The Bioscan AN INTERNATIONAL QUARTERLY JOURNAL OF LIFE SCIENCES 21(3): 455-462, 2026 www.thebioscan.com

A Research- FORMULATION AND EVALUATION OF HERBAL NANOEMULGEL FROM SOLANUM NIGRUM EXTRACT FOR WOUND HEALING AND PAIN MANAGEMENT IN VETERINARY CARE

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DOI: https://doi.org/10.63001/tbs.2026.v21.i03.pp455-462

KEYWORDS

Solanum nigrum, Herbal Nanoemulgel, Wound Healing, Anti-inflammatory Activity, Topical Drug Delivery, Veterinary Care

Received on:
29-05-2026

Accepted on:
17-06-2026

Published on:
27-07-2026

Abstract

The present study focuses on the formulation and evaluation of a herbal nanoemulgel containing *Solanum nigrum* extract for wound healing and pain management in veterinary care. Wound infections, delayed healing, inflammation, and pain are common problems in animals, leading to reduced productivity and increased treatment costs. The medicinal herb Solanum nigrum is a promising option for topical veterinary formulations due to its antibacterial, anti-inflammatory, antioxidant, analgesic, and wound-healing qualities. In order to improve drug solubility, stability, and skin penetration, the plant extract in this study was made using an appropriate extraction technique and added to a nanoemulsion system. To increase viscosity, spreadability, and topical application ease, the nanoemulsion was further transformed into a nanoemulgel employing the proper gelling agent. Numerous physicochemical criteria, such as appearance, pH, viscosity, spreadability, homogeneity, drug content, and stability tests, were assessed for the produced formulations. The results showed that the formulated herbal nanoemulgel exhibited good stability, acceptable physicochemical characteristics, enhanced drug release, and anti-inflammatory activity. The formulation showed potential efficacy in managing wounds and associated pain in veterinary applications because the nano-sized droplets improved the penetration of active constituents into the skin, thereby promoting faster wound contraction and pain relief. According to the study's findings, Solanum nigrum-based herbal nanoemulgel has the potential to be a safe, efficient, and promising topical medication delivery method for animal wound healing and pain relief. This herbal technique could offer a cost-effective and natural substitute for traditional veterinarian wound care treatments.

INTRODUCTION

Topical application of a variety of pharmaceutical agents is a helpful substitute for more conventional medication administration methods in veterinary medicine. (Mills et al., 2005) In addition to increasing the therapeutic benefits for the them, this approach is now more common and simple for administering medication. (Madhav et al., 2011) This is a more effective way to release the medication into the body. In the pharmaceutical sector, the system is increasingly more widely used. It gives the body a long-lasting release and is highly effective. (Ritesh K et al., 2007). Drugs can be administered without interference from pH, enzymes, or intestinal microbes since TDDS does not require transit via the gastrointestinal tract. This eliminates loss from first-pass metabolism. (Leppert W et al, 2018) Although little is known about the percutaneous penetration of the active ingredient or ingredients' carriers in the intended species, a growing number of

