Preparation and Evaluation of Topical Gel Containing Aloe Vera Exudate

Amina M. Al-Nima*

1Department of Pharmaceutics, College of Pharmacy, University of Mosul, Mosul, Iraq
Corresponding author: amnah.mudhafar@uomosul.edu.iq

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ABSTRACT

Background: Aloe vera is known as a miracle plant belonging to the Lily (Liliaceae) family. The exudate of Aloe vera is utilized as a natural drug as it can be used for many medicinal purposes.

Aims: The present work has been undertaken with the aim to prepare a topical gel containing Aloe vera exudate and to evaluate its physical properties in addition to its anti-microbial potency in light of the orientation towards plant-based treatment for being more acceptable and preferred in the belief that they are safer with fewer side effects than synthetic ones. Aloe vera has been proved its great medicinal and cosmetic benefits.

Materials and Methods: Two sets of gel formulas were prepared; the first set containing carbopol-934 (F1-F4) and the second set containing carboxymethylcellulose (F5-F10) with varied concentrations to achieve the best formula that has been selected according to various physicochemical evaluations; like homogeneity, transparency, appearance, consistency, color, odor, pH, phase separation and others. The selected formula was further subjected to in vitro study for its anti-microbial activity.

Results and Discussion: There was a significant difference (p≤0.05) in physical parameters between the two sets of formulas except in pH. Results showed that (F2) containing 1.5% m/v carbopol-934 is considered the promising formula which showed better physicochemical properties and exhibited good anti-microbial activity against both (E.coli) and (Staphylococcus aureus).

Conclusion and recommendations: The selected formula (F2) of Aloe vera exudate could be considered as a suitable medicinal herbal candidate for treatment of topical bacterial infections. In addition, it is recommended to use higher concentration of antioxidants to preserve the stability of the product, also it is recommended to add water soluble fragrance (perfume) for better odor.

Key words: Aloe vera, anti-microbial, exudate, gel, herbal.
النتائج والمناقشة: أظهرت النتائج فرق كبير (p≤0.05) في التقييمات الفيزيائية بين مجموعتي الصيغ باستثناء الرقم الهيدروجيني وتم اختيار الصيغة (F2) المحتوية على 1.5٪ Carbopol-934 كصيغة واعدة لما أظهرته من خصائص فيزائية مقبولة، وقد أظهرت هذه الصيغة نشاطاً جيداً مضادًا لكل من Staphylococcus و E.coli الالتهاب والانفلام: يمكن اعتبار الصيغة المختارة (F2) المحتوية على عصارة الصبار مرشحاً طبياً مناسبًا لعلاج الالتهابات البكتيرية الموضعية مع التوصية بأهمية استخدام تركيز أعلى من مضادات الأكسدة للحفاظ على ثباتية المنتج وكذلك إضافة عطر قابل للذوبان بالماء للحصول على رائحة أفضل.

الكلمات المفتاحية: هلام موضعي، مضاد للميكروبات، عصارة الصبار، عشبي.

INTRODUCTION:

Aloe vera is known as a miracle plant belonging to the Lily (Liliaceae) family. This plant has been used therapeutically, certainly before 6000 years old in Egypt. It was named the “Plant of immortality” and was gifted as a burial offering to the deceased pharaohs. It is also known as “the healing plant” because it has growth-promoting activities. There are nearly 250 known species of aloes, intimated by the scientific names of Aloe vera and Aloe barbadensis. Many of the health aids correlated with Aloe vera have been blamed on the polysaccharides accommodated in the gel of the leaves. However; it is rather a combination of compounds and not only polysaccharides that account for the health aids of Aloe vera. These health aids include improvement and acceleration of wound healing, antifungal potency, anti-inflammatory, hypoglycemic or antidiabetic actions, immunomodulatory, anticancer, and gastro-protective attributes as well as its famous use for cosmetic preparations. Commercially, it is possible to get three types of products from the leaves of Aloe; the dried exudate, the exudate, and the oil. The exudate is utilized as a natural drug as it can be used for cathartic effect. The exudate or juice of Aloe vera has many medicinal uses; if it is administered directly or with any liquid to give relief from many kinds of gastric disturbances like irritable bowel, Crohn’s disease, dyspepsia, acidity, reflux and many other gastric problems. It aids to preserve the balance of acids in the stomach which leads to a soothing effect in the stomach.
In recent years, the potential anti-microbial activity of plant extracts has been reported in many studies especially against bacterial pathogens.\textsuperscript{9-11} The activity of the \textit{Aloe vera} gel extract was reported in many studies and was attributed to the synergistic effect of various compounds in the Aloe exudate\textsuperscript{9}. It is shown to have a wide range of activity against Gram-positive and Gram-negative bacteria like \textit{Staphylococcus aureus, Klebsiella pneumoniae, Pseudomonas aeruginosa, Streptococcus pyogenes, Escherichia coli, Propionibacterium acnes, Salmonella typhi} and \textit{Helicobacter pylori}.\textsuperscript{12-14} So, the \textit{Aloe vera} as a plant could be recommended furtherly in the treatment of various bacterial diseases.\textsuperscript{13}

