To formulate and evaluate the *Carissa carandas* leaves extract cream for the management of diabetic foot ulcer.

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Abstract

The primary aim of this research was to develop and evaluate a topical herbal cream containing Carissa carandas leaf extract for the management and potential management of diabetic foot ulcers (DFUs). Given the plant's known medicinal properties—especially its antioxidant, anti-inflammatory, and antimicrobial effects—the research aimed to assess its suitability as a complementary medicinal substance that promotes wound healing in diabetic patients.

The research was conducted through Plant Collection and Extraction Carissa carandas leaves) which were collected and extracted using ethanol via maceration. Phytochemical Analysis

The extract underwent qualitative and quantitative phytochemical screening, including:

DPPH radical scavenging assay to evaluate antioxidant activity.

Ursolic acid quantification using spectrophotometric and chromatographic methods.

Three topical cream formulations (F1, F2, F3) were prepared using the emulsification method. These included varying concentrations of the plant extract in an oil-in-water base.

Evaluation of Formulations-Each formulation was subjected to a range of tests to assess:

Physical characteristics (color, texture, odor, pH), Irritancy (applied on human skin and checked for erythema or edema), Washability, Viscosity (using a viscometer), Spreadability, Phase separation, Greasiness

The DPPH assay showed that the Carissa carandas extract had high antioxidant properties, comparable to standard ascorbic acid, suggesting high free radical scavenging capacity. The extract exhibited a high concentration of ursolic acid, a triterpenoid compound with known wound-healing, anti-inflammatory, and antimicrobial effects.

Formulation Properties include All three formulations showed: Faint green color, smooth texture, and pleasant odor.. pH values between 7.94 and 7.97—suitable for skin application. No signs of phase separation and high stability over 30 hours.Non-greasy nature and good washability.High viscosity (ranging from 24,865 to 33,371 cP), with F1 having the highest viscosity. Effective spreadability, with F2 being the most spreadable. No signs of skin irritation on topical application

The results indicate that Carissa carandas is a promising botanical candidate for formulating herbal creams targeted at diabetic foot ulcers. The high antioxidant properties and significant ursolic acid content of the extract sample support its efficacy in promoting wound healing. Ursolic acid may enhance collagen synthesis, reduce inflammation, and support angiogenesis—key processes impaired in diabetic wound healing.

The formulated creams demonstrated excellent physical stability, non-irritant behavior, and good usability qualities, which make them suitable for topical application. Their non-greasy and easily spreadable nature further enhances patient compliance.

These results are in line with previous studies that highlight the contribution of oxidative stress and chronic inflammation in diabetic complications, particularly foot ulcers. Herbal formulations targeting these mechanisms may serve as effective adjuncts to conventional therapy.

However, limitations exist. The study did not assess the clinical efficacy of the cream in human diabetic patients or the bioavailability of active constituents upon skin application. Additionally, standardization and long-term stability of the herbal extract need further exploration.

Key words: *Carissa carandas*, diabetes foot ulcer, cream, anti oxidant activity, anti inflammatory activity

1. Diabetes, also known as diabetes mellitus, is a metabolic disease marked by hyperglycemia, or increased blood glucose levels. (Das, Roy, & Mishra) In order to move blood sugar into the body's cells, where it can be stored or used as fuel, pancreatic β-cells generate insulin. However, the pancreas either produces insufficient amounts of insulin or the body becomes unable of using the insulin that is generated in a person with diabetes. (organization)

Diabetes is divided into the following categories (level of hyperglycemia):

- Type 1 diabetes is seen in 10% of diabetics, which results in cell loss and the pancreas' production of insulin.
- When insulin resistance develops in elevated blood sugar levels in type 2 diabetes.

Because the placenta produces substances that inhibit insulin, gestational diabetes develops throughout pregnancy

.**Aetiopathogenesis:**

The three classic symptoms of DFU are infection, ischemia, and neuropathy. Diminished local angiogenesis, diminished peripheral blood flow, and diminished cell and growth factor response are merely some of the factors that participate to the compromised metabolic processes in diabetes mellitus, which also increase the possibility of infection and poor wound healing. (Brem H) Therefore, the feet are susceptible to gangrene, deformities, ulcerations, peripheral vascular disease, and injury to peripheral nerves.

Vasculopathy:

In peripheral arteries, hyperglycemia results in abnormalities of smooth cells and malfunction of endothelial cells. Nitric oxide, which is produced by endothelial cells, dilates blood arteries and shields them from endogenous damage. Nitric oxide's physiological characteristics, which typically control endothelium homeostasis, anticoagulation, leukocyte attachment and the growth of smooth muscle cells, and antioxidant capacity, are thus disrupted in hyperglycemia. Vasodilators and nitric oxide produced by the endothelium are reduced, which causes blood vessel constriction and a higher likelihood of atherosclerosis, which ultimately results in ischemia. (Creager MA, 2003) (BB, 2008) Additionally, ischemia can occur even when there are detectable pedal pulses present. Arteriolar-venular shunting also disrupts the microcirculation, lowering blood flow to the necrotic area. (Jeffcoate WJ H. K.-1., 2003)

Neuropathy:

Almost 60% of foot ulcers are caused by neuropathy, which can occur in patients with both type 1 and 2 diabetes mellitus. (Clayton W, 2009) When blood sugar levels increase, more enzymes like sorbitol dehydrogenase and aldose reductase are produced. These enzymes change glucose into fructose and sorbitol. The buildup of these sugar compounds reduces the production of myoinositol in nerve cells, which impacts nerve transmission. Moreover, reversible metabolic, immunologic, and ischemic damage to autonomic, motor, and sensory nerves results from hyperglycemia-induced microangiopathy. (Younger DS, 1998) Peripheral sensation is diminished consequently of this impairment of the nerve innervations of the small muscles in the foot and the sensitive vasomotor regulation of the pedal circulation. (Jeffcoate WJ H. K.-1., 2003) Additionally, diabetes mellitus impacts the autonomic nerve system, causing skin to become dry and fissured, which increases the risk of infection. Moreover, the autonomic nervous system controls the skin's microcirculation. In the end, these alterations lead to the emergence of ulcers, gangrene, and limb loss. (Vinik AI, 2003), (Boyko EJ, 1999)

Classifications of Diabetic foot

The Wagner-Ulcer Classification System and the University of Texas Wound Classification are the two most often used classification schemes. (FW, 1981), (Lavery LA, 1996)

The University of Texas Wound Classification is a straightforward categorization that considers the stage (the presence or absence of infection and ischemia) and grade (the depth of the lesion). The "grade" goes from 0 (totally epithelized lesion before or after ulceration) to III (bone or joint involvement). A indicates that neither infection nor ischemia is present, B indicates infection, C indicates ischemia, and D indicates both infection and ischemia. The ultimate classification is obtained by combining the "grade and stage."

