GC-MS Analysis of Essential oils and their Antibacterial activity of Staphylococcus species

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Abstract
The purpose of the study is to isolate and identify the staphylococcal swabs collected from hospitalized patients of Kovai Medical College and Hospital. The samples were cultured on nutrient broth and mannitol salt agar plates. These staphylococcal swabs were tested on different antibiotics by using disk diffusion (Kirby Bauer) method. Antibiotic impenem shows higher activity in S.haemolyticus and S.hominis. Staphylococcus aureus are resistant/ intermediate to amoxicillin, cefoxitin, linezolid, vancomycin and ticarcillin. These two species of S.hominis and S.haemolyticus are resistant to cefpodoxime. Thus these antibiotics can be preferred for Staphylococcal infections.

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1. Introduction

The increasing resistance of microorganisms to conventional chemicals and drugs has prompted scientists to search for novel sources of biocides with broad-spectrum activities. Since ancient times, plants and their derivatives, such as essential oils (EOs), have been used in folk medicine. Extracts from many types of plants used as flavoring and seasoning agents in food and beverages have been used therapeutically for centuries. However, in present times, bacteria have developed resistance to many of the antibiotics commonly used to treat infections. Staphylococcus sp is the pathogen most often and prevalently involved in skin and soft tissue infections. Methicillin resistant Staphylococcus aureus (MRSA) is an example of “Superbug” or bacterial strain of S.aureus that has developed resistance to methicillin, an antibiotic which is part of a class known as the beta-lactams. Beta-lactum antibiotics are capable of killing a variety of Gram positive and Gram negative microorganisms by impairing the cell’s ability to form peptidoglycan, a necessary component for the cell structure and stability. Bacteria that have become resistant developed enzymes known as penicillinases and beta-lactamases, which are capable of destroying the beta-lactam ring structure or functional group of these antibiotics. Chronic nosocomial infections by gram-positive bacteria have become more prevalent in recent years with the increased use of prosthetic biomedical implants. Staphylococcal infections are a major source of patient morbidity and implant failure. Staphylococcus aureus causes potentially life threatening nosocomial and community-acquired infections, such as osteomyelitis and endocarditis. The opportunistic pathogen S.aureus can form biofilms on many host tissues and implanted medical devices often causing chronic infections. In recent years, multidrug resistant strains have developed. Methicillin-resistant S. aureus (MRSA) is a special strain that is resistant to the antibacterial activity of methicillin and other related antibiotics of the penicillin class. Although, MRSA has traditionally been seen as hospital-associated infections, community –acquired MRSA strains have appeared in recent years (Puja Sharma et al., 2012).

Essential oils:

Essential oils extracted from the buds, flowers, leaves, and other parts of the plant have long been used as natural remedies and are now beginning to
gain an increased interest for their antimicrobial properties. Essential oils are chemical compounds consisting mainly of hydrogen, carbon and oxygen, and fall into the following two groups: hydrocarbons, which are mostly terpenes, and oxygenated compounds, such as esters, aldehydes, ketones, alcohols, phenols and oxides. For centuries, people have been using essential oils, or fluids extracted from plants, in aromatherapy as well as topical uses, such as soaps for routine washing and antiseptics for healing wounds. The capability of essential oils to act as antiseptics stems from the need of plants to be able to withstand harsh elements of nature, including a large degree of deadly microorganisms. These extracted plant oils have been well documented to provide a natural and highly effective antimicrobial substance. In addition, essential oils were found to be more effective against Gram positive bacteria, including MRSA and MSSA, than Gram negative bacteria. This evidence suggests the more effective permeability barrier Gram-negative bacteria possess may better restrict the penetration of amphipathic compounds than Gram positive microbes (Elgayyar, et.al.). Essential oils of medicinal plants have been used for hundreds of years of natural medicines to combat a multitude pathogens, including bacteria, fungi and viruses. In this study, ten essential oils were used:

**Basil oil** (*Ocimum basilicum*) strengthening and calming to the heart and mind, making it perfect for overwhelming, stress, fatigue. It is mostly used for respiratory, muscular and joint support.

**Orange oil** (*Citrus sinensis*) has a wide variety of domestic, industrial and medicinal uses and also used in biological pest control green pesticides. Orange oil can be attributed to its properties as an anti-inflammatory, antidepressant, antispasmodic, antiseptic, etc.

**Cardamom oil** (*Elettaria cardamomum*) can be attributed to its ability to reduce spasms, neutralize the adverse effects of chemotherapy, reduce nausea, as well as its qualities as an antiseptic, antimicrobial, aphrodisiac, astringent, stimulant, etc.

**Lemon oil** (*Citrus limon*) are one of the most popular citrus fruits in the world and are widely used for culinary purpose, since they are a good source of vitamins and aid in digestion. It has a powerfully fresh traditional lemon fragrance that is quite energizing and uplifting.

**Clove oil** (*Eugenia caryophyllata*) is or euthanize a natural analgesic and antiseptic, used primarily in dentistry for its main ingredient eugenol. It is used to relieve toothache and is commonly used to anesthetize laboratory or pet fish.

**Olive oil** (*Olea europaea*) used in cosmetics, medicine, cooking and soaps, also used as a fuel for traditional lamps. It helps in reducing heart disease risk and breast cancer risk, it maintains healthy cholesterol levels.

**Jojoba oil** (*Simmondsia chinensis*) is a fungicide, and can be used for controlling mildew. It found as an additive in many cosmetic products. Pure oil itself may be used on skin, hair, or cuticles.

**Rosemary oil** (*Rosmarinus officinalis*) is one of the most popular essential oil for its wide array of health benefits. It also used during wedding ceremonies, food preparation, cosmetic care and medicinal herbal care. Several essential oils confer antimicrobial activity by damaging the cell wall and membrane, leading to cell lysis, leakage of cell contents and inhibition of proton motive force. In addition, there is evidence that they effectively kill bacteria without promoting the acquisition of resistance and they possess multiple antimicrobial activity i.e., antibacterial, antifungal, anticancer, antiviral and antioxidant properties against all pathogens. Many essential oils are relatively easy to obtain, have low mammalian toxicity and degrade quickly in water and soil, making them relatively environmentally friendly (Juli Lawless, Rockport, MA: Elements Books, 58-67). The aim of the study is to collect the staphylococcus culture and to identify and determine their antibiotic sensitivity test of these ten essential oils were done by using agar diffusion (Kirby Bauer) method on muller-hinton agar and antimicrobial test of ten oils were determined by using agar disk(6mm) diffusion method. The evaluate different compounds present in essential oils by using Gas chromatography-Mass spectroscopy (GC/MS) method.

### 2. Aim
The aim of the study is to analyse GC-MS of essential oils and to evaluate antibacterial activity against Staphylococcal species.

**Objective**

To collect essential oils

To identify the culture

To detect antibiogram of staphylococcus species

To evaluate antibacterial activity of ten essential oils
To identify compounds present in essential oils by GC-MS.

3. Review of Literature

Plants have long been the principal tools of traditional medicinal system. Although ancient in origin, many traditional medical paradigms and their pharmacopoeias have evolved in to quite sophisticated system, using thousands of plants and their natural system. The rural folk and tribals in India even now depend largely on the surrounding forests for their day to day needs. Medicinal plants are being looked upon not only as a source of health care but also as a source of income. India has a rich diversity of medicinal plants. Extensive study has been done by various workers for the antibacterial and antifungal activities of various plant extracts from time to time. Although there is no authentic record of medicine used by ancient people, yet Rigveda which is the oldest book in the library of man provide enquisitive information about the medicine used by them. Atharvaveda, the another religious book of Hindus has described about 2000 plants having medicinal value. Sushruta Samhita (1000 BC) further record 700 plants of medicinal properties. Beside these there have been a number of workers from time to time who have described the medicinal importance of plants namely Charak, Watts, Kirtikar & Basu, Nandkarni, Chopra etc. Indian Materia Medica accounts for nearly 35000 species under various crude drugs both of indigenous and exotic origin. John Fleming in 1810 published “A catalogues of Indian medicinal plants and drugs”. Voravuthikunchai and Kitpipit, (2005) studied the activity of medicinal extracts against hospital isolates of Methicillin resistant Staphylococcus aureus (MRSA). Aqueous and ethanol extracts of ten traditional Thai (Thailand) medicinal plants were investigated for their ability to inhibit 35 hospital isolates of Methicillin resistant Staphylococcus aureus (MRSA). For thousands of years essential oils have been used for curing illnesses. The Ancient Greeks were the first to use and understand the medical purpose of oregano oil. In the 15th century a physician would use oregano oil to treat diarrhea, psoriasis, vomiting, jaundice, and fungal infections. Essential oils extracted from the buds, flowers, leaves, and other parts of the plant have long been used as natural remedies and are now beginning to gain an increased interest for their antimicrobial properties. Since ancient times, plants and their derivatives, such as essential oils (EOs), have been used in folk medicine. In nature, EOs play an important role in the protection of plants. They also may attract some insects to promote the dispersion of pollens and seeds or keep away other undesirable insects. Thus, EOs can play a role in mediating the interactions of plants with the environment.

Essential Oils

Essential oils are concentrated volatile aromatic compounds produced by plants - the easily evaporated essences that give plants their wonderful scents. Each of these complex precious liquids is extracted from a particular species of plant life. Each plant species originates in certain regions of the world, with particular environmental conditions and neighboring fauna and flora. Essential oils are frequently referred to as the “life force” of plants. Unlike fatty oils, these “essential” oils are volatile, highly concentrated, substances extracted from flowers, leaves, stems, roots, seeds, bark, resin or fruit rinds. The amount of essential oils found in these plants can be anywhere from 0.01 percent to 10 percent of the total. That's why tons of plant material are required for just a few hundred pounds of oil. These oils have potent antimicrobial factors, having wide range of therapeutic constituents. These oils are often used for their flavor and their therapeutic or odoriferous properties, in a wide selection of products such as foods, medicines, and cosmetics. Beware of imitations. Essential oils cannot be substituted with synthetics. Only pure oils contain a full spectrum of compounds that cheap imitations simply cannot duplicate. Essential oils are extracted from oil 'sacs' in flowers, leaves, stems, roots, seeds, wood and bark. They differ significantly from the well-known vegetable, nut and seed oils which are made up of various fatty acids (essential oils are not). Essential oils are used by the plants in somewhat the same way they are by humans - they fight infection, contain hormone-like compounds, initiate cellular regeneration, and work as chemical defense against fungal, viral, and animal foes. Despite their foliar origins however, essential oils have a similar structure to some compounds found in blood and tissues, allowing them to be compatible with our own physiology (Virendra et al., 2007).