Commercially, there are numerous products available that contains \textit{Aloe vera} to be used for medicinal purposes like burns as well as for cosmetic purposes.\textsuperscript{15}

This study has several aims, which include preparation of an exudate of \textit{Aloe vera} and \textit{Aloe vera} gel, studying the physicochemical properties of this gel and studying the anti-microbial activities against potential causes of topical infections including two types of bacteria Gram negative (\textit{E.coli}) and Gram positive (\textit{Staphylococcus aureus}).

**MATERIALS AND CHEMICALS**

The fresh \textit{Aloe vera} (\textit{Aloe barbadensis}) leaves were purchased from Local market, Mosul, Iraq. Carbopol-934 (CAR.934) was purchased from (HIMEDIA, India), carboxymethylcellulose (CMC) was procured from (SHARKCMC\textsuperscript{®} L, China). Propylene glycol (PG) was purchased from (THOMAS BAKER, India). Methyl paraben (MeP) and propyl paraben (PrP) were purchased from (Direvo, Germany), sodium sulphite and triethanolamine (TrE) was obtained from (Tedia, USA). All chemicals used were of analytical grade.

**METHODS**

**Preparation of Aloe vera exudate**

In this study, thick moist-rich leaves of \textit{Aloe vera} were used. To obtain \textit{Aloe vera} exudate or juice which is the mucilaginous jelly in the center (the parenchyma) of the plant leaf, the leaves were washed with water and were cut transversely with a knife into pieces. The thick green epidermis was selectively peeled off and the pulp containing the inner gel-like tissue in the center of the leaf was isolated with a large spoon, blended in an electric blender (Royal-Japan) for nearly 5 minutes at room temperature, then the resultant juice or exudate was sieved using a 200mm in diameter laboratory sieve and the resulted amount of juice was measured in volume using a 100-ml graduated cylinder. It is very necessary to add sodium sulphite as an antioxidant during this procedure so as to prevent discoloration of the exudate due to very high oxidation ability and the final mixture was stored in a tightly closed amber container and left in a refrigerator.\textsuperscript{16,17}

**Trials for the selection of suitable gelling agent**

One of the main ingredients of gel preparation is the gelling agent (polymer). Different gel formulations (F1-F10) were prepared using two different polymers for trials to select the suitable one. Formulas F1-F4 were prepared using different concentrations (1.0, 1.5, 2.0 and 3.0) % of CAR.934 as demonstrated in Table1, whereas Table2 demonstrates F5-F10 formulas which were prepared using different concentrations (2.5, 3.0, 4.0, 5.0 and 6.0) % of CMC concerning that CMC is used as a gelling agent at concentrations 3-6%.\textsuperscript{18} Finally, the gel formula that
showed a good appearance and consistency was selected.

**Preparation of Aloe vera gel using CAR.934**

As mentioned previously, an antioxidant material (sodium sulphite) was added and mixed thoroughly with the calculated amount of the Aloe vera exudate in a separate beaker to form the exudate solution (Alov Ex) which was kept in an amber container in refrigerator. Codes F1-F4 were given for formulas prepared using CAR.934 (Table 1). The CAR.934 dispersions were prepared by adding little amount of distilled water (D.W.) to the polymer with slight hand-mixing and then stirred by using a magnetic stirrer (Fisher Scientific, Korea) at moderate speed with slight heating at about 50-55°C for 2-3 hours. After cooling, the CAR.934 dispersion was covered with a slice of aluminum foil and left overnight to get rid of air bubbles. The preservatives (MeP) and (PrP) were dissolved within propylene glycol in a different beaker and stirred magnetically (Fisher scientific,400 rpm, 25-30°C, 2-3 hours) till forming clear solution. Then the preservative solution was added to CAR.934 dispersion using magnetic stirrer at moderate speed for 2-3 hours and left for overnight. In the next day, a calculated volume of (Alov Ex) was hand-mixed with the final polymer-preservative mixture and then stirred using a magnetic stirrer at nearly 350 rpm for 2-3 hours. Volume was completed with D.W. and stirred continuously till a uniform gel formula was formed and left in a refrigerator. As a final step of gel preparation, a few parts of milliliter of (TrE) were added and the resultant gel formula was covered with a piece of aluminum foil and left in a refrigerator.

