FORMULATION OF NATURAL MIRACLE BALM BY USING HERBAL PLANTS AND EVALUATION ON TOPICAL REGIONS

P. Geetha Devi, S. Yamuna^{*}, Sk. Nourin¹, K. Naveen², Sk. Salma³, D. swathi⁴, K. Gayathri⁵, P. subrahmanyam⁶

AUTHORS INFORMATION;

P. Geetha devi (MSC, BED) Associate professor, Department of microbiology

*S. Yamuna Student of 4th year b. pharmacy, Jagans college of pharmacy, Nellore MAIL.ID: <u>samadhiyamuna8049@gmail.com</u>

1.Sk. Nourin

Student of 4th year b. pharmacy, Jagans college of pharmacy, Nellore MAIL.ID: sknourinkousar336@gmail.com

2.K. Naveen kumar Student of 4th year b. pharmacy, Jagans college of pharmacy, Nellore MAIL.ID: k.naveenmarch282001@gmail.com

3.Sk. Salma Student of 4th year b. pharmacy, Jagans college of pharmacy, Nellore MAIL.ID: sksalma5842@gmail.com

4.D. Swathi

Student of 4th year b. pharmacy, Jagans college of pharmacy, Nellore MAIL.ID: swathidasari854@gmail.com

5.K. Gayathri

Student of 4th year b. pharmacy, Jagans college of pharmacy, Nellore MAIL.ID: gayathrikuncham57787@gmail.com

6.P. Subrahmanyam Student of 4th year b. pharmacy, Jagans college of pharmacy, Nellore

ABSTRACT;

There has been an increasing focus on development of new routes of drug administration to provide tailored treatments for patients, without decreasing efficacy of analgesia, in proportion to the progression of the knowledge of pain mechanisms. While acute pain acts as an alarm, chronic pain is a syndrome requiring meticulous selection of analgesic drugs of high bioavailability for long-term use. Such criteria are challenges that topical medications aim to overcome, allowing progressive delivery of active component, maintaining stable plasma levels, with a good safety profile. This review presents recent findings regarding topical formulations of the method of preparing said composition. The composition comprising extracts of organically certified herbs, organic essential oils and organic beeswax, wherein the essential oils used herein is obtained by cold pressed method. The oils used in it is used as a pain killer.

Key words; natural pain relief balm, vitex negundo oil, Eucalyptus oil.

INTRODUCTION

Herbal balm is an ayurvedic formulation of powerful essential oils for quick relief from head ache, back ache, cold and in relieving pain.

Herbal balm composition comprising organic essential oils, organic bees wax and other desired herbal components has medicated topical preparations for application to skin of human beings. Balms are topical preparations for application to skin to relieve pain and stiffness. These balm contains counter irritant chemical compounds such as methyl salicylate. Petroleum jelly is the common base for any kind of balms.

Pain is an unpleasant feeling often caused by intense or damaging stimuli, such has stubbing a toe, burning a finger, putting alcohol on a cut and bumping the funny bone. The international association for the study of pains widely used definition states, pain is an unpleasant sensory and emotional experience associated with actual or potential tissue damage, or described in terms of such damage.

Pain motivates the individual to withdrawn from damaging situations, to protect a damaged body parts while it kills and to avoid similar experiences in future. Most pain resolves promptly hence the pain stimulus is removed and the body has healed, but sometimes pain persists despite removal of stimulus and apparent healing of the body and sometimes pain arrive in the absence of any detectable stimulus, damage or disease.

MYTHS ABOUT PAIN AND CURE FOR PAIN;

A cure for pain doesn't exist. To cure pain, the condition causing your pain must be anatomically removed from your body through surgery and in most cases this simply is not feasible nor possible, nor label as a cure for pain. Pain results as a result of agitated in famed nurse at the point of injury or diseases. Any pain relief product that works will more often than not be unique to reach of us as individuals. In order to achieve pain relieve and pain control, your task is to find the wright product, methodology or pain relief treatment that allows here dramatic reduction of your pain full condition.

Pain balm is some thing that is comforting and soothing. It is one which leads the pain. Thus the word relief is inbuilt in the word pain balm a balm in the physical sense is defined as a semi solid preparation applied externally as a remedy or for soothing and irritation. It is also defined as any of various aromatic resinous substances contained in a preparation used for healing and soothing. When pain relief are rubbed, on the area where the pain exits, the pressure and movement produces excess of sensory in put that blocks the pain sensation.

