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Preparation of Topical Gel and Cream Formulations containing Lavender and Tea Tree Essential Oils and Evaluation of their Antimicrobial Activity

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ABSTRACT

Skin infections are among the most commonly occurring infections and hence effective topical formulations are continuously in demand. Often, these infections present therapeutic challenges to medical practitioners due to the growing concerns regarding multidrug-resistant pathogens. Keeping this in mind, the objectives of the current study were to formulate topical cream and gel containing essential oils of lavender and tea tree, and to study their antibacterial activity. Various batches of formulations were prepared in our study and evaluated for their organoleptic characteristics and physicochemical properties. The Minimum Inhibitory and Bactericidal Concentrations (i.e., MIC and MBC) of essential oils were determined by broth dilution method and viable count assay respectively. The MICs of lavender and tea tree oil were found to be 4% and 0.2%, and the MBCs were 3% and 0.1% respectively. Various topical formulations with 4% lavender oil and 0.2% tea tree oil were prepared and the one showing best pharmaceutical and physicochemical characteristics was further subjected to antibacterial assay. The batches C6 and C3C of cream and gel formulations respectively exhibited considerable antibacterial activity as observed by the zones of inhibition obtained in the range of 13-19mm.

Keywords: Cream, Formulation, Gel, Lavender oil, MBC, MIC, Tea Tree oil.

INTRODUCTION

The common skin infections are generally ignored by common people due to the low mortality rate associated with it. However, skin infections are among the main reasons requiring medical interventions and they are the 4th leading cause of non-fatal disease burden occurring globally[1]. Bacterial skin infections, alone, were recorded to infect 600 million individuals worldwide in 2013[2]. Factors like high population densities and poor sanitary practices combined with hot and humid climatic conditions, in developing countries like India, also increase the chances of skin infections[3]. A higher rate of skin infections are observed in people of old age[4], and belonging to poor socioeconomic background[5]. It can further present more complications in immuno-compromised individuals[6]. The seriousness of skin

infections can be contemplated from the fact that, in 2013, the US Food and Drug Administration (USFDA) classified a new category of "acute bacterial skin and skin structure infections" as a formal guideline for development of specific drugs[7].

Although skin itself acts as a natural barrier against the invasion of pathogens, through acidic sebaceous secretions and protective keratinous layer; cuts or breaks in skin can cause skin infections. These cuts may result from bruises, surgery, puncture, ulcer, animal or insect bites, thorn and needle pricks, or burns[8]. Sometimes, the normal flora of skin may act as an opportunistic pathogen and cause infections. *Staphylococcus aureus* is one such commensal mainly associated with skin infections and often with bacteraemia, infective endocarditis and respiratory infections[9]. A benign impetigo and uncomplicated cellulitis is the most common skin infection caused by *S. aureus*, but can also lead to life threatening complications. Currently, the majority of *S. aureus* strains have developed resistance to antistaphylococcal drugs, including the β-lactams, sulfonamides, tetracyclines, glycopeptides and others. Moreover, Methicillin Resistant *S. aureus* (MRSA) is increasingly spread from hospitals-settings to the community[10].

The treatment of skin infections typically involves the use of topical antimicrobial formulations. However, the emergence of antimicrobial resistance has led to the occurrence of complicated infections that require systemic antibiotics[11]. Many a times, a therapeutic unresponsiveness is observed towards known antibiotics during the treatment of complicated infections caused by multi-drug resistant pathogens. Thus, alternative treatment approaches are increasingly being explored.

Considering the current options, the essential oils are among the best alternatives of antimicrobial agents that can be included in topical formulation to treat bacterial skin infections[12]. India has a rich biodiversity, and Ayurveda, involving the use of plant based medicines, forms a traditional culture[13]. Over 100's of plant species have been reported in ayurvedic literature to treat dermatological disorders. Among these, the essential oils of lavender and tea tree are reported to possess extraordinary benefits in treatment of skin infections[14, 15].

In addition to being antiseptic and antimicrobial agent, lavender oil promotes granulation and tissue formation by collagen synthesis, differentiates fibroblasts and up-regulates TGF-β which accelerates wound healing and reduces scarring[16]. The two main constituents of the lavender oil, linalool and linalyl acetate, shows astringent and anti-inflammatory properties, and are highly effective in aromatherapy[16-18]. The presence of over 100 different bioactive compounds in tea tree oil allows its use as a safe alternative to broad spectrum antimicrobial agents. It is used in aseptic surgical and dental procedures, wound disinfection and inhalation therapies[19]. The primary constituent of tea tree oil i.e., terpenes (mainly monoterpenes and sesquiterpenes) are highly effective in skin rejuvenation[20]. They are also effective against other skin ailments like psoriasis, eczema etc.