### Table 1. Composition of Aloe vera gel containing CAR.934, F2 was selected.

<table>
<thead>
<tr>
<th>Code No.</th>
<th>Alov Ex (ml)</th>
<th>Sodium sulphite (%m/v)</th>
<th>CAR.934 %m/v</th>
<th>CMC %m/v</th>
<th>TEA (ml)</th>
<th>PG (%w/v)</th>
<th>MeP %m/v</th>
<th>PrP %m/v</th>
<th>D.W. q.s. to</th>
</tr>
</thead>
<tbody>
<tr>
<td>F1</td>
<td>70</td>
<td>0.1</td>
<td>1</td>
<td>--</td>
<td>q.s.</td>
<td>10</td>
<td>0.04</td>
<td>0.01</td>
<td>100</td>
</tr>
<tr>
<td>F2</td>
<td>70</td>
<td>0.1</td>
<td>1.5</td>
<td>--</td>
<td>q.s.</td>
<td>10</td>
<td>0.04</td>
<td>0.01</td>
<td>100</td>
</tr>
<tr>
<td>F3</td>
<td>70</td>
<td>0.1</td>
<td>2</td>
<td>--</td>
<td>q.s.</td>
<td>10</td>
<td>0.04</td>
<td>0.01</td>
<td>100</td>
</tr>
<tr>
<td>F4</td>
<td>70</td>
<td>0.1</td>
<td>3</td>
<td>--</td>
<td>q.s.</td>
<td>10</td>
<td>0.04</td>
<td>0.01</td>
<td>100</td>
</tr>
</tbody>
</table>

Alov Ex: Aloe vera exudate, CAR.934: Carbopol-934, CMC: carboxymethyl cellulose, TEA: Triethanolamine, PG: Propylene glycol, MeP: Methyl paraben, PrP: Propyl paraben,
Table 2. Composition of *Aloe vera* gel containing CMC, no one was selected.

<table>
<thead>
<tr>
<th>Code No.</th>
<th>Alov Ex (ml)</th>
<th>Sodium sulphite (%m/v)</th>
<th>CAR.934 (%m/v)</th>
<th>CMC (%m/v)</th>
<th>TEA (ml)</th>
<th>PO (%v/v)</th>
<th>MeP (%m/v)</th>
<th>PrP (%m/v)</th>
<th>D.W. q.s. to</th>
</tr>
</thead>
<tbody>
<tr>
<td>F5</td>
<td>70</td>
<td>0.1</td>
<td>--</td>
<td>1.5</td>
<td>q.s.</td>
<td>10</td>
<td>0.04</td>
<td>0.01</td>
<td>100</td>
</tr>
<tr>
<td>F6</td>
<td>70</td>
<td>0.1</td>
<td>--</td>
<td>2.0</td>
<td>q.s.</td>
<td>10</td>
<td>0.04</td>
<td>0.01</td>
<td>100</td>
</tr>
<tr>
<td>F7</td>
<td>70</td>
<td>0.1</td>
<td>--</td>
<td>2.5</td>
<td>q.s.</td>
<td>10</td>
<td>0.04</td>
<td>0.01</td>
<td>100</td>
</tr>
<tr>
<td>F8</td>
<td>70</td>
<td>0.1</td>
<td>--</td>
<td>3</td>
<td>q.s.</td>
<td>10</td>
<td>0.04</td>
<td>0.01</td>
<td>100</td>
</tr>
<tr>
<td>F9</td>
<td>70</td>
<td>0.1</td>
<td>--</td>
<td>4</td>
<td>q.s.</td>
<td>10</td>
<td>0.04</td>
<td>0.01</td>
<td>100</td>
</tr>
<tr>
<td>F10</td>
<td>70</td>
<td>0.1</td>
<td>--</td>
<td>6</td>
<td>q.s.</td>
<td>10</td>
<td>0.04</td>
<td>0.01</td>
<td>100</td>
</tr>
</tbody>
</table>


