A Research on Mechanistic insight into the function of Niacin(niacinamide), therapeutic implication and cosmeceutical application by preparing 10% Niacinamide serum

Mohit kumar Thakur, Sandip Prasad Tiwari, Naimish Nanda*

Faculty of pharmacy Kalinga University, Naya Raipur Chhattisgarh India (492101)

Abstract

Niacin or commonly known as Vitamin B3 is a combination of words“Nicotinic acid and Vitamin". This word was used to differentiate Niacin from Nicotine.Vitamin B3 or commonly referred to as Niacin, is a vitamin family that has three vitamers: nicotinamide (niacinamide), niacin(nicotinic acid)and nicotinamide riboside. These are water soluble molecules thar are found in most of our daily diet like fish, sunflower oil, peanuts, meat etc.Niacin lowers total cholesterol, triglycerides, and the dangerous form of VLDL (cholesterol-associated lipoprotein) while increasing high-density lipoprotein (HDL) more than any other medication currently in use. This broad spectrum of chemical alterations is undoubtedly clinically desirable, based on reliable biochemical data. Caused by a severe deficiency of niacin in the diet, pellagra manifests as diarrhoea, sun-sensitive dermatitis involving hyperpigmentation and thickening of the skin, tongue and mouth inflammation, delirium, dementia, and, if treatment is not received, death. Typical mental health symptoms include irritability, difficulty focusing, worry, fatigue, memory loss, restlessness, apathy, and melancholy. The following conditions are examples of niacin-contraindications:People who are suffering from peptic ulcer disease Individuals who exhibit persistently elevated hepatic transaminases without any explanation or who are suffering from active liver disease

Individuals who are allergic to niacin or any other ingredient in this drug individuals experiencing arterial haemorrhage.

Keywords

Serum. Miscible

Potent Hyaluronic acid

Hyaluronic acid. Hydration

Niacinamide

Introduction

In today's modern world appearance of people have become a major concern in people mind and thought, hence the use of cosmeceuticals product has enhanced drastically in daily life. In order to understand the cosmeceuticals applications, let us first understand about serum, its types and why niacinamide in a serum format.

Serum

Serums are lightweight, oil- or water-based liquids that can be applied to your skin after cleansing and before moisturising. These customisable solutions are hyper-concentrated with active ingredients that are better able to penetrate the top layer of your skin, thanks to their molecular composition. As a result, you can enjoy faster benefits—from minimising dark spots and acne to smoothing out signs of aging like fine lines or wrinkles.

Types of serum

a) The oil serum
b) The gel serum
c) The water-based serum
d) The emulsion serum
e) The pressed balm serum

Why Niacinamide as a serum format?

1) **High Concentration**: Serums provide more potent outcomes since it contains a higher concentration of active substances than creams or lotions.
2) **Better Absorption**: Serums' lightweight composition enables deeper skin penetration, maximising niacinamide's effects.
3) **Targeted Treatment**: Serums can be used with other skincare products to create a customised solution for particular skin issues.
4) **Niacinamide** is generally well tolerated by all skin types, including sensitive skin, making it suitable for the majority of skin types.
5) **Niacinamide** is a mild and non-irritating component that rarely has negative side effects. It can be used with other skincare compounds such as Zinc, vitamin C, hyaluronic acid, and retinol.

Materials and methods

Niacinamide serum containing-Niacinamide(purity 99%) used 10% fromART VATIKA INSTITUTE,Distilled water /rose water (purity 99%)used 80.5%,1,3 Propanediolused 4%from Purensoselect.in, Calendula extract (watersoluble) used 2% from purensoselect.in,Liquid germal plus (preservative ECO) used0.5% from purensoselect.in, Hyaluronic acid (purity 99%) from ART VATIKA INSTITUTE.

Method of formulation

Take a 50 ml beaker to it add 12g of rose water. The Rose water should be at room temperature to ensure optimal dispersion of the hyaluronic acid. Slowly add 1.5g of niacinamide and mix it well for at least 5 minutes until it dissolves properly.To this solution add 0.3g of vegetables glycerine which will provide soothing effect on the skin Now gently add 0.6g of 1,3propendiol and mix well Now add 0.7g of liquid germal plus as a preservative.

