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FORMULATION AND EVALUATION OF ALOE VERA HYDRO GEL USING COTTON SEED OIL

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ABSTRACT

The main objective of present work is to prepare and evaluate Aloe vera hydro gel using cotton seed oil and evaluate physical and microbial parameters for the formulated gel. Six different formulations were prepared using different concentrations of Sodium Carboxy methyl cellulose. After three days, the formulations were weighed, to check the percentage moisture loss for the gel and it was found to be less than 1gm. The pH of all formulations were checked initially after formulation and after 1 week. Viscosity also checked initially for all the formulations using Brookfield viscometer and found satisfactory for all formulations except Trial 1 and 2. Viscosity is an important feature to determine the resistance of flow of gel formulation so that it can spread on the skin properly. It was determined with the help of Brookfield viscometer using different spindles and concluded to use spindle number 2. Nutrient agar media was used for microbial growth study. Aloe Vera gel was aseptically transferred onto nutrient agar plates with blank and sample (n=2). Petri plates were used for microbial study and the gel samples were aseptically transferred on to the sample plates in a cross pattern. The inoculated plates were then incubated at optimal conditions for microbial growth. Daily microbial examination of the agar plates was checked and evaluated for a period of 14 days. Observations were checked for any visible growth, colony morphology and characteristics.

INTRODUCTION

Aloe vera is a type of succulent plant species of genus Aloe that comes under Asphodelaceae (Liliaceae) family. It is an evergreen perennial as it lives more than two years. It originates from Arabian Peninsula, situated northeast of Africa. It grows wild in tropical, subtropical and desert climates all over the world [1]. Aloe vera (syn. Aloe barbadensis Mill., Fam. Liliaceae), also known as Barbados or Curaçao Aloe, used in traditional and folk medicines for thousands of years to treat and cure variety of diseases [2]. As a potted plant, the species thrives inside and is visually pleasing [3]. Aloe vera grows to a height of 60–100 centimeters (24–39 inches) and spreads by offsets. It is a stemless or very short-stemmed plant. Thick and meaty, the leaves range in color from green to grey-green, and certain types have white speckles on the upper and bottom surfaces of the stems. The leaf's edge features tiny white teeth and is serrated [4].

It is grown for commercial usage, mostly for topical applications. Aloevera finds application in the food, pharmaceutical, and cosmetic sectors. It serves as a basic material for a variety of goods in the lavatory and cosmetic industries, including masks, skin moisturizers, soaps, shampoos, sun lotions, make-up creams, scents, shaving creams, and bath aids [5].

Identification of the phenolic components of aloe vera, including chrysofanol, Aloin A and B, Aloenin (B), and Aloesin [4]. Aloin, emodin, gum, and other components including essential oils are the main components of aloe vera fluid. Aloin is an antibacterial and antiseptic active component [5]. Three components make up Aloevera gel's carbohydrates: Mannan makes up 16.2% of the liquid, polysaccharide-rich microparticles (0.7%) include 0.7% of the liquid, and galactonic acid makes up 83.1% of the cell walls [6]. Mannose, glucose, galactose, galactose A, fructose, arabinose, and silosa make up the polysaccharides that were extracted from Aloe vera using alcohol. Their ratio is 120: 9: 6: 3: 2: 2: 1. The aloe vera gel's active polysaccharide and barbaloin components are most stable at 70 °C, and their stability decreases at temperatures above or below room temperature. When dissolved in methanol, barbaloin is an unstable component that changes into a number of unknown components [6].

This cells containing a transparent mucilaginous jelly which is referred as Aloe vera gel. Its composition is 99–99.5% water with the remaining 0.5–1.0% solid matter including over 75 distinct chemicals that may be active, such as organic acids, polysaccharides, phenolic compounds, sterols, salicylic acid, lignin, and saponin.

Active Constitutes of Aloe Vera:

About 98% of the aloe vera leaf gel is made up of water. Aloe vera gel has a total solid concentration of 0.66% and soluble solids of 0.56%, with significant seasonal variation. Polysaccharides make up 53% of the dry matter content of aloe gel, followed by sugars (17%), minerals (16%), proteins (7%), lipids (5%) and phenolic compounds (2%). The multifunctional action of aloe vera is caused by 200 potentially active ingredients, including vitamins, enzymes, minerals, carbohydrates, lignin, saponins, salicylic acids, and amino acids, which are responsible for the multifunctional activity of Aloe 7 - 9.