**Preparation of *Aloe vera* gel using CMC**

To prepare the suitable gel, other formulas coded as F5-F10 were prepared using different concentrations of CMC as a gelling polymer and they are presented in Table 2. Dispersions of CMC were prepared by mixing little quantity of D.W. with the polymer in one beaker at moderate speed for 2-3 hours (350 rpm, 30°C). Then the preservatives (MeP and PrP) were weighed and dissolved in PG and stirred at (400rpm, 25-30°C, 3 hours) till forming a solution. The polymer dispersion and the preservative solution were added to each other and mixed thoroughly. Finally, the calculated amount of Alov Ex was hand-mixed and stirred with the final polymer-preservative mixture and volume was made with D.W. to form the gel formula. The final *Aloe vera* gel formula was covered with a piece of aluminum foil and left in a white and dark background.

**EVALUATION OF GEL**

**Homogeneity and transparency**

All prepared formulas of *Aloe vera* gel were examined for homogeneity assessment. This was done visually after the settlement of gel in suitable beakers. Formulas were observed for their type of smear, after feel, how is the removal of gel, and transparency.19,22,25

**Appearance and consistency**

The prepared *Aloe vera* gel formulas were examined for their appearance and consistency by a visual inspection after filling the gel in the container against a white and dark background. Examinations have been made for their color, appearance as a gel and existence of aggregates.11,19,26

**Selection of the best formula**

It is essentially to consider that one of the basic ingredients of any gel formula is the gelling agent (polymer). Gel formulas were prepared to select the type and the appropriate concentration of polymer. This preparation occurs using two main polymers, CAR.934 and CMC. Many formulas were prepared using the same concentration of Alov Ex and different concentrations of CAR.934 and CMC were tried as represented in tables...
1 and 2. CAR.934 and CMC are generally recognized as safe and are used as thickeners. The selection of the best gel formula relies on many parameters like the gel appearance, consistency. The suitable gel formula was selected and used for further anti-microbial activity evaluation.

**pH measurement**

An accurate mass of nearly 0.5 g of Aloe vera gel formula was placed in a petri dish and dissolved in 50 ml distilled water, and the pH was measured after 2 hr. using digital pH meter (Eco Testr, Oakton Instruments, Singapore). The measurements of pH were done in triplicate and mean values were calculated. 

**Evaluation by in-vitro anti-microbial activity for the selected gel formula**

The microorganisms (Gram positive, Staphylococcus aureus) and (Gram negative, Escherichia coli) which were used in this work were obtained from microbiology laboratory of clinical laboratory sciences department at College of Pharmacy in Mosul University. The anti-microbial potency of the selected formula of Aloe vera gel was evaluated by using a well diffusion method. The obtained bacteria were sub-cultured on a selective media for each type of bacteria, then the inoculated plates were incubated aerobically for 24 hours at 37°C. Evaluating the antibacterial effect was determined by the zone diameter measured after 24 hours of incubation. 

**Preparation of stock solution**

In this study a stock solution (100 mg / ml) was prepared from the formulated Aloe vera gel, then serial dilution (0.5 – 2 mg / ml) from the stock solution was prepared.

**Susceptibility test**

A suspension of 0.5 MacFarland was prepared from bacteria mix with Muller-Hinton broth, then a sterile cotton swab soaked in bacterial suspension which used to inoculate the bacteria onto the surface of Muller–Hinton agar plates.

**Antibacterial assay**

Muller- Hinton agar was used in this study for susceptibility test, five wells were punched in each agar plates with 4 mm depth, after that 0.1 ml from each concentration of the selected formula of Aloe vera gel were added to each well, in addition to control antibiotic of gentamycin was used, the inoculated culture media were incubated at 37 °C for 24 hours, then zone diameter was measured for the determination the antibacterial effect.

**Statistical Analysis**

The results were statistically analyzed using t-test and given as mean ± standard deviation. P values ≤ 0.05 were considered significant.

**RESULTS AND DISCUSSION**

**Evaluation of gel**

It is recommended nowadays for greater percent of the world’s population to use Aloe vera for its health care and cosmetic benefit. The prepared gel formulas (F1-F10) were prepared from Aloe vera leaf exudates. The exudation method was carried out by simple method to obtain a fluid-like juice called Aloe vera exudate or Aloin. The method was easy and convenient, it can be achieved even at home and the yield value is relatively good, a nearly 100-ml of exudate can be obtained from each one leaf; however, the high tendency of Aloin to be oxidized which could be observed by its dark red color makes it difficult to overcome unless by the use of antioxidant. So the use of one or more than one antioxidant is necessary to overcome the problem.

