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Characterization and Antimicrobial Activity of Patchouli Essential Oil Extracted From Pogostemon cablin [Blanco] Benth. [lamiaceae]

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ABSTRACT

The physico-chemical properties of Philippine patchouli oil, hydro-distilled from fresh leaves and young shoots of *Pogostemon cablin* were characterized and found to be within the specifications set by the United States Essential Oils Society. Philippine patchouli oil and commercial patchouli oil have the same major components as shown by GC-MS analyses: patchouli alcohol, d-guaiene, a-guaiene, a-patchoulene, seychellene, [3-patchoulene, and transcaryophylene, with slightly lower concentrations in the Philippine oil. Using the disk diffusion method patchouli oil was found to be active against the gram-positive bacteria: Staphylococcus, Bacillus, and Streptococcus species. Fifty five percent [11/20] of community and only 14.8% [9/61] of hospital-Staphylococcus aureus isolates were susceptible to an MIC of 0.03% [v/v.] and Sixty-four percent or 23/36 of methicillin-resistant *Staphylococcus aureus* [MRSA] isolates was sensitive to patchouli oil at 0.06%, as opposed to only 44% or 11/25 of the sensitive strains. Philippine patchouli essential oil was also active against several dermatophytes at 0.25%. The bioactivity of the oil was stable when refrigerated at its natural pH in the dark. Calculations by linear regression [8] and extrapolation of data showed that the shelf-life [t 5o] of Philippine patchouli oil had 6 years of age when stored at 30°C.

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INTRODUCTION

Pogostemon cablin [Lamiaceae], or patchouli [puh-CHOO-lee] is a bushy herb which is a member of the mint family and hails from Indonesia, Malaysia, the Philippines. This plant is indigenous to the Philippines known as "patchouli" or "kabling" .It is also cultivated in southern China including Guangdong and Hainan Province and was called Guang Huo Xiang to differentiate it from the HuoXiang of the north due to the species Agastache rugosa that it resembles [27].

Pogostemon cablin is a tender perennial fragrant herb with soft, opposite, egg-shaped dark to medium green leaves that reach up to 3 inches [7.5 cm] long and about half as wide, and square stems; grows 2-3 feet in height. Plants in containers run about 12 inches [30 cm] tall with an equal spread giving a peculiar, exotic Oriental odor when rubbed. They are hardy in USDA zone 8-12. These plants survive in damp warm climate and are regarded as soil-exhausting plants [18].

Dried leaves of plant are steam-distilled to obtain the commercial "oil of patchouli." According to Zhao *et al*, Patchoulic oil is the volatile oil of Pogostemon cablin [Blanco] Benth. And the Chinese crude drug Herba Pogostemonis is widely used in the cosmetic and oral hygiene industries[28]. Patchoulic oil is one of the most important base materials used in perfumery with its strong fixative property. Thus, the strong-smelling oil taken from the leaves is used in perfumes, incense, detergents, and hair conditioners. It has been used in some cultures to prevent diseases. Aside from providing alluring oriental notes, patchouli oil imparts tenacity to the perfume. Patchouli oil is equally indispensable in soaps, cosmetics, and incense [5].

The leaves of patchouli plant contain 1.5-4% volatile oil composed mainly of patchouli alcohol [32-40%] and other sesquiterpenes such as pogostol, bulnesol, norpatchoulinol, a-guaiene, abulnesene and 13-patchoulene [1,7,10]. Patchoulic alcohol is commonly used as an indicator for the quality assessment of dried P. cablin. However, the complexity of the herbal constituents makes it difficult for using conventional gas chromatography [GC] for analytical purpose.[28] Other compounds found in the Patchouli oil include cycloseychellene, patchoulipyridine, epiguaipyridine, guaipyridine, benzaldehyde, cinnamaldehyde, limonene, camphene, a- pinene, 13-pinene, and eugenol [2, 5, 6, 22]. Patchouli alcohol and norpatchoulenol are mostly

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responsible for the odor of patchouli oil. In this study, the chemical composition, physico-chemical properties, antimicrobial activity against clinical isolates, and the bioactive property of patchouli oil are evaluated within a framework of standard pharmacological research.

