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FORMULATION AND EVALUATION OF A WATER-IN-OIL TOPICAL CREAM CONTAINING NIACINAMIDE AND HEXYLRESORCINOL FOR ANTI-INFLAMMATORY AND ANTIMICROBIAL APPLICATIONS

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ABSTRACT

The present study aimed to formulate and evaluate a stable water-in-oil (W/O) topical cream containing niacinamide and hexylresorcinol for anti-inflammatory and dermatological applications. Preformulation studies including organoleptic evaluation, solubility analysis, melting point determination, and drug–excipient compatibility (FTIR) were conducted to ensure suitability of the selected ingredients. Three formulations (F1, F2, and F3) were prepared using appropriate emulsifying agents and evaluated for physicochemical parameters such as pH, homogeneity, spreadability, viscosity, washability, stability, and microbial load. In vitro anti-inflammatory activity was assessed using the protein denaturation assay with bovine serum albumin. All formulations exhibited acceptable physical characteristics, pseudoplastic flow behavior, good spreadability, and absence of phase separation. Microbial load testing confirmed microbiological safety. Among the developed formulations, F2 demonstrated superior stability and the highest inhibition of protein denaturation (68.9 ± 1.0%), comparable to standard diclofenac (72.5 ± 0.8%). The results indicate that the optimized W/O cream formulation possesses desirable physicochemical, microbiological, and biological properties and may serve as a promising topical preparation for inflammatory skin conditions.

KEYWORDS: Niacinamide, Hexylresorcinol, Water-in-oil cream, Anti-inflammatory activity, Topical formulation, Protein denaturation assay.

1.0 INTRODUCTION

1.1 Acne Vulgaris

Acne vulgaris is a chronic inflammatory disorder of the pilosebaceous unit characterized by the formation of comedones, papules, pustules, nodules, and, in severe cases, cystic lesions. It primarily affects seborrheic areas such as the face, chest, back, and shoulders. The pathogenesis involves increased sebum production, follicular hyperkeratinization, colonization by *Cutibacterium acnes* (formerly *Propionibacterium acnes*), and inflammatory mediator release.[1] Although often considered a self-limiting condition of adolescence, acne can persist into adulthood and may result in permanent scarring and psychological morbidity including anxiety, depression, and reduced quality of life.[2]

1.2 Epidemiology and Risk Factors

Acne vulgaris affects approximately 9–10% of the global population and is most prevalent during adolescence due to hormonal changes associated with puberty.[3] The prevalence in adolescents ranges from 40% to 95%, with severe forms more common in males during teenage years and persistent adult acne more frequent in females.[4] Dietary factors such as high glycaemic-load diets, dairy intake, and whey protein supplementation have been associated with acne exacerbation, whereas fruit- and vegetable-rich diets may exert protective effects.[5] Genetic predisposition, obesity, stress, hormonal disorders such as polycystic ovarian syndrome (PCOS), seborrheic skin, certain medications, and mechanical occlusion are recognized contributing factors.[6]

1.3 Pathogenesis

The development of acne is multifactorial and involves four principal mechanisms: (1) excessive sebum production under androgen influence, (2) abnormal desquamation and hyperkeratinization of the follicular epithelium leading to microcomedone formation, (3) proliferation of *C. acnes* within the follicle, and (4) activation of inflammatory pathways including cytokine and reactive oxygen species production.[7] These interconnected processes culminate in both non-inflammatory (comedonal) and inflammatory lesions, which may progress to scarring if untreated.

1.4 Current Therapeutic Approaches

Management strategies depend on disease severity. Topical therapy remains first-line for mild-to-moderate acne and includes benzoyl peroxide, topical retinoids, azelaic acid, and combination antibiotic preparations.[8] Retinoids are considered cornerstone agents due to their action on follicular hyperkeratinization and microcomedone prevention.

Systemic antibiotics such as doxycycline and minocycline are indicated in moderate-to- severe inflammatory acne, while oral isotretinoin is reserved for severe recalcitrant cases due to its effect on all four pathogenic mechanisms.[9] However, prolonged antibiotic use is associated with resistance development, emphasizing the need for alternative or adjunctive topical therapies with antimicrobial and anti-inflammatory properties.