Grade 0	No ulcer in a high risk foot.
Grade 1	Superficial ulcer involving the full skin thickness but not underlying tissues.
Grade 2	Deep ulcer, penetrating down to ligaments and muscle,but no bone involvement or abscess formation.
Grade 3	Deep ulcer with cellulitis or abscess formation, often with osteomyelitis.
Grade 4	Localized gangrene.
Grade 5	Extensive gangrene involving the whole foot

Table :1.4.1 Wagner Classification of Diabetic Foot Ulcer (FW, 1981)

Stages	Grade			
	0	I	II	III
A	pre- or post-ulcerative completely epithelized lesion	superficial wound capsule	Wound penetration upto tendon or	Wound penetration upto bone or joint
B	Infection	Infection	Infection	Infection
C	Ischaemia	Ischaemia	Ischaemia	Ischaemia
D	Infection and Ischaemia	Infection and Ischaemia	Infection and Ischaemia	Infection and ischaemia

Table no.: 1.4.2 The University of Texas wound classification system. (Lavery LA, 1996)

1.7.2 Composition of creams:

Oil phase: provides emollient qualities and acts as a vehicle for substances that are lipophilic, or soluble in oil.

Aqueous phase: functions as a solvent for hydrophilic (water-soluble) components and supplies hydration.

Active ingredients: Offer medicinal benefits like anti-aging, anti-inflammatory, antioxidant, or moisturizing qualities.

Emulsifiers: Maintain the water-oil interface's stability and avoid phase separation.

Excipients: Improve the formulation of the cream's viscosity, stability, texture, and sensory qualities.

1.7.3 Advantages of Herbal Cream (de Koos PT, 1983), (Samim M, 2012)

1. Ease of Application: Creams are usually spreadable and smooth, which makes it simple to apply them to the skin without leaving a greasy residue or creating irritation.
2. Stability: Creams that are properly made can last a long period without losing their effectiveness. In contrast to other formulations like gels or lotions, they are less likely to oxidize or degrade.
3. Targeted Delivery: Localized disorders like psoriasis, acne, and eczema can be effectively treated using creams because they enable the precise distribution of active chemicals to certain skin regions.
4. Moisturizing Properties: Examples of substances that provide moisture include glycerin and hyaluronic acid, included in many lotions that help hydrate the skin and enhance its texture and look.
5. Versatility: Numerous active substances can be used into cream compositions, enabling customization to satisfy particular skincare requirements or therapeutic objectives.

1.7.4 Ideal Properties of Herbal Cream (Karaman S, 2014), (Kumar SW, 2013)

1. It must be in harmony with the pH of the skin.
2. It shouldn't have any harmful consequences when used.
3. It need to possess an ideal particle size.
4. It ought to have a nourishing impact.
5. It ought to be thicker than a lotion but yet holding its form, like a 50/50 oil and water emulsion.
6. It should be evenly distributed across the skin's surface.

Plant profile

Carissa congesta Wight, also known as Carissa carandas Auct. (previously commonly referred to as C. carandas L.), It is one of the Apocynaceae family members of dogbanes and is widely dispersed in India. (Deep, 1991) It is frequently used to Indian spices and pickles as a condiment. It is a hardy plant that can withstand drought and grows well in a variety of soil types. (Singh., 1965) Historically,this plant has been utilized to treat intestinal worms, biliousness, pruritus, scabies, and other conditions. Considerable biological activity that has been recorded include analgesic, anti-inflammatory, antipyretic, cardiotonic, and histamine-releasing properties. (C.S.I.R. Government of India, 1968.) (Singh., Dictionary of Economic Plants of India., 1965.)

4.2 Taxonomical Classification

Kingdom: Plantae
Divison: Magnoliophyta
Class: Magnoliopsida
Order: Gentianales
Family: Apocynaceae
Genus: Carissa
Species: C.carandas

4.1.1 Carissa carandas description

A perennial deciduous shrub belonging to the family Apocynaceae, Carissa carandas is referred to as karonda. It typically grows to a height of 2-4 meters. White latex abounds on the stem, and the branches have jagged spines. The tiny flowers are white and have a diameter of 3–5 cm. Berries are glabrous with 5 to 7 wings, woody and fibrous, and they are grouped in clusters of three to ten fruits with five, one hard angle curved upward. With numerous seeds, the fruit has a wide, ovoid, or globose shape. Different genotypes produce different colors of ripe fruit, such as white, green, or pinkish red. Three to five seeds per fruit, each flat, elliptical, blankish brown, and light in weight. January and February mark the beginning of flowering, and May and June mark the maturity of the fruits. For vegetable purposes, fruits are often gathered when still immature. (Youngken, 1950) , (Sumbul S, 2012) The conical, oblong leaves are 4-6 inches long and 2-3 inches wide, with a brown underside and a green top. This type of plant is known for its white, milky sap, which is visible if the leaves or stems are damaged. White or yellowish blooms are found in clusters. Gray, silky bark. The inner bark is crimson, velvety, and thick. (WC, 1996)

Chemical Constituents:

- Salicylic acid, an alkaloid, and 2-acetyl phenol, which has an odor like to that of Piper betel leaves, are among the volatile components found in the **root** (A. Zaki, 1983), (J. Pino, 2004). Carissone, carindone, carinol, lignin, undoside H, digitaloxigenin, glucose, D-digitalose, and 2 acetylphenol have all been identified in studies involving root materials. A mixture of sesqui terpenes, specifically carissone and carindone, has been isolated as a novel form of C31 terpenoid as a result of certain investigations. (B. Singh and R. P. Rastogi, 1972), (J. Reisch, 1990) (Joshi DV, 1957)
- In **stem bark**, alkaloids are found.
- Triterpenes, tannins, ursolic acid, and carissic acid have all been found in **leaves**. (S. Siddiqui, 2003), (Z. Naim, 1988) A study of fresh leaves revealed four pentacyclic triterpenoids, including two previously unidentified compounds and a novel ingredient called carissin (3beta-hydroxy-27-E-feruloyloxyurs-12-en-28-oic acid).
- A variety of volatile principles are produced by **fruits**, including ß-caryophyllene, 2-phenyl ethanol, linalool, iso amyl alcohol, benzyl acetate, lupeol, oxalic, tartaric, citric, malic, malonic, and glycolic acids, glycine, alaline, phenyl alaline, cerine, glucose, galactose, and a new triterpenic alcohol (carissol, an epimer of α-amyrin). An analysis of the volatile taste components of fruits grown in Cuba produced 150 compounds in the fragrance concentrate, with beta-caryophyllene, isobutanol, and isoamyl alcohol being the main elements.
- Fatty acids, including palmitic, stearic, oleic, arachidic, and linoleic acids, are produced by **seeds**. The plant's polar glycoside underwent modest enzymatic hydrolysis, producing the sugars D-glucose and D digitalose as well as the undoside H and digitoxigenin. Additionally, C. congesta contains 13.0% crude protein, 7.8% polyphenols, 5.3% fixed oil, 58% hydrocarbons, and 31.4% free acid. This species' higher gross hest values suggest that it has the potential to be exploited as a fuel source.

Material and Methods:

Determination of Ursolic Acid Content:

Five grams of powdered plant material are extracted using methanol or ethanol and macerated for six to eight hours or refluxed for two to three hours in order to determine how much ursolic acid is there.After that, the extract sample is reconstituted in methanol, filtered through a 0.45 µm membrane filter, and concentrated using a rotary evaporator. To create working standards at different concentrations (e.g., 10, 20, and 50 µg/mL), a stock solution of ursolic acid standard (1 mg/mL in methanol) is made and diluted.(Janicsák et al., 2003)

In HPLC analysis, a C18 reversed-phase column is employed to detect UV light at 210–220 nm and a mobile phase made of acetonitrile and water (80:20) with 0.1% formic acid. The flow rate is 1.0 mL/min. The sample extract is examined to determine and measure the amount of ursolic acid based on retention duration and peak area after standard solutions are injected to produce a calibration curve.(Olszewska, 2008) As an alternative, spectrophotometric analysis uses the calibration curve to calculate the ursolic acid content by calculating the standard's absorbance and sample solutions at 210 nm. (Baricevic et al., 2001)The following formula is used to determine the ursolic acid content:

$$\text{Ursolic Acid Content (mg/g)} = \frac{Concentration \times Dilution\ Factor \times Extract\ Volume}{Sample\ Weight}$$

5.5. Formulation of cream:

S.No.	Ingredients	F1	F2	F3	F4	F5
1.	Beeswax	5.2 gm	5.45 gm	5.5 gm	4.97 gm	5.5 gm
2.	Liquid paraffin	18 ml	18.1 ml	19.2 ml	21 ml	20 ml
3.	Borax	0.4 gm	0.36 gm	0.5gm	0.56 gm	0.4 gm
4.	Methylparaben	0.02 gm	0.03 gm	0.3 gm	0.59 gm	0.4 gm
5.	Distilled water	q.s	q.s	q.s	q.s	q.s
6.	Rose oil	q.s	q.s	q.s	q.s	q.s
7.	*Carissa carandas* extract	2.8 ml	2.7 ml	1.5 ml	1.45 ml	1.40 ml

Table 5.3 : Formulation of Cream

1. **Heating of Oil Phase:** Heat the beeswax and liquid paraffin to 75°C in a borosilicate glass beaker and keep it there. The oil phase of the cream is made up of this.
2. **Preparation of Aqueous Phase:** Completely dissolve the borax and methylparaben in distilled water in a separate beaker. After that, the mixture should be heated to 75°C in

order to create a translucent solution. The cream's aqueous phase is created as a result.

3. **Combining Oil and Aqueous Phases:** With constant stirring, gradually incorporate the aqueous phase into the heated oil phase. This slow addition makes the emulsification process easier.
4. **Incorporation of extract:** When the mixture is emulsified, add the indicated amounts of Carissa carandas extract. To guarantee even dispersion of the herbal ingredients and a homogenous mixture, give it a good stir.
5. **Addition of Fragrance:** To add fragrance to the cream, add a few drops of rose oil to the recipe.
6. **Smoothing and Texture Adjustment:** After transferring the cream onto a slab, add a few drops of distilled water to change the texture as needed. To get a smooth consistency and guarantee that all of the components are thoroughly blended, mix the cream on the slab in a geometric pattern.(Muthukumarasamy et al., 2016)
7. **Slab Technique or Extemporaneous Method:** This technique, also referred to as the slab technique, entails manually combining the ingredients for the cream on a slab to guarantee even dispersion and appropriate integration of every component.(Simões et al., 2018)

5.6 Evaluation Parameters (DHYANI et al., 2019) (Dudhe et al., 2023)

1. Physical Evaluation: This include examining the cream's color, stability, texture, and odor.

2. Irritancy: This test determines if the cream's ingredients are bad for the mucosa or skin. To do this test, mark a spot on the dorsal surface of your left hand, apply the cream there, and record the time. Check the region for indications of irritability after a few minutes.

3. Washability: Applying a tiny bit of the cream to the hand and then washing it off with tap water to see how easy it comes off is how this test evaluates the cream's quality.

4. pH Test: This gauges the cream's acidity levels, which ought to be within the typical pH

4–7 range. A digital pH meter or pH paper is capable of measuring pH.

5. Phase Separation: Over the course of 24 to 30 hours, this test looks for component separation in the cream. Keep the cream out of direct sunlight and in a covered container between 30 and 80°C.

6. Viscosity: This test mainly verifies the cream's effectiveness by forecasting how its ingredients will operate in real-world applications.

7. Greasiness: This test establishes if the cream is greasy or oily. The formulations can be categorized as non-greasy according to the results.