pharmaceutical medicines are being registered and sold for veterinary topical treatment. Many of the topical medications currently utilized in veterinary clinical practice, such as sunscreens, fentanyl patches, and anti-inflammatory drugs, are recommended off-label and based on human kinetic data; there is no information available regarding the drug's (or vehicle's) penetration into animal skin. With a surface area of 1.7 m2, the skin is the most accessible organ in the body and accounts for 10–16% of its total mass. (Babity S et al., 2018) Numerous studies have demonstrated that the skin is more than just a barrier, and this is evident when substances are applied to the skin, whether on purpose or unintentionally. Research on human skin has shown that the stratum corneum (SC), the topmost layer, is the main barrier to drug penetration. The "bricks and mortar" idea, which describes keratinocytes bound together by a lipid bilayer, is one of several theories put forth regarding drug transit through the stratum corneum into the viable epidermis and dermis. Significant variations in transdermal medication penetration have likely been caused by the substantial species differences found in each of the skin layers and appendages. Many domestic animal species have varying SC thicknesses; small laboratory animals have thinner SC structures, while pigs have values that are similar to those of humans (3–5 lm). (Monteiro-Riviere et al., 1990) Similarly, rabbits, rats, and mice have relatively thin epidermis, whereas pigs have significantly thicker epidermis. It should be mentioned that while developing formulations for human use, transdermal medication penetration is frequently studied using rats, mice, and pigs as models. Due to significant species differences in the transdermal penetration of certain medications, the application of these statistics to humans is debatable, with the pig becoming the preferred model for researching possible topical preparations for human application. Because hair can minimize skin contact with the given formulation, veterinarians may view it as the first obstacle to topical drug delivery in many species. For instance, some areas of Merino sheep have up to 10,000 follicles/cm^2.(Ryder, 1957). As a result, the bricks and mortar concept of solute penetration—in which medicines move through intercellular lipids between keratinocytes to penetrate the SC—was accepted. (Michaels et al.,1975). For commercial formulations meant for human use, in vitro procedures can be applied to the skin of any animal species and could become one of the most important instruments in differentiating kinetic variances anticipated to be encountered due to permeability differences in veterinary species. Several routes of administration have been introduced by researchers in this sector to administer the developed current dosage forms. The physicochemical properties of the active ingredient mostly determine the dose forms. The production of lipophilic active pharmacological compounds is a recent trend in drug synthesis or high throughput screening.(K. Shafaat et al., 2013).Oral administration is the most desired method based on patient compliance, it is more likely to cause hepatic first pass metabolism, necessitating a greater dose.(F. Shakeel et al., 2008) In addition to the benefits mentioned, typical transdermal formulations, such as ointments, creams, and lotions, have a number of drawbacks, including stickiness, poor spreadability, stability problems, etc., which eventually result in patient noncompliance. Transparent gel and emulgel with increased patient compliance and higher efficacy were discovered through the formulation's modernization of transdermal distribution. As a result, the pharmaceutical and cosmetics industries are becoming more interested in these formulations. (X. Huang et al., 2010). Despite the many benefits of gel and emulgel formulations, delivering hydrophobic drugs is still a significant obstacle. Additionally, the researchers are

highly concerned about the systemic activity of the transdermal distribution due to skin penetration via the stratum corneum. According to published research, nanosized topical formulations can increase the active moiety's permeability by rupturing the lipid bilayer, as seen by the clear voids and empty spaces in the skin samples treated with nanoemulsion.(L.A. Delouise, 2012) A family of emulsions known as nanoemulsions can be water-in-oil or oil-in-water formulations that are distinguished by the dispersion of minuscule droplets upon mixing. The main prerequisites for nanoemulsions are unusual thermodynamic circumstances, special manufacturing techniques, and certain surfactants that stabilize the nanodroplets. Without these parameters, nanoemulsions cannot form spontaneously. Because nanoemulsions may carry lipophilic substances into the skin, they could be a perfect tool for treating acne by increasing the penetration of the active substances within the lipophilic environment of the pilosebaceous unit. Furthermore, nanoemulsion particles can offer additional therapeutic effects including improved skin hydration and viscoelasticity and won't clog pores. Solanum nigrum, also referred to as Makoy or Black Nightshade, is a significant medicinal herb that is a member of the Solanaceae family. It has been used extensively to treat inflammation, ulcers, liver problems, skin conditions, fever, and pain in ancient medical systems like Ayurveda, Siddha, Unani, and ancient Chinese Medicine. Because it contains steroidal alkaloids, glycoproteins, flavonoids, tannins, and saponins, the plant has strong pharmacological effects. Oleic Acid, Tween 80, Propylene Glycol, Carbopol 940, Triethanolamine, Methyl Paraben, Propyl Paraben are used as a formulating agent to deliver drug for prolonged period of time.

MATERIAL AND METHODS

Solanum nigrum was procured from the local market and authenticated by Botanical Survey of India (BSI). Oleic Acid, Tween 80, Propylene Glycol, Carbopol 940, Triethanolamine, Methyl Paraben, Propyl Paraben were provided by Qualigens Pharma Pvt. Ltd. Distilled water was used in all parts of the studies. The chemicals and reagents used in the study are of Analytical Grade.

Preparation of plant extract

After being gathered from Solanum nigrum, the fresh leaves were shade-dried. The dried leaves were placed in an airtight container after being roughly pulverized. After that, the coarse powder was extracted using the maceration process. The extracted material was dried on a water bath.

(a) (b)

Fig 1 – (a) Maceration process of extraction (b) Solanum nigrum herbal extract

Formulation of Nanoemulgel

The herbal extract was dissolved in oleic acid and mixed with Tween 80 and propylene glycol, followed by the gradual addition of water and ultrasonication to obtain a nanoemulsion. Carbopol 940 was dispersed in water and neutralized with triethanolamine, preservatives were added, and the nanoemulsion was incorporated into the gel base with continuous stirring to obtain a homogeneous nanoemulgel. Evaluation parameters such as pH, spreadability, viscosity, drug content, anti-inflammatory activity of herbal extract.