**Homogeneity and transparency**
All developed gel formulas were evaluated for homogeneity. The prepared F1, F2 and F3 gel formulas containing 1.0, 1.5 and 2 % m/v CAR.934 respectively were homogeneous with no aggregates or clumping as mentioned by Syed Umar Farooq et al who used also the polymer CAR.934 in preparing herbal gel formulas. However; F2 gel formula which contains 1.5 % m/v CAR.934 found more homogeneous than all CAR.934 prepared formulas as represented in figure 2. Aggregates or clumps where found after preparing (F4) gel formula as shown in Figure 3 which may be due to high concentration of CAR.934 (3% m/v). For F5, F6 and F7 gel formulas which were prepared using 1.5, 2.0, 2.5 % m/v CMC showed little homogeneity while (F8) and (F9) which were prepared using 3 and 4 % m/v CMC showed bad homogeneity besides phase separation of gel contents and precipitation of white material in beakers as appeared in Figure 4. Due to rapid hydration behavior of CMC which may cause agglomeration and lump formation when the CMC powder is introduced into aqueous solution. This might be a cause of lumping and formation of thick gel in (F10) which contains 6 % m/v CMC, this formula showed good homogeneity with no phase separation but aggregate formation was clear. Lump creation can be eliminated by applying high agitation while the CMC is added into the aqueous Aloin solution. All prepared Aloe vera gel formulas (F5-F10) using CMC polymer are pictured in Figure 4.

Figure (2): The accepted Aloe vera gel formulas (F2) containing 1.5 % m/v CAR.934
Figure (3): Aloe vera gel formulas (F4) containing 3% m/v CAR.934

Figure (4): Aloe vera gel formulas (F5-F10) containing CMC in various concentrations: 1.5, 2.0, 2.5, 3.0, 4.0, and 6.0.

Appearance and consistency

Appearance and consistency are one of the most important characteristic features of gel formulations. The prepared (F1) gel formula containing 1.0% m/v CAR.934 was opaque, with white or pale-pink color, thin consistency, and no gel appearance. Whereas (F2) containing 1.5% m/v CAR.934 was opaque, with white or pale-pink color, smooth texture, glossy appearance and was accepted as gel formula. This result is in agreement with Alkhalidi HM et al. 30 This accepted (F2) formula is shown in Figure 2. While (F3) containing 2% m/v CAR.934 was opaque, with pale-pink color, smooth texture, and glossy appearance. Conversely, the prepared (F4) gel formula which contains 3% (m/v) showed transparency, yellow color, thick texture, sticky with glossy appearance which may be due to high CAR.934 concentration, this formula is pictured in Figure 3. All prepared Aloe vera gel formulas containing CAR.934 polymer shared in that they were smooth in texture, with no odor and no phase separation. All results of observations of Aloe vera gel containing CAR.934 are illustrated in Table 3.
### Table 3. Evaluation of *Aloe vera* gel containing CAR.934

<table>
<thead>
<tr>
<th>Parameter</th>
<th>F1</th>
<th>F2</th>
<th>F3</th>
<th>F4</th>
</tr>
</thead>
<tbody>
<tr>
<td>Homogeneity</td>
<td>++</td>
<td>+++</td>
<td>+++</td>
<td>-</td>
</tr>
<tr>
<td>Transparency</td>
<td>Opaque</td>
<td>Opaque</td>
<td>Opaque</td>
<td>Transparent</td>
</tr>
<tr>
<td>Gel appearance</td>
<td>Thin</td>
<td>Very good</td>
<td>good</td>
<td>Thick</td>
</tr>
<tr>
<td>Gel consistency</td>
<td>Smooth</td>
<td>Smooth</td>
<td>Smooth</td>
<td>Smooth</td>
</tr>
<tr>
<td>Color</td>
<td>white or pale-pink</td>
<td>white or pale-pink</td>
<td>white or pale-pink</td>
<td>yellow</td>
</tr>
<tr>
<td>Odor</td>
<td>No</td>
<td>No</td>
<td>No</td>
<td>No</td>
</tr>
<tr>
<td>Phase separation</td>
<td>No</td>
<td>No</td>
<td>No</td>
<td>No</td>
</tr>
<tr>
<td>After feel</td>
<td>Poor</td>
<td>Good</td>
<td>Good</td>
<td>Poor</td>
</tr>
<tr>
<td>Type of smear</td>
<td>Non-greasy</td>
<td>Non-greasy</td>
<td>Non-greasy</td>
<td>Sticky</td>
</tr>
<tr>
<td>Removal</td>
<td>Easy</td>
<td>Easy</td>
<td>Easy</td>
<td>Not easy</td>
</tr>
</tbody>
</table>

