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CLINICAL PRESENTATION AND WOUND HEALING FEATURES OF ONION PEEL EXTRACT-BASED EMULGEL

Iffra Naqvi[1], Syeda Rida Nisar Bukhari[2], Syed Anns Nisar Mustafa[3], Huzaira Zainab[4], Aousaf Ahmad[*5], Syed Nisar Hussain Shah[6]

[1,2,3,4]*Clinical Pharmacy Services, University of Lahore Hospital, Lahore, Pakistan*
[*5,6]*Quaid-e-Azam College of Pharmacy, Sahiwal, Pakistan*

[*5]aousaf.321@gmail.com, [6]nisarhussain@bzu.edu.pk

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Corresponding Author: *
Aousaf Ahmad

Abstract

The objective of this study was to assess the frequency and effectiveness of Onion peel extract-based formulations containing Quercetin for the treatment of different kind of wounds. It was an analytical prospective and clinical assessment study which were done in Department of Clinical Pharmacy Services at The University of Lahore Teaching Hospital, Lahore and duration of study was 12 months from 2023-24. Clinical studies were carried out in two male and one female patient (age 21±4) suffered with chronic diabetic wound, insect bite and burn wound, in order to observe the therapeutic effects of our formulated Onion peel extract-based emulgels using different natural oils in all conditions. The three (03) different patients were given topical formulated Onion peel extract-based emulgels who showed chronic wounds to be healed up and they were instructed to apply topically thrice a day on the affected area. After one to three weeks of treatment, the patients showed improvements in wound healing and considerable reduction in inflammation. The results suggested that Onion peel extract-based formulations could be an effective treatment for different kind of wounds.

INTRODUCTION

Quercetin shows strong antioxidant action due to its primary chemical structure, which may help to stop free radicals from creating resonance-stabilized Phenoxyl radicals. However, due to low oral bioavailability, chemical instability and poor water solubility Quercetin applicability is limited. In the human body, Quercetin can be glucuronidated, sulfated or methylated as a result of digestion, hence, Quercetin is Nano-encapsulated/ topical use with the aim to get over these problems and hypothetically greatly increase its stability, effectiveness, and bioavailability (Das et al., 2020).

The onion, *Allium cepa* L. (Alliaceae), contains a variety of organosulfur-containing substances, including flavonoids, saponins, and vitamins B1, B2, B6, C, and E. These ingredients have antibacterial, antiviral, antioxidant and anti-inflammatory properties together. Quercetin is the primary active ingredient in onion extract with wound-healing properties. The onion's tannins and flavonoids are what aid in the healing process of wounds, and it also demonstrates the ability to scavenge free radicals (Ahmed et al., 2023).

Although the medicinal benefits of traditional wet and dry dressings are well-established, their benefits in closing wounds, preventing infections and serving as physical barriers are somewhat restricted. Furthermore, adhesion to these dressings may cause tissue injury during removal. Consequently, in order to promote patient compliance, prevent

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complications from the wound, and hasten the healing process, multiple therapeutic approaches are employed concurrently in the management of chronic wounds (Gorain et al., 2022). Because of their low bioavailability, rapid disintegration rate, and poor water solubility, the majority of naturally occurring bioactive compounds have limited application in clinical settings. In recent years, these issues have been resolved by encapsulating/ topical Nano-formulations (Kant et al., 2021).

The integration of the emulsions with the gels increase emulgel stability and enables it to operate as a means of delivery for dual-controlled delivery systems, which are becoming more and more well-liked as a novel topical drug delivery technology (Sharma et al., 2018).