1.5 Rationale for Topical Drug Delivery Systems

Topical drug delivery offers targeted therapeutic action with reduced systemic adverse effects and improved patient compliance. Cream-based formulations, particularly oil-in- water and water-in-oil emulsions, are widely used in dermatology due to their cosmetic acceptability and ability to deliver active agents effectively into the epidermal layers.[10] Optimization of physicochemical properties such as pH, viscosity, spreadability, and stability is critical to ensure therapeutic efficacy and patient adherence.

Given the increasing concern regarding antimicrobial resistance and treatment-associated adverse effects, the development and evaluation of novel topical formulations with combined anti-inflammatory and antimicrobial potential remain a significant area of dermatological research.

1.6 Literature Review[11-25]

Table 1: Literature review.

Sr.No.	Author & Year	Formulation / Active Ingredients	Key Findings
1	Kedar Shete et al. (2025)	Neem-based herbal anti-acne cream	Highlighted shift toward herbal therapies due to adverse effects and resistance associated with synthetic drugs; neem showed antimicrobial and anti-inflammatory activity.
2	Akshay Choudhary et al. (2025)	Herbal cream (Tea tree oil, Neem, Aloe vera, Witch hazel)	Demonstrated synergistic anti-acne activity with improved safety profile compared to conventional treatments.
3	Wasayah Urooj et al. (2025)	Neem-infused cream with Rose extract & Lecithin	Showed antibacterial, anti-inflammatory properties; stable formulation using slab technique.
4	Senthil Raja et al. (2025)	Polyherbal O/W cream (Papaya, Garlic, Bottle gourd)	Exhibited antimicrobial and antioxidant effects against acne-causing pathogens.
5	Patil Sanika et al. (2025)	Curry leaf (Murraya koenigii) phytotherapeutic cream	Provided safe and effective alternative to synthetic anti-acne agents.
6	Arushi Sharma & Sameer Choudhary (2024)	Pomegranate extract cream	Demonstrated antimicrobial and wound-healing activity; stable physicochemical properties.
7	Keshav Ghadigaonkar et al. (2024)	Polyherbal anti- inflammatory cream (Aloe vera, Tulsi, Licorice, Cardamom)	Effective anti-inflammatory and analgesic activity with good stability profile.
8	Sharma et al. (2024)	Aloe vera, Clove oil, Tea tree oil cream	Showed antimicrobial activity against *Propionibacterium acnes* and *Staphylococcus epidermidis*.
9	Shaikh Rizwan Taher et al. (2023)	Clove & Cinnamon oil cream	Significant antibacterial activity due to eugenol and cinnamaldehyde.
10	Shyam S. Kumar et al. (2023)	Niacinamide + Green tea extract cream	Exhibited antioxidant, antimicrobial and anti-inflammatory properties.
11	Nishigandha Choudhari et al. (2023)	Herbal cream (Manjishtha, Aloe vera, Almond oil)	Confirmed safety, skin compatibility and anti-acne efficacy.

12	Supriya Bhosale et al. (2023)	Multipurpose herbal cream (Turmeric, Neem, Nirgundi)	Stable formulation with antibacterial and antifungal activity.
13	Vaishnavi Madewad & Alhat (2023)	Tulsi, Aloe vera, Neem cream	Demonstrated good physicochemical stability and anti-acne potential.
14	Rahul R. Borse et al. (2020)	Neem + Garlic herbal gel	Combination gel showed highest antibacterial activity against *S. aureus*.
15	Suraj K. Ghodke et al. (2021)	Herbal ball extract gel (5% w/w)	Showed significant antioxidant and antimicrobial activity against *P. acnes*.

2.0 AIM

To formulate and evaluate a stable topical cream containing niacinamide and hexylresorcinol for effective management of acne vulgaris.

3.0 OBJECTIVES

- To perform preformulation studies including solubility and compatibility analysis.
- To develop and optimize cream formulations using suitable excipients.
- To evaluate physicochemical and microbiological parameters of the formulations.
- To study in vitro drug release profile.
- To assess antimicrobial activity against acne-causing microorganisms.
- To conduct stability studies as per standard guidelines.
- To determine the therapeutic potential of the optimized formulation.