HOW DOES THE PAIN RELIEF BALM WORK;

Pain relief balm works on the principle of counter irritant instead of actually relieving the pain they work on the principle of suppressing the pain by causing irritation on the point where the pain relief balm is applied. Pain balms generally contains 3 components namely methyl salicylate, menthol and camphor all these are easily absorbed through the skin. A combination of these three active ingredients is useful in case of head ache and rheumatic pains. The other ingredients in the pain the pain relief balm are eucalyptus oil, petroleum jelly, negundo oil, bees wax.

Although these pain relief balms have a special pharmacological effect in relieving pain, it is actually the amount of pressure applied and the movement that plays a significant role. Role of the balm includes a local anesthetic effect and finally provides a comfortable stage. These products do not have any side effect or allergic reactions such as irritation or darkening of the skin or cause inflammation on the point of application. Hence the consumer develops a liking for the chosen product.

Petroleum jelly or petrolatum is the semi solid mixture of hydrocarbons and has become house hold preparation for various medical purposes. Petrolatum has associated with some of negative effects due to improper use including lipid pneumonia when inhaled from the nose. Petrolatum is not a material from renewable sources and not biodegradable which may be a cause of concern for environmental pollution etc.

Non organic essential oils are processed from herbal materials cultivated using pesticides and chemical fertilizers. Non organic essential oils are processed by chemical extraction and therefore all chances of presence of solvent residue and pesticides in normal oils.

In the recent years, the old is moving towards natural that are free from chemical substances which may be safe and save the planet from the pollution. organic forming is a step forward in this direction to grow herbal materials with out using any pesticides and chemical fertilizers to produce crops that are safe and natural with out any side effects.

Herbal balm composition comprising extracts of organically certified herbs, organic essential oils and organic bees wax which are mixed to form a medicated topical application. These application is used for quick relieve from pain. Such as head ache, back ache, arthritic pain.

The preparation of balms which can be certified organic balms as per organic certification standards using organic essential oils and organic bees wax. The organic vegetable oil in the formula is of Indian origin like coconut oil. These balms comprise cold balm, lip balm, body balm and foot balm etc.

Essential oils are well known for their therapeutic properties in relieving pain and common cold. Organic essential oils are distilled or cold press from the organically grown herbal materials that are free from pesticides and very low heavy metals.

Beeswax is a natural wax produced in the bee hives of honeybees belonging to the species Apis dorsata, Apis florea, Apis cerana indica, Trigona iridipennis. It is a tough wax and formed from a mixture of several compounds. Its main components are palmitate, palmitoleate, hydroxy palmitate and oleate esters of long chain aliphatic alcohols. It is a natural and developed from renewable sources and biodegradable.

The analgesic and anti- inflammatory potentials of essential oils of eucalyptus leaf. Twenty chemical compounds, which were isolated from the leaves essential oil eucalyptus used against cyclooxygenase two, tumor necrosis factor- α and interleukin- 1 β convertase to elucidate the analgesic and anti- inflammatory activity.

Eucalyptus is one of the major fast growing exotics in Bangladesh, is a tree of the family Myrtaceae. the plants of Myrtaceae possess essential oils that have different biological activities including antimicrobial, antifungal, cytotoxic and anti- inflammatory effects. The oils were used conventionally for the treatment of colds, influenza, cystitis, diabetes, gastritis, kidney disease, laryngitis.

The essential oil of eucalyptus leaves possess an inhibitory effect on inflammation in rats. They inhibit cyclooxygenase enzyme and pro-inflammatory cytokines to clarify analgesic and anti- inflammatory activity.

Eucalyptus leaves and oil extracted from it, have been extensively used for treating common respiratory diseases and other infections including cough and cold, fever, sore throat, congestion, joint pain, wounds etc.

The tree which goes by the botanical name eucalyptus globulus, though native to Australia is cultivated all over the world now a days. The tree has over 300 species all known for its profound medicinal properties.

The curative property of the tree which is also known as 'Blue gum ' comes from the oil extracted from its oval shaped leaves. These leaves are dried, crushed and passed through the stem distillation method or cold- pressed to haul out the essential oil. The oil is colorless but possesses a strong woody aromatic fragrance which is generally diluted before using it directly. In ancient times, the aborigines of Australia made extensive use of the tree. Be it chewing on the roots of the tree are sipping on eucalyptus leaf spiked tea, the medicinal properties of the oil present in the leaf or root helped people in managing fever, cold, body pain and hence eucalyptus became famous as the" Australian fever tea".