Statement of problem:

As an aromatic crop, Pogostemon cablin yields an essential oil and is widely cultivated in South-eastern Asia. [26]

The volatile oil compositions of Herba Pogostemonis are collected from various of cultivation regions and the harvesting times are obviously different. Based on the chemical differences of the volatile oil compositions, Pogostemon cablin is divided into two chemotypes, Pogostone-type and Patchouliol-type. The former was cultivated in Guangzhou and Gaoyao regions, locally named as "Shipai Huoxiang"; the latter was locally named as "Hainan Huoxiang", cultivated in Wuchuan, Suixi and Leizhou regions of Guangdong Province and Wanning region of Hainan Province. The Pogostone-type contains rich oxygenated components, especially pogostone in the volatile oil compositions and poor non-oxygenated composition with patchouliol. The above chemical data may be used as the evaluation standard for the authentic Shipai Huoxiang. The Patchouliol-type contains similar quantities of oxygenated and non-oxygenated composition especially rich patchouliol with poor pogostone in oxygenated compositions, rich delta-guaiene and alpha-guaiene in non-oxygenation compositions.[23]

Preparations contain patchouli oil, patchouli alcohol; and/or relevant derivatives thereof are effective for inhibiting the asexual propagation of fungi and preventing the adhesion of microorganisms to surfaces, and filtering the media, adhesives, building materials, building auxiliaries, and for laundry detergents, cleaning compositions, rinse agents, fabric treatment compositions, hand washing compositions, manual dishwashing detergents, machine dishwashing detergents, cosmetic compositions, pharmaceutical compositions, oral hygiene compositions, dental care compositions, and denture care compositions that contain such preparations.[24]

According to Zhao [2005], Patchoulic oil is the volatile oil of Pogostemon cablin [Blanco] Benth. And the Chinese crude drug Herba Pogostemonis is widely used in the cosmetic and oral hygiene industries. Patchoulic alcohol is commonly used as an indicator for the quality assessment of dried P. cablin. However, the complexity of herbal constituents makes it difficult for using conventional gas chromatography [GC] for analytical purpose.[25]

Patchouli oil is a useful home remedy agent for skin ailments such as acne, athlete's foot, cracked and chapped skin, dandruff, eczema and other fungal infections [l2]. Investigations on the bioactivity of patchouli oil are limited. Locally, Torres *et al*. [20] reported on the activity of steam-distilled patchouli oil against reference strains *Staphylococcus aureus* ATCC 25923 [15.0 mm], *Escherichia coli* ATCC 25922 [18.1 mm], and *Pseudomonas aeruginosa* ATCC 27853

[7.0 mm]. Patchouli oil was also found to inhibit the growth of *Mycobacterium tuberculosis* H37Rv and *M. bovi.s ATCC* 35724 [17].

There have been numerous studies regarding the physico-chemical properties, chemical composition, antimicrobial activity against clinical isolates, and the stability of the bioactive property of patchouli oil in medical and pharmaceutical literature, respectively, but most of them have not focus on these properties in a cumulative experimental study. This study will provide a novel range of result regarding the objectives above.

Research importance:

Nowadays, most of the innovative methods have sought to find the herbal medical factors to include them in cosmetic and dermatologic ointments and lotion because of their little side effect and complications among patients in different races. Patchouli oil has been under a close attention especially in eastern traditional and herbal medical centers and its extracted derivatives were utilized in several different medical products, despite a progressive need for new biochemical effects of this oil in several standard centers to establish a novel production line in this field. Therefore, the result of this study may be beneficial for all cosmetic and dermatologic medical centers.

literature Review:

Numerous different studies are performed on the antimicrobial and metabolic activities of Patchouli oil as follows;

Feng *et al* [1999] analyzed the chemical constituents of the volatile oil of the stems and leaves of Pogostemon cablin collected from Leizhou county by means of GC-MS. They observed that the main constituents of Patchouli oil were patchouli alcohol, delta-guaiene, alpha-guaiene, seychellene, alpha-patchoulene, aciphyllene, trans-caryophyllene.[29]

Luo *et al* [1999] also analyzed the chemical constituents of the volatile oil of the stems and leaves of Pogostemon cablin collected from Gaoyao county, Guangdong province by means of GC-MS. They identified Sixty four compounds among them. The main constituents were pogostone [30.99% in stems, 21.31% in leaves, the same below], patchouli alcohol [10.26%, 37.53%], transcaryophyllene [4.92%, 6.75%], alpha-guaiene [2.27%, 6.18%] and seychellene [1.56%, 1.99%], etc. [30]

The antifungal and pesticide activity of Patchouli oil were also tested in different studie:.