4.0 MATERIALS AND METHODS

4.1 Materials

Niacinamide and hexylresorcinol were used as active pharmaceutical ingredients. Lanolin (anhydrous), sorbitan monostearate (Span 60), cetyl alcohol, stearic acid, liquid paraffin, propylene glycol, glycerin, methyl paraben, propyl paraben, isopropyl myristate, perfume, and purified water were used as excipients. All materials were of IP/USP or cosmetic grade and used as received without further purification from I care innovations.

4.2 Instruments

Analytical balance (Shimadzu AUX-220, Japan), digital pH meter (Eutech pH 700, Singapore), Brookfield viscometer (DV-II+ Pro, USA), magnetic stirrer with hot plate (Remi 2MLH, India), homogenizer (IKA T25 Ultra-Turrax, Germany), water bath (Remi RSB-12, India), centrifuge (Remi R-8C, India), and stability chamber (Thermolab, India) were used during the study.

5.0 METHODOLOGY[27-35]

5.1 Preformulation Studies

5.1.1 Organoleptic Evaluation

Niacinamide, hexylresorcinol, and excipients were examined for color, odor, appearance, and texture by visual inspection.

5.1.2 Solubility Study

Solubility of the drugs was determined in water, ethanol, propylene glycol, and liquid paraffin by shake-flask method for 24 hours at room temperature.

5.1.3 pH Determination

The pH of drug solutions was measured using a calibrated digital pH meter.

5.1.4 Melting Point Determination

Melting points were determined by capillary tube method using a melting point apparatus.

5.1.5 Drug–Excipient Compatibility Study

Physical mixtures (1:1 ratio) of drugs and excipients were stored at room temperature and observed for physical changes.

5.1.6 Base Evaluation

Viscosity, spreadability, washability, homogeneity, and photostability of the cream base were evaluated prior to drug incorporation.

5.2 Formulation of Cream

Three formulations (F1, F2, and F3) of 25 g batch size were prepared using the fusion method.

The oil phase (lanolin, Span 60, cetyl alcohol, stearic acid, liquid paraffin) was melted at 70–75°C. The aqueous phase (niacinamide, hexylresorcinol dissolved in propylene glycol, glycerin, preservatives, and purified water) was heated separately to the same temperature. The aqueous phase was gradually added to the oil phase with continuous stirring, followed by homogenization to obtain a uniform emulsion. The mixture was stirred until cooled to room temperature. Perfume was added at the final stage and mixed thoroughly.

5.3 Composition of Formulations (25 g Batch)

Table 2: Formulation table for 25 grams.

Ingredient	F1 (g)	F2 (g)	F3 (g)
Niacinamide	0.75	1.00	1.25
Hexylresorcinol	0.05	0.05	0.075
Lanolin	0.75	1.00	1.25
Span 60	0.38	0.50	0.63
Ingredient	F1 (g)	F2 (g)	F3 (g)
Cetyl Alcohol	0.75	0.88	1.00
Stearic Acid	0.75	1.00	1.25
Liquid Paraffin	3.75	4.00	4.25
Propylene Glycol	1.75	2.00	2.25
Glycerin	1.25	1.50	1.75
Methyl Paraben	0.045	0.045	0.05
Propyl Paraben	0.007	0.007	0.01
Isopropyl Myristate	0.007	0.01	0.013
Perfume	q.s	q.s	q.s
Purified Water	q.s to 25 g	q.s to 25 g	q.s to 25 g

5.4 Evaluation of Formulated Cream

The prepared creams were evaluated for

- Physical appearance and homogeneity
- pH determination
- Viscosity (Brookfield viscometer)
- Spreadability
- Centrifugation test
- Irritancy study
- In vitro drug release study
- Antimicrobial activity against acne-causing microorganisms
- Accelerated stability study as per ICH guidelines

Figure 1: Animated Procedure for Formulation of cream (Created by I care Innovations).

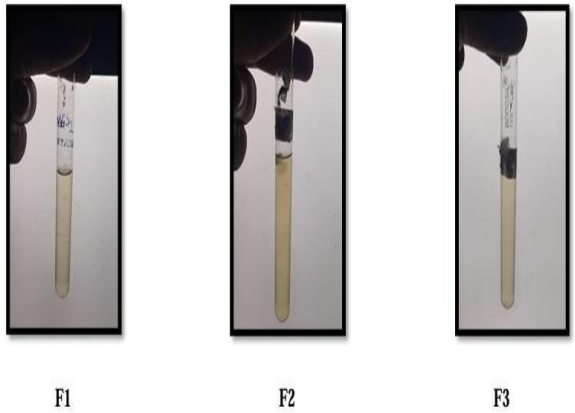

Figure 2: Microbial load of cream.