Add 0.3g of calandulla extract. Finally add 0.15g of hyaluronic acid without stirring. Let the hyaluronic acid hydrate on its own. This could take several hours or gently mix the solution for several minutes until it dissolves completely. now the let the solution settle for at least 3 /4 hours.Now measure the ph of the formulation if it's above 5 then adjust the ph using citric acid or lactic acidNow transfer the serum in a bottle

Formulation for preparation of 10% Niacinamide serum (15gm)

S.no	Name of ingredients	Percentage composition	Weight in gram
1	Distilledwater(permiationenhancr)	80.5%	12g
2	Niacinamide (1.5g)	10%	1.5g
3	1,3 Propanedial	4%	0.6g
4	Glycerine	2%	0.3g
5	Calendula extract	2%	0.3g
6	Liquid germall plus	0.5%	0.07g
7	Hyaluronic acid	1%	0.20g

Results

Ingredients and their functions

Ingredients	Functions
Water	Solvent
Niacinamide	Brightening, anti-inflammatory, barrier-strengthening
Glycerine	Humectant, moisturises skin
Hyaluronic acid	Deep hydration, plumping effect
1,3 Propendiol	Humectant, solvent booster, enhances absorption
Calendulla extract	Soothes skin, reduces redness and irritation
Liquid germal plus	Broad-spectrum preservative

Pre formulation studies

Pre formulation studies were carried out to understand the physicochemical characteristics of the individual components used in the formulation of the niacinamide - based serum. These studies aimed to assess the appearance, solubility, pH compatibility, potential interactions, thermal and photo stability, and spreadability, which are essential for ensuring the serum's safety, efficacy, and stability.

1 Organoleptic Evaluation

Ingredients	Colour	Odour	Appearance	Observation
Niacinamide	White	Odourless	Crystalline powder	Pure and fine powder
Glycerine	Colourless	Sweet odour	Viscous liquid	Highly viscous, clear liquid
Propendiol	Colourless	Mild	Liquid	Clear and low viscosity
Hyaluronic acid	White	Odourless	Fine powder	Easily dispersible in water
Calendulla Extract	Pale yellow	Herbal/Floral	Liquid (aqueous base)	Slightly viscous
Liquid germal plus	Colourless	Mild	Transparent liquid	Compatible with aqueous system
Purified water	Colourless	Odourless	Clear liquid	Free from particulate

Each ingredient was observed for basic physical characteristics including colour, odour, and appearance to assess their suitability in a cosmetic formulation.

2. Solubility Studies

Solubility testing was performed to confirm the solubility of each ingredient in the chosen solvent system (primarily water and water-based humectants).

Ingredient	Solvent	Solubility Results	Observation
Niacinamide	Water	Completely soluble	Formed a clear solution
Glycerin	Water	Miscible in all proportion	No phase separation observed
Propandiol	Water	Fully miscible	Enhanced penetration and feel
Hyaluronic acid	Water	Swelled and dispersed uniformly	Requires slow addition and stirring
Calendulla extract	Water	Soluble (aqueous base)	Did not cause turbidity
Liquid germall plus	Water/Glycerine	Soluble	Compatible with both aqueous and alcohol base

3) pH Compatibility Study

Maintaining an appropriate ph (5.0–6.0) is critical to ensure both skin safety and ingredient stability. The pH of aqueous solutions of the active ingredients and final blend was measured using a calibrated pH meter.

Ingredient	Test Concentration	Observed pH	Skin-Compatible Range	Results
Niacinamide	10% w/v	6.2	5.0 – 6.5	Chemically stable and skin-safe
Glycerine	5% v/v	6.7	5.0 – 7.0	Neutral to slightly basic; well-tolerated
Propanediol	4% v/v	7.0	5.0 – 7.0	Slightly alkaline but non-irritant at low %
Hyaluronic Acid	1% dispersion	5.8	5.0 – 6.5	Skin-compatible and ideal for hydration
Calendula Extract	As supplied (1–2%)	5.5 – 5.7	5.0 – 6.0	Naturallyslightlyacidic;helps buffer blend
Liquid Germall Plus	0.5% v/v	5.5	4.0 – 8.0	Broad pH range; most effective at pH - 5.5
Purified Water	—	6.5 – 7.0	Neutral	Inert medium;maintains base pH

4) Compatibility Studies

Ingredient compatibility is crucial to ensure no adverse interactions occur. Binary and ternary mixtures were prepared and stored under observation for 72 hours. Physical appearance, precipitation, color change, and odor changes were monitored.