Considering the scope of essential oils in treatment of bacterial skin infections, the objectives of the current study were determination of antibacterial activity of lavender and tea tree essential oils against *S. aureus*, and formulation of stable topical cream and gel containing these essential oils.

MATERIALS AND METHODS

Determination of Minimum Inhibitory and Minimum Bactericidal Concentration of lavender and tea tree essential oil

A 20% stock of lavender and tea tree oil each was prepared in Mueller Hinton (MH) broth, using 0.5% tween 20 as an emulsifier. Different concentrations of oil were prepared by broth dilution method in test tubes, and inoculated with 0.1mL of 24h old *S. aureus* (0.02 $O.D_{540nm}$) culture. These tubes were

incubated at 37°C for 24h. Two positive controls were set up to confirm the growth of test cultures in MH medium alone and in presence of tween 20, respectively. Similarly, 2 negative controls i.e., MH medium and the same containing tween 20, were also set up. Since the test tubes containing emulsified oils were turbid, the Minimum Inhibitory Concentration (MIC) and Minimum Bactericidal Concentration (MBC) of essential oils could not be determined by observing visible growth in test tubes. Hence, the MIC was determined by spot inoculating the samples from each tube on MH agar plates and incubating it at 37°C for 24h to check for inhibition of test culture. After obtaining the results of MIC,a viable count of bacteria was also performed from each tube to determine the MBC of essential oils. The MIC was defined as the lowest concentration of oil that inhibited the visible growth of bacteria after overnight incubation, and the MBC was defined as the lowest concentration of oil that killed over 99.9% test bacteria.

Determination of synergistic effect of lavender and tea tree oil

The two concentration each of lavender oil (3% and 4%)and tea tree oil (0.1% and 0.2%) were mixed in 4 combinations in MH broth. The synergistic effect in terms of MIC and MBC of lavender and tea tree oil was determined similarly as mentioned above.

Preparation of gel bases containing selected essential oils

A gel base was prepared by using different concentrations of xanthan gum, HPMC and Carbopol 970 as gelling agents. The preservatives (methyl paraben and propyl paraben) were dissolved in boiling water, cooled, and added to a suitable concentration of polymer dispersed in purified water using a magnetic stirrer. For Carbopol 970, an adequate amount of triethanolamine was added to neutralize its free carboxylic acid groups to obtain a pH of 5.5. The observed MBC of essential oils were incorporated into the prepared gel by continuous stirring with the help of a magnetic stirrer. Table 1 represents the formulation code for different batches of gelling agents. After preparation, the gel was transferred into a sterile container and stored for further evaluation of quality control characteristics.

Table 1: Composition of gel formulations

Sr. No.	Ingredients	Category	Formulation Code		
			X1A/H1A/C1A	X2B/H2B/C2B	X3C/H3C/C3C
1	Xanthan gum/ HPMC/ Carbopol 970	Gelling agent	0.4%/ 1%/ 1.5%	0.8%/ 2%/ 2%	1.6%/ 3%/ 2.5%
2	Methyl Paraben	Preservative	0.2%	0.2%	0.2%
3	Propyl Paraben	Preservative	0.02%	0.02%	0.02%
4	Tea tree oil	Anti-microbial agent	0.2%	0.2%	0.2%
5	Lavender oil	Anti-microbial agent	4%	4%	4%
6	Triethanolamine	Neutralizing agent	q.s	q.s	q.s
7	Distilled Water	Vehicle	q.s to 100g	q.s to 100g	q.s to 100g

Key: q.s- Quantity Sufficient

Preparation of cream bases containing selected essential oils

Table 2 represents the composition of cream formulations. The oil emulsion phase was prepared by mixing olive oil with stearic acid and cetyl alcohol (emulsifier) mixture. The aqueous phase was prepared by dissolving other ingredients i.e., glycerine, propylene glycol, methyl and propyl paraben, in water. Both the phases were heated at 75°C. The aqueous phase was added to the oil phase, with continuous stirring at 100rpm, once both the phases attained same temperature. The mixture was stirred continuously until it was cooled to room temperature, and the pH was adjusted between 5.5-5.7 with the help of triethanolamine. The observed MBC of essential oils were incorporated into prepared creams by continuous stirring with the help of magnetic stirrer. After preparation, the cream was transferred to a sterile container and stored for further evaluation of quality control characteristics.