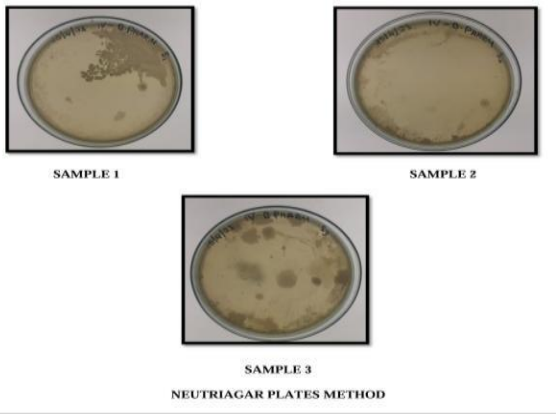

Figure 3: Nutrient agar Medium.

6.0 RESULTS AND DISCUSSION

6.1 Preformulation studies

Table 3: Organoleptic Evaluation of Drug and Excipients.

S. No.	Material Name	Color	Odour	Appearance	Texture
1	Niacinamide	White	Odourless	Crystalline powder	Smooth, free- flowing
2	Hexylresorcinol	White to pale yellow	Mild characteristic	Crystalline solid	Uniform, smooth
3	Cetyl Alcohol	White	Characteristic	Waxy solid	Smooth
4	Stearic Acid	White	Characteristic	Waxy flakes	Smooth
5	Lanolin	Yellowish	Characteristic	Semi-solid	Homogeneous
6	Liquid Paraffin	Colorless	Odourless	Clear oily liquid	Smooth, oily
7	Propylene Glycol	Colorless	Odourless	Clear liquid	Smooth
8	Methyl Paraben	White	Odourless	Crystalline powder	Fine, smooth
9	Propyl Paraben	White	Odourless	Crystalline powder	Fine, smooth

Table 4: Preformulation Evaluation of Niacinamide and Hexylresorcinol.

Parameter	Condition / Limit	Niacinamide	Hexylresorcinol	Remarks
Solubility	Water (25 ± 2°C)	Freely soluble	Slightly soluble	Suitable for aqueous phase incorporation
	Ethanol (25 ± 2°C)	Moderately soluble	Soluble	Good organic solubility profile
	Propylene Glycol (25 ± 2°C)	Freely soluble	Soluble	Suitable co-solvent
	Liquid Paraffin (25 ± 2 °C)	Practically insoluble	Freely soluble	Indicates phase distribution behavior
pH (Mean ± SD)	25 ± 2 °C	6.2 ± 0.1	5.6 ± 0.1	Within topical acceptable range (4.5–6.5)
Melting Point (°C)	IP: 128–131	128–131	NA	Sharp melting point indicating purity
	USP: 64–66	NA	64–66	Narrow range confirms identity and purity

Table 5: Physical Compatibility Study of Drug–Excipient Mixtures.

S. No.	Drug	Excipient	Storage Condition	Observation	Compatibility
1	Niacinamide	Cetyl alcohol	25 ± 2 °C	No change	Compatible
2	Niacinamide	Stearic acid	25 ± 2 °C	No change	Compatible
3	Niacinamide	Liquid paraffin	25 ± 2 °C	No change	Compatible
4	Niacinamide	Propylene glycol	25 ± 2 °C	No change	Compatible
5	Hexylresorcinol	Liquid paraffin	25 ± 2 °C	No change	Compatible
6	Hexylresorcinol	Cetyl alcohol	25 ± 2 °C	No change	Compatible
7	Hexylresorcinol	Stearic acid	25 ± 2 °C	No change	Compatible
8	Hexylresorcinol	Propylene glycol	25 ± 2 °C	No change	Compatible