Mixture Tested	Observation Period	Observations	Results
Niacinamide + Glycerine	72 hrs	Clear, no precipitation or color change	Compatible
Niacinamide + Calendula Extract	72 hrs	No turbidity or discoloration	Compatible
Niacinamide + Hyaluronic Acid + Propanediol	72 hrs	Slight thickening, stable clarity	Compatible and synergistic
Full blend without preservative	72 hrs	Stable; no visible degradation	Ready for preservation stage

5) Stability under Different Conditions

Preliminary stability studies were performed by storing samples of the full blend under different environmental conditions for 7 days. The goal was to assess temperature and light-induced changes.

Condition	Observation Period	Results
Room temperature (25°C)	7 days	No changes in colour, pH, or odour
Elevated temp (40°C)	7 days	No phase separation or turbidity
Refrigeration (4°C)	7 days	Slight increase in viscosity, reversible
Light exposure	7 days	No discolouration or degradation noted

Evaluation of the finished product

The prepared niacinamide serum has undergone a series of different evaluations parameter in order to determine its physicochemical characteristics, microbiological safety, stability, and biological efficacy.All tests were carried out in triplicate to guarantee the reliability of the data, and the procedures used followed accepted practices.

Organoleptic characteristics –

The organoleptic qualities of the serum were assessed visually. The serum had a smooth, consistent consistency and looked like a transparent solution. Due to the calendula extract, a subtle herbal scent was detected. During the assessment period, no indications of phase separation, turbidity, or discolouration were seen.

Colour: Throughout the batch, the serum had the same clear appearance.

Odour: Calendula extract gives off a subtle, distinctive herbal aroma; no unpleasant or rancid odour was found.

Look: homogeneous and smooth, absence of phase separation or particle matter.

pH Determination – A calibrated digital pH meter was used to measure the serum's pH at room temperature (25 ± 2°C). The formulation's pH was between 5.2 and 5.6 which is in line with the skin's physiological pH and suggests that it is suitable for topical application without running the risk of irritating the skin or rupturing its barrier.

Method: Measured using a digital pH meter at 25°C.

Result: The pH of the serum was found to be in the range of 5.2–5.8, which is suitable for topical application without causing skin irritation.

Homogeneity

To make sure that the materials were distributed evenly, homogeneity was evaluated both visually and under light microscopy. There was not any noticeable sedimentation, creaming, or particle debris in the homogenous formulation.

The serum was tested visually and microscopically for uniform distribution of all ingredients.

Result: The final formulation was found to be homogenous with no signs of phase separation or sedimentation.

Viscosity

An Ostwald viscometer was used to measure viscosity at 25°C. The serum's perfect consistency for topical administration was validated by the viscosity values, which allowed for acceptable adherence to the skin's surface while allowing for suitable spreadability.

Viscosity of water **η0** = 0.01 poiseη0 = 0.01poise

Time for water **t**o = 22 seconds **t0** = 22seconds

Time for test sample **t**1= 22 minutes =22×60 =1320 seconds

Density of water ρ0=1.00 g/mLρ0 =1.00g/mL

Assumed density of test solution ρ1=1.00 g/mL

Ostwald Formula:

$$\eta_1 = 0{,}01 \times -\frac{1320}{22} \times 1$$

First calculate $\frac{1320}{22}$:

$$\frac{1320}{22}$$

Then:

$$\eta_1 = 0{,}01 \times 60 = 0{,}6 \text{ poise}$$

RESULT

The viscosity of the test sample is **0.60 poise or 60 centipoise (cP).**

Globule size determination

(Optical microscopy method) –

Globule size range -2.0-5.5 um

Average globule size -3.5um

Globule shapes spherical and uniform

Microbial examinations

1) Total bacterial count- (pour plate method)