There is an urgent need of effective formulation containing Quercetin to speed up the healing process in wounds by modulating antioxidant system of wound, cytokines, growth factors, other proteins and cells involved in healing. Several flavonoids like Hesperidin, Curcumin, Quercetin, Rutin, Naringin, and Luteolin have shown their potential in managing healing wounds. Natural products that maintain glucose haemostatic, exert anti-inflammatory activity, inhibit microbial growth, modulate cytokines, inhibit matrix metalloproteinase (MMP), stimulate angiogenesis and extracellular matrix, and modulate growth factor can be considered as a potential therapeutic lead to treat diabetic wound.(Vinay 2020)

The main objective of management is to improve the rapid growth of the epidermis and to promote healing of different type of wounds. The treatment of the patients must be of that which could easily be understood and a complete observance could be achieved easily. It may also be acceptable by patients and cosmetically nonirritant. The patience and time are necessary for treatment related to skin, so patients must be enquired to have some patience and adherence towards treatment. Like any other skin treatments, topical is far best to treat skin ailments. It is advisable to ask patient to use given treatment external only and apply on the affected area.

These Clinical study trials related substantial evaluation of particular mechanisms of Quercetin extracted from Onion peels and natural oils in the form of Emulgel when applied to wounded patients would significantly vital for patient compliance (1). It could also be credible the recovery of wounds by regular checking of their blood cells effecting the inflammation concerning the results for patients for wound healing which could be improve in future, with more attributes.

Formulation of Onion peels Extract-based Emulgel:

Onion peels: Onions peels were cultivated in Sindh, Pakistan and collected from the local market of Lahore, Pakistan (Figure 1). For the purpose of removing polyphenol components, only orange onion peels were utilized. The peels were dried for ten hours at 60°C in a drying oven, resulting in moisture concentrations of 4% to 5% (w/w). After that, the dried onion peels had beed pulverized with a high-speed mixer (Blender 7012S; Waring, Torrington, USA) to a particle that range in size of 1– 10 mm, and they were kept at 4°C until needed (Manog et al., 2022).

Onion peel extract: The *Allium cepa* L. (Liliaceae) plant, also known as the onion, is eaten all over the world. The medicinal effects of onion and its derivatives, such as saponins, aglycones, quercetin, cepaenes, flavonoids, organosulfurs, and phenolic substances, were demonstrated by a variety of pharmacological qualities. The outer layers of onions are particularly high in antioxidants. Onions because of their prebiotic properties, aid in digesting as well as promote the beneficial bacteria in the intestines. In addition to minerals like selenium and vitamin C, onions contain a sulfur component. Onions should definitely be a part of your diet given all of these benefits. Some individuals use onions as a component in skin care products because of the same qualities that draw them in onion proponents assert the following benefits for skin, along with the onion properties that support those claims:

- Antioxidants have the potential to minimize surgical scars and impart a smoother texture to the skin.
- Phytochemicals contribute to skin softness and a radiant complexion.
- The antibacterial properties work to combat inflammation-causing bacteria.
- Antiseptic qualities aid in defending against viruses and fungi.

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Antioxidants are abundant in onions. These substances work to prevent free radicals from causing inflammation. In the body, substances called free radicals can damage cells and result in wrinkles as well as premature aging symptoms. Radicals without chains are neutralized by antioxidants so they cannot cause harm. Quercetin is among the most widely recognized anti-oxidants in extract of onions. (Marefati et al., 2021).

Onion peels (20 g) were combined with a 200 mL of 70% (v/v) ethanol and boiled in a water bath for three hours at 60 degrees Celsius to extract ethanol. Following the extraction of ethanol, the mixture was filtered via filter paper to extract the solid residue more than thrice. After the rotary evaporator was used to evaporate the solvent (Figure 2). Subsequently, the extract's solubility in methanol, ethanol, and distilled water was assessed (Manog et al., 2022).