In the late 1800s when the concentrated oil was extracted, doctors reportedly noted that the oil stimulated sweating and providing relief from chest congestion and sore throat. Later with more researches, doctors started prescribing this oil and formulations containg eucalyptus oil for various respiratory anomalies including asthma, bronchitis, flue and coughs.

Imbued with natural bio active components and volatile oils like eucalyptol, globulol, alpha and beta-pinene, camphele, limonene, sadinene, p-cymene, camphor, citronellal, alphaphellandrene, aromadendril and piperidole, the oil portrays multifaceted curative properties like decongestant, anti inflammatory, anti spasmodic, disinfectant, antimicrobial and insecticidal properties. Owing to its characteristic aroma, the benefits of the oils are known available in the form of liniments for tired, tender muscles, chest rubs for cold and congestion, cough and cold drops and lozenges, sour throat sprays, pain relief oil for join pain, rash creams, inhalers, mouth washes, sops, shampoos and many more. A part from the medicinal benefits, the magic oil also has preservative properties and hence it is used in several foods and beverages to prevent spoilage. It is also used as a natural floor cleaner owing to its disinfectant property to kill germs having a strong aromatic fragrance, it has also found its place has a natural deodorizer and used in several room sprays and potpourris.

Inhale hot water infused with a few drops of eucalyptus oil to get relief from cough and cold congestion. One can also use warm imbued with this oil to gargle and reduce pain and irritation in the case of a sore throat.

Negundo, also called a five-leaved chaste tree, is a potent ayurvedic plant, that possesses noteworthy therapeutic properties and heals several ailments including asthma, muscle spasms and anxiety. It is scientifically termed vitex negundo and commonly known as ''Nishinda'' in Bengali, ''Nallavalli'' in Telugu, ''Nagod'' in Gujarati and ''Nallanochi'' in Tamil.

Negundo's natural habitats are chiefly in the southern parts of Asia and Africa, being widely cultivated in the tropical environments of China, India, Indonesia, Tanzania, and Madagascar. It is a deciduous shrub, usually 2 to 8 meters in height, with a brown bark and green leaves that hold five leaflets. The flowers are white or blue in color and upon developing, give rise to succulent, oval-shaped, purple fruits or drupes, with a fleshy pulp and seed in the interior.

Negundo, also called a five-leaved chaste tree, is a potent ayurvedic plant, that possesses noteworthy therapeutic properties and heals several aliments including asthma, muscle spams and anxiety. It is scientifically termed vitex negundo and commonly known as "Nishinda" in Bengali, 'Nallavalli' in Telugu, in gujarati and 'nallanochi' in Tamil.

Negundo natural habits are chiefly in the southern parts of Asia and Africa, being widely cultivated in the tropical environments of China, India, Indonesia, Tanzania, and Madagascar. It is a deciduous shrub, usually 2 to 8 meters in height, with a brown bark and green leaves that hold five leaflets. The flowers are white or blue in color and upon developing, give rise to succulent, oval-shaped, purple fruits or drupes, with a fleshy pulp and seed in the interior.

Ancients ayurvedic texts praise the curative traits of 'Negundo', which in Sanskrit literally translates to that which protects the body from diseases. True to its name, this wonderful gift of mother nature offers some fantastic rewards for overall human health.

The roots, leaves, flowers, fruits, and bark of the negundo plant are utilized in herbal concoctions in the form of oil, pastes, juices, and powders, to cure disorders ranging from widely prevalent fevers to the very rare leprosy. Today, this magical herb is being naturalized and propagated world- wide, including American and Australia, so the global population can reap the excellent advantages that negundo offers, for overall well-being.

Biochemical Components of Negundo;

A vast array of beneficial plant-based compounds with antioxidant and anti-inflammatory traits are present in negundo. These include flavonoids and phenols which display cardioprotective qualities for heart wellness, besides Terpenoids and organic fatty acids that are laden with calming and analgesic properties to relieve mental stress, joint pain and muscle aches. Negundo is also bestowed with alkaloids such as Nishindine, Vitricine, which confer useful anticancer and antimicrobial effects on the body, thus combatting tumor growths and stomach infections.

Profuse amounts of vitamin C and vitamin E in negundo leaf and stem extracts showcase natural antioxidant capabilities. This helps protect cells, tissues from oxidative damage by harmful free radicals and also enriches skin texture, nurtures the growth of thick, long silky hair. Housing valuable bioactive constituents such as camphene, pinene, dulcitol, negundo effectively lessens inflammation in the body, which is turn alleviates symptoms of asthma, arthritis, anxiety and enhances digestion, metabolism.