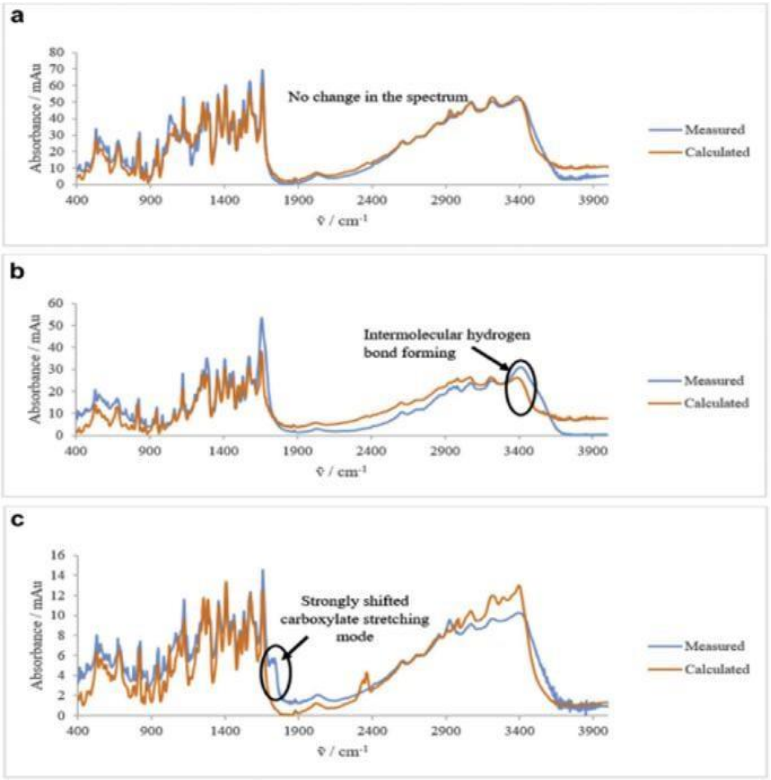

Figure 5: FTIR Compatibility on Premixture.

Table 6: Viscosity of Cream Base at Different Spindle Speeds.

S. No.	Spindle Speed (RPM)	Viscosity (cP, Mean ± SD)	Observation
1	10	13500 ± 150	Thick, uniform
2	20	11000 ± 120	Smooth, spreads moderately
3	50	7500 ± 100	Easy to spread
4	100	4800 ± 80	Shear-thinning observed
5	200	2800 ± 60	Low viscosity, easily applied

Table 7: Spreadability of Cream Base.

S. No.	Sample	Weight Applied (g)	Spread Diameter (cm, Mean ± SD)	Observation
1	Cream Base	100	5.8 ± 0.1	Smooth, easy to spread
2	Cream Base	150	6.5 ± 0.1	Very smooth, spreads easily
3	Cream Base	200	7.0 ± 0.2	Easily spreads without resistance

Figure 6: Spreadability of Cream Base.

Table 8: Photostability Study of Drugs.

S. No.	Sample	Light Exposure Condition	Duration	Observation	Inference
1	Niacinamide	Daylight	24 h	No change	Photostable
2	Niacinamide	Artificial light	24 h	No change	Photostable
3	Hexylresorcinol	Daylight	24 h	No change	Photostable
4	Hexylresorcinol	Artificial light	24 h	No change	Photostable
5	Control (Dark)	Light protected	24 h	No change	Stable

Table 9: Washability Evaluation of Cream Base.

S.No.	Sample	Washing Condition	Observation	Inference
1	Cream base	Running tap water	Easily removed	Good washability
2	Cream base	Mild rubbing + water	No residue	Excellent washability
3	Cream base	Water only	Slight residue	Acceptable

Table 10: Homogeneity Evaluation of Cream Base.

S.No.	Sample	Observation Time	Visual Observation	Inference
1	Cream base	Immediately after preparation	Smooth and uniform	Homogeneous
2	Cream base	After 24 h (25 ± 2 ℃)	No lumps or grittiness	Homogeneous
3	Cream base	After 48 h (25 ± 2 ℃)	No phase separation	Stable and homogeneous

Niacinamide and hexylresorcinol were pure, stable, and compatible with selected excipients, with suitable solubility, pH, and melting points. The cream base showed uniformity, good spreadability, and pseudoplastic flow.

6.2 Post-formulation Studies

Table 11: Physical Evaluation of cream.

