

Original Article

## Formulation and Evaluation of Wound Healer Waterproof Gel Spray containing *Cymbopogon nardus* L. and *Musa paradisiaca* L. Leaves Wax

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**Abstract:** Red lemongrass (*Cymbopogon nardus* L.) is an Indonesian plant which has potential as a wound healer. Red lemongrass contain essential oil which has the potential effect to treat wounds, such as citral. The citral content in every 1 mL of this essential oil is 32.28%. A wound is an injury-induced disturbance of the tissues' normal anatomical relationships. Wound is basically prone to infection, and it can infect bacteria, such as *Staphylococcus aureus* bacteria. Water splashes exposed the *Staphylococcus aureus* bacteria, which then caused an infection. Water is a medium for bacterial growth. In this research, kepok banana leaves wax (*Musa paradisiaca* L.) is added as a waterproof base to prevent wounds from being splashed by water, because wounds on skin tissue are protected by a layer of wax originating from kepok banana leaves (*Musa paradisiaca* L.). This research aimed was to make a waterproof spray from red lemongrass essential oil using wax of banana leaves as a waterproof base. The result showed waterproof gel spray preparation has yellowish white colour; a distinctive aromatic odor, pH 6.28; viscosity 135 cps, and dry time 7.5 minutes and it has waterproof properties.

**Keywords:** red lemongrass, wax banana leaves, waterproof gel spray, wound healer

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### 1. INTRODUCTION

Red lemongrass (*Cymbopogon Nardus* L.) is a plant originally from Indonesia and it has antibacterial effect as a wound healer. Essential oil content of red lemongrass, such as citral has potential effect as a wound healer agent. Yield of essential oil content of red lemongrass during the distillation process from eight kilogram wet red lemongrass could produce ten milli litres essential oil. One milli litre lemon grass essential oil which is harvested in three months contained 32.23% citral [1]. Essential oils are found in red lemongrass (*Cymbopogon nardus* L.) plants. The average amount of essential oil found in lemongrass leaves is 0.7% (it can reach 1.2% in the dry season and about 0.5% in the wet season). The colour of red lemongrass oil is a light yellow. Aldehyde compounds (citronellol-C<sub>10</sub>H<sub>16</sub>O), alcoholic compounds (citronellol-C<sub>10</sub>H<sub>20</sub>O and geraniol-C<sub>10</sub>H<sub>18</sub>O), and other compounds such geraniol, citral, nerol, mentol, heptanone, and dipentene are the major active components generated [2]. From 300 grams of plant material, the root of the lemongrass plant contains about 0.52% alkaloids. The flavonoids luteolin, luteolin 7-O-glucoside (cynaroside), isoscoparin, and 2''-O-rhamnosyl isoorientin are found in the leaves and roots of lemongrass.

Quercetin, kaempferol, and apigenin are other flavonoid substances that have been identified from the aerial sections of the lemongrass plant [3]. From the results of the compound content obtained, carried out with a thin layer chromatography screening test.

Wound can be described as a disruption of the normal anatomical relationship of tissues as a result of injury. The injury may be intentional such as a surgical incision or accidental following trauma [4]. There are so many type of wound, such as incised wound, laceration, abrasion, and surgical wound. Infection of wound caused by bacterial activity including *Staphylococcus aureus*. Previous research explained that concentration of 3% essential oil obstructed *Staphylococcus aureus* activity.

The research aimed was to evaluate and formulate of waterproof gel spray containing Red Lemongrass (*Cymbopogon nardus* L.) and Kepok Banana (*Musa paradisiaca* L.) leaves wax as a waterproof base. A novel waterproof gel spray preparation was made by combining red lemongrass essential oil (*Cymbopogon nardus* L.), an antibacterial agent, combined by wax from kepok banana leaves (*Musa paradisiaca* L.). This antimicrobial helps to hasten the healing of wounds. A concentration of three grammes of wax from the extraction of kepok banana leaves (*Musa paradisiaca* L.) was shown in a research from Ramadhan et al. (2020) to have the capacity to create a waterproof layer. The wax material found in Kepok banana leaves was used to create a waterproof gel spray because it has the ability to create a waterproof coating and it shows that after spraying, the waterproof layer can stick for 30 minutes. This waterproof gel spray can protect wounds from water, and can speed up wound healing due to the presence of essential oil from red lemongrass (*Cymbopogon nardus* L.).

## 2. MATERIALS AND METHODS

### 2.1. Collection red lemongras (*Cymbopogon nardus* L.)

Red lemongrass is harvested in Sorogenen, Ambarketawang, Gamping, Sleman, Indonesia. To get rid of dirt and bacteria, the stems of red lemongrass are cleaned. The red lemongrass stems are dried at 60°C in an oven until they are free of any remaining impurities [5]. This parameters used to detect which red lemongrass had been dried, and the weight remained consistent [6].