8. Spreadability Test: Two glass slides with a small bit of cream in between were used as part of the spreadability test. The spreading action on the skin was then replicated by carefully moving

one glass slide horizontally over the other. A smoother movement and shorter duration indicated that the cream was more spreadable.

Spread ability= m × l/t

Where, m= Standard weight which is tied to or placed over the upper slide (30g) l= length of a glass slide (5 cm) t= time taken in second

6.1. Collection of plant was successfully done.

6.2. Extract preparation of *Carissa carandas* was successfully prepared.

6.3. Antioxidant Potential of Extract Through DPPH Assay:

Table 6.1 : DPPH Assay

Groups	***Radical scavenging activity (%)***				
	20(µg/ml)	***40(µg/ml)***	***60(µg/ml)***	***80(µg/ml)***	***100(µg/ml)***
Ascorbic Acid	**85.2**	**89.5**	**88.67**	**98.2**	**98.3**

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Carissa carandas	**88.7**	**91.81**	**95.47**	**96.23**	**97.99**

Carissa Carandas showed the better antioxidant properties when compared with the standard Ascorbic Acid.

6.4. Determination of Ursolic Acid Content:

Formulation	Concentration	Urosolic Content
F1	10 µg/mL	186.4
F2	20 µg/mL	212.1
F3	50 µg/mL	234.4

Table 6.2 : High urosolic content signifies the high medicinal property in the plant.

6.5 Formulation of Cream

Figure 6.1 : Cream formulation

4.6 Evaluation Parameter

4.6.1. Physical Evaluation:

Parameters	F1	F2	F3
Colour	Faint green	Faint green	Faint green
Odor	Pleasant	Pleasant	Pleasant
Texture	Smooth	Smooth	Smooth
State	Semi Solid	Semi Solid	Semi Solid

Table 6.3 : Physical evaluation

4.6.2. Irritancy Test:

Parameters	F1	F2	F3
Irritant effect	_	_	_
Erythema	_	_	_
Edema	_	_	_

Table 6.4 : Irritancy

4.6.3. Washability:

Formulation	Washability
F1	Easily washable
F2	Easily washable
F3	Easily washable

Table 6.5 : Washability

4.6.4. pH Test:

FORMULATION	pH
F1	7.96
F2	7.94
F3	7.97

Table 6.6 : pH

4.6.5: Phase Separation:

FORMULATION	Phase Separation
F1	Not observed
F2	Not observed
F3	Not observed

Table 6.7: phase sepeartion

4.6.7. Viscosity:

FORMULATION	Viscosity (cP)
F1	33371
F2	31689
F3	24865

Table 6.8: Viscosity

4.6.7. Greasiness:

FORMULATION	Greasiness
F1	Non greasy
F2	Non greasy
F3	Non greasy

Table 6.9 : Greasiness

4.6.7. Spreadability Test:

Formulation	Time (sec)	Spreadability
F1	7	2.14
F2	5	3
F3	6	2.5

Table 6.10 : spreadability

REFERENCES

1. Bahadoran, Z., Mirmiran, P., & Azizi, F. (2013). Dietary polyphenols as potential nutraceuticals in management of diabetes: A review. *Journal of Diabetes and Metabolic Disorders*, *12*(1), 1–9. https://doi.org/10.1186/2251-6581-12-43
2. Baricevic, D., Sosa, S., Della Loggia, R., Tubaro, A., Simonovska, B., Krasna, A., & Zupancic, A. (2001). Topical anti-inflammatory activity of Salvia officinalis L. leaves: the relevance of ursolic acid. *Journal of Ethnopharmacology*, *75*(2–3), 125–132. https://doi.org/10.1016/S0378-8741(00)00396-2

3. Caturano, A., D'Angelo, M., Mormone, A., Russo, V., Mollica, M. P., Salvatore, T., Galiero, R., Rinaldi, L., Vetrano, E., Marfella, R., Monda, M., Giordano, A., & Sasso, F. C. (2023). Oxidative Stress in Type 2 Diabetes: Impacts from Pathogenesis to Lifestyle Modifications. *Current Issues in Molecular Biology*, *45*(8), 6651–6666. https://doi.org/10.3390/cimb45080420
4. Chen, Z., Bertin, R., & Froldi, G. (2013). EC50 estimation of antioxidant activity in DPPH* assay using several statistical programs. *Food Chemistry*, *138*(1), 414–420. https://doi.org/10.1016/j.foodchem.2012.11.001
5. de Paulo Farias, D., de Araújo, F. F., Neri-Numa, I. A., & Pastore, G. M. (2021). Antidiabetic potential of dietary polyphenols: A mechanistic review. *Food Research International (Ottawa, Ont.)*, *145*, 110383. https://doi.org/10.1016/j.foodres.2021.110383
6. DHYANI, A., Chander, V., & Singh, N. (2019). Formulation and evaluation of multipurpose herbal cream. *Journal of Drug Delivery and Therapeutics*, *9*(2), 341–343. https://doi.org/10.22270/jddt.v9i2.2540
7. Dudhe, A. R., Choudhari, N., Dudhe, R., Katole, G., Mahajan, R., Pathak, N., Darode, A., Devnani, D., & Rd, H. (2023). *International Journal of Newgen Research in Pharmacy & Healthcare*. *1*, 129–134.
8. Ebrahimzadeh, A., Ebrahimzadeh, A., Mirghazanfari, S. M., Hazrati, E., Hadi, S., & Milajerdi, A. (2022). The effect of ginger supplementation on metabolic profiles in patients with type 2 diabetes mellitus: A systematic review and meta-analysis of randomized controlled trials. *Complementary Therapies in Medicine*, *65*(January), 102802. https://doi.org/10.1016/j.ctim.2022.102802
9. Gulcin, İ., & Alwasel, S. H. (2023). DPPH Radical Scavenging Assay. *Processes*, *11*(8). https://doi.org/10.3390/pr11082248
10. Janicsák, G., Veres, K., Kállai, M., & Máthé, I. (2003). Gas chromatographic method for routine determination of oleanolic and ursolic acids in medicinal plants. *Chromatographia*, *58*(5–6), 295–299. https://doi.org/10.1365/s10337-003-0058-y
11. Kolimi, P., Narala, S., Nyavanandi, D., Youssef, A. A. A., & Dudhipala, N. (2022). Innovative Treatment Strategies to Accelerate Wound Healing: Trajectory and Recent Advancements. *Cells*, *11*(15). https://doi.org/10.3390/cells11152439
12. Li, J., Bai, L., Wei, F., Zhao, J., Wang, D., Xiao, Y., Yan, W., & Wei, J. (2019). Therapeutic