Table 2: Composition of cream formulations

Sr. No	Ingredients	Formulation Code					
		C1	**C2**	**C3**	**C4**	**C5**	**C6**
1	Stearic acid	2g	2g	4g	4g	3g	3g
2	Cetyl Alcohol	1.5g	1.5g	2g	2g	2.5g	2.5g
3	Olive Oil	5mL	5mL	7mL	7mL	6mL	6mL
4	BHT	0.02g	0.02g	0.02g	0.02g	0.02g	0.02g
5	Methyl Paraben	0.2g	0.2g	0.2g	0.2g	0.2g	0.2g
6	Propyl Paraben	0.02g	0.02g	0.02g	0.02g	0.02g	0.02g
7	Disodium EDTA	0.02g	0.02g	0.02g	0.02g	0.02g	0.02g
8	Distilled water	20mL	20mL	20mL	20mL	20mL	20mL
9	Glycerin	-	-	-	-	-	5mL
10	TEA	1mL	1mL	1mL	1mL	1mL	1mL
11	Tea Tree oil	0.8mL	0.8mL	0.8mL	0.8mL	0.8mL	0.8mL
12	Lavendar oil	0.04mL	0.04mL	0.04mL	0.04mL	0.04mL	0.04mL

Evaluation of physical parameters of topical gel and cream formulation

Physical examination

The optimized gel and cream formulations were inspected visually for their appearance, colour, consistency and phase separation. The texture, grittiness and homogeneity were tested by visual inspection, and by applying a small amount on skin to sense its feel on application. In addition to above characteristics, the cream formulations were applied on the skin and assessed manually for the ease and extent of washability with water.

Transparency and smoothness

The 5g gel formulation was taken in a 10mL (Borosil) test tube and visually checked for its transparency. The smoothness of the gel formulation was tested by rubbing between the fingers. Similarly, other observations like formation of clumps, homogeneity and/or roughness were also noted.

Evaluation of pharmaceutical parameters of topical gel and cream formulation

Measurement of pH

The pH of gel and cream formulations were measured using a digital pH meter which was calibrated before each use with standard buffer solutions at pH 4.0 and 9.2. The measurement of pH of each formulation was done in triplicate and average values were calculated.

Viscosity

Viscosity measures the flow characteristics of topical formulations. The changes in viscosity of the formulations are indicative of poor stability of the product. The viscosity of gel and cream was measured by using a Brook-field viscometer with spindle number LV-61 and LV-64, respectively. It was lowered perpendicularly into the gel/ cream placed in a beaker, taking care that the spindle does not touch the bottom of the beaker. The spindle was rotated at a speed of 100 torque and the readings were recorded after 30sec for determining the viscosity of the gel. Similarly, the spindle was rotated at a speed of 20, 50, 60 and 100rpm to determine the viscosity of the cream.

Spreadability

A good spreadability depends on the viscosity of the cream/gel, and is one of the basic criteria in its formulation, to meet the quality standards. The efficacy of a topical therapy is directly proportional to, and is expressed in terms of, the extent to which it can spread readily on skin or affected area. This is extremely important to deliver an accurate dose of drugs to the target tissues. In the current study, the spreadability of topical formulations was measured on the basis of its slip and drag characteristic. For this purpose, the spreadability apparatus was used as reported by Chatterjee et al.[21] with slight modifications. The apparatus consisted of a wooden block with a pulley attached at one end, and a ground glass slide fixed on to it. A predetermined amount of formulation was placed on a ground glass slide. The formulation was sandwiched between this plate and another glass slide of the same dimensions attached to the other side of the wooden block with a hook. A 300g weight was placed on top of these slides to expel air and allow the formation of a uniform layer of gel/ cream between the slides. The excess formulation was wiped off from the sides of the slides. The upper slide was then subjected to a pull of 120g, with the help of a string attached to the hook, and the time required by the top plate to cover a distance of 10 cm was noted. The lesser the time interval to cover this distance, the lesser is the friction between glass slides; which indicates better spreadability. It was calculated using the formula

$$S = \frac{M\ x\ L}{T}$$

Where, “S” denotes spreadability, “M” is the weight tied to the upper plate, “L” is the length moved by the glass slide and “T” is the time taken to separate the glass slide.