Joint pain is one of the most common complaints with many possible causes. Some medicines used for joint pain relief such as NSAID_s have substantial and frequent side effects. Topical route possibly reduces adverse reactions by maximizing local delivery and minimizing systemic toxicity. Plants have been the most important sources of medicines for human health and Iranian traditional medicine is well known for its extensive use of herbal medicines to treat diseases accompanied with joint pain for centuries

PLANT PROFILE;

COCONUT OIL:



Fig 1; coconut oil

BIOLOGICAL NAME : Cocos nucifera

KINGDOM	: Plantae
ORDER	: Arecales
FAMILY	: Arecaceae
SUB-FAMILY	: Arecoideae
GENUS	: Cocos L.
SPECIES	: nucifera

Coconut oil or Copra oil is an edible oil extracted from the kernel or meat of mature coconuts harvested from the coconut palm (Cocos nucifer). It has various applications. Because of its high saturated fat content, it is slow to oxidize and thus resistant to rancidification. Coconut oil

is one of natures super foods and a truly essential nutrient in any diet or beauty regime. Coconut oil is unique when compared to other oils because it is composed predominantly of a group of fat molecules known as medium chain fatty acids.

DESCRIPTION:

The coconut palm, Cocos nucifera, is an erect palm in the which is grown its fruits, used primarily for the extraction of coconut oil for use in cooking. The coconut palm has an erect or slightly curved stem which grows from a swollen base. The stem is smooth, light gray in colour and has prominent leaf scars, the stem is topped with a crown of 60-70 spirally arranged leaves. The leaves are long (up to7m\23ft).

pinnately divided and composed of 200to250 tapering leaf let. The inflorescence is a spike produced at the leaf axiry with 20 to 60 branch, each with a female flower at the base and many male flowers. The fruit is a drupe containing a single seed. It is ovoid in shape with 3 sides divided by ridges.

The exocarp and the mesocarp make up the husk of the coconut. The seed is protected by a thick, stony shell or endocarp and is partially filled with a liquid known as coconut water. The edible endosperm is white and meaty and can be between 1.0 and 2.5cm (0.4 to 1.0 in) thick. Coconut palms can reach a height of 30m (98 ft). produce up to 75 fruits a year and live for up to 90 years. The origin of the coconut is unknown although the center of genetic diversity lies in south east Asia.

EUCALYPTUS OIL:



Fig 2; Eucalyptus oil

SCIENTIFIC NAME: eucalyptus globulus labill		
KINGDOM	: plantae	
ORDER	: myrtales	
FAMILY	: myrtaceae	
SUB-FAMILY	: Myrtoideae	
GENUS	: eucalyptus L her	
SPECIES	: eucalyptus obliqua L'her	

DESCRIPTION;

Eucalyptus globulus labill. Is an aromatic tree in the myrtle family which commonly attains a height of 150-180 feet and a diameter of height 4-7 feet. It has a straight trunk up to two-third of its total height and a well-developed crown. The central trunk and tap root are fringed with many lateral stems and roots. The tap root rarely exceeds a length of 10 feet. The light, yellow-brown bark is deciduous.

The leaves of the older branches are narrowly lanceolate, often curved, alternate and hung vertically. They are glossy, dark green, thick and leathery. They average in length from 1.5-2 dm. the leaves of the young shoots are ovate, opposite, sessile, and horizontal. They are covered with a gray waxy bloom which is much thicker on the bottom surface of the leaf. Young stems are squared or winged.

The white flowers are solitary in the axil on flattened stalks. They approximately 4-5.5 cm wide. The fruit is 2-2.5 cm across. The the numerous seeds are approximately 2×1 mm. seeds are dark brown with a brownish red chaff.

USES;

Augments Skin and Hair;

Eucalyptus oil being a powerhouse of antioxidants and volatile oils, diminishes skin inflammation, scalp infections and promotes overall skin and hair health. It is also extremely effective in treating eczema, burns and itching. The high amount of eucalyptol present in the oil decreases sebum production and controls the growth of acne, pimples, zits etc.

Mix a few drops of eucalyptus oil with a carrier oil like virgin coconut oil or almond oil and use it as a mask on the face to prevent skin infections and get a clear, glowing complexion. The potent anti -inflammatory and anti-microbial properties also prevent the growth of lice and dandruff on the hair scalp, stimulates blood circulation which in turn prevents hair fall and promotes the growth of new hair.