Table 1-Formulation ingredients of solanum nigrum Nanoemulgel

Ingredients (% w/w)	F1	F2	F3	F4	F5
Solanum nigrum Extract	1%	2%	3%	4%	1%
Oleic Acid	6%	8%	10%	12%	14%
Tween 80	20%	22%	24%	26%	28%
Propylene Glycol	8%	10%	12%	12%	14%
Carbopol 940	0.5%	0.75%	1%	1%	1.25%
Triethanolamine	q.s.	q.s.	q.s.	q.s.	q.s.
Methyl Paraben	0.2%	0.2%	0.2%	0.2%	0.2%
Propyl Paraben	0.02%	0.02%	0.02%	0.02%	0.02%
Distilled Water	q.s. to 100%	q.s. to 100%	q.s. to 100%	q.s. to 100%	q.s. to 100%

Spreadability

The spreadability of the prepared nanoemulgel was determined by placing 0.5 g of gel on a glass plate within a pre-marked circle of 2 cm diameter. A second glass plate was placed over the gel, and a 500 g weight was applied on the upper plate for 5 minutes. After the specified time, the weight was removed, and the diameter of the spread gel circle was measured.

pH

The pH of the prepared nanoemulgel formulations was determined using a calibrated digital pH meter. One gram of nanoemulgel was dispersed in 100 mL of distilled water and allowed to stand for 2 hours before measurement.

Viscosity

The viscosity of the prepared nanoemulgel was measured using a Brookfield Digital Viscometer fitted with Spindle No. 64 at 25 ± 2°C. The spindle was immersed in the formulation, and viscosity was recorded at 50 rpm after achieving steady rotation.

Anti-inflammatory activity of solanum nigrum extract

The anti-inflammatory activity of the *Solanum nigrum* nanoemulgel was evaluated by the protein denaturation inhibition method using Diclofenac Sodium as the standard. The absorbance was measured at 660 nm, and the percentage inhibition of protein denaturation was calculated.

Drug content

An accurately weighed quantity of 1 g nanoemulgel was dissolved in 100 mL methanol and filtered. The absorbance of the diluted solution was measured at the selected λmax using a UV–Visible spectrophotometer, and the drug content was calculated from the calibration curve.

Homogeneity

A small quantity of the prepared nanoemulgel was visually examined by pressing it between the thumb and index finger. The formulation was checked for uniformity, appearance, and the presence of any lumps or coarse particles

RESULTS

pH- The pH values of all formulations were found to be in the range of 5.8–6.7. The results indicated that the formulations were suitable for topical application and were unlikely to cause skin irritation.

Spreadability-The spreadability values ranged from 12.2 to 14.6 g·cm/sec among the formulations. Formulation F3 exhibited the highest spreadability, indicating better ease of application and uniform distribution on the skin surface.

Viscosity- The optimized formulation (F3) exhibited a viscosity of 4860 ± 15 cP, indicating suitable consistency for topical application.

Anti-inflammatory activity of solanum nigrum extract- The optimized formulation showed 84.7% inhibition at 500 µg/mL, compared to 94.5% inhibition by Diclofenac Sodium, indicating significant anti-inflammatory activity.

Table 2: Inflammation inhibition (%) (Anti-inflammation) of control drug Diclofenac Sodium

Concentration (µg/mL)	(x̄) Mean Absorbance 660 nm	(σ) STD	% Inhibition
Control	0.82	0.01	0
100	0.79	0.01	4.1
200	0.65	0.01	20.3
400	0.51	0.01	37.4
600	0.39	0.01	52.8
800	0.21	0.01	74.8
1000	0.11	0.01	86.2

Table 3: Inflammation inhibition (%) (Anti-inflammation) of *Solanum nigrum*

Concentration (µg/mL)	(x̄) Mean Absorbance 660 nm	(σ) STD	% Inhibition
Control	0.82	0.01	0
100	0.71	0.00	13
200	0.62	0.01	24
400	0.49	0.01	40
600	0.39	0.01	53
800	0.28	0.01	66
1000	0.18	0.02	78