Extrudability

Another requisite for an ideal gel/ cream is the measure of force required for its extrusion from packaged tubes. This is because the packing of gels and cream have considerable importance in delivery of desired quality. The extrudability test was carried out by using Pfizer hardness tester. For this purpose, each 15g sample of gel and cream was filled in an aluminium tube and a plunger was adjusted to hold the tube in

place. The pressure of 1kg/cm^2 was applied for 30sec and the quantity of gel and cream extruded were weighed. The percentage of gel and cream extruded from the tube was calculated and the formulations were graded accordingly (+++ Excellent, ++ Good, + fair). The procedure was repeated at three equidistant places of the tube and the tests were carried out in triplicates.

Determination of antimicrobial activity of cream and gel formulations

The antibacterial activity of cream and gel preparations was determined by the agar cup method. To carry out this technique, 0.2mL of culture was inoculated in sterile and molten MH agar butt and poured into the petriplates. The plates were allowed to solidify for 15min and cups were bored in the medium using a sterile cork borer. 100gms each of cream and gel formulations were dispersed in 3mL of DMSO, to allow diffusion of active ingredients in the medium, and a fixed volume (50µL) of this emulsion was added to the cups along with addition of DMSO to the control cup. The plates were incubated at 37°C for 24h and observed for zones of inhibition. Different concentrations of oils used in our study are indicated in Table 3.

Table 3: Different concentrations of oil dispersed in cream/gel formulations

Sr. No	Concentration (%)		Final concentration of essential oils (%)
	Lavender oil	Tea tree oil	
1	3	0.1	3.1
2	3	0.2	3.2
3	4	0.1	4.1
4	4	0.2	4.2
5	4	0.3	4.3
6	4	0.4	4.4

RESULTS AND DISCUSSION

Determination of MIC and MBC of essential oils and evaluation of synergistic activity

The MIC of essential oils was confirmed by observing growth inhibition on MH agar plates, and the MBC was evaluated on the basis of viable count. Table 4 and Table 5 represent the observation for the MIC and MBC values of essential oils. The MIC of lavender oil and tea tree oil was confirmed to be 4% and 0.2% respectively. The MBC of lavender oil and tea tree oil was observed to be 3% and 0.1% respectively. Table 6 represents the synergistic effect of lavender oil and tea tree oil in terms of MIC and MBC. and the combination of concentrations of 3 % lavender oil with 0.1% tea tree oil was both MIC and MBC against the test culture.

Table 4: MIC and MBC of test isolate at different concentrations of Lavender oil

Tube No.	Concentration of oils/ culture	MIC results as growth observed on MH plates	Viable count (cfu/ml)	Reduction in viability (%)
1	*S. aureus* (0.02 $O.D_{540nm}$)	+	$4.73x10^8$	
2	1% lavender oil	+	Uncountable	
3	2% lavender oil	+	Uncountable	
4	3% lavender oil	+	$2.43x10^5$	99.95
5	4% lavender oil	-	-	100
6	5% lavender oil	-	-	
7	6% lavender oil	-	-	
8	7% lavender oil	-	-	
9	8% lavender oil	-	-	
10	9% lavender oil	-	-	
11	10% lavender oil	-	-	
12	11% lavender oil	-	-	
13	12% lavender oil	-	-	

***Key: Growth +, No growth -**

Table 5: MIC and Viable count of test isolate at different concentrations of Tea Tree oil

Tube No.	Concentration of oils/ culture	MIC results as growth observed on MH plates	Viable count (cfu/mL)	Reduction in viability (%)
1	*S. aureus* (0.02 $O.D_{540nm}$)	+	$4.73x10^8$	
2	0.1% Tea Tree oil	+	$2.37x10^5$	99.95
3	0.2% Tea Tree oil	-	-	100
4	0.4% Tea Tree oil	-	-	
5	0.8% Tea Tree oil	-	-	
6	1.6% Tea Tree oil	-	-	
7	3.2% Tea Tree oil	-	-	
8	6.4% Tea Tree oil	-	-	
9	12.8% Tea Tree oil	-	-	

***Key: Growth +, No growth -**

Table 6: Viable count of test isolate at different concentrations of essential oils

Tube No.	Concentration of oils/ culture	MIC results as growth observed on MH plates	Viable count (cfu/mL)	Reduction in viability (%)
1	*S. aureus* (0.02 $O.D_{540nm}$)	+	$4.73x10^8$	
2	3% of lavender oil + 0.1% Tea Tree oil	-	$1.42x10^5$	99.97
3	3% of lavender oil + 0.2% Tea Tree oil	-	-	
4	4% of lavender oil + 0.1% Tea Tree oil	-	-	
5	4% of lavender oil + 0.2% Tea Tree oil	-	-	