Figure 1. Crushed form of onion peels

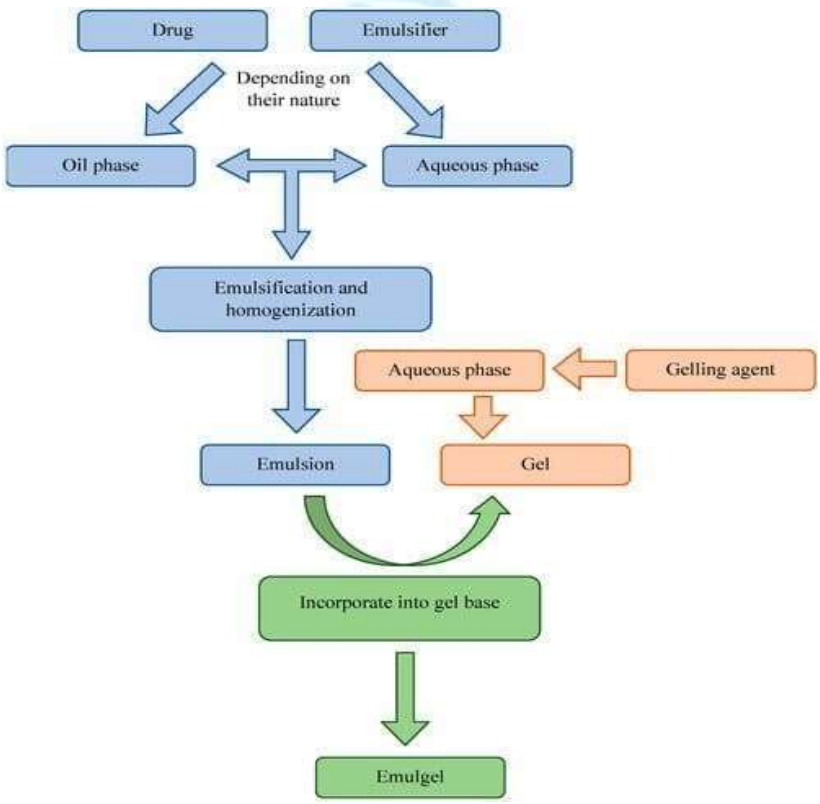

Figure 2. Steps in the process of Emulgel preparation (Milutinov et al., 2023)

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Emulgel preparation: Emulgel can be done in a number of ways, but it usually consists of three phases. Usually, the procedure is simple: gel and emulsion are made independently and then blended at the end. Here's an overview of the steps:

Step 1; When the aqueous and oil phases were prepared independently, they were combined to create an emulsion that may be oil-in-water or water-

Step 3; the produced emulsion and gel were mixed and homogenized in the last phase (Milutinov et al., 2023).

in-oil. When necessary, this included heating stages and adding the medication, often into the interior phase of the emulsion. Added materials to different phases according to their properties; for example, add materials that are hydrophobic to the oil phase and hydrophilic substances to the watery phase.

Step 2; added a gelling ingredient to the water to create the gel.