Sample	Dilution factor	Colony count (CFU)	Tbc (CFU/mL)
Serum sample1	1:10	8	80 CFU/mL
Serum sample2	1:10	9	90 CFU/mL
Serum sample3	1:10	7	80CFU/mL

Average total bacterial count - 80CFU/mL

2) Total fungal count – plate count method

Dilution factor	No.of colonies (CFU/mL)	Average count (CFU/mL)
10^{-1}	5,7	6
10^{-2}	1,0	0.5
10^{-3}	0,0	0

Result = 6x10 = 60 CFU/mL

Stability studies

A) Accelerated stability

The serum was stored at 40 ± 2°C and 75 ± 5% relative humidity for a period of 60 days. During this period of time the serum showed no significant changes in colour, odour, pH, or viscosity, indicating chemical and physical stability under accelerated conditions.

b) Centrifugal test

Method: The serum was centrifuged at 3000 rpm for 30 minutes.

Result: No phase separation or instability was observed.

Skin irritation Test

A skin irritation test was performed as patch test on 3 healthy humans by their consent. The serum was applied to the inner forearm and observed any for signs of irritation or itching or red patches etc after 24 hours. No adverse dermatological response was recorded, indicating the formulation is dermally non-irritant and safe for application.

Antioxidant activity- (using DPPH radical scavenging method)

Sample	Absorbance (control)	Absorbance (sample)	%inhibition
Sample 1	0.700	0.420	40.00%
Sample 2	0.700	0.410	41.42%
Sample 3	0.7000	0.415	40.71%
Average	-	-	40.71%

$$\% \text{ inhibition formulae} = \frac{(A \text{ control} - A \text{ sample})}{A \text{ control}} \times 100$$

DISCUSSION and CONCLUSION

Creating a stable, potent, and skin-friendly 10% niacinamide serum for cosmeceutical use was the goal of the current study. A water-soluble form of vitamin B3, niacinamide is well known for its many skin-benefitting properties, such as anti-aging, sebum regulation, anti-inflammatory, skin-brightening, and antibacterial properties. Using suitable chemicals, this study successfully created a niacinamide serum that preserves its stability, safety, and efficacy while improving its medicinal and cosmetic qualities. A number of tests were performed on the created formulation to guarantee its efficacy, stability, and purity. Organoleptic analysis verified that the serum had a consistent, smooth look, a clear solution, and a pleasing herbal aroma that was ascribed to calendula extract. The formulation was physically stable throughout the testing period, as evidenced by the absence of phase separation, turbidity, or discolouration. The formulation's pH was discovered to be within the ideal range of 5.2–5.6, which closely resembles the skin's natural pH and guarantees no irritation when applied. A homogeneity

analysis showed that the ingredients were distributed uniformly and that there was no discernible creaming or sedimentation. With a measured viscosity of 60 cP, the serum is perfect for topical applications because it is easy to apply and adheres well to the skin. The average globule size, as determined by optical microscopy, was around 3.5 µm, with homogeneous spherical particles. This uniformly tiny particle size improves skin absorption and adds to the serum's overall aesthetic attractiveness. A total bacterial count of about 80 CFU/mL and a fungal count of 60 CFU/mL were found by microbiological investigation; both counts were within permissible bounds, indicating the product's microbiological safety. Strong physical and chemical stability was demonstrated by stability investigations, which included centrifugation and accelerated stability testing, which verified that the serum maintained its organoleptic and physicochemical characteristics over time. The structural integrity of the formulation was supported by the centrifugal test, which revealed no phase separation. The serum is safe for human usage and non-irritating, as evidenced by the absence of any allergic reaction, redness, or irritation symptoms. Using the DPPH radical scavenging method, the serum also showed a considerable antioxidant activity of about 40.71%, suggesting potential for reducing oxidative stress and aiding in anti-aging benefits. The 10% niacinamide serum is a promising formulation with desired cosmetic and therapeutic effects, according to the study's overall findings. It provides a steady, non-irritating, and efficient way to treat a variety of skin issues, including pigmentation, microbial infections, excess sebum, and aging symptoms. Future research and commercialization of this formulation in the cosmeceutical sector are highly promising. To confirm its long-term safety and effectiveness, more clinical trials with a larger population are advised.