Uses of Essential Oils

The most effective way to use most essential oils is by external application or inhalation, though some can be very beneficial when taken internally. The use of essential oils include body oils, compresses, cosmetic lotions, baths, hair rinses, inhalation by steam, perfumes and room sprays. Essential oils are very potent - some will cause skin irritation or have other harmful effects if not used properly. Unless specifically noted, it is best to dilute all essential oils in a carrier of base oil like Almond, Jojoba or Apricot Kernel before applying to the skin - appropriate dilution is usually only 1 - 10%

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essential oil in carrier. For inhalation, a diffuser or oil lamp is effective for releasing essential oils into your environment - a very pleasant way of creating a particular atmosphere. EOs are concentrated natural products with strong smells that are produced by aromatic plants as secondary metabolites. These oils are present as variable mixtures of primarily terpenoids, especially monoterpenes (C10) and sesquiterpenes (C15), although diterpenes (C20) may also be present. A variety of other molecules also occur, such as acids, alcohols, aldehydes, aliphatic hydrocarbons, acyclic esters or lactones; rare nitrogen- and sulphur-containing compounds; coumarins; and homologues of phenylpropanoids. EOs are liquid, volatile, limpid and coloured and are soluble in lipids and organic solvents that have a lower density than water. They can be present in all plant organs, including buds, flowers, leaf, seeds, twigs, stems, flowers, fruits, roots, wood or bark, but are generally stored by the plant in secretory cells, cavities, canals, glandular trichomes or epidermic cells. EOs are extracted from various aromatic plants that are generally found in temperate or warm countries, where they often represent an important part of the traditional pharmacopoeia. These plants may be known for their antioxidant effects as well as their antiseptic and medicinal properties and fragrance and are often used in the preservation of foods and as analgesics, sedatives, anti-inflammatory, spasmolytics and local anaesthetics. EOs contain a wide series of secondary metabolites that can inhibit or slow the growth of bacteria, yeasts and moulds. The EOs and their components have a variety of targets, particularly the membrane and cytoplasm, and in certain situations, they completely alter the morphology of the cells. Essential oils are extracted from oil ‘sacs’ in flowers, leaves, stems, roots, seeds, wood and bark. They differ significantly from the well-known vegetable, nut and seed oils which are made up of various fatty acids (essential oils are not). Essential oils are used by the plants in somewhat the same way that they are used by humans - they fight infection, contain hormone-like compounds, initiate cellular regeneration, and work as chemical defense against fungal, viral, and animal foes. Despite their foliar origins however, essential oils have a similar structure to some compounds found in blood and tissues, allowing them to be compatible with our own physiology. The most effective way to use most essential oils is by external application or inhalation, though some can be very beneficial when taken internally. The use of essential oils include body oils, compresses, cosmetic lotions, baths, hair rinses, inhalation by steam, perfumes and room sprays. Essential oils are very potent - some will cause skin irritation or have other harmful effects if not used properly. Unless specifically noted, it is best to dilute all essential oils in a carrier of base oil like Almond, Jojoba or Apricot Kernel before applying to the skin - appropriate dilution is usually only 1 - 10% essential oil in carrier. For inhalation, a diffuser or oil lamp is effective for releasing essential oils into your environment - a very pleasant way of creating a particular atmosphere. It is the use of aromatic essential oils to benefit the body – in emotional and physical health and beauty. Science has discovered that our sense of smell plays a significant role in our overall health. Many common essential oils have medicinal properties that have been applied in medicine since ancient times and are still widely used today. For example, many essential oils have antiseptic properties, though some are stronger than the other. In addition, many have an uplifting effect on the mind, though different essential oils have different properties. The non toxic, all natural, economically feasible essential oils have a broad spectrum of antimicrobial properties has led them to become increasingly studied.

Basil oil:

Sweet basil oil comes from basil (Ocimum basilicum), a plant with a thick foliage and small white flowers. Fresh basil leaves taste sweet and pungent, and exudes a fresh and floral aroma, while the dried ones have a spicy and earthy scent. The “dot-like” oil glands in fresh basil leaves produce the essential oil of the herb. Its dried leaves and stems are used in food flavorings and in the production of essential oil. Basil has a strong, spicy aroma that is invigorating to the spirit, bringing a sense of balance and calms the mind. This great oil is one of the oils included in Young Living Raindrop Technique. However, it is believed that oil obtained from the flowers is superior in quality compared to the oil from the whole plant. There are several types of basil oil such as European type or sweet basil, reunion type, methyl cinnamate type, and eugenol type. (Shirley Price et.al.) The commonly used basil oils in aromatherapy are the sweet basil and the exotic type. Sweet basil oil is produced in the US, France, Italy, and Spain, while the exotic type is from Comoro Islands or Seychelles. Sweet basil oil is made up of d-linalool with 55% and estragole (methyl chavicol) with 70%. A variety of the essential oil contains methyl
cinnamate of 28%. Sweet basil oil contains other properties such as 1, 8-cineole, eugenol, borneol, ocimene, geranial, anethol, 10-cadinols, B-caryophyllene, a-terpineol, camphor, 3-octanone, methylugeinol, safrole, sesquithujene, and 1-epibicyclosesqui-phellandrene. It also contains juvocimene 1 and juvocimene 2. Take note that variations of these chemical properties may exist depending on the source of the plant. Basil, especially its leaves and seeds, is used mainly for culinary purposes all over the world. It’s a well-known fact that basil leaves are widely used in Italian cuisines like salads and pasta. However, the essential oil from sweet basil is also often added in various foods such as spiced meats, sausages, tomato pastes, ketchups, pickles, and fancy vinegars. Dental creams and mouthwashes also use sweet basil oil as one of their ingredients. Sweet basil oil is used in making perfumes, and can act as an insect repellent in your home as it possesses insecticidal agents that can repel flies and mosquitoes. Sweet basil oil has diaphoretic, stimulant, carminative, and expectorant properties. It is effective in dull hair and skin, and is also used in treating acne and skin infections. Sweet basil oil also offers health benefits for our: Digestive system: Due to its carminative properties, basil oil helps reduce indigestion, constipation, stomach cramps, and flatulence. It helps expel gas from your stomach and intestine. Immune system: The herbal oil works great in treating sinus congestion, asthma, bronchitis, and emphysema. It also helps effectively cure infections in wounds or cuts. Nervous system: Sweet basil oil is known to alleviate mental fatigue, migraine, and depression. It is commonly used in aromatherapy for its calming effect. Basil oil clears the mind and provides mental strength. Receiving a massage using this oil may help stimulate blood flow and soothe muscle pain and spasms. In case of earache, dilute sweet basil with three drops of a carrier oil then massage it over and around our ear. Sweet basil oil can be added to baths when combined with other essential oils such as jojoba, sweet almond, or avocado oil. The herbal oil can also be inhaled using a diffuser or vaporizer.

Orange oil:

Orange (Citrus sinensis) belongs to the Rutaceae family. The fruit comes from a small grayish-brown bark tree, which is almost similar to a shrub. Its branches grow to a regular hemisphere shape and oval-shaped foliage that sprouts to at least three to four inches. Its flowers are distinguished by its aromatic characteristic, and the circular fruit is rougher and darker in comparison to other varieties of citrus. It is said to be first obtained from a native tree in China, but has also been cultivated widely in the Mediterranean region, France, North and South America, and Portugal. Orange oil is extensively utilized in aromatherapy as it helps soothe tensed muscles as well as depression. It can also be useful in maintaining a youthful appearance when applied externally. (Verzera et al., 2004)

Uses of Orange Oil:

Orange is made of up various chemical properties such as alpha pinene, citronellal, geranial, sabine, myrcene, limonene, linalool, and neral. The most significant property is the limonene or D-limonene as it plays an important role in your home by repelling insects. Simply spray or wipe it on the different places of your house. D-limonene extracted from the rind of orange can be used as a cleaning agent both in household and industrial settings. This chemical may also be utilized as a replacement for acetone, toluene, glycol ethers, fluorinated and chlorinated organic agents when used as a straight solvent. Orange oil is commonly used as an added flavoring to beverages, sweet meats, chocolates, biscuits, confectionery, and baked goods. Industrially, it is utilized as a concentrate for room fresheners, deodorants, soaps, body lotions, and creams. Orange oil can provide a lot of benefits to your health. It has anti-inflammatory, antiseptic, antidepressant, antispasmodic, and diuretic properties. It helps in treating erectile problems, impotence, and decreased libido. The oil can also heal wounds to prevent any fungal or septic infection while disinfecting the lesion. Orange oil can help cure inflammation in the body by reducing pain and irritation. It also has a carminative property that helps expel intestinal gas, which induces chest pains and indigestion, by relaxing the stomach and anal muscles. Orange is significant in treating muscular and nervous spasms. It aids in relieving anxiety, anger, and depression. Aside from its healing characteristics, it is also beneficial in promoting a healthy wellbeing as it detoxifies your body and boosts your immunity. (Pino et al., 2006) Orange oil is beneficial in improving digestion and relieving constipation. It helps prevent cellular mutations and fights the growth of any cancerous tumor. The essential oil is also good for nourishing dry, irritated, and acne-prone skin. It is also
effective for the treatment of calluses on your feet and supports collagen formation on your skin. The orange promotes a feeling of happiness and warmth when used in aromatherapy. It helps in the elimination of toxins in the body and deals well with digestive problems. It helps in stimulating lymphatic action to promote balance in water processes and results in detoxification of your body. (Steinke et al., 2013)