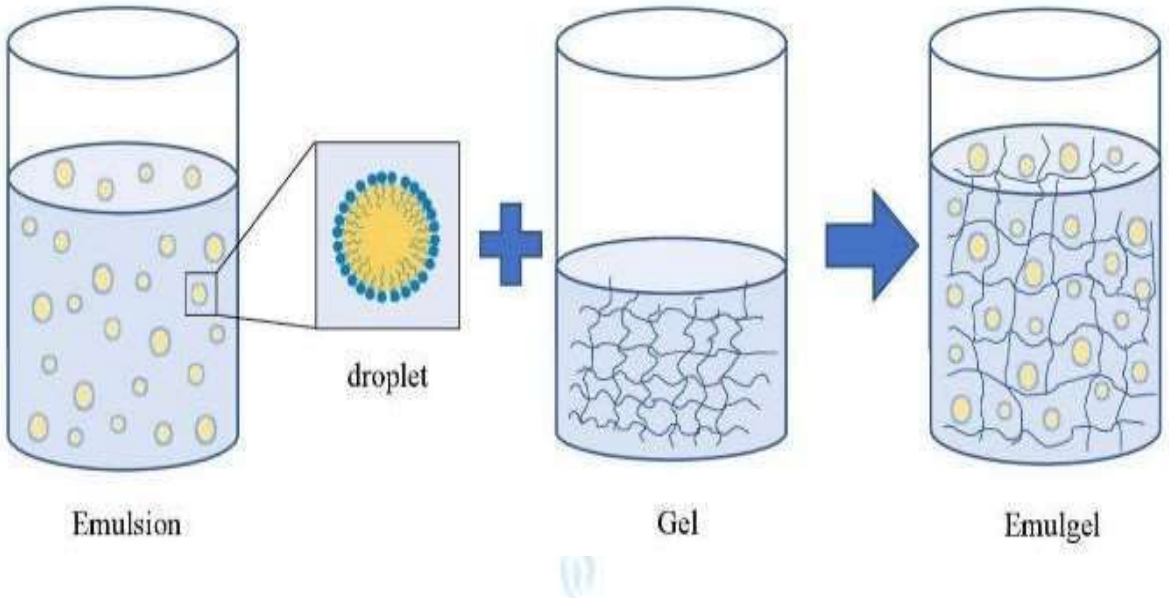

Figure 3. Schematic process diagram of Emulgel formation (Milutinov et al., 2023).

Table 1: Formulations of onion peel extract based Emulgel by using various oils

Different PG and PEG concentrations were used to create 100 g of onion extract oil-based Emulgel. The gel phase was first created by adding the required amount of Carbopol 940 to water and swirling continuously with a mechanical stirrer at the right speed. To create the oil phase, Span20 was combined with liquid paraffin after onion extract and oil were added. After dissolving Tween 20 in distilled or deionized water to create an aqueous phase, methylparaben and polyethylene glycol-1000 were then added. After cooling to ambient temperature and heating both stages to between 70° and 80°C, the emulsion was combined. The emulsion was then mixed with gel in a 1:1 ratio and lightly swirled to create Emulgel (Table 1) (Saeed et al., 2023).

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Formulations										
F1 (Oleic oil)	5	5	15	6	4	0.5	0.1	0.03	0.02	84.15
F2 (Safflower oil)	5	5	11	6	4	0.5	0.1	0.03	0.02	84.15
F3 (Silicone oil)	5	5	5	6	4	0.5	0.1	0.03	0.02	84.15

Figure 4. Emulgel formulations were thick, creamy pinkish color preparations that had a glossy, smooth, uniform texture. pH of these formulations were 6.30 ± 0.031, 5.56 ± 0.031 and 6.03 ± 0.031 respectively.

Discussion:

Quercetin in Onion peels extract and its skin benefits: A type of secondary metabolites presents in plants called flavonoids includes Quercetin. Natural sources of Quercetin include apples, onions, red wine and green tea, among different vegetables and fruit varieties. Recognized for its potent ability to scavenge radicals that are free and stop lipids from oxidizing, it is among the most common antioxidants found in food. Because Quercetin is a strong anti-inflammatory, it can aid in reducing redness and inflammation brought on by contaminants, pollution, and other environmental stresses. Moreover, antiviral, antibacterial, and perhaps anticancer effects of Quercetin have been demonstrated. When administered topically, Quercetin has additional skin-benefiting properties. It can aid in promoting healing, lowering inflammation, and shielding the skin from sun damage. In addition, studies have demonstrated the efficacy of topical Quercetin in treating acne and in minimizing the appearance of wrinkles and age spots. Quercetin, a flavonoid with diverse health benefits, also offers several notable advantages for the skin:

- UV Radiation Protection: Acts as an all-natural sunscreen,
  rotecting the skin from harm caused by UV rays. Quercetin has demonstrated efficacy in protecting against photo-aging, sunburn, and various forms of skin damage caused by sun exposure.
- Anti-Inflammatory Properties: Works as a strong anti-inflammatory, assisting in the decrease of redness and inflammation caused by exposure to the sun and other environmental stresses. Its effectiveness extends to treating skin conditions such as eczema, dermatitis, acne, and psoriasis.
- Wound Healing and Scar Reduction: Facilitates accelerated and effective healing of wounds and scars by stimulating the growth of new skin cells and promoting collagen production. This dual action contributes to a more efficient recovery process.

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- Anti-Aging Effects: Diminishes the appearance of wrinkles and other signs of aging. Quercetin's ability to stimulate collagen production is particularly crucial in maintaining skin firmness and smoothness, thereby combating the visible effects of aging.