Vitamins: It has antioxidant vitamins A (beta-carotene), C, and E. Choline, folic acid, and vitamin B12 are also present. Free radicals are neutralized by antioxidants.

Enzymes: aliase, alkaline phosphatase, amylase, bradykinase, carboxy-peptidase, catalase, lipase, cellulase, and peroxidase are the eight enzymes found in it. When administered topically to the skin, brady kinase helps to minimize excessive inflammation, and other enzymes aid in the breakdown of fats and sugars.

Minerals: Zinc, magnesium, manganese, copper, selenium, potassium, sodium, and chromium are available. Although few of them are antioxidants, they are essential to many different enzyme systems in different metabolic pathways functioning correctly.

Sugars: It contains polysaccharides like polymannose and glucomannans as well as monosaccharides like glucose and fructose. They are called mucopolysaccharides and come from the plant's mucilage layer. Aloe vera has yielded novel anti-inflammatory chemical C-glucosyl chromone and a glycoprotein with anti-allergic characteristics named alprogen.

Anthraquinones:

It contains twelve anthraquinones, which are phenolic substances that have historically been used as laxatives. Aloin and emodin have antiviral, antibacterial, and analgesic properties.

Fatty acids:

It produces four plant steroids: campesterol, β -sisosterol, lupeol, and cholesterol. Each of them has anti-inflammatory qualities, while lupeol also has analgesic and antibacterial qualities.

Hormones: Gibberellins and auxins, which have anti-inflammatory and wound-healing properties.

Others: It offers seven of the eight essential amino acids and twenty of the twenty-two amino acids that humans need. Salicylic acid, which has antibacterial and anti-inflammatory qualities, is another ingredient in it. When added to topical medicines, the inert component lignin improves the other chemicals' ability to penetrate the skin. About 3% of the gel is made up of soapy

chemicals called saponins, which have antibacterial and cleaning qualities [6].

Aloe barbadensis is another name for aloe vera. Aloe vera gel is a whitish, mucilaginous gel. It is made from recently harvested aloe vera leaves. The leaves are meaty, prickly, marinated, and packed with a colorless gel that is 10 cm thick at the base and 30 to 60 cm long. Aloe vera contains a variety of chemicals with varying structural characteristics. Three components make up the leaf: rind, gel, and latex. The Aloe vera leaf tissue produces the gel. 98% of raw aloe vera pulp is made up of water. Mucilage is the transparent, viscous liquid that makes up 99.5% of parenchyma cells. Minerals, water- and fat-soluble vitamins, polysaccharides, organic acids, enzymes, and phenolic compounds make up the remaining 0.5 to 1% solid components. In addition, it has lipids, sterols, vitamins, and amino acids. Glycosides, anthraquinones, and yellow sap are present in latex. The gel matrix's outer, protective layer is known as the rind. The rind produces both protein and carbohydrates. Polysaccharides make up 77% of its dry matter content, followed by minerals (16%), sugar (17%), lipids (4%), protein (7%), phenolic compounds (1%), and other vitamins. Because of its ability to heal wounds, aloe vera gel is utilized to create a variety of products in the pharmaceutical and cosmetics industries.

Aloe vera gel has been used to treat a variety of skin conditions, including pigmentation, discoloration, sunburns, rashes, dry skin, acne, pimples, and scars or dark patches on the skin.

Sodium Carboxymethyl cellulose: Sodium carboxymethyl cellulose (Sodium CMC) is used as viscosity improving agent and it stops lotions and creams from separating and controls the thickness and consistency of liquids, creams and gels.

Disodium EDTA: Disodium EDTA is a salt of ethylene diamine tetra acetic acid (commonly known as EDTA). In cosmetics, it functions primarily as a chelating agent, meaning it prevents ingredients in a formula from binding with trace elements (mainly minerals) that can be present in water or other ingredients.

Glycerin: Glycerin is a denaturant, an ingredient in fragrances, a hair conditioner, a humectant, a medicine for oral health care, an oral care agent, a skin protectant, a humectant, and a viscosity-reducing agent.