Abe *et al* studied the effects of 12 essential oils popularly used as antifungal treatments in aromatherapy on growth of Candida albicans. Mycelial growth of C. albicans, which is known to give the fungus the capacity to invade mucosal tissues, was inhibited in the medium containing 100 micro g/ml of the oils: lemongrass [Cymbopogon citratus], thyme [Thymus vulgaris], patchouli [Pogostemon cablin] and cedarwood [Cedrus atlantica].[31]

Zhu *et al*. [2003] tested Patchouli oil obtained from Pogostemon cablin [Blanco] Benth and its main constituent, patchouli alcohol, for their repellency and toxicity against Formosan subterranean termites [Coptotermes formosanus Shiraki]; both of them were found to be toxic and repellent. Unusual tissue destruction was noted inside the exoskeleton of the termite after patchouli alcohol was topically applied to the dorsum [32].

Haze *et al*. [2002] investigated the effects of fragrance inhalation on sympathetic activity in normal adult subjects using both power spectral analysis of blood pressure fluctuations and measurement of plasma catecholamine levels. Fragrance inhalation of essential oils such as pepper oil, estragon oil, fennel oil or grapefruit oil, resulted in 1.5 to 2.5 fold increased in relative sympathetic activity representing low frequency amplitude of systolic blood pressure [SBP-LF amplitude] compared with inhalation of an odorless solvent, triethyl citrate [$P<0.05$, each]. In contrast, fragrance inhalation of rose oil or patchouli oil caused a 40% decrease in relative sympathetic activity [$P<0.01$, each]. Fragrance inhalation of pepper oil induced a 1.7-fold increase in plasma adrenaline concentration compared with the resting state [$P= 0.06$], while fragrance inhalation of rose oil caused a 30% decrease in adrenaline concentration [$P<0.01$]. their results indicated that fragrance inhalation of essential oils might modulate the sympathetic activity in normal adults[33].

Pattnaik *et al* tested the essential oils of aegle, ageratum, citronella, eucalyptus, geranium, lemongrass, orange, palmarosa, patchouli and peppermint for antibacterial activity against 22 bacteria including Gram-positive Cocci and rods and Gram-negative rods, and twelve fungi [3 yeast-like and 9 filamentous] by disc diffusion method. Lemongrass, eucalyptus, peppermint and orange oils were effective against all the 22 bacterial strains. Aegle and palmarosa oils inhibited 21 bacteria; patchouli and ageratum oils inhibited 20 bacteria and citronella and geranium oils were inhibitory to 15 and 12 bacterial strains, respectively. All twelve fungi were inhibited by seven oils [aegle, citronella, geranium, lemongrass, orange, palmarosa and patchouli]. Eucalyptus and peppermint oils were effective against eleven fungi. Ageratum oil was inhibitory to only four fungi tested. The MIC of eucalyptus, lemongrass, palmarosa and peppermint oils ranged from 0.16 to > 20 microliters ml-1 for eighteen bacteria and from 0.25 to 10 microliters ml-1 for twelve fungi[34].

like many plants within the Lamiaceae, Pogostemon cablin [patchouli] accumulates large amounts of essential oil. Patchouli oil is unique because it consists of over 24 different sesquiterpenes rather than a blend of different mono-, sesqui- and di-terpene compounds. To determine if this complex mixture of sesquiterpenes arises from an equal number of unique sesquiterpene synthases, Deguerry *et al* [2006] developed a RT-PCR strategy to isolate and functionally characterize the respective patchouli oil synthase genes. Unexpectedly, only five terpene synthase cDNA genes were isolated. Four of the cDNAs encode for synthases catalyzing the biosynthesis of one major sesquiterpene, including a gamma-curcumene synthase, two germacrene D synthases, and a germacrene A synthase. The fifth cDNA encodes for a patchoulol synthase which catalyzes the conversion of FPP to patchoulol plus at least 13 additional sesquiterpene products. Equally intriguing, the yield of different in vitro reaction products resemble quantitatively and qualitatively the property of sesquiterpenes found in patchouli oil extracted from plants, suggesting that a single terpene synthase is responsible for the bulk and diversity of terpene products produced in planta[34].