Wound healing mechanism of Quercetin: Quercetin exhibits a diverse range of biological actions that render it particularly beneficial for wound care. Collagen accumulation, anti-inflammatory and antioxidant abilities, vascular development, and fibroblast expansion are some of its effects. When topically applied at a concentration of 0.3%, it has been noted that Quercetin greatly speeds up wound closure, promotes epidermal layer regrowth, and lowers the level of oxidative stress (Nongmaithem Randhoni Chanu et al 2023). Additionally, it participates in reactive oxygen species recycling, prevents the depletion of naturally occurring antioxidants, hinders lipid peroxidation following UV radiation exposure, and helps shield keratin cells from external oxidative agents. Quercetin exhibits remarkable anti-inflammatory characteristics; it is more powerful than other flavonoids at blocking NF-kB activity and the release of cytokines that are pro-inflammatory. The transcription factor NF-kB is essential for controlling immunological responses and inflammation. When it comes to wound healing, NF-kB stimulates the inflammation response by encouraging the production of chemical messengers and cytokines that promote inflammation like TNF-α and IL-β. Through the recruitment of lymphocytes to the site of injury, these chemicals start the process of tissue healing. Moreover, NF-kB is vital for the proliferating stage of healing wounds, which involves fibroblast migration and the creation of new blood vessels, both of which are necessary for tissue regeneration. It continues to play a role throughout the remodeling phase, when it increases the synthesis of matrix proteins and elastin to support and fortify newly formed tissue. On the other hand, prolonged inflammation can be brought on by persistent or overactive NF-kB signals, which can impede the healing of wounds. Consequently, in order to maximize the process of healing wounds, appropriate control of the signaling pathway associated with NF-kB is essential (Panthi et al., 2023).

Stability study and skin irritation test

The stability testing results indicated that all onion peel extract based Emulgel formulations (Figure 5) were maintained in a stability chamber for six months at a temperature of 40°C ± 1°C. The appearance of every onion peel extract based Emulgel formulation was found to be excellent, and there were no notable changes seen in terms of drug content, drug release behavior, viscosity, pH, or moisture content.

Figure 5. Images showing skin irritation test

Human volunteers participated in a 30-day skin irritation test for each onion peel extract based emulgel formulation. The results showed (Figure 5) that there were no skin irritations, lesions, or scrapes (Javed et al., 2018).

Case Repots

1. Diabetic foot wound: Twenty-three 23 years' young male, living in Punjab (Lahore) presenting with complaint of diabetic foot. There was history of using many allopathic treatments containing parenteral broad-spectrum antibiotics, antibiotic ointments for wound healing, however, none were thought to be the best for wound healing (Vinay et al 2021). Prior to

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beginning treatment, the hematological tests (CBC, thyroid profile) revealed that all of the tests had been cleared. Thus, after receiving written consent from the patient and his guardian, the patient was instructed to apply a useful amount of safflower oil-based onion peel extract Emulgel to the affected parts of diabetic foot thrice a day. On the first day, pictures of his diabetic foot were obtained (Figure 1). The patient has kept up their topical Emulgel use. By the first week, diabetic foot wound was observed to be improving and by the third week, it had been healed.

Treatment Plan: Safflower oil-based onion peel extract Emulgel was applied on the infected areas of patient thrice a day (Once in the morning, noon and in the night before sleep) with follow up appointment in first to third week and reviewed after 21 days (Figure 6). Like other medication, Safflower oil-based onion peel extract only contained Quercetin and Safflower oil having no side effects.

Treatment progress: Following the treatment, the patient was asked to take photographs throughout the treatment course to evaluate the wound healing of Safflower oil-based onion peel extract Emulgel.

During first week of a treatment, patient reported slight improvement in chronic diabetic wound. Following another week the wound continues to show improvement, while after third week, there was a profound overall improvement, with reduced inflammation and marked improvement in skin color. After that, patient asked to apply the Safflower oil-based onion peel extract just one time a day at night.

