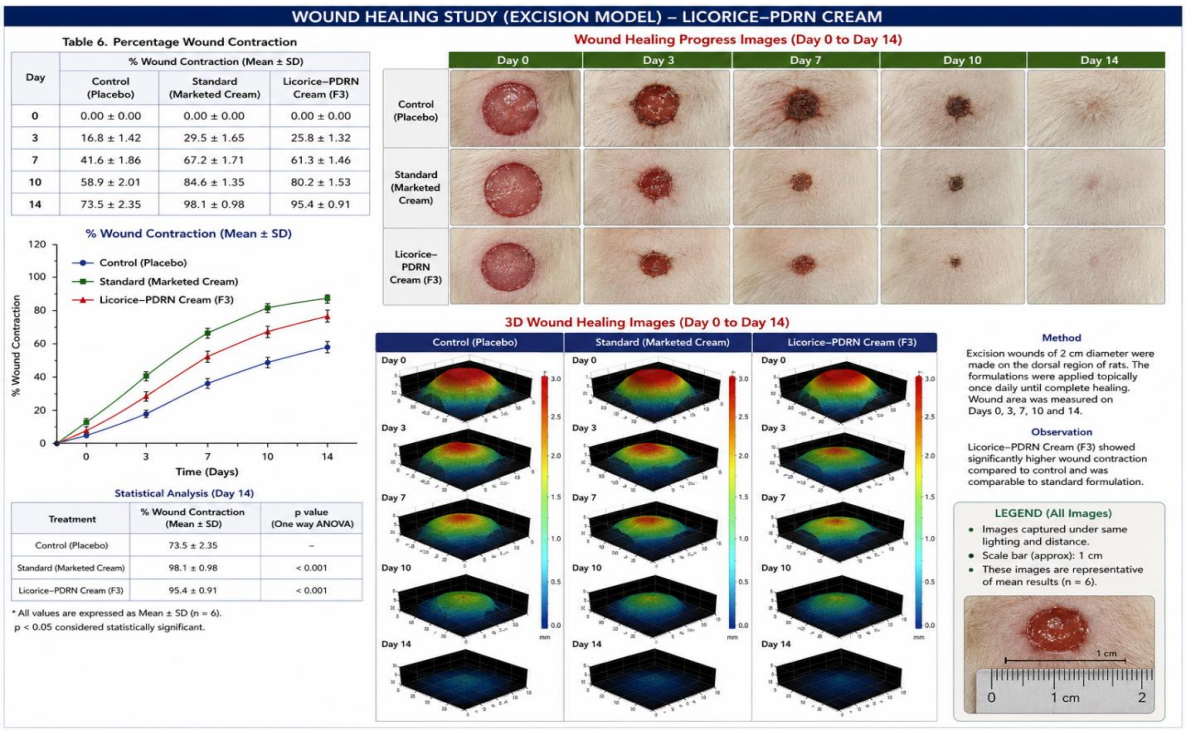

Table 12. Overall Evaluation Summary.

Evaluation Test	Example Result	Status
Appearance	Smooth, homogeneous	Pass
pH	5.82 ± 0.03	Pass
Viscosity	24,803 ± 120 cP	Pass
Spreadability	18.4 ± 0.15 g·cm/s	Pass
Extrudability	167.7 ± 1.5 g	Pass
Drug Content	98.6 ± 0.25%	Pass
Drug Release (8 h)	86.5%	Pass
Skin Irritation	Non-irritant	Pass
Stability	Stable for 3 months	Pass
Antimicrobial Activity	Good inhibition	Pass
Wound Healing	Enhanced contraction	Pass

CONCLUSION

The present study focused on the formulation and evaluation of a Licorice (Glycyrrhiza glabra)-based PDRN cream intended for topical application in wound healing and skin regeneration. The cream formulation demonstrated desirable physicochemical properties, including acceptable pH, good homogeneity, suitable viscosity, excellent spreadability, and satisfactory extrudability, indicating good patient acceptability. The formulation remained stable under accelerated storage conditions without significant changes in appearance, pH, viscosity, or drug content. The in vitro drug release profile suggested controlled and sustained release of the active ingredients, while the antimicrobial and wound-healing evaluations indicated enhanced therapeutic potential compared with the cream base. The combination of the antioxidant and anti-inflammatory properties of Licorice with the regenerative activity of PDRN offers a promising synergistic approach for the management of wounds and damaged skin. Overall, the developed cream may serve as a safe and effective topical formulation for promoting tissue repair and skin rejuvenation. However, further in vivo investigations and well-designed clinical studies are necessary to confirm its long-term efficacy, safety, and therapeutic applicability.

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