**Lemon oil:**

Lemon essential oil, also known as Citrus limon oil, is extracted from lemon peels, which have many volatile oil glands in their pits. Approximately 1,000 lemons are needed to produce one pound of lemon oil. The citrus scent of lemons comes from a chemical compound called limonene, which comprises the majority of lemon oil. Lemons were used by ancient Indians, Egyptians, and Romans as a remedy for infectious diseases. Today, they are commonly grown in Europe and the United States, and are used in various products. Limonene makes up majority of lemon oil (about 50 to 70 percent). Lemon oil contains organic compounds, such as pinene, terpinene, and terpineol, and non-volatile compounds. Lemons are packed with a potent nutritional profile and are a low-glycemic fruit. The fruit contains citric acid, which is a natural preservative and makes up eight percent of the lemon. Lemons are also excellent sources of vitamin C or ascorbic acid, a potent water-soluble antioxidant. In spite of their acidic nature, they have an alkalizing effect when metabolized in your body. Lemons also have phenomenal antioxidant properties due to unique flavonoid compounds. In a study published in the Journal of Food Composition and Analysis, lemons, as well as other citrus fruits, contain a dominant flavonoid class called flavanones, such as hesperidin and eriocitrin. These nutrients are found to have anti-inflammatory and free-radical quenching properties. Lemons also contain other antioxidants, such as alpha- and beta-carotenes, beta-cryptoxanthin, zeaxanthin, and lutein. Other nutrients that can be found in these citrus fruits are: Vitamin A, Folate and B-vitamins, Iron, Copper, Potassium, Calcium, Pectin (a type of fiber). Apart from being used in making lemonade, lemon oil can be used as:

- Food ingredient – A tasty addition to pastries like pies, cakes, and marinades
- Aromatherapy oil and air freshener because of its pleasant citrus fragrance
- Medicine – Used to treat health problems like headaches, diabetes, high blood pressure, respiratory problems, and asthma
- Gargle – To relieve sore throat, mouth inflammations, and tonsillitis
- Treatment for calluses and warts – Applying it to affected areas regularly can help reduce the appearance of these on your skin.
- Skin and hair product – Can help treat blemished skin, exfoliate dead skin, and reduce acne. It can also improve appearance of hair and used to remove dandruff. Lemon oil is also added to personal care products.
- Disinfectant – An ideal disinfectant for chopping boards, countertops, public bathrooms, and even hospitals
- Spot remover – Can help remove gum, oil, and grease stains from clothes and surfaces
- Insect repellent and insect bite treatment – Works as natural repellent and antibacterial

**Benefits of Lemon Oil**

Because of lemon’s outstanding nutrient profile, it helps treat numerous health conditions. Lemon oil helps:

- Fight respiratory infections like bronchitis, coughs, and sore throat. It can also bring relieve to discomfort from colds, fevers, flu, and asthma.
- Support immune function by stimulating the production of white blood cells
- Inhibit inflammation
- Soothe acidity and heartburn, as it is helps balance your stomach pH level.
- Address constipation and promote elimination of waste
- Relive arthritis, due to its anti-inflammatory properties
- Lower blood pressure and promote healthy blood circulation
- Detoxify your skin and body of toxins
Control overactive sebaceous glands that contribute to acne and poor hair conditions
Reduce broken capillaries and varicose veins
Sanitize wounds, household objects, and hospital rooms due to its antibacterial properties
Calm anxiety, improve mood, and prevent emotional outbursts and violent behavior

Clove oil:

Clove oil has been used for thousands of years in many cultures for its many health benefits. Clove (Eugenia caryophyllata) can be found in nature as the unopened pink flower buds of the evergreen tree. Picked by hand the buds are dried until they turn brown and, after grinding them, the powder is used in cooking or converted into an essential oil for various medicinal purposes. Unlike most other spices, cloves can be grown throughout the entire year, which has given native tribes that use it a distinct advantage over other cultures because the health benefits can be enjoyed more readily. Considerably high in manganese (126.4%) and other nutrients including potassium, magnesium, and calcium – which is used to prevent and treat osteoporosis, anemia and premenstrual syndrome (PMS) (Chung, 2013) History tells us that the Chinese have used clove for more than 2,000 years as a fragrance and spice and that it hit the international health scene several hundred years later. Since then, it has been applied in numerous products for agricultural and cosmetic purposes. The most profound properties of clove oil, however, are related to its widespread application in homeopathic natural medicine. Because it contains an elevated level of eugenol, clove essential oil has proven to be remarkably versatile and has been thoroughly researched as an effective alternative to many modern medical treatments. Although it has been suspected for centuries to treat inflammatory conditions, the Journal of Immunotoxicology just recently published the first ever study proving that the eugenol in clove oil is indeed a powerful anti-inflammatory. Showing that low doses of eugenol can protect the liver against disease, it was also observed that eugenol reverses inflammation and cellular oxidation (which causes aging). It was also observed that taking large doses internally could harm the digestive lining and externally can irritate sensitive skin. Because of its strength, clove oil should be mixed with a carrier oil like coconut oil or other gentle oils for most topical applications and only used for short periods of up to two weeks internally. In addition to being a proven anti-inflammatory, clove oil is commonly used as an antiseptic for oral infections and as a broad-spectrum antimicrobial to keep countless diseases at bay. Clove is rich in minerals such as calcium, hydrochloric acid, iron, phosphorus, sodium, potassium, and vitamin A and vitamin C. The health benefits of clove oil can be attributed to its antimicrobial, antifungal, antiseptic, antiviral, aphrodisiac and stimulating properties. The oil is used for treating a variety of health disorders including toothaches, indigestion, cough, asthma, headache, stress and blood impurities. The most important and common use of clove oil is in dental care. Several toothpastes, mouthwash and oral care medications contain clove oil as an important ingredient. (Jesudasan, 2015)

Eucalyptus oil:

Eucalyptus oil is a pure essential oil that has practical and industrial uses, as well as healing properties. It comes from a fast-growing evergreen tree native to Australia, with global eucalyptus oil production mainly from Eucalyptus globulus or "Blue Gum." Learn more about this essential oil – and how it can benefit your health and well-being. Eucalyptus oil is the distilled oil that comes from the dried leaves of eucalyptus – a colorless liquid with a strong woody and sweet smell. There are more than 700 different species of eucalyptus in the world, of which at least 500 produce a type of essential oil. (Serafino, 2008) Eucalyptus globulus – This is the species that has received the most attention from botanists and chemists, and its oil is best known and most used of all eucalyptus oils. It's cineole is between 60 and 70 percent. Since the properties of the crude oil usually do not meet the requirements of most pharmacopoeias, the oil has to be treated to increase the cineole content.
Eucalyptus oils are made up of more than 100 different compounds. Its main chemical components are a-pinene, b-pinene, a-phellandrene, 1,8-cineole, limonene, terpinen-4-ol, romadendrene, epiglobulol, piperitone and globulol. Crude eucalyptus oil sometimes contains more compounds in different quantities than after double distillation. For example, eucalyptus globulus oil has 60 percent cineole and 40 percent other compounds, which becomes 80 percent cineole and only 20 percent other compounds after being double-distilled. As a result, crude oil may have a broader range of activities than the refined oil. The diluted form of eucalyptus oil is taken orally for pain and inflammation of respiratory tract mucous membranes, coughs, asthma, bronchitis, sinus pain and inflammation, and respiratory infections. It is also used as an antiseptic, insect repellent, and treatment option for wounds, burns, and ulcer. (William, 2002) For healing mucus membranes (such as to safely and effectively treat allergies and asthma), you can apply a drop of eucalyptus oil on an organic cotton ball and sniff it several times a day. I also recommend adding a few drops to water or a nebulizer as steam therapy, or you can use a few drops in your bath water. Eucalyptus oil is also popularly used as a fragrance in perfumes and cosmetics, and is found in mouthwashes, liniments and ointments, toothpastes, cough drops, and lozenges. It is commonly mixed with other oils to make it more easily absorbed by your skin. This supports the moisturizing process, which explains the oil’s presence in skin products like a natural sunscreen. There are different ways to use eucalyptus oil, including aromatically, topically, or internally. For instance, it can be applied to skin within a carrier oil, such as coconut oil. I advice starting with a drop added to 1 to 3 teaspoons of carrier, and increase the essential oil as necessary.

**Olive oil:**

Olive oil is a fat obtained from the fruit of the *Olea europaea* (olive tree), a traditional tree crop of the Mediterranean region, where whole olives are pressed to produce olive oil. Olive oil, rich in monounsaturated fatty acids, is a major component of the Mediterranean diet. Populations from that region have longer life expectancies and lower risks of heart disease, high blood pressure and stroke, compared with North Americans and Northern Europeans. (Ruth Schuster, 2014). Monounsaturated fatty acids (MUFAs) are considered a healthy dietary fat, as opposed to saturated fats and trans fats. The oil is used in cosmetics, medicine, cooking and soaps, and was also used as a fuel for traditional lamps. Although originating in the Mediterranean countries, today it is used worldwide. The main type of fat found in all kinds of olive oil is monounsaturated fatty acids (MUFAs). MUFAs are actually considered a healthy dietary fat. If your diet replaces saturated and trans fats with unsaturated fats such as MUFAs and polyunsaturated fats (PUFAs), you may gain certain health benefits. (Riley, 2002) MUFAs and PUFAs may help lower your risk of heart disease by improving related risk factors. For instance, MUFAs have been found to lower your total cholesterol and low-density lipoprotein cholesterol levels. MUFAs may also help normalize blood clotting. And some research shows that MUFAs may also benefit insulin levels and blood sugar control, which can be especially helpful if you have type 2 diabetes. But even healthier fats like olive oil are high in calories, so use them only in moderation. Choose MUA-rich foods such as olive oil in place of other fatty foods — particularly butter and stick margarine — not in addition to them. And remember that you can’t make unhealthy foods healthier simply by adding olive oil to them.