***Key: Growth +, No growth -**

The essential oils typically possess the characteristic of hydrophobicity which allows them to partition between the lipid bilayer of the bacterial cell membrane. This allows increased cell permeability, leakage

of cellular components and ultimately cell death. Several studies have indicated the potency of essential oils as antibacterial agents. Our study showed a considerable reduction of 58%-59% in the viable count of test bacteria when a combination of essential oils was used (Tube 2, Table 6) as compared to the viable count obtained in presence of individual oils (Tube 5, Table 4 and Tube 3, Table 5). This indicates a potential synergistic effect between the components of essential oils of tea tree and lavender. The antifungal activity of tea tree and lavender essential oils has been reported, against *Trichophyton rubrum* and *Trichophyton mentagrophytes,* in an earlier study[22]. The antibacterial activities of these two oils are also reported previously. Both studies also reported synergistic activity between tea tree and lavender essential oils[23]. Among other sources, a similar study reported the MIC of lemongrass and thyme essential oils to be 30µL/mL and 4µL/mL, respectively, against MRSA strains[24]. The essential oil of the herb *Bidens tripartite* was found to be a promising agent against skin candidiasis[25]. The MICs of *Elettaria cardamomum* and *Cinnamomum zeylanicum* were reported to be below 20mg/mL against pathogenic bacterial and fungal isolates[26]. The essential oil of *C. martini* showed broad-spectrum antimicrobial activity with MIC value ranging from 0.65µg/mL-10µg/mL against bacterial (*K. pneumoniae*, *P. aeruginosa*, *S. aureus* and *S. pyogen*) and fungal (*A. niger*, *C. albicans, M. canis, T. mentagrophytes*, *T. rubrum* and *T. verrucosum*) skin pathogens[12].

Preparation of topical formulations and evaluation of quality parameters

The formulations were selected on the basis of observed physical and pharmaceutical quality parameters represented in Table 7 (for gel) and Table 8 (for cream). In the current study, the batch C3C showed good efficiency to entrap lavender oil (4%) and tea tree oil (0.1%) as compared to other batches. Hence the batch C3C represented in Table 1 was selected as ideal formulation for gel (Figure 1). Among the formulations of cream, batches C1 and C2 were found to be of very thin consistency and batches C3 and C4 were sticky. These batches did not show good homogeneous characteristics as well. Batch 5 was less sticky as compared to previous batches and also showed better homogeneity. However, the addition of glycerine in batch C6 resulted in smooth consistency, and no air entrapment, indicating uniform homogeneity. Hence batch C6 represented in Table 2 was selected as the ideal formulation for cream (Figure 2).

The physical parameters evaluated in our study can be characterised only on the basis of sensory perceptions of the product quality. There are no regulatory or selection criteria for standardising these parameters and evaluating the observations. As against other parameters that are required to comply with strict FDA protocols and standards available as specific formulation guidelines[27]. The pH of the gel and cream formulations were slightly in the acidic range, which is similar to the pH of skin. Hence, the risk of skin irritation due to changes in pH is negligible with these preparations. All excipients and their concentrations used in the formulations were also approved by the USFDA for topical applications[27]. The therapeutic efficacy of topical formulations is highly affected by characteristics like viscosity and spreadability. They were found to be in acceptable range in all formulations reported in our study. Also, the viscosities of the gels were found to be directly proportional to concentration of polymers used in our study. This observation is also in agreement with Shukr and Metwally[24] and Pena[28].

Table 7: Observed characteristics of gel formulations

Batch Code	pH	Viscosity (Cps)	Homogeneity	Spreadability ($g.cm^2/Sec$)	Colour	Feel on application	Extrudability	Consistency
X1A	6.32	45.27	Good	12.25	White	Smooth	++	Very Thick
X2B	6.45	54.38	Good	13.68	White	Smooth	+	Thick
X3C	6.40	57.62	Satisfactory	17.62	White	Smooth	+	Hard
H1A	6.2	19.50	Good	5.20	White	Smooth	++	Extremely thin
H2B	6.34	20.10	Good	7.13	White	Smooth	+	Very thin
H3C	6.29	24.56	Good	9.58	White	Smooth	+	Very thin
C1A	5.5	17.56	Good	4.98	White	Smooth	++	Extremely thin
C2B	5.6	18.69	Good	6.54	White	Smooth	+	Very thin
C3C	5.5	22.18	Good	8.94	White	Smooth	+	Thin