References

1) American Academy of Dermatology Association. 10 Skin Care Secrets for Healthier Looking Skin*(https://www.aad.org/public/everyday-care/skin-care-secrets/routine/healthier-looking-skin)*. Accessed 3/16/2021.
2) American Academy of Dermatology Association. What Kids Should Know About Skin*(https://www.aad.org/public/parents-kids/healthy-habits/parents/kids/about-skin)*. Accessed 3/16/2021.
3) National Cancer Institute. Layers of the Skin*(https://training.seer.cancer.gov/melanoma/anatomy/layers.html)*. Accessed 3/16/2021.
4) National Institute of Arthritis and Musculoskeletal and Skin Disease. Healthy Skin Matters *(https://www.niams.nih.gov/health-topics/kids/healthy-skin)*. Accessed 3/16/2021.
5) National Institute on Aging. Skin Care and Aging*(https://www.nia.nih.gov/health/skin-care-and-aging)*. Accessed 3/16/2021.
6) Alibardi, Lorenzo (15 August 2003). "Adaptation to the land: The skin of reptiles in comparison to that of amphibians and endotherm amniotes". *Journal of Experimental Zoology*.
7) Nasoori, Alireza (August 2020). "Formation, structure, and function of extra-skeletal bones in mammals". *Biological Reviews*. 95 (4): 986–1019.
8) Proksch E, Brandner JM, Jensen JM (2008). "The skin: an indispensable barrier". *ExpDermatol*.
9) Madison, Kathi C. (August 2003). "Barrier Function of the Skin: 'La Raison d'Être' of the Epidermis". *Journal of Investigative Dermatology*.
10) Thornton, M. J. (December 2002). "The biological actions of estrogens on skin: Estrogens and skin". *Experimental Dermatology*. 11 (6): 487–502. doi:10.1034/j.1600-0625.2002.110601.x. PMID 12473056.
11) Ashcroft, Gillian S.; Greenwell-Wild, Teresa; Horan, Michael A.; Wahl, Sharon M.; Ferguson, Mark W.J. (October 1999). "Topical Estrogen Accelerates Cutaneous Wound Healing in Aged Humans Associated with an Altered Inflammatory Response". *The American Journal of Pathology*. 155 (4): 1137–1146. doi:10.1016/S0002-9440(10)65217-0. PMC 1867002. PMID 10514397.
12) Oh, Desiree May; Phillips, Tania J. (2006). "Sex Hormones and Wound Healing". *Wounds*. 18 (1): 8–18.
13) "fur". Archived from the original on 3 March 2017. Retrieved 4 March 2017 – via The Free Dictionary.
14) Clarke, B. T. (August 1997). "The natural history of amphibian skin secretions, their normal functioning and potential medical applications". *Biological Reviews of the Cambridge Philosophical Society*.
15) 3.Göçeri E. 2020 Tenth International Conference on Image Processing Theory, Tools and Applications (IPTA) 2020. Impact of deep learning and smartphone technologies in dermatology: automated diagnosis;