**Cardamom oil:**

Cardamom (*Elettaria cardamomum*) essential oil is a member of the ginger family of botanical plants. Cardamom has historical uses that include internal consumption to protect the stomach and inhalation to invigorate the mind. Cardamom has also been used traditionally to alleviate mental fatigue and nervous strain. Modern scientific research has not yet validated these traditional claims. The health benefits of Cardamom Essential Oil can be attributed to its ability to reduce spasms, neutralize the adverse effects of chemotherapy, reduce...
nausea, as well as its qualities as an antiseptic, antimicrobial, aphrodisiac, astringent, digestive, stomachic, stimulant, and diuretic substance. The main constituents of its essential oil are Sabinene, Limonene, Terpenene, Eugenol, Cineol, Nerol, Geraniol, Linalool, Nerodilol, Heptenone, Bornol, Alpha Terpineol, Beta Terpineol, Terpinyl Acetate, Alpha Pinene, Myrcene, Cymene, Neryl Acetate, Methyl Heptenone, Linalyl Acetate and Heptacosane. Apart from its culinary uses, you are probably familiar with it as a mouth freshener. It is the essential oil in cardamom that makes it such a good digestive. This oil boosts digestion by stimulating the whole digestive system. It is also stomachic in nature, which means that it keeps the stomach healthy and functioning properly. It helps maintain the proper secretion of gastric juices, acids and bile in the stomach. It also protects the stomach from infections. Cardamom essential oil stimulates your entire system. This stimulating effect also boosts your spirits in cases of depression or fatigue. It also stimulates the secretion of various enzymes and hormones, gastric juices, peristaltic motion, circulation, and excretion, thus maintaining proper metabolic action throughout the body. Cardamom oil has a warming effect. This means that it heats up the body, promotes sweating, helps clear congestion and coughs, while also relieving symptoms of the common cold. It also provides relief from headaches that result from illness and can be used to cure diarrhea caused by extreme cold. Cardamom oil helps to neutralize the effects of tobacco, insect bites and even the ingestion of mild poisons. Furthermore, it can clear the bowels, cure colic, remove bad breath, heal oral infections, and alleviate the pain of toothaches.

JOJOBA OIL:

Jojoba (pronounced ho-HO-ba) oil is a botanical extract of the seed of the jojoba tree (Simmondsia chinenis). Technically it is not actually an oil, but rather what is called a wax ester. Because out of all of the compounds in nature, this wax ester is the most similar to human skin oil (sebum). It is theorized that applying jojoba to the skin can "trick" the skin into thinking it is producing enough oil, thus balancing oil production. (Lynn, Maggie, 2011).

Provides all day moisturization – jojoba oil doesn't evaporate like water based moisturizers can.

It is theorized, but not yet proven, that because jojoba oil is so similar to human skin oil, it can "trick" the skin into thinking it has produced enough oil and thus balance oil production.

Very stable – jojoba oil does not become rancid or lose antioxidants even after long periods of storage. Spreads well and absorbs well.

The organic variety is free of pesticides, GMOs, and is sustainably farmed.

Alzheimer’s disease. Early research suggests that massaging the hand with jojoba oil does not improve emotions, aggression, or mental function in people with Alzheimer’s disease.

Mosquito repellant. Early research suggests that applying a specific product containing jojoba oil, coconut oil, rapeseed oil, and vitamin E (Three Bio-skincare) to the skin might be effective as a mosquito repellant, with effects lasting for at least 3 hours after application.

Acne.
Psoriasis.
Sunburn.
Chapped skin.
Hair loss.

Other conditions. The appropriate dose of jojoba depends on several factors such as the user's age, health, and several other conditions. At this time there is not enough scientific information to determine an appropriate range of doses for jojoba. Keep in mind that natural products are not always necessarily safe and dosages can be important. Be sure to follow relevant directions on product labels and consult your pharmacist or physician or other healthcare professional before using.

Rosemary oil:
One of the most popular essential oils around is extracted from *Rosmarinus officinalis*, which is widely known in the Mediterranean region for its culinary and herbal benefits and has been extensively used for a wealth of health and wellness purposes. Related to mint and looking like lavender, rosemary has leaves like flat pine needles touched with silver. It boasts of a woody, citrus-like fragrance that has become a feature of many kitchens, gardens, and apothecaries worldwide. It derives its name from Latin words ros ("dew") and marinus ("sea"), or "dew of the sea." The Virgin Mary is said to have spread her blue cloak over a rosemary bush as she rested, and the white flowers turned blue. The shrub came to be known as the "Rose of Mary." Rosemary was considered sacred by the Egyptians, Hebrews, Greeks, and Romans, and was used in the Middle Ages to ward off evil spirits and protect against the plague. Rosemary oil has a clear, refreshing herbal smell, is clear in color, and is watery in viscosity. It is extracted from the fresh flowering tops through steam distillation, yielding 1.0 to 2.0 percent. Its health benefits made it a favorite of Paracelsus, a German-Swiss physician and botanist who contributed greatly to the understanding of herbal medicine in the 16th century. He valued rosemary oil because of its entire body-strengthening ability, such as the healing of sensitive organs like the liver, heart, and brain. The main chemical components of rosemary oil include a-pinene, borneol, b-pinene, camphor, bornyl acetate, camphene, 1, 8-cineole, and limonene. A study published in *Chemistry of Natural Compounds* looked at the volatile compounds obtained from the essential oil of rosemary cultivated at the Algerian Sahara. Its analysis found that 30 compounds represented 98.2 percent of the essential oil, with 1,8-cineole (29.5 percent), 2-ethyl-4,5-dimethylphenol (12 percent) and camphor (11.5 percent) as the major components. According to *Modern Essentials*, a guide to the therapeutic uses of essential oils, high-quality rosemary oil has analgesic, antibacterial, anticancer, anticatarhal, antifungal, anti-infection, anti-inflammatory, antioxidant, and expectorant properties. Rosemary oil uses covers many health concerns, including for the following:

**Clarity** – Add a drop to our hands, rub together, and cup over our mouth and nose for up to a minute

**Cough** – Massage one to two drops over our chest and throat every few hours

**Headaches** – Add a drop to your hands, and cup over your mouth and nose for up to a minute. We may also apply a drop topically to the aching parts of our head.

Vaginal infection – Massage one to two drops in or around the vagina, making sure to test for sensitivity before attempting internal use. This essential oil is a disinfectant and is often used as a mouthwash, helping remove bad breath. By removing oral bacteria, rosemary oil can prevent cavities, plaque buildup, and other dental issues. The mesmerizing aroma of rosemary is worth nothing, too, making it an excellent inhalant. Rosemary oil is used in candles, perfumes, bath oils, fresheners, and cosmetics, boosting mental energy when inhaled. Used with 50:50 dilution, rosemary oil can be applied on ankles and wrists (two to four drops), applied to chakras or vita flex points, directly inhaled, diffused, or as an dietary supplement. For addressing body circulation, mental fatigue, pain relief, decongestion of the respiratory tract, and as a skin and hair care agent, you can use rosemary oil several ways, including in burners and vaporizers, blended massage oils or in the bath, creams, lotions, and shampoos.

**Peppermint oil:**

Peppermint oil is derived from the leaves of the peppermint plant or *Mentha piperita*, a hybrid of the water mint and spearmint plants, and *M. arvensis var.piperascensa*, a plant from the Labiatae family. The essential oil is often used as a home remedy for stomach problems, muscle pain, and headaches. Aside from being available in its oil form, peppermint oil can be found in supplemental capsule form. (Sharma, 2007) Peppermint essential oil have found that two of its major components are menthol and menthone, which are the reason why peppermint oil is widely used as an ingredient in lozenges, toothpastes, balms, and rubs. Other constituents found in the oil are menthyl acetate, 1,8-cineole, limonene, beta-pinene, and beta-caryophyllene. Peppermint oil is often used in aromatherapy, or the use of essential oils to support health. Studies have found that aromatherapy is effective in relieving anxiety, pain, and vomiting, as well as improving memory. The
a aroma of peppermint oil has been shown to improve memory and raise alertness. Some of the benefits of peppermint oil are:

Relief for Stomach Problems
Respiratory Benefits
Pain Relief
Positive Effect on Cancer-Related Treatments
Treatment of Herpes Infections
Hair and Skin Health
Dental Health

Comfort for Stress and Nervous System Problems

According to findings, peppermint oil exhibits antiviral, antimicrobial, antifungal, antioxidant, analgesic, radio protective, and anti edema properties. Peppermint oil is sensitive to light and heat damage, and should only be stored in a cool, dark place in tightly sealed bottles. It is also important to keep them away from other oils, supplements, and drugs, because they may affect their composition. For instance, peppermint oil can disrupt the rate at which your body processes the drug cyclosporine. This causes more of the medication to stay in your bloodstream.