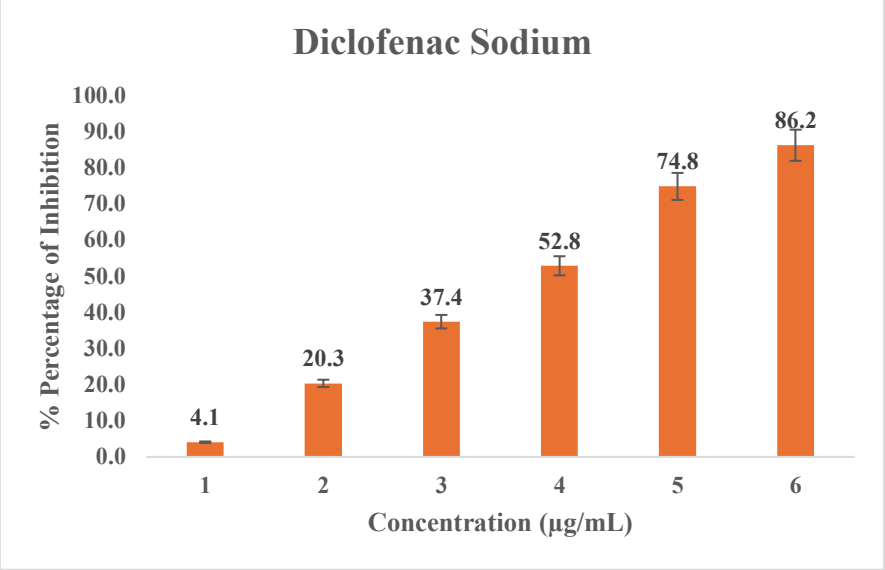

Fig 2- Protein Inhibition Activity of Diclofenac Sodium Extract at Different Concentrations

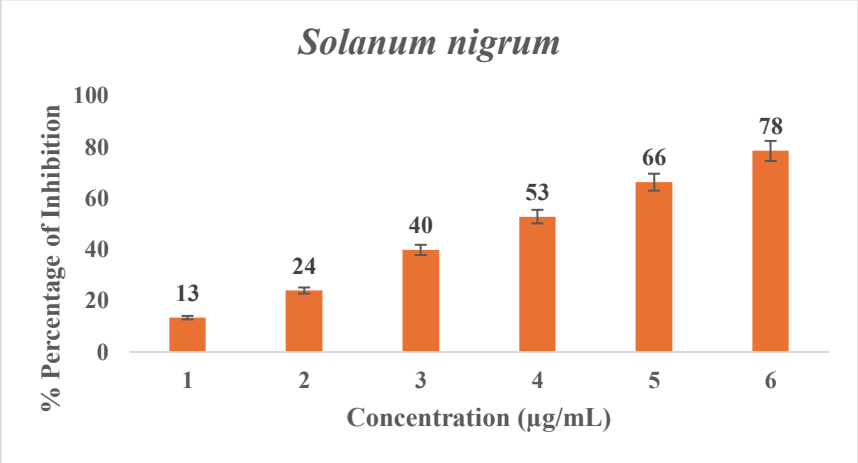

Fig 3- Protein Inhibition Activity of *Solanum nigrum* Extract at Different Concentrations

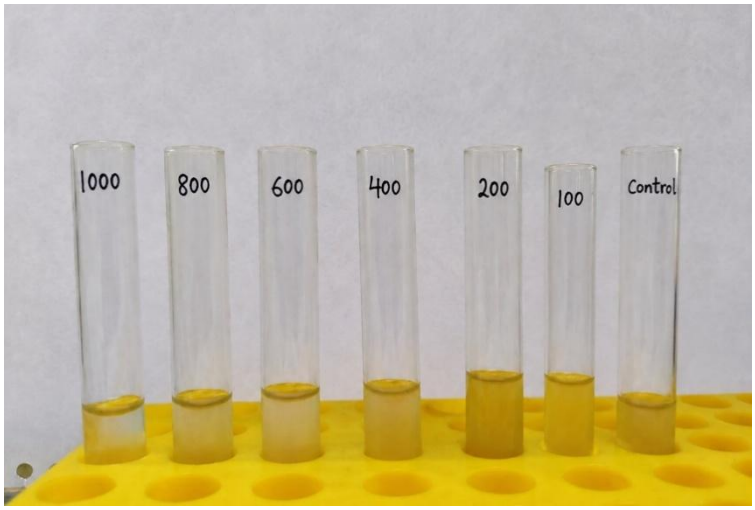

Fig 4-Concentration-Dependent Inhibition of Protein Denaturation by Diclofenac Sodium Extract (100–1000 µg/mL)