Table 8 : Observed characteristics of cream formulations

Batch Code	Physical appearance	pH	Texture	Grittiness	Spreadability (g.cm2/sec)	Extrudability	*Consistency	Feel on application
C1	Off white	5.5	Good	-	26.35	+	++	Sticky
C2	Off white	5.6	Good	-	25.69	+	++	Sticky
C3	Yellowish	5.5	Good	-	24.74	+	+	Sticky
C4	Yellowish	5.6	Good	-	23.45	+	+	Sticky
C5	Off white	5.6	Smooth	-	22.34	+	++	Sticky
C6	Off white	5.5	Very Smooth	-	19.25	++	+++	Smooth

***Key: Excellent +++, Good ++, Satisfactory +**

As indicated in our study, 0.75% Carbopol 940 was reported to be a good gelling agent in formulation containing lemon grass oil as antibacterial agent[24]. Similar findings are also reported by Pandey et al[26]. In another study, the carrageenan and HPMC were reported to be optimal gelling agents as compared to xanthan gum and guar gum for the metallic active ingredients i.e., copper and zinc sulphate. These formulations were

further reported to be stable up to 12 weeks after preparation and their consistency and appearance was comparable to the marketed products[29].

Figure 1: Formulation of gel Figure

2: Formulation of cream

Determination of antimicrobial activity of cream and gel formulations

Table 9 represents the antibacterial activities of cream and gel formulations. The antibacterial activity of cream formulation was not observed in our study. However, the gel formulation showed significant zones of inhibition indicating effective antibacterial activity. The gel formulation containing 4% Lavender oil and 0.2% Tea tree oil showed a zone of inhibition of 15mm. Interestingly, a higher concentration of tea tree oil (0.3%) along with 4% lavender oil showed a smaller zone of inhibition (10mm). Proper zones were not observed on further increase of the concentration of tea tree oil. This indicates the possible antagonistic effect of the essential oils or specific components of the gel. Hence, 4% of lavender oil and 0.2% of Tea tree oil was finalised for incorporation in formulation of gel.

Table 9: Antibacterial activity of cream (C6) and gel formulations (C3C)

Sr. No.	Final concentration of oil in the cream/gel mixture %	Zone of inhibition (mm)	
		Gel	Cream
1	3.1	12.4	-
2	3.2	13.6	-
3	4.1	14.2	-
4	4.2	15.7	-
5	4.3	10.3	-
6	4.4	No proper zone	-
7	Control	-	-

***Key: No zone of inhibition -**

A gel containing *Bidens tripartite* showed significant zones of inhibition against pathogenic *Candida* sp.[25]. The safety of this formulation was further reported with the help of MTT and Comet assays. A synergistic activity between zinc and copper sulphate has been reported in a previous study, which was used in cream and gel formulations[29]. A synergistic activity has also been reported between lemongrass and thyme essential oil. The study indicated that the zone of inhibition of formulations containing combinations of essential oils was almost double as compared to formulations containing individual oils[24]. The ointment formulation containing essential oil of *Eucalyptus citriodora* was reported to be more effective as compared to cream formulations in another study. The former showed growth inhibition in the range of 10-27mm, whereas the latter showed growth inhibition in the range of 05-20mm against pathogenic bacteria and fungi tested in their study[30]. Significant zones of inhibition was observed against *S. aureus* (15-23mm), *E. coli* (18-22mm), *A. niger* (15-25mm), *A. varies* (15-25mm) and *C. albicans* (14-20mm) with topical formulations containing essential oils of camphor and *C. sativum*, or plant extracts of *Elettaria cardamomum*, *Cinnamomum zeylanicum* and *L. Bipinnata*[26]. The observed activity was not affected when these plant-extracts and essential oils were incorporated in topical gels.

CONCLUSION

The formulation of gel and cream containing specific concentrations of essential oils are difficult because it may interfere with the consistency and thereby affect its stability. Our study successfully prepared stable formulations of cream and gel containing lavender and tea tree essential oils. However, the antibacterial activity of these essential oils was lost in cream formulation indicating the antagonist effect of one or more ingredients of the formulation and essential oils. Hence, further studies need to be carried out specifically to determine the ingredients responsible for the loss of antibacterial activity. The gel formulation showed promising antibacterial activity against *S. aureus*, thus suggesting it to be a promising topical agent that can be used against skin infections.

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