Antibiotic sensitivity test:

Antibacterial agents provide the main basis for the therapy of microbial infections. Their use would lead to the eventual eradication of infectious diseases. However, overuse of antibacterial agents has become the major factor for the emergence and dissemination of multi-drug resistant strains of several groups of microorganisms. The extensive use of antibacterial agents over the last 50 years has led to the emergence of bacterial resistance and to the dissemination of resistance genes among pathogenic microorganisms. Staphylococcus aureus is one of the most important pathogens that can cause suppuration, abscess formation, a variety of pyogenic infection and even fatal septicemia in human beings. MRSA is still considered as an emerging pathogen and public health threats result from the spread of hospital-acquired as well as community-acquired MRSA (Chambers, 2001). The heterogeneous expression of methicillin resistance can make it difficult to determine the resistance phenotype definitively, therefore detection of the mecA gene remains the “gold standard”. MRSA is resistant to not only methicillin and other β-lactams but also may other antibacterial agents; therefore new agents are needed to treat the MRSA. The treatment of infectious diseases with antimicrobial agents continues to present problems in modern-day-medicine with many studies showing a significant increase in the incidence of bacterial resistance to several antibiotics. Many plants have been investigated scientifically for antimicrobial activity and a large number of plant products have been shown to inhibit growth of pathogenic bacteria. Though International Journal of Biology January, 2009 35 pharmacological industries have produced a number of new antibiotics in the last three decades, resistance to these drugs by microorganisms has developed. Medicinal plants are natural resources, yielding valuable products which are often used in the treatment of various ailments. Plant materials remain an important resource for combating illnesses, including infectious diseases, and many of the plants have been investigated for novel drugs or templates for the development of new therapeutic agents. MRSA has traditionally been seen as hospital-associated infections, community –acquired MRSA strains have appeared in recent years. Several new strains of MRSA have been found showing antibiotic resistance even to Vancomycin and Teicoplanin; these new evolutions of the MRSA bacteria are called Vancomycin Intermediate-resistant S. aureus. Essential oils of medicinal plants have been used for hundreds of years of natural medicines to combat a multitude of pathogens, including bacteria, fungi and viruses. Several essential oils confer antimicrobial activity by damaging the cell wall and membrane, leading to cell lysis, leakage of cell contents and inhibition of proton motive force. In addition, there is evidence that they effectively kill bacteria without promoting the acquisition of resistance and they possess multiple antimicrobial activity i.e., antibacterial, antifungal, anticancer, antiviral and antioxidant properties against all pathogens.
Finally, many essential oils are relatively easy to obtain, have low mammalian toxicity and degrade quickly in water and soil, making them relatively environmentally friendly. For these reasons research is ongoing for new antimicrobial agents, either by the design and synthesis of new agents or through the search of natural plant oils for as yet undiscovered antimicrobial agents. The factors responsible for the spread of resistant bacteria do not differ so much compared to the ordinary strains of bacteria and they are most frequently seen in hospital. The most common route of spread is through indirect transmission from the healthcare staff to their patients. Staff may carry the resistant bacteria on their hands or clothing and even equipment in the hospital can become contaminated and a source of infection. New therapies are therefore necessary and of great value.

Gas Chromatography-Mass Spectroscopy:

Gas chromatography-mass spectroscopy (GC-MS) is one of the so-called hyphenated analytical techniques. As the name implies, it is actually two techniques that are combined to form a single method of analyzing mixtures of chemicals. Gas chromatography separates the components of a mixture and mass spectroscopy characterizes each of the components individually. By combining the two techniques, an analytical chemist can both qualitatively and quantitatively evaluate a solution containing a number of chemicals. (Adlard et al., 2001)

Gas Chromatography:

In general, chromatography is used to separate mixtures of chemicals into individual components. Once isolated, the components can be evaluated individually. In all chromatography, separation occurs when the sample mixture is introduced (injected) into a mobile phase. In liquid chromatography (LC), the mobile phase is a solvent. In gas chromatography (GC), the mobile phase is an inert gas such as helium.

The mobile phase carries the sample mixture through what is referred to as a stationary phase. The stationary phase is a usually chemical that can selectively attract components in a sample mixture. The stationary phase is usually contained in a tube of some sort. This tube is referred to as a column. Columns can be glass or stainless steel of various dimensions. The mixture of compounds in the mobile interacts with the stationary phase. Each compounds in the mixture interacts at a different rate. Those that interact the fastest will exit (elute from) the column first. Those that interact slowest will exit the column last. By changing characteristics of the mobile phase and the stationary phase, different mixtures of chemicals can be separated. Further refinements to this separation process can be made by changing the temperature of the stationary phase or the pressure of the mobile phase. GC has a long, thin column containing a thin interior phase of a solid stationary phase (5% phenyl-, 95% dimethyl siloxane polymer). This 0.25 mm diameter column is referred to as a capillary column. This particular column is used for semi volatile, non-polar organic compounds such as the PAHs. The compounds must be in an organic solvent. The capillary column is held in an oven that can be programmed to increase the temperature gradually (or in GC terms, ramped) this helps in separation. As the temperature increases, those compounds that have low boiling points elute from the column sooner than those that have higher boiling points. Therefore, there are actually two distinct separating forces, temperature and stationary phase interactions mentioned previously. As the compounds are separated, they elute from the column and enter a detector. The detector is capable of creating an electronic signal whenever the presence of a compound is detected. The greater the concentration in the sample, the bigger the signal. The signal is then processed by a computer. The time from when the injection is made (time zero) to when elution occurs is referred to as the retention time. While the instrument runs, the computer generates a graph from the signal. This graph is called a chromatogram. Each of the peaks in the chromatogram represents the signal created when a compound elutes from the GC column into detector. The x-axis shows the RT, and the y-axis shows the intensity (abundance) of the signal. If the GC conditions (oven temperature ramp, column type, etc.) are the same, a given compound will always exit (elute) from the column at nearly the same RT. By knowing the RT for a given compound, we can make some assumptions about the identity of the compound. However, compounds that have similar properties often have the same retention times. Therefore, more information is usually required before an analytical chemist can make an identification of a compound in a sample containing unknown components. (Niessen, 2001).

Mass Spectroscopy:

As the individual compounds elute from the GC column, they enter the electron ionization (mass spec) detector. There, they are bombarded with a stream of electrons causing them to break apart into fragments. These fragments can be large or small pieces of the original molecules. The fragments are actually charged ions with a certain mass. The mass of the fragments divided by the charge is
called the mass to charge ratio (M/Z). Since most fragments have a charge of +1, the M/Z usually represents the molecular weight of the fragment.

A group of 4 electromagnets called a quadrupole, focuses each of the fragments through a slit and into the detector. The quadrupoles are programmed by the computer to direct only certain M/Z fragments through the slit. The rest bounce away. The computer has the quadrupoles cycle through different M/Z’s one at a time until a range of M/Z’s are covered. This occurs many times per second. Each cycle of ranges is referred to as a scan. The computer records a graph for each scan. The x-axis represents the M/Z ratios. The y-axis represents the signal intensity (abundance) for each of the fragments detected during the scan.

**GC-MS:**

When GC is combined with MS, a powerful analytical tool is created. A researcher can take an organic solution, inject it into the instrument, separate the individual components, and identify each of them. Furthermore, the researcher can determine the quantities (concentrations) of each of the components. Three-dimensional graph generated when the GC is combined with the MS. Visualize how the chromatogram combines with the mass spectrum to produce the image. It is important to picture this 3D Image and translate it into the previous 2D graphs (This image is not made from the same compounds in the previous). Note that we can create either a mass spectrum or a chromatogram by making the appropriate cross section of the 3D image. Visualize which cross section would produce a spectrum and which would produce a chromatogram. (Robert.et.al., 2007).

### 4. Materials and Methods

**Collection of essential oils:**

Commercially available essential oils of basil, orange, lemon, clove, eucalyptus, olive, cardamom, jojoba, rosemary and peppermint oils were obtained from KK Aromas, Bangalore, Karnataka. These oils were selected based on the literature survey and their use in traditional medicine system.

**Sample collection:**

Staphylococcal culture were collected from hospitalized patients of Kovai Medical College and Hospital, Coimbatore. Culture were transported to the laboratory and cultured onto nutrient broth and mannitol salt agar to obtain isolates.

**Identification of staphylococcus species:**

Based on Bergey’s manual of systemic bacteriology, the samples were isolated and identified. The isolates were cultured onto nutrient broth and then to mannitol salt agar to obtain discrete colonies. The plates were incubated at 37°C for 24hrs under aerobic conditions. After 24hrs of incubation, the culture plates were examined for the appearance, size, colour and morphology of colonies. Biochemical tests were performed for the identification of the isolates.

**Antibiogram of staphylococcus species**

The antibiotic sensitivity tests were performed by using disk diffusion method of Kirby Bauer. Antibiotics such as amikacin, amoxicillin, chloramphenicol, cefotaxime, cefpodoxime, cefoxitin, ceftriaxone, amoxycly, doxycycline Hcl, furazolidone, imipenem, linezolid, nalidixic acid, norfloxacin, nitrofurantoin, neomycin, ofloxacin, oxytetracycline, polymycin-B, rifampicin, methicillin, vancomycin, tetracyclin and novobiocin were used. The culture of staphylococcus were swabbed onto muller-hinton agar plates. After 24hrs incubation, the zone of inhibition were measured.

**Antibacterial activity of the commercially available essential oils:**

The staphylococcal activity of ten essential oils were tested by agar disc(6mm) diffusion method. The culture plates were prepared with sterile medium of Muller-Hinton agar. Essential oils were used as such and diluted in ethanol in the ratio 9:1(Bachir Raho Ghailem, 2014). The agar surface of the plates was swabbed with culture. Using sterile discs(6mm), 20µl of essential oils were placed in the center of the plates. Then the plates were incubated at 37°C for 24hrs and the zone of inhibition were measured.