16) Comparison between method of One factor at a time (OFAT) and Design of Experiment (DOE); A thesis submitted in fulfillment of the requirements for the award of the degree of Bachelor of hemical& Natural Resources Engineering University Malaysia Pahang; December 2010
17) Indian Dermatology Online Journal Vitamin C in dermatology PumoriSaokarTelang; Indian Dermatology Online Journal:Apr-Jun 2013 - Volume 4
18) Anti-Inflammatory and Skin Barrier Repair Effects of Topical Application of Some Plant Oils by Lily Zhong,andJuan Luis Santiago; DOI: 10.3390/ijms19010070; PMCID: PMC5796020;
19) EfficacyEvaluation of a Topical Hyaluronic Acid Serum in Facial Photoaging By Zoe Diana;
Isabell Diaz; Thomas Boyd PMID:34176098;PMCID:PMC8322246;DOI: 10.1007/13555021-00566-0; August 2021Antioxidants in Dermatology By Flavia
20) AlvimSantannaAddorPMCID:PMC5514576; PMID: 29180248; DOI:10.1590/abd1806-4841.20175697; 2017 May June PMID:29280987; Received2017 Oct 31; Accepted 2017 Dec 20.
21) Niacin: the Real Story1st edition ; Edition: 1st edition ; ISBN-13: 978-1591202752 ; Format: Paperback/softback ; Publisher: Turner Publishing Company (11/26/2015)
22 Chambers, E.S.; Vukmanovic-Stejic, M. Skin barrier immunity and ageing. Immunology 2020, 160, 116–125. [Google Scholar] [CrossRef] [PubMed]
23 Fisher, G.J.; Kang, S.; Varani, J.; Bata-Csorgo, Z.; Wan, Y.; Datta, S.; Voorhees, J.J. Mechanisms of photoaging and chronological skin aging. Arch. Dermatol. 2002, 138, 1462–1470. [Google Scholar] [CrossRef] [PubMed]
24 Rittie, L.; Fisher, G.J. Natural and sun-induced aging of human skin. Cold Spring Harb. Perspect. Med. 2015, 5, a015370. [Google Scholar] [CrossRef] [PubMed]
25 Tzaphlidou, M. The role of collagen and elastin in aged skin: An image processing approach. Micron 2004, 35, 173–177. [Google Scholar] [CrossRef]
26 Pizzino, G.; Irrera, N.; Cucinotta, M.; Pallio, G.; Mannino, F.; Arcoraci, V.; Squadrito, F.; Altavilla, D.; Bitto, A. Oxidative Stress: Harms and Benefits for Human Health. Oxid. Med. Cell. Longev. 2017, 2017, 8416763. [Google Scholar] [CrossRef]
27 Gu, Y.; Han, J.; Jiang, C.; Zhang, Y. Biomarkers, oxidative stress and autophagy in skin aging. Ageing Res. Rev. 2020, 59, 101036. [Google Scholar] [CrossRef] [PubMed]
28 Kammeyer, A.; Luiten, R.M. Oxidation events and skin aging. Ageing Res. Rev. 2015, 21, 16–29. [Google Scholar] [CrossRef]
29 Shah, A.A.; Sinha, A.A. Oxidative stress and autoimmune skin disease. Eur. J. Dermatol. 2013, 23, 5–13. [Google Scholar] [CrossRef]
30 Bickers, D.R.; Athar, M. Oxidative stress in the pathogenesis of skin disease. J. Investig. Dermatol. 2006, 126, 2565–2575. [Google Scholar] [CrossRef] [Green Version]
31 Awad, F.; Assrawi, E.; Louvrier, C.; Jumeau, C.; Giurgea, I.; Amselem, S.; Karabina, S.A. Photoaging and skin cancer: Is the inflammasome the missing link? Mech. Ageing Dev. 2018, 172, 131–137. [Google Scholar] [CrossRef]

32 Boo, Y.C. Natural Nrf2 Modulators for Skin Protection. Antioxidants 2020, 9, 812. [Google Scholar] [CrossRef]
33 Baek, J.; Lee, M.G. Oxidative stress and antioxidant strategies in dermatology. Redox Rep. 2016, 21, 164–169. [Google Scholar] [CrossRef]
34 Hegyi, J.; Schwartz, R.A.; Hegyi, V. Pellagra: Dermatitis, dementia, and diarrhea. Int. J. Dermatol. 2004, 43, 1–5. [Google Scholar] [CrossRef]
35 Kirkland, J.B. Niacin Status, NAD Distribution and ADP-Ribose Metabolism. Curr. Pharm. Des. 2009, 15, 3–11. [Google Scholar] [CrossRef] [PubMed]
36 De Figueiredo, L.F.; Gossmann, T.I.; Ziegler, M.; Schuster, S. Pathway analysis of NAD^+ metabolism. Biochem. J. 2011, 439, 341–348. [Google Scholar] [CrossRef] [Green Version]
37 Mattiussi, A.J.; Blais, D. Niacin Versus Niacinamide. Can. Med Assoc. J. 1992, 147, 990–991. [Google Scholar]
38 MacKay, D.; Hathcock, J.; Guarneri, E. Niacin: Chemical forms, bioavailability, and health effects. Nutr. Rev.2012, 70, 357–366. [Google Scholar] [CrossRef]
39 Surjana, D.; Damian, D.L. Nicotinamide in dermatology and photoprotection. Skinmed 2011, 9, 360–365. [Google Scholar] [PubMed]