Mechanisms of Herbal Medicines Against Insulin Resistance: A Review. *Frontiers in Pharmacology*, *10*, 661. https://doi.org/10.3389/fphar.2019.00661

13. Muthukumarasamy, R., Ilyana, A., Fithriyaani, N. A., & Najihah, N. A. (2016). *Formulation and evaluation of natural antioxidant cream comprising Formulation and Evaluation of Natural Antioxidant Cream Comprising Methanolic Peel Extract of Dimocarpus longan. September*.
14. Olszewska, M. (2008). Optimization and validation of an HPLC-UV method for analysis of corosolic, oleanolic, and ursolic acids in plant material: Application to Prunus serotina Ehrh. *Acta Chromatographica*, *20*(4), 643–659. https://doi.org/10.1556/AChrom.20.2008.4.10
15. Simões, A., Veiga, F., Vitorino, C., & Figueiras, A. (2018). A Tutorial for Developing a Topical Cream Formulation Based on the Quality by Design Approach. *Journal of Pharmaceutical Sciences*, *107*(10), 2653–2662. https://doi.org/10.1016/j.xphs.2018.06.010
16. Xie, J., & Schaich, K. M. (2014). Re-evaluation of the 2,2-diphenyl-1-picrylhydrazyl free radical (DPPH) assay for antioxidant activity. *Journal of Agricultural and Food Chemistry*, *62*(19), 4251–4260. https://doi.org/10.1021/jf500180u
17. Yee, H.-Y., Yang, J.-J., Wan, Y.-G., Chong, F.-L., Wu, W., Long, Y., Han, W.-B., Liu, Y.-L., Tu, Y., & Yao, J. (2019). [Molecular mechanisms of insulin resistance and interventional effects of Chinese herbal medicine]. *Zhongguo Zhong yao za zhi = Zhongguo zhongyao zazhi = China journal of Chinese materia medica*, *44*(7), 1289–1294. https://doi.org/10.19540/j.cnki.cjcmm.20181105.003
18. Bahadoran, Z., Mirmiran, P., & Azizi, F. (2013). Dietary polyphenols as potential nutraceuticals in management of diabetes: A review. *Journal of Diabetes and Metabolic Disorders*, *12*(1), 1–9. https://doi.org/10.1186/2251-6581-12-43
19. Baricevic, D., Sosa, S., Della Loggia, R., Tubaro, A., Simonovska, B., Krasna, A., & Zupancic, A. (2001). Topical anti-inflammatory activity of Salvia officinalis L. leaves: the relevance of ursolic acid. *Journal of Ethnopharmacology*, *75*(2–3), 125–132. https://doi.org/10.1016/S0378-8741(00)00396-2
20. Caturano, A., D'Angelo, M., Mormone, A., Russo, V., Mollica, M. P., Salvatore, T., Galiero, R., Rinaldi, L., Vetrano, E., Marfella, R., Monda, M., Giordano, A., & Sasso, F. C. (2023). Oxidative Stress in Type 2 Diabetes: Impacts from Pathogenesis to Lifestyle Modifications. *Current Issues in Molecular Biology*, *45*(8), 6651–6666. https://doi.org/10.3390/cimb45080420

21. Chen, Z., Bertin, R., & Froldi, G. (2013). EC50 estimation of antioxidant activity in DPPH* assay using several statistical programs. *Food Chemistry*, *138*(1), 414–420. https://doi.org/10.1016/j.foodchem.2012.11.001
22. de Paulo Farias, D., de Araújo, F. F., Neri-Numa, I. A., & Pastore, G. M. (2021). Antidiabetic potential of dietary polyphenols: A mechanistic review. *Food Research International (Ottawa, Ont.)*, *145*, 110383. https://doi.org/10.1016/j.foodres.2021.110383
23. DHYANI, A., Chander, V., & Singh, N. (2019). Formulation and evaluation of multipurpose herbal cream. *Journal of Drug Delivery and Therapeutics*, *9*(2), 341–343. https://doi.org/10.22270/jddt.v9i2.2540
24. Dudhe, A. R., Choudhari, N., Dudhe, R., Katole, G., Mahajan, R., Pathak, N., Darode, A., Devnani, D., & Rd, H. (2023). *International Journal of Newgen Research in Pharmacy & Healthcare*. *1*, 129–134.
25. Ebrahimzadeh, A., Ebrahimzadeh, A., Mirghazanfari, S. M., Hazrati, E., Hadi, S., & Milajerdi, A. (2022). The effect of ginger supplementation on metabolic profiles in patients with type 2 diabetes mellitus: A systematic review and meta-analysis of randomized controlled trials. *Complementary Therapies in Medicine*, *65*(January), 102802. https://doi.org/10.1016/j.ctim.2022.102802
26. Gulcin, İ., & Alwasel, S. H. (2023). DPPH Radical Scavenging Assay. *Processes*, *11*(8). https://doi.org/10.3390/pr11082248
27. Janicsák, G., Veres, K., Kállai, M., & Máthé, I. (2003). Gas chromatographic method for routine determination of oleanolic and ursolic acids in medicinal plants. *Chromatographia*, *58*(5–6), 295–299. https://doi.org/10.1365/s10337-003-0058-y
28. Kolimi, P., Narala, S., Nyavanandi, D., Youssef, A. A. A., & Dudhipala, N. (2022). Innovative Treatment Strategies to Accelerate Wound Healing: Trajectory and Recent Advancements. *Cells*, *11*(15). https://doi.org/10.3390/cells11152439
29. Li, J., Bai, L., Wei, F., Zhao, J., Wang, D., Xiao, Y., Yan, W., & Wei, J. (2019). Therapeutic Mechanisms of Herbal Medicines Against Insulin Resistance: A Review. *Frontiers in Pharmacology*, *10*, 661. https://doi.org/10.3389/fphar.2019.00661
30. Muthukumarasamy, R., Ilyana, A., Fithriyaani, N. A., & Najihah, N. A. (2016). *Formulation and evaluation of natural antioxidant cream comprising Formulation and Evaluation of Natural Antioxidant Cream Comprising Methanolic Peel Extract of Dimocarpus longan*.