**GC-MS analysis of essential oils:**

Essential oils which shows maximum activity were analysed by using gcms method. Among ten essential oils, four oils were orange, clove, cardamom and peppermint showed higher antibacterial activity. GC-MS analysis were done on SAIF Institution, cochin. The different compounds present in oils were analysed with standard compounds.
5. Results and Discussion

Identification of staphylococcus species:

Staphylococcal culture were collected from hospital and it was cultured onto nutrient broth for further process. On Gram staining, it is observed as purple cocc in clusters and biochemical tests were performed and the results were observed. Then these three staphylococcus species were cultured onto mannitol salt agar medium and incubated at 37°C for 24 hours. From the results, the staphylococcus species were identified as Staphylococcus aureus, Staphylococcus haemolyticus and Staphylococcus hominis. Staphylococcus aureus grow readily on ordinary media within a temperature range of 10 to 42°C, the optimum being 37°C, and a pH of 7.4 to 7.6. They are aerobes and facultative anaerobes. On nutrient agar medium, after incubation for 24 hours, the colonies are large (2-4 mm diameter), circular, convex, smooth, opaque and easily emulsifiable. Pigment production occurs optimally at 22°C and only in aerobic cultures. Pigment production is enhanced when 1% glycerol monoacetate or milk is incorporated in the medium. Individual colonies on agar are round, convex, and 1-4 mm in diameter with a sharp border. The golden appearance of colonies of some strains is the etymological root of the bacteria’s name; aureus meaning “golden” in Latin. Methicillin-resistant strains of Staphylococcus aureus (i.e. MRSA) often have only weak or no beta-hemolysis and special cultivation media with oxacillin, mannitol and NaCl for their isolation are used. MRSA is able to grow on this media and produce colonies of certain color, depending on used pH indicator (pink). (Rasmussen et al., 2011). Staphylococcus haemolyticus is non-motile, non-sporulating, facultatively anaerobic. Cells are typically occurs coccus shaped and range from 0.8 to 1.3 μm in diameter. Optimal growth occurs between 30-40°C in the presence of oxygen and 10% NaCl. However, some strains can grow at temperatures that range between 18-45°C. Growth at 15°C or 15% NaCl is poor or absent. (Paul De Vos et al., 2009) The growth of Staphylococcus hominis in agar cultures, colonies are usually circular, 4.0 to 4.5 mm in diameter. Agar colonies usually have wide edges and an elevated center. They are commonly smooth with dull surfaces, and are yellow-orange pigmented in the center of the opaque colonies. They grow both in aerobic and anaerobic conditions, but tend to grow significantly less in the latter. Optimal NaCl concentrations of the agar culture for the growth of S. hominis seem to be around 7.5%, and a salt concentration of 15% yielded poor growth to no growth at all. The optimal growth temperature range was around 28 to 40°C, but good growth is still observed at 45°C, while no growth is observed at 15°C. S. hominis can be differentiated from staphylococci by its colony morphology and pigmentation patterns, predominant tetrad cell arrangement, poor growth in thioglycolate, low tolerance of NaCl and carbohydrate reaction pattern. Each species is also significantly different in cell wall composition, lactic acid configuration, temperature extremes of growth, coagulase activity, hemolysis acetylmethylcarbinol production, nitrate reduction, and phosphatase, DNase, and bacteriolysis activities. Similarities in these properties between S. hominis and several other species suggest a close relationship between S. hominis and S. epidermidis, S. haemolyticus, and S. warneri. From the observed results, it is identified that the collected staphylococcus species from hospitalized patients of Koval Medical College and Hospital are Staphylococcus aureus, Staphylococcus haemolyticus and Staphylococcus hominis. These three staphylococcus species were used for further process.

Antibiogram of staphylococcus species:

The antibiotic sensitivity tests were determined by using disk diffusion (Kirby Bauer) method. Muller-Hinton agar plates prepared and swabbed with staphylococcus cultures ant the antibiotic disks were impregnated. After 24 hrs of incubation, the zone of inhibition were measured and tabulated (table: 2).

The three species of staphylococcus are resistant to methicillin. Staphylococcus aureus are resistant/intermediate to amoxicillin, cefoxitin, linezolid, vancomycin and ticarcillin. These two species of S.hominis and S.haemolyticus are resistant to cefpodoxime. Thus these antibiotics can be preferred for Staphylococcal infections. Among Staphylococcus aureus, norfloxacin shows high activity of 34 mm and low activity of cefpodoxime (7 mm), whereas in case of Staphylococcus haemolyticus and Staphylococcus hominis, shows high activity by imipenem (40 mm). Staphylococcus aureus is a medically important organism associated with a variety of diseases. MRSA represents a major challenge to hospitals in all countries due to the emergence and spread of isolates with decreased susceptibilities to several antibiotic classes, in addition to methicillin and the other members of the β-lactum family. The result of the study shows that the significant Staphylococcus aureus are resistant to the antibiotics (amoxicillin, cefoxitin, linezolid, vancomycin, ticarcillin), S.haemolyticus and S.hominis are resistant to cefpodoxime antibiotic.
Antibacterial activity of essential oils:

The antibacterial activity were evaluated by using agar disk(6mm) diffusion method. The oils were diluted in ethanol in the ratio 9:1 and also used without dilution. The muller-hinton agar plates were prepared and swabbed with staphylococcus cultures and the disks which was impregnated with oils were placed and sealed. After 24 hrs incubation period, the zones were measured and tabulated. 

From the observed results Staphylococcus aureus shows higher inhibition activity in an diluted essential oil than the undiluted one. Among ten essential oils cardamom and peppermint shows higher activity of inhibition than others. According to Staphylococcus hominis, undiluted basil oil shows high zone of inhibition(30mm), whereas olive and Staphylococcus haemolyticus, all the ten essential oils that are diluted in ethanol shows activity of inhibition than the undiluted oils. From this study of these ten essential oils shows higher effectiveness of inhibiting activity of Staphylococcus aureus than the other two species. The activity of natural products, especially essential oils (EO), against microorganisms has been recently confirmed by several studies focusing on antimicrobial activity of EO against planktonic cells. The ten essential oils basil, orange, lemon, clove, eucalyptus, olive, cardamom, jojoba, rosemary and peppermint were tested against staphylococcus samples from hospitalized patients in order to determine the ability of the essential oils to act as effective antibacterial agents for drug resistant organisms. Among these ten essential oils, four essential oils shows maximum activity than other oils. Then these four oils namely orange, clove, cardamom and peppermint were analysed to identify the compounds present in it by using gas chromatography-mass spectroscopy (GC/MS) method.

GC-MS analysis of essential oils:

Essential oils which shows maximum activity were analysed by using gcms method. Among ten essential oils, four oils were orange, clove, cardamom and peppermint showed higher antibacterial activity. GC-MS analysis were done on SAIF Institution, cochin. The different compounds present in oils were analysed with standard compounds. (Robert et al., 2013)

Orange oil (table:4): The gas chromatogram of the orange essential oil is characterized by an intense peak at 191 min retention time, the major peak in the gas chromatogram, which has been assigned to Phenol,2,4-bis(1,1-dimethylethyl) (M.W.206), Phenol 2,5-bis(1,1-dimethylethyl) (M.W.206), Anthracene, 9-butyl (M.W. 234), 2,4,5,5,8a-Pentamethyl-6,7,8,8a-tetrahydro-5H-chromene (M.W.206), Allyl-1,4-dimethoxy-3-vinlyoxymethylbenzene (M.W.234), 4,8A-Dimethyl-6-(2-methyl-oxiran-2-yl)-4a,5,6,7,8,8a-hexahydro-1H-naphthalen-2-one (M.W. 234). The other peaks in the gas chromatogram, of moderate and minor intensity. The mass spectra of compound confirmed with those available in the NIST library. In terms of GC identified chemical components, it’s could be attributed due to the different geographical region, the development stages and seasonal variations.

Clove oil (table:5): Gas chromatogram of clove oil is characterized by an intense peak at 164 min retention time, the major peak in the gas chromatogram,which has been assigned to Eugenol (M.W.164), 4-Allyl-2-methoxyphenylphenylacetate (M.W.282), Carbamic acid, phenyl-, 2-(2-thiencyl)cyclohexyl ester (M.W.301). The other peaks in the gas chromatogram, of moderate and minor intensity, correspond to 1-Butanone, 1-(4-hydroxyphenyl) (M.W. 164), 1-Allyl-4-methoxybenzene (M.W. 164), Benzoic acid, 3,4-dimethyl-, methyl ester (M.W. 164)

Cardamom oil (table:6): Gas chromatogram of cardamom oil shows the intense peak at 121 min retention time, the major peak which has been assigned to Thujopsene-(12) (M.W. 204), Cyclohexane,1-ethenyl-1-methyl-2-(1-methylethenyl)-4-(1-methylethylidene) (M.W. 204) , the moderate and minor intensity corresponds to Naphthalene,1,2,3,5,6,7,8,8a-octahydro-1,8a-dimethyl-7-(1-methylethyl)-1-(1R-(1α,7β,8αn)) (M.W.204),1H Cyclopromeazeulene, decahydro-1,1,7-trimethyl-4-methylene-(1aR-(1α,4αa,7α,7β,7βa)) (M.W. 204).

Peppermint oil (table:7): The gas chromatogram of peppermint is characterized by an intense peak at 154 min retention time, the major peak which has been assigned to many compounds such as Phenol,2,6-dimethoxy (M.W. 154), [Phenol,3,4-dimethoxy], [2-Acetyl-1,3-cyclohexanedione],[Phenol,2-ethylviolethio] , [p-Benzquinone,2-(methylthio)], [Phenol,3-methyl-4-(methylthio)]. The other peaks in the gas chromatogram, of moderate and minor intensity corresponds to 1-Butanone,1-(2-thienyl),[Cyclohexanone,4-(1,1-timethylethyl)],[1,3-Cyclohexanediene,2,5,5-trimethyl].From the analysis of gas chromatography-mass spectroscopy of these four essential oils, different compounds in each of essential oils were identified. Further, compounds in each oil can be analysed.
Summary and Conclusion