Research Objectives:

This study is seeking to focus on the following objectives:

1- Finding out the chemical composition of patchouli oil;
2- Defining the physico-chemical properties of patchouli oil;
3-Evaluatung the antimicrobial activity against clinical isolates;
4- Evaluating the stability of patchouli oil bioactive properties

Research Hypotheses:

The following hypotheses are proposed in this study:

1-Physico-chemical properties of patchouli oil may be regarded as an effective dermatological and antimicrobial treatment.
2- Patchouli oil has an effective antimicrobial activity against clinical isolates.
3- The stability and bioactive properties of patchouli oil make it as a suitable material in medical clinical application.

MATERIALS AND METHODS

Plant material:

The identification of plant was authenticated by the Philippine National Museum. Fresh leaves and shoots of *Pogostemon cablin* were obtained from Nagcarlan, Laguna, air-dried in the shade for 7 days, pre-weighed, placed in an Erlenmeyer flask, and submerged in tap water. *A* clavenger tube was attached to the condenser, and the hydro-distillation [16] was carried out slowly for 4 hrs until no more oily drops were collected. The oil was dehydrated with anhydrous sodium sulfate. The patchouli oil was stored in amber bottles and refrigerated prior to use.

Determination of physico-chemical properties:

Using the USP standard procedures [21], the physicochemical properties of patchouli oil were determined and compared with standard specifications.

Determination of chemical composition:

The components of patchouli oil were determined using gas chromatography and gas chromatography-mass spectroscopy.

Gas chromatographic analysis [GC]:

Two milliliter-aliquots of tested oil and a commercial foreign oil [Aromaworld, USA] were loaded separately with a split-type injector into the injection port of a Shimadzu GC 14B with a Supelco omegawax 250, 30 x 0.25 mm, 0.25 um film capillary column. A Flame Ionization Detector [FID] was used and programmed at 70-190°C with 5°C/min increase in temperature. Peak spiking was done.

Gas chromatography-mass spectroscopy analysis [GS-MS]:

Prior to the analysis, both oils were extracted with methanol: ethyl acetate [90:10]. One microliter-aliquot of either oils was injected into the Hp 6890 with 5972 MSD, column RTX-5, 30 x 0.32 mm x 320 um film capillary column, detector [FID], programmed at 60-280°C, 15°C/min; intel pressure of 0.28 kg/cm ^{2}; Helium as carrier gas; and 0.2 ml/min constant flow. Identification of mass spectra was done through the database and retention indices.

Determination of antimicrobial activity- Preliminary screening on 4 reference and 9 clinical strains were done using the modified disk diffusion method [14, 16]. Twenty microliters of oil was dispensed into 6.35 mm-diameter disks. The oil disks were pressed at equidistant points on the surface of 4-mm deep Mueller Hinton agar seeded with approximately 108 cfus/ml of inoculum in the logarithmic phase. The oil was allowed to diffuse through the medium for 60 minutes. The plates were incubated at 37°C for 24 hrs. The zones of inhibition were measured to the nearest tenth of a millimeter with a Fisher-Lilly antibiotic zone reader.

The modified agar dilution method [15] was used to assay the activity of the oil against 61 hospital [25] methicillin-sensitive and 36 methicillin-resistant] and 20 community *Staphylococcus*

aureus strains and 6 dermatophytes. Serial two-fold dilutions of patchouli oil, emulsified in 20% Tween 80, were prepared and mixed with Mueller Hinton agar or Sabouraud dextrose agar to final concentrations of 0.0078-2.0% [v/v]. A density of approximately 10^{4} cfus/ml per organism in the logarithmic phase was spot-inoculated on the surface of Mueller Hinton agar: patchouli plates [MHA] for bacteria, and Sabouraud dextrose agar:patchouli plates [SDA] for dematophytes. The MHA plates were incubated at 35°C for 24 hours, while the SDA plates were incubated at room temperature for 5 days. The minimum inhibitory concentration [MIC] was read as the last clear spot or the minimum concentration of patchouli oil that inhibited the growth of organism.

Determining the stability of patchouli oil bioactive properties- Patchouli oil was stored for three months under different conditions: Ph levels (pH 2, 3.5, 4.8, 7, 9), temperatures (4°, 28°, 37°, 45°C), flint and amber bottles for presence and absence of light. Every 14 days, the samples were withdrawn and the effects at these different conditions on the bioactivity of oil were assayed by disk diffusion method using *Staphylococcus aureus* ATCC 25923.