 +++ Excellent, ++ Good, + Satisfactory, - Bad

Appearance and consistency of the second group of gel formulations which were prepared using CMC can be summarized as follows: The prepared (F5) gel formula containing 1.5% m/v CMC was opaque, pale-yellow color, thin consistency, and no gel appearance. Whereas (F6) and (F7) containing 2.0 and 2.5 % m/v CMC were opaque, with white or pale yellow color, smooth texture, thin gel and no glossy appearance and were not accepted as gel formulas. (F8) containing 2 % m/v CAR.934 was opaque, with pale-pink color, smooth texture, and glossy appearance. (F9) containing 4.0 %m/v CMC was opaque, thin and white or pale pink color. Whereas (F10) containing 6.0 %m/v CMC showed opacity, dark pink color, thick consistency, no glossy appearance as shown in Figure 5. All prepared *Aloe vera* gel formulas containing CMC polymer shared in that they were opaque, smooth, no odor, poor after feel, non-greasy. All results of observations of *Aloe vera* gel containing CMC are represented in Table 4.
### Table 4. Evaluation of *Aloe vera* gel containing CMC

<table>
<thead>
<tr>
<th>Parameter</th>
<th>F5</th>
<th>F6</th>
<th>F7</th>
<th>F8</th>
<th>F9</th>
<th>F10</th>
</tr>
</thead>
<tbody>
<tr>
<td>Homogeneity</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>++</td>
</tr>
<tr>
<td>Transparency</td>
<td>Opaque</td>
<td>Opaque</td>
<td>Opaque</td>
<td>Opaque</td>
<td>Opaque</td>
<td>Opaque</td>
</tr>
<tr>
<td>Gel Appearance</td>
<td>thin</td>
<td>thin</td>
<td>thin</td>
<td>thin</td>
<td>thin</td>
<td>thick</td>
</tr>
<tr>
<td>Gel Consistency</td>
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<td>Smooth</td>
<td>Smooth</td>
<td>Smooth</td>
<td>Smooth</td>
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</tr>
<tr>
<td>Color</td>
<td>pale-yellow</td>
<td>pale-yellow</td>
<td>pale-yellow</td>
<td>pale-yellow</td>
<td>white or pale-pink</td>
<td>dark pink</td>
</tr>
<tr>
<td>Odor</td>
<td>No</td>
<td>No</td>
<td>No</td>
<td>No</td>
<td>No</td>
<td>No</td>
</tr>
<tr>
<td>Phase separation</td>
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<td>No</td>
<td>No</td>
<td>Yes</td>
<td>Yes</td>
<td>No</td>
</tr>
<tr>
<td>After feel</td>
<td>Poor</td>
<td>Poor</td>
<td>Poor</td>
<td>Poor</td>
<td>Poor</td>
<td>Poor</td>
</tr>
<tr>
<td>Type of smear</td>
<td>Non-greasy</td>
<td>Non-greasy</td>
<td>Non-greasy</td>
<td>Non-greasy</td>
<td>Non-greasy</td>
<td>Non-greasy</td>
</tr>
<tr>
<td>Removal</td>
<td>Easy</td>
<td>Easy</td>
<td>Easy</td>
<td>Easy</td>
<td>Easy</td>
<td>Not Easy</td>
</tr>
</tbody>
</table>

+++ Excellent, ++ Good, + Satisfactory, - Bad

### Selection of the best formula

For the preparation of *Aloe vera* gel using two polymers (CAR.934 and CMC), the appropriate formula of *Aloe vera* gel was reviewed in terms of homogeneity, transparency, appearance, consistency, color, odor, phase separation, after-feel, type of smear and how is the removal of gel. Significant difference (p≤0.05) in physical parameters between the two sets of formulas was observed. The set of formula which were prepared using CAR.934 were preferred more than the set containing CMC. Among all the prepared formulas of *Aloe vera* gel; the selected one was (F2) (1.5 % m/v CAR.934) (Fig.3) as the most accepted formula for further evaluations and this result was in agreement with Rajasekaran Aiyalu *et al* who found that 1.5 % of carbopol (934 or 940) containing gels was the best percent concentration of carbopol with the requirements of gel preparations.\textsuperscript{31}