September.

31. Olszewska, M. (2008). Optimization and validation of an HPLC-UV method for analysis of corosolic, oleanolic, and ursolic acids in plant material: Application to Prunus serotina Ehrh. *Acta Chromatographica*, *20*(4), 643–659. https://doi.org/10.1556/AChrom.20.2008.4.10
32. Simões, A., Veiga, F., Vitorino, C., & Figueiras, A. (2018). A Tutorial for Developing a Topical Cream Formulation Based on the Quality by Design Approach. *Journal of Pharmaceutical Sciences*, *107*(10), 2653–2662. https://doi.org/10.1016/j.xphs.2018.06.010
33. Xie, J., & Schaich, K. M. (2014). Re-evaluation of the 2,2-diphenyl-1-picrylhydrazyl free radical (DPPH) assay for antioxidant activity. *Journal of Agricultural and Food Chemistry*, *62*(19), 4251–4260. https://doi.org/10.1021/jf500180u
34. Yee, H.-Y., Yang, J.-J., Wan, Y.-G., Chong, F.-L., Wu, W., Long, Y., Han, W.-B., Liu, Y.-L., Tu, Y., & Yao, J. (2019). [Molecular mechanisms of insulin resistance and interventional effects of Chinese herbal medicine]. *Zhongguo Zhong yao za zhi = Zhongguo zhongyao zazhi = China journal of Chinese materia medica*, *44*(7), 1289–1294. https://doi.org/10.19540/j.cnki.cjcmm.20181105.003
35. Bahadoran, Z., Mirmiran, P., & Azizi, F. (2013). Dietary polyphenols as potential nutraceuticals in management of diabetes: A review. *Journal of Diabetes and Metabolic Disorders*, *12*(1), 1–9. https://doi.org/10.1186/2251-6581-12-43
36. Baricevic, D., Sosa, S., Della Loggia, R., Tubaro, A., Simonovska, B., Krasna, A., & Zupancic, A. (2001). Topical anti-inflammatory activity of Salvia officinalis L. leaves: the relevance of ursolic acid. *Journal of Ethnopharmacology*, *75*(2–3), 125–132. https://doi.org/10.1016/S0378-8741(00)00396-2
37. Caturano, A., D'Angelo, M., Mormone, A., Russo, V., Mollica, M. P., Salvatore, T., Galiero, R., Rinaldi, L., Vetrano, E., Marfella, R., Monda, M., Giordano, A., & Sasso, F. C. (2023). Oxidative Stress in Type 2 Diabetes: Impacts from Pathogenesis to Lifestyle Modifications. *Current Issues in Molecular Biology*, *45*(8), 6651–6666. https://doi.org/10.3390/cimb45080420
38. Chen, Z., Bertin, R., & Froldi, G. (2013). EC50 estimation of antioxidant activity in DPPH* assay using several statistical programs. *Food Chemistry*, *138*(1), 414–420. https://doi.org/10.1016/j.foodchem.2012.11.001
39. de Paulo Farias, D., de Araújo, F. F., Neri-Numa, I. A., & Pastore, G. M. (2021). Antidiabetic potential of dietary polyphenols: A mechanistic review. *Food Research*

International (Ottawa, Ont.), *145*, 110383. https://doi.org/10.1016/j.foodres.2021.110383

40. DHYANI, A., Chander, V., & Singh, N. (2019). Formulation and evaluation of multipurpose herbal cream. *Journal of Drug Delivery and Therapeutics*, *9*(2), 341–343. https://doi.org/10.22270/jddt.v9i2.2540
41. Dudhe, A. R., Choudhari, N., Dudhe, R., Katole, G., Mahajan, R., Pathak, N., Darode, A., Devnani, D., & Rd, H. (2023). *International Journal of Newgen Research in Pharmacy & Healthcare*. *1*, 129–134.
42. Ebrahimzadeh, A., Ebrahimzadeh, A., Mirghazanfari, S. M., Hazrati, E., Hadi, S., & Milajerdi, A. (2022). The effect of ginger supplementation on metabolic profiles in patients with type 2 diabetes mellitus: A systematic review and meta-analysis of randomized controlled trials. *Complementary Therapies in Medicine*, *65*(January), 102802. https://doi.org/10.1016/j.ctim.2022.102802
43. Gulcin, İ., & Alwasel, S. H. (2023). DPPH Radical Scavenging Assay. *Processes*, *11*(8). https://doi.org/10.3390/pr11082248
44. Janicsák, G., Veres, K., Kállai, M., & Máthé, I. (2003). Gas chromatographic method for routine determination of oleanolic and ursolic acids in medicinal plants. *Chromatographia*, *58*(5–6), 295–299. https://doi.org/10.1365/s10337-003-0058-y
45. Kolimi, P., Narala, S., Nyavanandi, D., Youssef, A. A. A., & Dudhipala, N. (2022). Innovative Treatment Strategies to Accelerate Wound Healing: Trajectory and Recent Advancements. *Cells*, *11*(15). https://doi.org/10.3390/cells11152439
46. Li, J., Bai, L., Wei, F., Zhao, J., Wang, D., Xiao, Y., Yan, W., & Wei, J. (2019). Therapeutic Mechanisms of Herbal Medicines Against Insulin Resistance: A Review. *Frontiers in Pharmacology*, *10*, 661. https://doi.org/10.3389/fphar.2019.00661
47. Muthukumarasamy, R., Ilyana, A., Fithriyaani, N. A., & Najihah, N. A. (2016). *Formulation and evaluation of natural antioxidant cream comprising Formulation and Evaluation of Natural Antioxidant Cream Comprising Methanolic Peel Extract of Dimocarpus longan*. *September*.
48. Olszewska, M. (2008). Optimization and validation of an HPLC-UV method for analysis of corosolic, oleanolic, and ursolic acids in plant material: Application to Prunus serotina Ehrh. *Acta Chromatographica*, *20*(4), 643–659. https://doi.org/10.1556/AChrom.20.2008.4.10
49. Simões, A., Veiga, F., Vitorino, C., & Figueiras, A. (2018). A Tutorial for Developing a