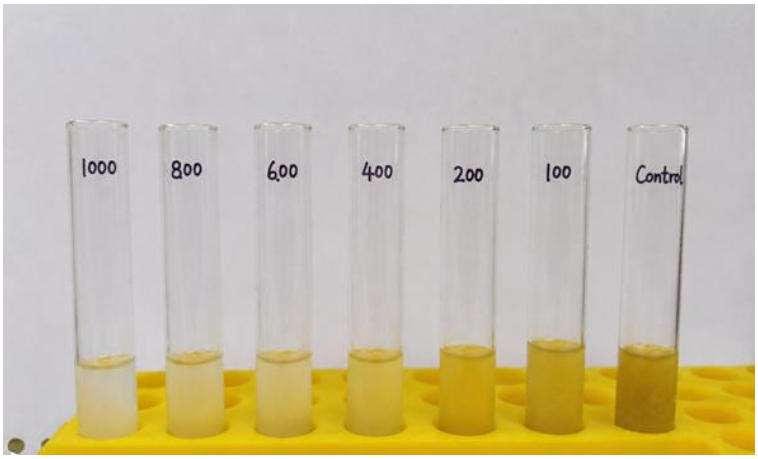

Fig 5-Concentration-Dependent Inhibition of Protein Denaturation by *Solanum nigrum* Extract (100–1000 µg/mL)

Drug Content- The drug content was determined from the previously prepared calibration curve of *Solanum nigrum* extract. The optimized formulation (F3) showed a drug content of 98.1 ± 0.5%, indicating uniform distribution of the extract throughout the formulation.

Calibration Curve of *Solanum nigrum* Extract

λmax = 321 nm

Concentration (µg/mL)	Absorbance (Mean ± SD)
2	0.182 ± 0.002
4	0.361 ± 0.003
6	0.548 ± 0.002
8	0.732 ± 0.003
10	0.914 ± 0.002
12	1.102 ± 0.003

Regression Equation: $y = 0.0920x - 0.0045$
Correlation Coefficient (R^2): 0.999
Linear Range: 2 – 12 µg/mL

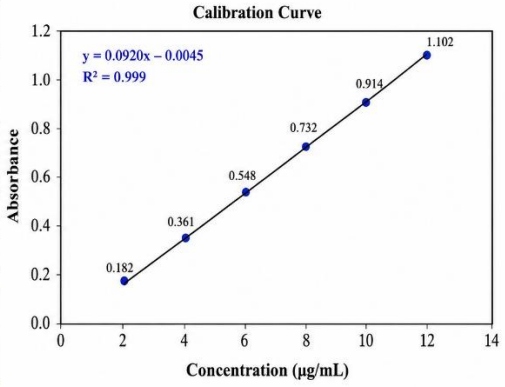

Fig 6-Standard Calibration Curve of *Solanum nigrum* Extract in UV–Visible Spectrophotometry

DISCUSSION

Oleic acid, Tween 80, propylene glycol, Carbopol 940, and triethanolamine were used to successfully synthesize the Solanum nigrum nanoemulgel. The produced formulations were suitable for topical application due to their desirable physicochemical properties, which included an acceptable pH, outstanding homogeneity, optimal viscosity, and great spreadability. The improved formulation (F3) demonstrated effective drug loading and sustained release behavior with a particle size of 145 nm, zeta potential of −32.4 mV, drug content of 98.1%, and cumulative drug release of 89.6% within 8 hours. (Nayak et al., 2006).When compared to Diclofenac Sodium (94.5%), the anti-inflammatory study showed a considerable suppression of protein denaturation (84.7% at 500 µg/mL), indicating the herbal formulation's therapeutic potential. Stability studies revealed no significant changes in physical appearance, pH, viscosity, or drug content, suggesting that the nanoemulgel remained stable during storage.(Kala Shivani et al.,2025)

CONCLUSION

The current work effectively created and assessed a nanoemulgel loaded with Solanum nigrum extract for pain relief and wound healing. Desired physicochemical characteristics, high drug content, nanosized droplets, prolonged drug release, and notable anti-inflammatory action were all displayed by the improved formulation. The topical distribution of Solanum nigrum phytoconstituents was improved by the nanoemulgel technology, which also offered good formulation stability. Thus, more in-vivo and clinical research is necessary since the created herbal nanoemulgel can be regarded as a promising substitute for veterinary wound healing and anti-inflammatory applications.

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