### 2.2. Preparation of red lemongrass extract

Extraction of red lemongrass is carried out using the steam distillation method to obtain essential oil which contains citral. It started by weighing the stems and leaves of red lemongrass which have been cut into small pieces (eight kilograms). Then put it in a distillation apparatus with the addition of distilled water as a solvent. Turn on the heater according to the temperature variable (100-105°C). The distillate is collected, then transferred into a separating funnel to separate the essential oil from the air. The essential oil is transferred into a flacon and stored in the freezer.

### 2.3. Preparation Wax Kepok Banana Leaves (*Musa paradisiaca* L.)

Preparing banana leaves that have been cleansed from the midrib and allowed to air dry for a week. The leaves are then divided into little bits. 100 grams of banana leaves should be weighed before being rubbed with n-hexane solvent. The filtering and separated filtrate were left at room temperature for 2 hours after the filtering and reflux process, which took place at 60°C for 10 minutes [7].

#### 2.4. Thin Layer Chromatography (TLC) Screening of Citral from red lemongrass (*Cymbopogon nardus* (L.) Rendl.)

Sample preparation started from the distillation of red lemongrass (*Cymbopogon nardus* L.). The mobile phase preparation used is n-Hexane:Ethyl acetate (8:2). Saturate the chamber using filter. Elute the sample to the limit mark. Dry the plate by airing it and then detect it with UV 254 light.

#### 2.5. Formulation of Wound Healer Waterproof Gel Spray

Formulation of wound healer waterproof spray could be seen in Table 1.

**Table 1.** Formulation of Waterproof Gel Spray

Material	Amount
PVA	1 gram
Span 80	0.375 %
Tween 80	0.125%
Wax Kepok Banana Leaves	5%
Essential Oil Red Lemongrass	5%
Ethanol	10%
Distillated water	Ad 20 mL

#### 2.6. Evaluation of Wound Healer Waterproof Gel Spray

##### 2.6.1. Organoleptic Test

Organoleptic testing is carried out by sense. Organoleptic testing includes smell, color and taste which are directly tested using the senses.

##### 2.6.2. pH Test

pH testing was carried out using a pH meter (Lutron pH-208), before using the pH meter, calibration was carried out using a pH 4.0 and pH 10.0 calibrator, after calibration the pH meter was dipped in the formula sample that had previously been made. The pH of a good preparation for wounds is 4.5 – 6.5 [8].

##### 2.6.3. Viscosity Test

Viscosity testing was carried out using a Brookfield viscosimeter, when testing the viscosimeter the tool was set at a speed of 100 rpm [9].

##### 2.6.4. Dry Time Test

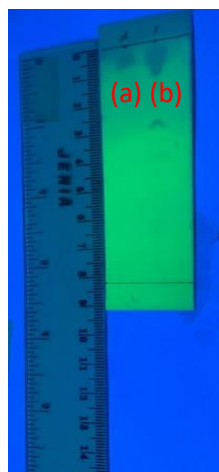
Testing the preparation dry time of less than 5 minutes can minimize the growth of microorganisms and evaluation of the volume of one waterproof gel spray is carried out to estimate how many times to spray the preparation to obtain a uniform volume. Dry time testing is carried out directly on the skin by spraying the sample solution, and waiting until the sample solution dries and the drying time is recorded.

### 3. RESULTS AND DISCUSSION

Red lemongrass (*Cymbopogon nardus* L.) is a Poaceae (grasses) perennial herb [10]. In almost all continents, lemongrass has been used extensively as a traditional medicine by making "tea" or an infusion from fresh or dried leaves. It has a wide range of indications, from mild ailments like the flu, fever, cuts, coughing, and headaches to more serious illnesses like rheumatoid arthritis, bladder disorder, diabetes, and malaria [11]. It is a powerful antibacterial, antitussive, antiseptic, analgesic, and anti-inflammatory agent [12].

### 3.1. Thin Layer Chromatography (TLC) Screening of Citral from red lemongrass (*Cymbopogon nardus* (L.) Rendl.)

TLC was used to determine the optimal eluent for separating citral, employing Silica Gel 60 GF254 as the stationary phase and one type of eluent pair as the mobile phase, namely n-hexane: ethyl acetate (8:2) [13]. Spots formed during the elution process were identified using a 254 nm UV light. The polarity of the eluent used determines spot mobility. The optimal eluent is chosen from a mixture of eluents that can get the component up to the end line of the silica plate, resulting in a polarity difference between the solvents and a genuine spot. Figure 1 shows the spots formed on the TLC plate.