Topical Cream Formulation Based on the Quality by Design Approach. *Journal of Pharmaceutical Sciences*, *107*(10), 2653–2662. https://doi.org/10.1016/j.xphs.2018.06.010

50. Xie, J., & Schaich, K. M. (2014). Re-evaluation of the 2,2-diphenyl-1-picrylhydrazyl free radical (DPPH) assay for antioxidant activity. *Journal of Agricultural and Food Chemistry*, *62*(19), 4251–4260. https://doi.org/10.1021/jf500180u
51. Yee, H.-Y., Yang, J.-J., Wan, Y.-G., Chong, F.-L., Wu, W., Long, Y., Han, W.-B., Liu, Y.-L., Tu, Y., & Yao, J. (2019). [Molecular mechanisms of insulin resistance and interventional effects of Chinese herbal medicine]. *Zhongguo Zhong yao za zhi = Zhongguo zhongyao zazhi = China journal of Chinese materia medica*, *44*(7), 1289–1294. https://doi.org/10.19540/j.cnki.cjcmm.20181105.003
52. Bahadoran, Z., Mirmiran, P., & Azizi, F. (2013). Dietary polyphenols as potential nutraceuticals in management of diabetes: A review. *Journal of Diabetes and Metabolic Disorders*, *12*(1), 1–9. https://doi.org/10.1186/2251-6581-12-43
53. Baricevic, D., Sosa, S., Della Loggia, R., Tubaro, A., Simonovska, B., Krasna, A., & Zupancic, A. (2001). Topical anti-inflammatory activity of Salvia officinalis L. leaves: the relevance of ursolic acid. *Journal of Ethnopharmacology*, *75*(2–3), 125–132. https://doi.org/10.1016/S0378-8741(00)00396-2
54. Caturano, A., D'Angelo, M., Mormone, A., Russo, V., Mollica, M. P., Salvatore, T., Galiero, R., Rinaldi, L., Vetrano, E., Marfella, R., Monda, M., Giordano, A., & Sasso, F. C. (2023). Oxidative Stress in Type 2 Diabetes: Impacts from Pathogenesis to Lifestyle Modifications. *Current Issues in Molecular Biology*, *45*(8), 6651–6666. https://doi.org/10.3390/cimb45080420
55. Chen, Z., Bertin, R., & Froldi, G. (2013). EC50 estimation of antioxidant activity in DPPH* assay using several statistical programs. *Food Chemistry*, *138*(1), 414–420. https://doi.org/10.1016/j.foodchem.2012.11.001
56. de Paulo Farias, D., de Araújo, F. F., Neri-Numa, I. A., & Pastore, G. M. (2021). Antidiabetic potential of dietary polyphenols: A mechanistic review. *Food Research International (Ottawa, Ont.)*, *145*, 110383. https://doi.org/10.1016/j.foodres.2021.110383
57. DHYANI, A., Chander, V., & Singh, N. (2019). Formulation and evaluation of multipurpose herbal cream. *Journal of Drug Delivery and Therapeutics*, *9*(2), 341–343. https://doi.org/10.22270/jddt.v9i2.2540
58. Dudhe, A. R., Choudhari, N., Dudhe, R., Katole, G., Mahajan, R., Pathak, N., Darode, A.,

Devnani, D., & Rd, H. (2023). *International Journal of Newgen Research in Pharmacy & Healthcare*. *1*, 129–134.

59. Ebrahimzadeh, A., Ebrahimzadeh, A., Mirghazanfari, S. M., Hazrati, E., Hadi, S., & Milajerdi, A. (2022). The effect of ginger supplementation on metabolic profiles in patients with type 2 diabetes mellitus: A systematic review and meta-analysis of randomized controlled trials. *Complementary Therapies in Medicine*, *65*(January), 102802. https://doi.org/10.1016/j.ctim.2022.102802
60. Gulcin, İ., & Alwasel, S. H. (2023). DPPH Radical Scavenging Assay. *Processes*, *11*(8). https://doi.org/10.3390/pr11082248
61. Janicsák, G., Veres, K., Kállai, M., & Máthé, I. (2003). Gas chromatographic method for routine determination of oleanolic and ursolic acids in medicinal plants. *Chromatographia*, *58*(5–6), 295–299. https://doi.org/10.1365/s10337-003-0058-y
62. Kolimi, P., Narala, S., Nyavanandi, D., Youssef, A. A. A., & Dudhipala, N. (2022). Innovative Treatment Strategies to Accelerate Wound Healing: Trajectory and Recent Advancements. *Cells*, *11*(15). https://doi.org/10.3390/cells11152439
63. Li, J., Bai, L., Wei, F., Zhao, J., Wang, D., Xiao, Y., Yan, W., & Wei, J. (2019). Therapeutic Mechanisms of Herbal Medicines Against Insulin Resistance: A Review. *Frontiers in Pharmacology*, *10*, 661. https://doi.org/10.3389/fphar.2019.00661
64. Muthukumarasamy, R., Ilyana, A., Fithriyaani, N. A., & Najihah, N. A. (2016). *Formulation and evaluation of natural antioxidant cream comprising Formulation and Evaluation of Natural Antioxidant Cream Comprising Methanolic Peel Extract of Dimocarpus longan*. *September*.
65. Olszewska, M. (2008). Optimization and validation of an HPLC-UV method for analysis of corosolic, oleanolic, and ursolic acids in plant material: Application to Prunus serotina Ehrh. *Acta Chromatographica*, *20*(4), 643–659. https://doi.org/10.1556/AChrom.20.2008.4.10
66. Simões, A., Veiga, F., Vitorino, C., & Figueiras, A. (2018). A Tutorial for Developing a Topical Cream Formulation Based on the Quality by Design Approach. *Journal of Pharmaceutical Sciences*, *107*(10), 2653–2662. https://doi.org/10.1016/j.xphs.2018.06.010
67. Xie, J., & Schaich, K. M. (2014). Re-evaluation of the 2,2-diphenyl-1-picrylhydrazyl free radical (DPPH) assay for antioxidant activity. *Journal of Agricultural and Food Chemistry*, *62*(19), 4251–4260. https://doi.org/10.1021/jf500180u