Results:

Philippine patchouli oil- Hydrodistillation of plant material yielded 3.4% of patchouli essential oil. The extracted oil was clear, greenish-yellow, with an aromatic, woody Oriental odor.

Physico-chemical properties of Philippine patchouli oil- Table 1 presents the physico-chemical properties of local patchouli oil. It also shows that the values are within the specifications set by the Essential Oil Association [EOA] of the United States of America [5].

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Composition of Philippine patchouli oil:

The gas chromatographic analysis of local and commercial patchouli oil was conducted with two reference standards: Transcaryophyllene and limonene. Transcaryophyllene was detected in both oils at 20.54 and 34.97%, respectively. Limonene was not detected in either oils. Using GC-MS, 26 chemical components were detected; some of them present in one, but absent in the other oil. This could be attributed to factors such as age, season of harvest of source plants, climate, type of soil, fertilization and/or duration of oil extraction. Figure 1 shows the major composition of Philippine patchouli oil as compared with the commercial oil: patchouli alcohol, d-guaiene, a-guaiene, a-patchoulene, seychellene, [3patchoulene, transcaryophylene]. The results indicate that the major components present in both local and commercial oils, with slightly lower concentrations in the former.

Table 1: Physico-chemical properties of Philippine patchouli oil.

Properties	Philippine patchouli oil	EOA specifications [5]
Physical		,
Specific gravity, 20°C	0.9724	0.950-0.975
Refractive index, 20°C	1.5098	1.5070-1.5151
Optical rotation	-52" 4'	No data
Solubility in 90% alcohol v/v	1/2	up to 1 /10
Chemical		
Acid value, mg/g	1.64	Max. 5
' Ester value, mg/g	4.45	10, max ISO
Saponification value	11.5	No data

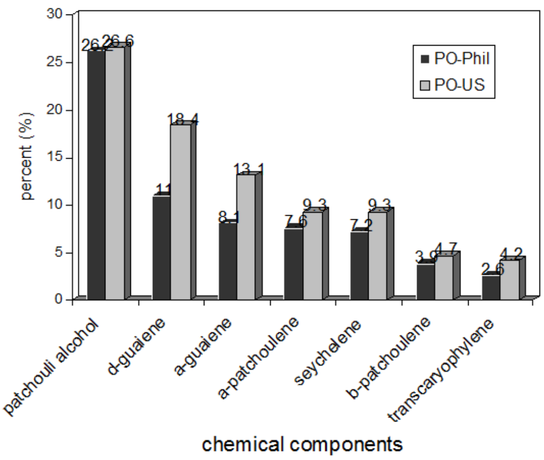

Fig. 1: Major chemical components of Philippine and US patchouli oils [PO].

Arrtimicrobial activity: Preliminary screening analyses on the antimicrobial activity of patchouli oil using the modified disk diffusion method shows that patchouli oil is active against gram positive bacteria: Staphylococcus, Bacillus and Streptococcus species [Table 2]. The growth of gram negative bacteria and yeast isolates were not inhibited in any degree. The results indicate that patchouli oil has a narrow spectrum of activity, being active only against gram-positive organisms.

Table 2: Antimicrobial spectrum of Philippine patchouli oil.

Organisms	Mean zone of inhibition [mm]
Reference strains	
Staphylococcus aureus ATCC 25923	22.5
Escherichia coli ATCC 25922	N I
Pseudomonas aeruginosa ATCC 27853	NI
Candida albicans ATCC 10231	N I
Clinical isolates	
Bacillus subtilis	27.0
Staphylococcus epidermidis	17.2
Streptococcus oralis	18.5
S. pneumoniae	16.6
S. constellatus	18.0
S. pyogenes	18.3
S. mitis	18.2
Salmonella typhi	NI
Klebsiella pneumoniae	NI

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Disk diffusion method, Disk diameter: 6.35 mm, NI: No inhibition:

Difference between the susceptibility of hospital and community S. aureus isolates to Philippine patchouli oil: A total of 81 clinical Staphylococcus aureus isolates, 61/81 or 75.3% from hospital patients [25/61 methicillin-sensitive, MSSA; 36/61 methicilin-resistant, MRSA] and 20/81 or 24.7% from community cases were tested for sensitivity to Philippine patchouli oil using the modified agar dilution method. Figure 2 shows that 65% of both hospital 39/61 and community 13/20 clinical isolates were sensitive to an MIC range of 0.03-0.06%, v/v.