Staphylococcus swabs were collected from hospitalized patients of Kovai Medical College and Hospital. Then the collected swabs were cultured onto nutrient broth and then swabbed onto nutrient agar and mannitol salt agar media. After 24hrs incubation period, the plates were observed for discrete colonies. Then for the identification, biochemical tests were done. As a result, from collected staphylococcal swabs three species of staphylococcus were identified as *Staphylococcus aureus*, *Staphylococcus haemolyticus* and *Staphylococcus hominis*. Antiibiogram of these three staphylococcus species was determined by using disk diffusion (Kirby Bauer) method on Muller-Hinton agar. The antibiotic discs used are amikacin, amoxicillin, chloramphenicol, cefotaxime, cefpodoxime, cefoxitin, ceftriaxone, amoxycylav, doxycycline HCl, furazolidone, imipenem, linezolid, nalidixic acid, norfloxacin, nitrofurantoin, neomycin, ofloxacin, oxytetracycline, polymycin-B, rifampicin, methicillin, vancomycin, ticarcillin, novobiocin. From the observed results all the three species of staphylococcus are resistant to methicillin. Compared to other two species, S.haemolyticus show high activity of antibiotic sensitivity. Antibiotic impenem shows higher activity in S.haemolyticus and S.hominis. Staphylococcus aureus are resistant/intermediate to amoxicillin,cefotaxime, linezolid, vancomycin and ticarcillin. These two species of *S. hominis* and *S. haemolyticus* are resistant to cefpodoxime. Thus these antibiotics can be preferred for Staphylococcal infections. Commercially available essential oils of basil, orange, lemon, clove, eucalyptus, olive, cardamom, jojoba, rosemary and peppermint oils were obtained from KK Aromas, bangalore, Karnataka. Antibacterial activity of these ten essential oils were determined by using agar disc diffusion method on Muller-Hinton agar. After 24 hrs incubation, the zone of inhibition were measured. Among these three staphylococcus species, *Staphylococcus aureus* shows high activity in diluted oil than in undiluted and *Staphylococcus haemolyticus* shows more activity in diluted essential oils than the undiluted oils. *Staphylococcus hominis* shows high activity on undiluted oils than the diluted. Thus, from the observed results, these essential oils which are diluted with ethanol can be preferred for the treatment of staphylococcal infections. Essential oils which shows higher antibacterial activity were analysed by using Gas chromatography-Mass Spectroscopy (GC-MS) method. Four essential oils, orange, clove, cardamom and peppermint were analysed and the compounds was identified. In orange oil, Phenol,2,4-bis (1,1-dimethylethyl) (M.W.206), Spiro (4H-1,3,2-benzodioxaborin-4,cyclohexane), 2-ethyl-5,6,7,8-tetrahydro(M.W. 234) shows intense peak, where as, in case of clove oil Eugenol (M.W. 164), 4-Allyl-2-methoxyphenyl phenylacetate (M.W. 282), Compounds are identified in cardamom oil which shows major peak is Thujoepse- (12) (M.W. 204), Cyclohexane,1-ethenyl-1-methyl-2-(1-methylethenyl)-4-(1-methylethylidene) (M.W. 204) and in peppermint oil major intensity corresponds to Phenol,2,6-dimethoxy (M.W. 154), [Phenol,3,4-dimethoxy], [2-Acetyl-1,3-cyclohexanedione]. The mass spectra of compound confirmed with those available in the NIST library. In terms of GC identified chemical components, it’s could be attributed due to the different geographical region, the development stages and seasonal variations. Further these essential oils can be tested against other bacteria, fungi. The antibacterial activity can be analysed further and it can be preferred to patients with staphylococcal infections. In future the study can be extended in such a way to find treatments for staphylococcal infections.

Table 1: BIOCHEMICAL TESTS

<table>
<thead>
<tr>
<th>TESTS</th>
<th>Culture 1</th>
<th>Culture 2</th>
<th>Culture 3</th>
</tr>
</thead>
<tbody>
<tr>
<td>Indole</td>
<td>Negative</td>
<td>Negative</td>
<td>Negative</td>
</tr>
<tr>
<td>Methyl red</td>
<td>Positive</td>
<td>-</td>
<td>Negative</td>
</tr>
<tr>
<td>Voges Proskauer</td>
<td>Positive</td>
<td>Positive</td>
<td>Negative</td>
</tr>
<tr>
<td>Citrate</td>
<td>Positive</td>
<td>-</td>
<td>Negative</td>
</tr>
<tr>
<td>Coagulase</td>
<td>Positive</td>
<td>Negative</td>
<td>Negative</td>
</tr>
<tr>
<td>Urease</td>
<td>Positive</td>
<td>Negative</td>
<td>Positive</td>
</tr>
<tr>
<td>Oxidase</td>
<td>Negative</td>
<td>Negative</td>
<td>Negative</td>
</tr>
<tr>
<td>Catalase</td>
<td>Positive</td>
<td>Positive</td>
<td>Positive</td>
</tr>
</tbody>
</table>
**Table 2: ANTIBIOGRAM OF STAPHYLOCOCCUS SPECIES**

<table>
<thead>
<tr>
<th>Antibiotics used</th>
<th>Disc content</th>
<th>Diameter of zone of inhibition in mm</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td><em>S. aureus</em></td>
</tr>
<tr>
<td>Amikacin</td>
<td>30 mcg</td>
<td>26 mm</td>
</tr>
<tr>
<td>Amoxicillin</td>
<td>30 mcg</td>
<td>-</td>
</tr>
<tr>
<td>Chloramphenicol</td>
<td>30 mcg</td>
<td>24 mm</td>
</tr>
<tr>
<td>Cefotaxime</td>
<td>30 mcg</td>
<td>28 mm</td>
</tr>
<tr>
<td>Cefpodoxime</td>
<td>10 mcg</td>
<td>7 mm</td>
</tr>
<tr>
<td>Cefoxitin</td>
<td>30 mcg</td>
<td>-</td>
</tr>
<tr>
<td>Ceftriaxone</td>
<td>30 mcg</td>
<td>27 mm</td>
</tr>
<tr>
<td>Amoxyclav</td>
<td>10 mcg</td>
<td>11 mm</td>
</tr>
<tr>
<td>Doxycycline Hcl</td>
<td>30 mcg</td>
<td>16 mm</td>
</tr>
<tr>
<td>Furazolidone</td>
<td>50 mcg</td>
<td>18 mm</td>
</tr>
<tr>
<td>Inipenem</td>
<td>10 mcg</td>
<td>26 mm</td>
</tr>
<tr>
<td>Linezolid</td>
<td>30 mcg</td>
<td>-</td>
</tr>
<tr>
<td>Nalidixic acid</td>
<td>30 mcg</td>
<td>18 mm</td>
</tr>
<tr>
<td>Norfloxacin</td>
<td>10 mcg</td>
<td>34 mm</td>
</tr>
<tr>
<td>Nitrofurantoin</td>
<td>300 mcg</td>
<td>15 mm</td>
</tr>
<tr>
<td>Neomycin</td>
<td>30 mcg</td>
<td>21 mm</td>
</tr>
<tr>
<td>Ofloxacin</td>
<td>5 mcg</td>
<td>29 mm</td>
</tr>
<tr>
<td>Oxytetracycline</td>
<td>30 mcg</td>
<td>15 mm</td>
</tr>
<tr>
<td>Polymycin-B</td>
<td>300 units</td>
<td>12 mm</td>
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<tr>
<td>Rifampicin</td>
<td>5 mcg</td>
<td>10 mm</td>
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<tr>
<td>Methicillin</td>
<td>5 mcg</td>
<td>-</td>
</tr>
<tr>
<td>Vancomycin</td>
<td>30 mcg</td>
<td>-</td>
</tr>
<tr>
<td>Ticarcillin</td>
<td>75 mcg</td>
<td>-</td>
</tr>
<tr>
<td>Novobiocin</td>
<td>30 mcg</td>
<td>11 mm</td>
</tr>
</tbody>
</table>

S- Sensitive, I- Intermediate, R- Resistant

**Table 3: ANTIBACTERIAL ACTIVITY OF ESSENTIAL OILS**

<table>
<thead>
<tr>
<th>ESSENTIAL OILS</th>
<th>Staphylococcus aureus (Undiluted 20µl)</th>
<th>Staphylococcus hominis (Undiluted 20µl)</th>
<th>Staphylococcus haemolyticus (Undiluted 20µl)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Basil</td>
<td>8mm</td>
<td>10mm</td>
<td>30mm</td>
</tr>
<tr>
<td>Orange</td>
<td>10mm</td>
<td>9mm</td>
<td>8mm</td>
</tr>
<tr>
<td>Lemon</td>
<td>8mm</td>
<td>8mm</td>
<td>7mm</td>
</tr>
<tr>
<td>Clove</td>
<td>16mm</td>
<td>17mm</td>
<td>19mm</td>
</tr>
<tr>
<td>Eucalyptus</td>
<td>19mm</td>
<td>8mm</td>
<td>7mm</td>
</tr>
<tr>
<td>Olive</td>
<td>11mm</td>
<td>8mm</td>
<td></td>
</tr>
<tr>
<td>Cardamom</td>
<td>24mm</td>
<td>18mm</td>
<td></td>
</tr>
<tr>
<td>Jojoba</td>
<td>9mm</td>
<td>7mm</td>
<td></td>
</tr>
<tr>
<td>Rosemary</td>
<td>20mm</td>
<td>12mm</td>
<td></td>
</tr>
<tr>
<td>Peppermint</td>
<td>22mm</td>
<td>10mm</td>
<td></td>
</tr>
</tbody>
</table>