68. Yee, H.-Y., Yang, J.-J., Wan, Y.-G., Chong, F.-L., Wu, W., Long, Y., Han, W.-B., Liu, Y.-L., Tu, Y., & Yao, J. (2019). [Molecular mechanisms of insulin resistance and interventional effects of Chinese herbal medicine]. *Zhongguo Zhong yao za zhi = Zhongguo zhongyao zazhi = China journal of Chinese materia medica*, *44*(7), 1289–1294. https://doi.org/10.19540/j.cnki.cjcmm.20181105.003
69. Bahadoran, Z., Mirmiran, P., & Azizi, F. (2013). Dietary polyphenols as potential nutraceuticals in management of diabetes: A review. *Journal of Diabetes and Metabolic Disorders*, *12*(1), 1–9. https://doi.org/10.1186/2251-6581-12-43
70. Baricevic, D., Sosa, S., Della Loggia, R., Tubaro, A., Simonovska, B., Krasna, A., & Zupancic, A. (2001). Topical anti-inflammatory activity of Salvia officinalis L. leaves: the relevance of ursolic acid. *Journal of Ethnopharmacology*, *75*(2–3), 125–132. https://doi.org/10.1016/S0378-8741(00)00396-2
71. Caturano, A., D'Angelo, M., Mormone, A., Russo, V., Mollica, M. P., Salvatore, T., Galiero, R., Rinaldi, L., Vetrano, E., Marfella, R., Monda, M., Giordano, A., & Sasso, F. C. (2023). Oxidative Stress in Type 2 Diabetes: Impacts from Pathogenesis to Lifestyle Modifications. *Current Issues in Molecular Biology*, *45*(8), 6651–6666. https://doi.org/10.3390/cimb45080420
72. Chen, Z., Bertin, R., & Froldi, G. (2013). EC50 estimation of antioxidant activity in DPPH* assay using several statistical programs. *Food Chemistry*, *138*(1), 414–420. https://doi.org/10.1016/j.foodchem.2012.11.001
73. de Paulo Farias, D., de Araújo, F. F., Neri-Numa, I. A., & Pastore, G. M. (2021). Antidiabetic potential of dietary polyphenols: A mechanistic review. *Food Research International (Ottawa, Ont.)*, *145*, 110383. https://doi.org/10.1016/j.foodres.2021.110383
74. DHYANI, A., Chander, V., & Singh, N. (2019). Formulation and evaluation of multipurpose herbal cream. *Journal of Drug Delivery and Therapeutics*, *9*(2), 341–343. https://doi.org/10.22270/jddt.v9i2.2540
75. Dudhe, A. R., Choudhari, N., Dudhe, R., Katole, G., Mahajan, R., Pathak, N., Darode, A., Devnani, D., & Rd, H. (2023). *International Journal of Newgen Research in Pharmacy & Healthcare*. *1*, 129–134.
76. Ebrahimzadeh, A., Ebrahimzadeh, A., Mirghazanfari, S. M., Hazrati, E., Hadi, S., & Milajerdi, A. (2022). The effect of ginger supplementation on metabolic profiles in patients with type 2 diabetes mellitus: A systematic review and meta-analysis of randomized

controlled trials. *Complementary Therapies in Medicine*, *65*(January), 102802. https://doi.org/10.1016/j.ctim.2022.102802

77. Gulcin, İ., & Alwasel, S. H. (2023). DPPH Radical Scavenging Assay. *Processes*, *11*(8). https://doi.org/10.3390/pr11082248
78. Janicsák, G., Veres, K., Kállai, M., & Máthé, I. (2003). Gas chromatographic method for routine determination of oleanolic and ursolic acids in medicinal plants. *Chromatographia*, *58*(5–6), 295–299. https://doi.org/10.1365/s10337-003-0058-y
79. Kolimi, P., Narala, S., Nyavanandi, D., Youssef, A. A. A., & Dudhipala, N. (2022). Innovative Treatment Strategies to Accelerate Wound Healing: Trajectory and Recent Advancements. *Cells*, *11*(15). https://doi.org/10.3390/cells11152439
80. Li, J., Bai, L., Wei, F., Zhao, J., Wang, D., Xiao, Y., Yan, W., & Wei, J. (2019). Therapeutic Mechanisms of Herbal Medicines Against Insulin Resistance: A Review. *Frontiers in Pharmacology*, *10*, 661. https://doi.org/10.3389/fphar.2019.00661
81. Muthukumarasamy, R., Ilyana, A., Fithriyaani, N. A., & Najihah, N. A. (2016). *Formulation and evaluation of natural antioxidant cream comprising Formulation and Evaluation of Natural Antioxidant Cream Comprising Methanolic Peel Extract of Dimocarpus longan. September*.
82. Olszewska, M. (2008). Optimization and validation of an HPLC-UV method for analysis of corosolic, oleanolic, and ursolic acids in plant material: Application to Prunus serotina Ehrh. *Acta Chromatographica*, *20*(4), 643–659. https://doi.org/10.1556/AChrom.20.2008.4.10
83. Simões, A., Veiga, F., Vitorino, C., & Figueiras, A. (2018). A Tutorial for Developing a Topical Cream Formulation Based on the Quality by Design Approach. *Journal of Pharmaceutical Sciences*, *107*(10), 2653–2662. https://doi.org/10.1016/j.xphs.2018.06.010
84. Xie, J., & Schaich, K. M. (2014). Re-evaluation of the 2,2-diphenyl-1-picrylhydrazyl free radical (DPPH) assay for antioxidant activity. *Journal of Agricultural and Food Chemistry*, *62*(19), 4251–4260. https://doi.org/10.1021/jf500180u
85. Yee, H.-Y., Yang, J.-J., Wan, Y.-G., Chong, F.-L., Wu, W., Long, Y., Han, W.-B., Liu, Y.-L., Tu, Y., & Yao, J. (2019). [Molecular mechanisms of insulin resistance and interventional effects of Chinese herbal medicine]. *Zhongguo Zhong yao za zhi = Zhongguo zhongyao zazhi = China journal of Chinese materia medica*, *44*(7), 1289–1294. https://doi.org/10.19540/j.cnki.cjcmm.20181105.003