**Figure 1.** TLC Profile. (a) standard of citral (b) essential oil of Red Lemongrass

As a result of steam distillation, which yielded pure essential oil, the elution results revealed the presence of a citral compound in the red lemongrass sample. The  $R_f$  values of the sample and the reference compound were calculated; the sample's  $R_f$  value was 0.925 cm and the citral standard's  $R_f$  value was 0.925 cm.

### 3.2. Formulation of Wound Healer Waterproof Gel Spray

Topical sprays have gained popularity in clinical practice over the past few decades as a method of applying hydrogels and or cell suspensions to treat both acute and chronic wounds because of their benefits, including the ability to treat large wounds or spray over areas with unfavorable topography, the shortened application time, and the uniform distribution of the sprayed suspensions [14]. This formula use PVA as a gelling agent, Span 80 and Tween 80 as surfactant, ethanol as a solvent, wax kepok banana leaves as a waterproof base, essential oil of red lemongrass as an active substance which include citral.

### 3.3. Physical Characterization of Waterproof Gel Spray

Topical spray usually contains ethanol and water as solvents. Other excipients are co-solvent, which is used to dissolve the drug substance to obtain a clear solution, film forming agent, and humectants. *Cymbopogon nardus* L. in combination with *Musa paradisiaca* L. through topical spray was developed by complexing citral as an essential oil. Polyvinyl alcohol (PVA) was a film former with good adhesion as well as a stabilizing agent. The wax of banana was also an auxiliary film-forming

agent. Span 80 and Tween 80 were used to promote moisture retention on the skin. Tween 80, when applied as a surfactant, can create a stable nano emulsion [15]. Tween 80 is not only employed as a surfactant, but it can also reduce the globule size of lipophilic compounds in microemulsion and nanoemulsion systems. Essential oils prepared with tween 80 can keep active ingredients stable and effective [16].

### 3.3.1. Organoleptic

The human senses are used in organoleptic testing. Organoleptic testing covers smell, color, and taste, all of which are assessed directly with the nose, eyes, and mouth. The results of the tests revealed a unique fragrant odor, a yellowish-white color, and a bitter taste.

### 3.3.2. pH

pH testing was carried out using a pH meter (Lutron pH-208), before using the pH meter, calibration was carried out using a pH 4.0 and pH 10.0 calibrator, after calibration the pH meter was dipped in the formula sample that had previously been made. The pH of a good preparation for wounds is 4.5 – 6.5 [8]. The results of the pH test were 6.28. This result is still acceptable because it is in accordance with the pH standard for the skin.

### 3.3.3. Viscosity

Viscosity testing was performed using a Brookfield viscosimeter, with the toolset to a speed of 100 rpm and the spindle inserted directly into the sample solution. An excellent spray gel preparation has a viscosity of less than 150 cps [9]. The viscosity testing results were obtained 135 cps this result meets the requirements for skin spray gel formulations.

### 3.3.4. Dry Time

A dry time of less than 5 minutes for the preparation can minimize microbe growth, and an evaluation of the volume of one spray is performed to predict how many times to spray the preparation to get a uniform volume. Dry time testing is performed directly on the skin by spraying the sample solution and waiting until the sample solution dries before recording the drying time. The preparation test results revealed that the drying time was 7.5 minutes, which it takes longer so further testing is needed.

Citral, one of lemongrass' key components, has been proven to decrease the production of interleukin (IL)-1b, IL-6, and tumor necrosis factor (TNF) $\alpha$  [17]. Thus, the surface of banana leaves has a waxy covering that provides hydrophobic qualities that act as a water repellent. The waxy surface of the leaves, according to Jetter and Kunst (2008), serves as a protective layer against water loss, ultra-purple light, infections, and insects [18]. Wax are also valuable raw materials for industrial uses such as high-value lubricants, cosmetics and medicines, and high-powered fuels. Wax and leaf microstructure also contribute to leaf surface roughness [19]. Furthermore, lemongrass has antioxidant activity and can scavenge free radical molecules [20].

Inflammation is involved in the phases of wound healing. Inflammatory is a two-edged sword with both positive and negative repercussions, and during the inflammatory phase, leucocytes create oxygen radicals as oxygen-dependent processes. These free radicals are excreted as a weapon to destroy bacteria, but they can also be harmful to body cells. Excess free radicals and/or antioxidant deficiency can cause oxidative stress, which can lead to cellular malfunction and, in extreme cases,

cell death. In this case, antioxidants serve to protect cells from harm caused by free radicals created during inflammation [21].

#### 4. CONCLUSION

The results of formulation and evaluation showed waterproof gel spray preparation has yellowish white colour; a distinctive aromatic odor, pH 6.28, viscosity of 135 cps, and dry time 7.5 minutes and it has waterproof properties.

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