Propylene glycol: Propylene glycol is used as a humectant. It is added to cosmetics to increase moisture retention in skin and hair. Propylene glycol is well accepted by the skin and shouldn't cause redness or irritation.

Sorbitol: Sorbitol serves as a humectant, flavoring, and skin-conditioning ingredient.

Methyl paraben and Propyl Paraben: Methyl paraben and Propyl Paraben used as

preservatives in cosmetic formulation. In combination both act as better preservative compared with individual use. Both in combination used to prevent the growth of harmful bacteria and mold that protect both the products and consumers.

Borax: It is used as emulsifier, buffering agent, and preservative for moisture product, cream, shampoo, gels, lotions, bath bombs and scrubs.

Borax is used in cosmetic industry to prevent or slow bacterial growth in moisturising products.

Cotton seed oil: It is used as skin softening emollient, rich in antioxidants, hydrating for the skin.

Rose Oil: Rose oil is used as flavouring agent.

MATERIALS:

The fresh Aloe vera (*Aloe barbadensis*) leaves were collected from our Agriculture garden, Rai University. Sodium Carboxy

methyl cellulose, Methyl paraben, propyl paraben, Sodium EDTA, Cotton seed oil, Propylene Glycol, Glycerin, Sorbitol, Rose Oil were collected from our store, School of Pharmacy, Rai University.

Extraction of Aloe vera extract:

We used thick, succulent Aloe vera (*Aloe barbadensis*) leaves that are collected from the Agriculture Garden at Rai University in Ahmedabad. Aloe vera leaves were gathered, cleaned with water and a moderate chlorine solution, and then sliced transversely into pieces in order to remove the mucilaginous jelly from the center (the parenchyma) of the plant. The leaf's interior, gel-like pulp was separated with a spoon, chopped, and homogenized in a mixer after the thick skin was carefully peeled off with a vegetable peeler.

Formulation Development of Aloevera Gel:
Formula

S N	Name of the Ingredients	Quantity in gm/100 gm					
		F1	F2	F3	F4	F5	F6
1	Aloevera Extract	75	75	75	75	75	75
2	Sodium CMC	3.0	3.0	3.0	3.0	4.0	4.0
3	Borax	-	-	-	0.5	-	-
4	Methyl Paraben	0.9	0.9	0.9	0.9	0.9	0.02
5	Propyl Paraben	0.1	0.1	0.1	0.1	0.1	0.002
6	Disodium EDTA	1.5	1.5	1.5	1.5	1.5	1.5
7	Propylene glycol	2.0 mL	2.0 mL		4.0 mL	-	-
8	Cottonseed Oil	2.0 mL	2.0 mL	4.0 mL	2.0 mL	4.0 mL	2.0 mL
9	Glycerine	5.0 gm	5.0 gm	10.0 gm	12.0 gm	10.0 gm	5.0 gm
10	Sorbitol	5.0 gm	5.0 gm	-	-	-	-
11	Rose Oil	1.0	1.0	0.2 mL	0.1 mL	0.1 mL	0.1 mL
12	Haladi Powder	-	2.0 gm	-	-	-	-
13	Distilled Water	Up to 100 gm	Up to 100 gm	Up to 100 gm	Up to 100 gm	Up to 100 gm	Up to 100 gm

Manufacturing Procedure:

Preparation of Aloe vera Extract:

1. Aloe vera leaf was washed properly with water, cut the upper portion with a sharp knife or peeler.
2. Collect the Aloe vera pulp in a beaker using a spoon.
3. Heat it for 30 minutes at 40°C using hot plate.
4. Cool it to room temperature and Aloe vera extract is ready to use.

Procedure for Aloe vera Gel:

1. Clean all the glassware, apparatus and equipment properly.
2. Weigh all the ingredients as per the manufacturing formula.
3. Transfer required quantity of Aloe vera extract in a suitable beaker (250 mL).
4. Gently kept it under mechanical stirrer and continue stirring to produce vortex.
5. Slowly transfer Sodium CMC to the beaker under stirring to avoid lumps formation.
6. Transfer Borax to the above beaker under stirring.