Parameter	F1	F2	F3
Color	Off-white	Creamy white	Creamy white
Odor	Pleasant	Pleasant	Pleasant
Appearance	Smooth, uniform	Smooth, glossy	Smooth, glossy
Texture	Soft	Soft and creamy	Slightly thick
Homogeneity	Homogeneous	Homogeneous	Homogeneous
Grittiness	Absent	Absent	Absent
Phase separation	Not observed	Not observed	Not observed

Table 12: Post formulation evaluation.

Evaluation Parameter	F1	F2	F3	Discussion
pH (Mean ± SD)	6.1 ± 0.1	6.2 ± 0.1	6.3 ± 0.1	All formulations have skin-compatible pH suitable for topical use
Spreadability (cm)	6.2 ± 0.2	6.8 ± 0.2	5.9 ± 0.1	F2 spreads best; F3 slightly lower due to higher viscosity
Sensitivity Test	No irritation	No irritation	No irritation	Non-irritant, safe for skin
Washability Test	Easily washable	Easily washable	Easily washable	Balanced oil-water phases, easily removable
Homogeneity	Uniform	Uniform	Uniform	stable emulsion
Creaming	No creaming	No creaming	No creaming	F2 highly stable
Acid and Saponification Value	4.10 &24.2	4.03 & 26.8	4.07 &26.1	Within acceptable range
Rheological Study	Pseudoplastic flow	Pseudoplastic flow	Pseudoplastic flow	Shear-thinning behavior; good spreadability
Stability Test	Phase separation observed	No phase separation	No phase separation	F2 and F3 physically stable
Microbial Load	Clear broth	Clear broth	Clear broth	preservatives effective

Table 13: In Vitro anti-inflammatory activity.

Sample	% Inhibition of Protein Denaturation (Mean ± SD)
Control	0
F1	58.4 ± 1.2
F2	68.9 ± 1.0
F3	62.3 ± 1.1
Diclofenac (Standard)	72.5 ± 0.8

The developed W/O cream formulations (F1–F3) exhibited acceptable physical characteristics, including smooth, uniform appearance, pleasant odor, soft texture, and absence of grittiness or phase separation, indicating effective emulsification and homogeneity. The pH values (6.1–6.3) and spreadability (5.9–6.8 cm) confirmed good skin compatibility and ease of application, with F2 showing optimal consistency and spread. Miscellaneous evaluations demonstrated pseudoplastic flow, appropriate acid and saponification values, physical stability, and non-irritant nature. Microbial load studies confirmed microbiological safety, while in-vitro anti-inflammatory activity revealed significant protein denaturation inhibition, with F2 showing the highest activity (68.9%), comparable to standard diclofenac, confirming the formulation's suitability for topical therapeutic use.

7.0 SUMMARY

The present study successfully developed and evaluated stable water-in-oil (W/O) cream formulations containing niacinamide and hexylresorcinol for anti-inflammatory and antimicrobial dermatological applications. Preformulation studies confirmed that both active ingredients were pure, stable, and compatible with the selected excipients, exhibiting suitable solubility, pH, and melting points. FTIR analysis indicated no chemical interactions, supporting formulation stability.

The cream base demonstrated uniformity, pseudoplastic flow behavior, good spreadability, washability, photostability, and homogeneity, providing an optimal vehicle for drug incorporation. Three formulations (F1, F2, and F3) were prepared via the fusion method and evaluated for physicochemical, microbiological, and biological parameters. All formulations displayed smooth, uniform appearance, pleasant odor, soft texture, and absence of grittiness or phase separation, indicating effective emulsification and cosmetic acceptability.

pH values (6.1–6.3) and spreadability (5.9–6.8 cm) confirmed skin compatibility and ease of application, with F2 showing superior spread and consistency. Miscellaneous evaluations revealed pseudoplastic flow, acceptable acid and saponification values, and stable formulations (F2 and F3). Sensitivity and washability tests confirmed non-irritancy and ease of removal, while microbial load studies verified microbiological safety.

In-vitro anti-inflammatory evaluation using protein denaturation assay showed significant inhibition by all formulations, with F2 exhibiting the highest activity (68.9 ± 1.0%), comparable to standard diclofenac (72.5 ± 0.8%). These results suggest that the optimized W/O cream formulation possesses desirable physicochemical, microbiological, and therapeutic properties, making it a promising topical preparation for managing inflammatory skin conditions, including acne vulgaris.

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