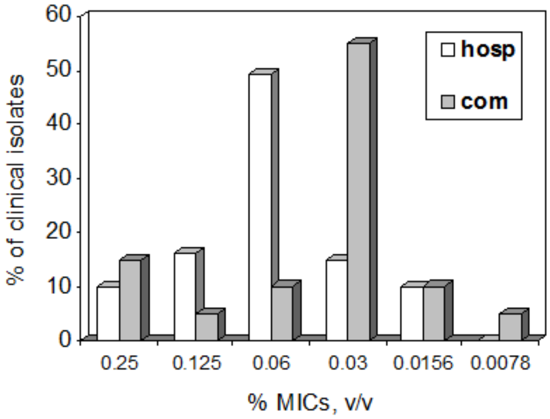

Fig. 2: Susceptibility of patchouli oil to hospital and community *S. aureus isolates.*

Difference between the susceptibility of hospital MSSA and MRSA isolates to Philippine patchouli oil- Figure 3 presents the comparative susceptibility of methicillin-sensitive and methicillin-resistant S. *aureus* isolates from hospital samples. The results indicate that the majority of isolates were sensitive to 0.03%-0.06% of Philippine patchouli oil: 75% 27/36 of MRSAs and 45% 12/25 of MSSAs.

The overall results suggest that Philippine patchouli oil is more active against the community isolates than hospital isolates; and better than MRSAs and then MSSAs.

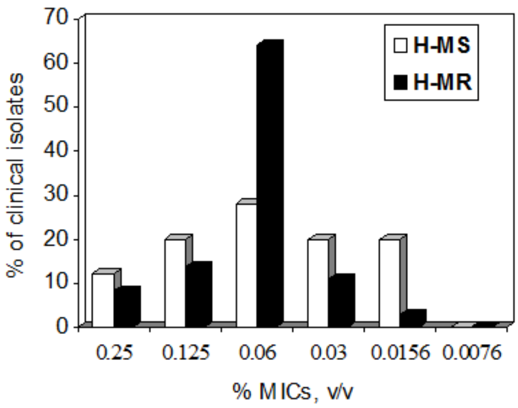

Fig. 3: Susceptibility of patchouli oil to hospital MSSA [H-MS] and MRSA [H-MR] isolates.

Activity of patchouli oil against dermatophytes:

The modified agar dilution was used to test the activity of patchouli oil against dermatophytes. The results [Table 3] indicate that the local patchouli oil is also active at 2.0%, v/v, against filamentous fungi isolated from clinical cases. The further findings suggest that patchouli oil is not active against *Candida albicans,* which was also not inhibited in the disk diffusion method.

Table 3: Antidermatophytic activity of Philippine patchouli oil.

Dermatophytes	MIC [%]
Epidermophyton floccosum	2.0
Microsporum gypseum	2.0
Microsporum canis	>2.0
Trichophyton metagrophyte	2.0
Trichophyton rubrurn	2.0
Candida alhicans	>2.0

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Stability of the bioactivity of Philippine patchouli essential oi:-

The biological stability of Philippine patchouli oil against S. *aureus* ATCC 25923 under different parameters of pH, temperature, and light during 12 weeks of storage showed that the oil is most stable when stored in amber bottles on its original pH 4.8 and under refrigeration as shown in Figures 3 a-c.

Figures 3a. Effect of temperature on the anti-staphylococcal activity of patchouli oil

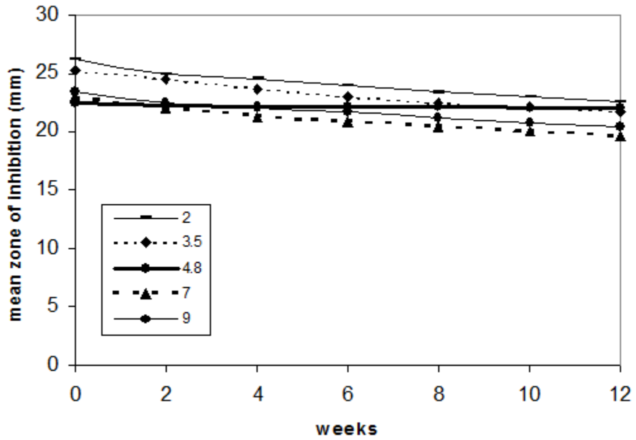

Fig. 3b: Effect of pH on the anti-staphylococcal activity of oil.