7. Transfer Methyl paraben, Propyl paraben and Disodium EDTA under stirring.
8. Continue stirring for 10 minutes.
9. Mix Cottonseed oil in required quantity of Propylene glycol, Glycerine and Sorbitol. Transfer the mixture solution to the above beaker under stirring.
10. Mix the Haldi powder in required quantity of Distilled water. Filter the solution using 80# mesh and transfer the solution to the beaker under stirring.
11. Add Rose oil to the above beaker dropwise for uniform mixing.
12. Continue stirring for 10 minutes.
13. Check the weight of the gel. Make the weight up to 100 grams using distilled water.
14. Continue stirring for another 10 minutes to form Aloe vera gel.
15. Cover the beaker using Aluminium foil and Store the final formulation gel in Freeze.

RESULT AND DISCUSSION:

Evaluation parameters:

Parameters	F1	F2	F3	F4	F5	F6
Homogeneity	-	++	+++	+++	+++	+++
Transparency	Opaque	Opaque	Opaque	Opaque	Opaque	Opaque
Gel appearance	Thin	Thin	Good	Good	Good	Good
Gel Consistency	Watery	Smooth	Smooth	Smooth	Smooth	Smooth
Color	Pale White	Yellow	Pale White	Pale White	Pale White	Pale white
Odour	No	No	No	No	No	No
Phase separation	Yes	No	No	No	No	No
After feel	Very Poor	OK	Good	Good	Good	Good
Type of Smear	Non-greasy	Non-greasy	Non-greasy	Non-greasy	Non-greasy	Non-greasy
Removal	Easy	Easy	Easy	Easy	Easy	Easy

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

Aloe vera is now advised for use by the majority of people on the planet due to its health and cosmetic benefits. The prepared gel formulas (F1-F6) were formulated from Aloe vera leaf exudates. Aloe vera exudate, a fluid-like juice, was obtained by the exudation process, which was carried out using a simple method. 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. To overcome this problem, Disodium EDTA was used as antioxidant in all the formulations.

All developed gel formulas were evaluated for homogeneity. The prepared F3, F4, F5 and F6 gel formulas containing 3.0, 3.0, 4.0 and 4.0 % w/w Sodium Carboxy Methyl cellulose respectively were homogeneous with no aggregates or clumping.

Appearance and consistency:

Appearance and consistency are most important characteristic features of gel formulations. The prepared (F4) gel formula containing 3.0% w/w Sodium CMC shows opaque, with pale white color, smooth texture, glossy appearance and was accepted as gel formulation.

Selection of the best formula

The proper Aloe vera gel formula was examined in terms of homogeneity, transparency, appearance, consistency, color, odor, phase separation, after-feel, kind of smear, and gel removal technique for the Aloe vera gel formulation with Sodium CMC as the polymer. Among all the prepared formulas of Aloe vera gel; the selected one was F4, which contains 3.0% w/w Sodium CMC and 2% v/v of cottonseed oil were best percent combination for preparation of gel.

pH: pH of the Aloe vera gel formulations were checked using suitable pH meter and was found within a range of 5.5 to 7 pH.. The pH of Aloe vera gel should be such to assure gel stability and at the same time to ensure no risk of skin irritancy when applying to the skin. The pH was determined for the selected gel formula (F4) was determined to be 5.9.

Microbial Study: After the 14-day incubation period, the microbial analysis revealed no observable growth of microorganisms on the agar plates inoculated with Aloe Vera gel formulation in batch F4. No microbial colonies were detected, indicating the absence of microbial contamination in the sample.

CONCLUSION:

Aloe vera was successfully formulated as a topical gel. This formula will help in the proper use of this herb for medicinal use and

for cosmetic use for public. Among all formulations prepared, one formula was selected for further investigations. Sodium CMC used as a gelling agent revealed desirable results of morphological observations. 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. Color changes to pale white color because of oxidation of the formulation. So recommend to use proper concentration of antioxidants to overcome this issue or to preserve the natural color of Aloe vera exudate. Moreover, Cottonseed oil was added to protect the formulation from oxidation, as cottonseed oil is rich in antioxidant. Furthermore, the chosen gel formula demonstrated strong anti-microbial action against *S. aureus*, the most frequent cause of skin infections, at all gel doses, and good activity against *E. coli*, albeit only at high concentrations. The results of this microbial study suggest that the Aloe Vera gel sample for Batch F4 is free from microbial contamination. This suggests that it can be used safely in a variety of applications without running the danger of contamination or microbial deterioration. For all above, the formulated Aloe vera

topical gel 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.

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