Figure 6. The images of diabetic foot wound are displayed. Prior to receiving therapy, the patient in figure 1 (a) complained of pus-filled wound. Figure 1 (b) Continuous improving after two weeks Figure 1 (c) showed that the wound condition was healed after three months of therapy with safflower oil-based onion peel extract Emulgel

2. Insect bite wound: Twenty-one 21 years' male living in the southern Punjab (Student of UoL) complained itching and infection that was due to some insect bite during sleeping on floor in hostel room. He was given Silicon oil-based onion peel extract Emulgel which he applied twice a day to that area, and then covered it with muslin cloth. Skin inflammation decreased by 50% within a day and wound healed after one week and vanished entirely after three weeks as shown in figure 7.

Figure 7 Photographs of insect bite infected patient (a) before treatment (b) after getting treatment (c) after 3 weeks of treatment, completely recovered

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Treatment Plan: Silicon oil-based onion peel extract Emulgel was applied on the infected wound of patient thrice a day with follows up appointment in the first week and reviewed after second week which was recovery phase. Like other medication, Silicon oil-based onion peel extract only contained Quercetin and Silicon oil having no side effects.

Treatment progress: Following the treatment, the patient was asked to take photographs throughout the treatment progression to evaluate the wound healing of Silicon oil-based onion peel extract Emulgel. During first week of a treatment, patient reported good improvement in his insect bite wound and there was no itching and redness around the wound area. There was significant improvement in wound healing following another week the wound continues to appear a profound improvement, with reduced inflammation and marked change in normal skin color. After that, patient was asked to apply the Silicon oil-based onion peel extract Emulgel till complete recovery.

3. Burn wound: Twenty-five 25 years' young female living in Punjab (Lahore), complained inflammation and burning wound in her fingers. She was given olive oil-based onion peel extract Emulgel, to apply on finger wound area twice a day. After one week, skin inflammation was reduced and wound started to heal and the patient was recovered within a month as shown in figure 8 (a, b).

Figure 8 The images of burn wounds are displayed in figure. Prior to receiving therapy, the patient in figure (a) complained of unhealed wound of burn and figure (b) showed that the wound condition was healed after one week of therapy with olive oil-based onion peel extract Emulgel.

Treatment Plan: Olive oil-based onion peel extract Emulgel was applied on the infected burn wound of patient thrice a day with follow up appointment in one week and reviewed after treatment period. Like other medication, Olive oil-based onion peel extract Emulgel only contained Quercetin and Olive oil having no side effects.

Treatment progress: The burning sensation on the burn wound was so awful that the patient was completely unable to sleep sometime, and had continuously been scratching his skin worsening the lesions. Likewise a chronic burn wound female patient, following the treatment, and was asked to take photographs throughout the treatment course to evaluate the anti-inflammatory efficacy of olive oil-based onion peel extract Emulgel.

During first three days of a treatment, patient reported intermittent reduction in itching just after application of Emulgel and profound improvement in burn wound healing, however, after one week treatment. So patient was asked to continue the treatment for next few days which once again showing the improvement with better skin condition.

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Conclusion: Consequently, the objective of the above clinical case studies for wound healing formulations derived from plant sources has been successfully achieved. The comprehensive evaluation of formulations, including their physicochemical properties, drug release profiles and therapeutic efficacy demonstrated their potential as effective treatments for skin infections and wound healing. This study underscores the promising role of natural plant extracts in the development of innovative pharmaceutical formulations for therapeutic application even in chronic patients.

Limitation of Study and Recommendations:
Based on the findings of this study, further investigation into the potential synergistic effects of combining onion peel extract with other natural extracts like Aloe Vera or pharmaceutical agents could enhance the therapeutic efficacy of the formulated Emulgel. Additionally, conducting in vivo studies to evaluate the safety and efficacy of these formulations on animal models would provide valuable insights into their clinical applicability. Furthermore, exploring alternative methods for enhancing the stability and shelf-life of the formulations, such as transdermal drug delivery systems, could contribute to their commercial viability. Lastly, considering the growing interest in sustainable and eco-friendly healthcare natural products, future research efforts could focus on optimizing the formulation processes to ensure minimal environmental impact while maintaining therapeutic effectiveness. These recommendations aim to further advance the development and utilization of plant-derived formulations for wound healing and skincare applications cost effectively.

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