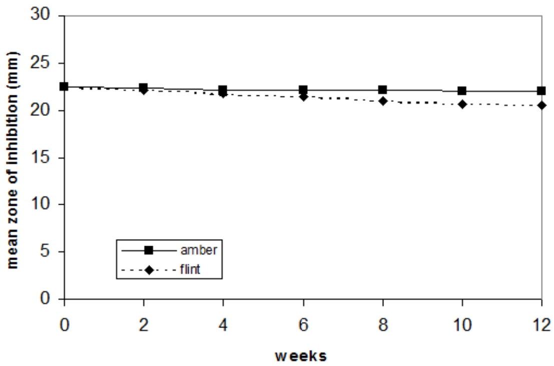

Fig. 3c: Effect of light on the anti-staphylococcal activity of the oil.

The decrease in potency may be due to the photo-oxidative effect of light and the effect of high temperatures on the active volatile components of patchouli oil. Calculations by linear regression [4,8] and extrapolation of data show that the shelf-life [t5o] of Philippine patchouli oil is 6 years when stored at 30°C.

Discussion:

Essential oils are the aromatic principles responsible for fragrance of plants and have found their ways in industrial applications. Aside from being applied in the perfumery and cosmetic products, the essential oils are also used medicinally. In the Philippines, eucalyptus oil is dropped in a cotton ball which is hung beside a sleeping child having difficulty of breathing due to colds. In Australia, the tea tree oil is being sold over the counter for skin cuts and bruises. Essential oils are generally recognized as having antimicrobial activity; a property that is really well known and being used regularly. Thyme oil and clove oils are applied for teeth cavities to prevent the infection. Pogostemon cablin [patchouli], like many plants within the Lamiaceae, accumulates large amounts of essential oil. Patchouli oil is unique because it consists of over 24 different sesquiterpenes, rather than a blend of different mono-, sesqui- and di-terpene compounds.[35]. According to Feng [1999], essential oil of P. cablin mainly contributes to the pharmacological activities and the therapeutic properties of essential oils are directly correlated with their qualitative and quantitative composition.[29]

The in vitro antimicrobial property of essential oils has been well documented [3, 11, 19,]. Pattnaik *et al* [1996] observed that patchouli oil inhibited 20 bacteria. [34]

This study focuses on the antimicrobial property of patchouli oil hydrodistilled from leaves of plants grown in the Philippines. It has been found to have the same major chemical components as the commercial oil. Its physico-chemical characteristics are within the standard specifications set by the Essential Oils Association of the USA. In a study by Hammer *et al*. [9], patchouli oil was found to be active against standard strains of S *aureus* and C. *albicans* at MICs of 0.25% [v/v] and 0.5% [v/v], respectively, using the agar dilution method. However, using the tube dilution method, the same study indicated a >2% MIC against C. *albicans*, which seems to contradict his findings with the agar dilution method. Abe *et al* also proved that Mycelial growth of C. albicans, which is known to give the fungus the capacity to invade mucosal tissues, was inhibited in the medium containing 100 micro g/ml of the oils: lemongrass [Cymbopogon citratus], thyme [Thymus vulgaris], patchouli [Pogostemon cablin] and cedarwood [Cedrus atlantica].[31]

In our study, the findings on the inactivity of patchouli oil against *Candida albicans* were corroborated in both disk diffusion and agar dilution methods. The patchouli oil MICs against S. *aureus* clinical isolates, exhibited in this study, are similar to those reported by Hammer *et al*. [9]. Patchouli oil was also found to be active against food-borne *Listeria monocytogenes [13]*. According to the above findings, efforts should be made to explore the use of patchouli oil as alternative raw material in pharmaceutical preparations.

Conclusion:

This oil is effective against gram positive organisms and several dermatophytes collected from wound and skin infections. The patchouli oil has advantage of possessing both antibacterial and antifungal activities. Furthermore, patchouli oil can be used as an alternative medication in cases where the skin infections are caused by strains that have develop resistance to antibiotics available in the market.

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