### pH measurement

The pH of any herbal gel should be such to assure gel stability and at the same time to ensure no risk of skin irritancy when applying the formula on skin. The pH for *Aloe vera* gel formulas was determined for CMC gel and CAR.934 formulas was in the range 5.0-7.0 and there was no significant difference (p≤0.05) in pH between the two sets of formulas. The pH was determined for the selected gel formula (F2) was determined to be 5.4.
Evaluation by *in-vitro* anti-microbial activity for the selected gel formula

In this study, gel formula F2 was selected for further anti-microbial efficacy investigation. The measurement of inhibition zone of F2 formula against bacteria was shown in Table5 and Figure 5) which revealed that zone of inhibition were ranged from (0-30 mm) against *E.coli* with high antibacterial activity against *S.aureus*.

Table5. The antibacterial activity of F2 Aloe vera gel against *E.coli* and *S. aureus*

<table>
<thead>
<tr>
<th>Microorganisms</th>
<th>F2 gel concentration mg/ml</th>
<th>Control</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>0.5</td>
<td>1.0</td>
</tr>
<tr>
<td>Zone of inhibition in (mm)</td>
<td></td>
<td></td>
</tr>
<tr>
<td><em>E.coli</em></td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td><em>S. aureus</em></td>
<td>30</td>
<td>30</td>
</tr>
</tbody>
</table>

Figure (5): The antibacterial activity of F2 Aloe vera gel (containing 1.5% m/v Carbopol.934) represented zone of inhibition against *E.coli* and *S.aureus* after incubation at 37±2°C for 1 days. Plate on left side represents antimicrobial activity of the gel against *E.coli* and plate on right side represents antimicrobial activity of the gel against *S.aureus*
According to the results of the activity of F2 Aloe vera gel against two types of bacteria (Gram positive, S. aureus, and Gram negative, E.coli) by using well-diffusion method, all the investigated Aloe vera concentrations inhibited the growth of S.aureus. However, there was no activity against Gram-negative bacteria E.coli except for the higher concentration of the gel (2mg/ml) as shown in Figure 5. Results of this study indicated that the prepared Aloe vera gel had higher anti-microbial activity against S.aureus than against E.coli. These results were in support with V.C. Pawar et al who observed a complete inhibition of the growth of S. aureus by the use of Aloe vera gel extract. In addition our results were agreed with Sahu RK et al who compared different plant extracts including Aloe vera against different types of bacteria including E.coli and they reported that the majority of bacterial species were susceptible to almost all the extracts of Aloe vera when used in high concentration.

CONCLUSION

Aloe vera was successfully prepared as a topical gel; this formula will help in the proper use of this herb for medical use as well as for cosmetic use for any person. Among all formulas that prepared, one formula was selected for further investigations, CAR.934 as a gelling agent revealed desirable results of morphological observations more than CMC which formed poor Aloe vera gel formulas. The selected gel formula exhibited good physicochemical characteristics including the homogeneity, appearance, consistency, color, no odor, no phase separation, good after feel, non-greasy type of smear and easy removal of gel and accepted pH. However; some challenges were facing this work including the high tendency for oxidation of Alov Ex which might lead to darkening the color to brown, but this problem could be resolved by the addition of antioxidant. It is recommended to use higher concentration of sodium sulphite or use a combination of two antioxidants to preserve the natural color of Aloe vera exudate. Moreover, it is better to add a natural perfume (fragrance) like Natural pearl essence or any other water soluble perfume for more acceptability of the gel to be topical with nice odor. In addition, the selected gel formula exhibited high anti-microbial activity in all concentrations of the gel against S.aureus which is the most common cause of skin infections, and good activity against E.coli, but only if it is in high concentration. For all above, the formulated topical gel of Aloe vera could be prepared easily and for its high anti-microbial activity. It could be regarded as a satisfactory candidate of use as a medicinal pharmaceutical preparation to be an efficient and safe alternative to the synthetic anti-microbial topical gel preparations to admit into the market.

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