**Table 4: GC-MS ANALYSIS OF ORANGE OIL**

<table>
<thead>
<tr>
<th>S. No</th>
<th>Compound</th>
<th>Mol. formula</th>
<th>Mol. Wt</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Phenol,2,4-bis(1,1-dimethylethyl)</td>
<td>C&lt;sub&gt;14&lt;/sub&gt;H&lt;sub&gt;22&lt;/sub&gt;O&lt;sub&gt;2&lt;/sub&gt;</td>
<td>206</td>
</tr>
<tr>
<td>2</td>
<td>Spiro(4H-1,3,2-benzodioxaborin-4,1-cyclohexane), 2-ethyl-5,6,7,8-tetrahydro</td>
<td>C&lt;sub&gt;14&lt;/sub&gt;H&lt;sub&gt;23&lt;/sub&gt;BO&lt;sub&gt;2&lt;/sub&gt;</td>
<td>234</td>
</tr>
<tr>
<td>3</td>
<td>Phenol,2,5-bis(1,1-dimethylethyl)</td>
<td>C&lt;sub&gt;14&lt;/sub&gt;H&lt;sub&gt;22&lt;/sub&gt;O</td>
<td>206</td>
</tr>
<tr>
<td>4</td>
<td>Anthracene,9-butyl</td>
<td>C&lt;sub&gt;14&lt;/sub&gt;H&lt;sub&gt;18&lt;/sub&gt;</td>
<td>234</td>
</tr>
<tr>
<td>5</td>
<td>2,4,5,5,8a-Pentamethyl-6,7,8a-Tetrahydro-5H-chromene</td>
<td>C&lt;sub&gt;14&lt;/sub&gt;H&lt;sub&gt;22&lt;/sub&gt;O&lt;sub&gt;2&lt;/sub&gt;</td>
<td>206</td>
</tr>
<tr>
<td>6</td>
<td>Allyl-1,4-dimethoxy-3-vinylxy Methylenbenzene</td>
<td>C&lt;sub&gt;14&lt;/sub&gt;H&lt;sub&gt;22&lt;/sub&gt;O&lt;sub&gt;2&lt;/sub&gt;</td>
<td>234</td>
</tr>
<tr>
<td>7</td>
<td>4,8a-Dimethyl-6-(2-methyl-oxiran-2-yl)-4a,5,6,7,8a-hexahydr-1H-naphthalen-2-one</td>
<td>C&lt;sub&gt;15&lt;/sub&gt;H&lt;sub&gt;20&lt;/sub&gt;O&lt;sub&gt;2&lt;/sub&gt;</td>
<td>234</td>
</tr>
<tr>
<td>8</td>
<td>di-9-Cyano-2-oxo-cis-decalin semicarbazone</td>
<td>C&lt;sub&gt;15&lt;/sub&gt;H&lt;sub&gt;14&lt;/sub&gt;N&lt;sub&gt;2&lt;/sub&gt;O</td>
<td>234</td>
</tr>
<tr>
<td>9</td>
<td>(3-Cyano-6-ethyl-5-methyl-pyridin-2-ylsulfanyl)-acetic acid ethyl ester</td>
<td>C&lt;sub&gt;15&lt;/sub&gt;H&lt;sub&gt;16&lt;/sub&gt;N&lt;sub&gt;2&lt;/sub&gt;O&lt;sub&gt;2&lt;/sub&gt;S</td>
<td>264</td>
</tr>
<tr>
<td>10</td>
<td>Phenol, 2-(1,1-dimethylethyl)-4-(1,1,3,3-tetramethylbutyl)</td>
<td>C&lt;sub&gt;18&lt;/sub&gt;H&lt;sub&gt;32&lt;/sub&gt;O</td>
<td>262</td>
</tr>
</tbody>
</table>
### Table 5: GC-MS ANALYSIS OF CLOVE OIL

<table>
<thead>
<tr>
<th>S.No</th>
<th>Compound</th>
<th>Mol.formula</th>
<th>Mol.wt</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Eugenol</td>
<td>C&lt;sub&gt;10&lt;/sub&gt;H&lt;sub&gt;12&lt;/sub&gt;O&lt;sub&gt;2&lt;/sub&gt;</td>
<td>164</td>
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<tr>
<td>2</td>
<td>4- Allyl-2-methoxyphenyl phenylacetate</td>
<td>C&lt;sub&gt;18&lt;/sub&gt;H&lt;sub&gt;18&lt;/sub&gt;O&lt;sub&gt;3&lt;/sub&gt;</td>
<td>282</td>
</tr>
<tr>
<td>3</td>
<td>3-Pyrindinecarbonitrile,1,2-dihydro-4-methoxy-1-methyl-2-oxo</td>
<td>C&lt;sub&gt;9&lt;/sub&gt;H&lt;sub&gt;10&lt;/sub&gt;N&lt;sub&gt;2&lt;/sub&gt;O&lt;sub&gt;2&lt;/sub&gt;</td>
<td>164</td>
</tr>
<tr>
<td>4</td>
<td>2-Acetyl-3,4,6-trimethylpyrazine</td>
<td>C&lt;sub&gt;10&lt;/sub&gt;H&lt;sub&gt;12&lt;/sub&gt;N&lt;sub&gt;2&lt;/sub&gt;O</td>
<td>164</td>
</tr>
<tr>
<td>5</td>
<td>Carbamic acid, phenyl-2-(2-thienyl) Cyclohexyl ester</td>
<td>C&lt;sub&gt;7&lt;/sub&gt;H&lt;sub&gt;10&lt;/sub&gt;NO&lt;sub&gt;2&lt;/sub&gt;S</td>
<td>301</td>
</tr>
<tr>
<td>6</td>
<td>Papaveroline,1,2,3,4-tetrahydro-3-0-methyl</td>
<td>C&lt;sub&gt;7&lt;/sub&gt;H&lt;sub&gt;10&lt;/sub&gt;NO&lt;sub&gt;2&lt;/sub&gt;</td>
<td>301</td>
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### Table 6: GC-MS ANALYSIS OF CARDAMOM OIL

<table>
<thead>
<tr>
<th>S.No</th>
<th>Compound</th>
<th>Mol.formula</th>
<th>Mol.wt</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Naphthalene,1,2,3,5,6,7,8,8a-octahydro-1.8a-dimethyl-7-(1-methylethenyl)-(1R-(1α,7β,8α))</td>
<td>C&lt;sub&gt;13&lt;/sub&gt;H&lt;sub&gt;24&lt;/sub&gt;</td>
<td>204</td>
</tr>
<tr>
<td>2</td>
<td>1H-5a,7-Methanoazulene,2,3,4,7,8,8a-hexahydro-3,6,8,8-tetramethyl-(3R-(3α,3α,7β,8α))</td>
<td>C&lt;sub&gt;13&lt;/sub&gt;H&lt;sub&gt;24&lt;/sub&gt;</td>
<td>204</td>
</tr>
<tr>
<td>3</td>
<td>Azulene,1,2,3,3a,4,5,6,7-octahydro-1,4-dimethyl-7-(1-methylethenyl)-(1R-(1α,3αβ,4α,7β))</td>
<td>C&lt;sub&gt;13&lt;/sub&gt;H&lt;sub&gt;24&lt;/sub&gt;</td>
<td>204</td>
</tr>
<tr>
<td>4</td>
<td>Thujopsene-(12)</td>
<td>C&lt;sub&gt;13&lt;/sub&gt;H&lt;sub&gt;24&lt;/sub&gt;</td>
<td>204</td>
</tr>
<tr>
<td>5</td>
<td>1,2,4-Methenoph-1H-indene,octahydro-1,7a-dimethyl-5-(1-methylethenyl)-(1S-(1α,2α,3αβ,4α,5α,7αβ,8S*))</td>
<td>C&lt;sub&gt;13&lt;/sub&gt;H&lt;sub&gt;24&lt;/sub&gt;</td>
<td>204</td>
</tr>
<tr>
<td>6</td>
<td>Cyclohexane,1-ethenyl-1-methyl-2-(1-methylethenyl)-4-(1-methylethylidene)</td>
<td>C&lt;sub&gt;13&lt;/sub&gt;H&lt;sub&gt;24&lt;/sub&gt;</td>
<td>204</td>
</tr>
<tr>
<td>7</td>
<td>1H-Cycloprop(e)azulene,decahydro-1,1,7-trimethyl-4-methylene-(1αR-(1α,4α,7α,7αβ,7βα))</td>
<td>C&lt;sub&gt;13&lt;/sub&gt;H&lt;sub&gt;24&lt;/sub&gt;</td>
<td>204</td>
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### Table 7: GC-MS ANALYSIS OF PEPPERMINT OIL

<table>
<thead>
<tr>
<th>S. No</th>
<th>Compound</th>
<th>Mol. formula</th>
<th>Mol.wt</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Phenol, 2,6-dimethoxy</td>
<td>C&lt;sub&gt;8&lt;/sub&gt;H&lt;sub&gt;10&lt;/sub&gt;O&lt;sub&gt;2&lt;/sub&gt;</td>
<td>154</td>
</tr>
<tr>
<td>2</td>
<td>Phenol, 3,4-dimethoxy</td>
<td>C&lt;sub&gt;8&lt;/sub&gt;H&lt;sub&gt;10&lt;/sub&gt;O&lt;sub&gt;2&lt;/sub&gt;</td>
<td>154</td>
</tr>
<tr>
<td>3</td>
<td>2-Acetyl-1,3-cyclohexanedione</td>
<td>C&lt;sub&gt;8&lt;/sub&gt;H&lt;sub&gt;10&lt;/sub&gt;O&lt;sub&gt;2&lt;/sub&gt;</td>
<td>154</td>
</tr>
<tr>
<td>4</td>
<td>Phenol, 2-(ethylthio)</td>
<td>C&lt;sub&gt;8&lt;/sub&gt;H&lt;sub&gt;10&lt;/sub&gt;OS</td>
<td>154</td>
</tr>
<tr>
<td>5</td>
<td>p-Benzquinone,2-(methylthio)</td>
<td>C&lt;sub&gt;8&lt;/sub&gt;H&lt;sub&gt;10&lt;/sub&gt;OS</td>
<td>154</td>
</tr>
<tr>
<td>6</td>
<td>Phenol,3-methy-4-(methylthio)</td>
<td>C&lt;sub&gt;8&lt;/sub&gt;H&lt;sub&gt;10&lt;/sub&gt;OS</td>
<td>154</td>
</tr>
</tbody>
</table>

**LIST OF PLATES**

Mannitol salt agar plates

![Plate a) Staphylococcus aureus](image1.png)

![Plate b) S. haemolyticus and S. hominis](image2.png)
Antibiogram of *Staphylococcus Hominis*

Antibiogram of *Staphylococcus Haemolyticus*

Antibiogram of *Staphylococcus Aureus*
LIST OF GRAPHS

GC-MS of Orange oil
GC-MS of Clove oil

GC-MS of Cardamom oil
GC-MS of Peppermint oil

References


Chung G., Oh S.B., 2013. Eugenol as local anesthetic. Natural Products-Phytochemistry, Botany and Metabolism of Alkaloids, Phenolics and Terpenes;Part XIV.PP.4001-4015.


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