Relieves pain and inflammation ;

Owing to the strong analgesic, anti-inflammatory and pain relieving properties of the active component eucalyptol, this oil is extensively used for providing relief from pain and inflammation in case of arthritis and joint pain. Being a natural vasodilator, it is also used to treat painful muscle spasms, sore muscles as well as neuralgia. Its property to calm and soothe the mind also makes it useful in case of headache and other inflammatory conditions.

To Treat Rheumatoid Arthritis;

Eucalyptus oil infused toothpaste, mouthwashes or dental gels hold high significance in battling the bad bacteria and hence preventing plaque build-up within the oral cavities that may ultimately lead to painful conditions like gingivitis, toothache etc. being rich in eucalyptol and cineole, an antiseptic and a potent bioactive component, the oil also helps in preventing tooth infection and bestows an instant fresh breath.

VITEX NEGUNDO;



Fig 3; vitex negundo oil

SCIENTIFIC NAME : vitex negundo		
KINGDOM	: plantae	
ORDER	: Lamiales	
FAMILY	: Lamiaceae	
GENUS	: vitex	
SPECIES	: vitex negundo	

Negundo also known as the five-leaved chaste tree is used popularly in ayurveda, unani, siddha, homeopathy and allopathy to treat a number of aliments. It is effective in treating headaches, venereal diseases such as syphilis, rheumatism, sprains, fever, cough, urinary problems, boils and various other ailments.

Negundo oil is an effective analgesic, anti-inflammatory, anti-catarrhal and appetizer among many other attributes that benefit us to stay healthy and recover from various illness.

BENEFITS OF NEGUNDO OIL;

INFECTIONS;

Negundo essential oil showed antibacterial activity against B.subtilis, S. aueus, E.coil, and P. aeruginosa.

PAIN;

The anti- inflammatory and pain suppressing activities of fresh leaves of negundo are attributed to prostaglandin synthesis inhibition.

ARTHRITIS;

The head leaf is used for managing rheumatoid arthritis related pain and sprains. The oil prepared from the leaves can also be used to cure arthritis.

LUNGS;

Negundo leaves decoction can be used for managing bronchitis and asthma.

SKIN;

Negundo leaf juice may be beneficial in managing bacterial and parasitic skin infections. Negundo seeds are used to deal with skin diseases and leprosy.

ANTIOXIDANT;

The water- soluble constituents of vitex negundo possess potent antioxidant activity. Human studies might demonstrate its efficacy in humans.

DESCRIPTION:

Negundo is a herb having quite similar look as basil. It is an ayurvedic herb with numerous health benefits in it. Many of us are still unaware of it. Appearance of five chaste. It is 6-12 ft tall, multibranch bushy plant covered with very fine hairs. The bark of stem is thin and the leaf stalk is long and 3-5 leaves grow at its tip.

The edges of the leaves are plain or serrated. It bears small flowers, in 2-3 inch long inflorescence and are blue or white in color with purple tint. The fruits are small, round and of mixed color. White and black. The bark of the root is green outside and yellow inside.

OTHER INGREDIENTS; METHYL SALICYLATE;



Fig 4; methyl salicylate

Methyl salicylate is a colorless yellowish or reddish liquid with odor

of wintergreen.

Odor; liquid having the characteristic odor of wintergreen.

Taste ; liquid having the characteristic taste of wintergreen.

Density; 1.174

Boiling point; 423° at 760 mm Hg **Melting point;** 16.5° F **Solubility;** less than 1 mg\ml at 66°F **USES;**

- Food additives: flavoring agents
- ✤ Industry uses;
- 1. Not known or reasonably ascertainable
- 2. Odor agents
- ✤ Consumer uses;
- 1. cleaning and furnishing care products
- 2. not known or reasonably ascertainable
- 3. personal care products

SODIUM BENZOATE;



Fig 5; sodium benzoate

Sodium benzoate powder is accepted as a preservative by some of the worlds toughest natural product certification. Using sodium benzoate in shampoo and conditioner as a preservative is a safe and effective technique to protect against bacteria and mold forming the bottles.

TYPE OF INGREDIENT; preservative

MAIN BENEFITS; sodium benzoate is also an effective product stabilizer commonly used in pain.

MENTHOL;



Fig 6; menthol

Menthol is an organic compound, more specifically a monoterpenoid, made synthetically or obtained from the oils of corn mint, peppermint, or other mints. It is a waxy, clear or white crystalline substance, and melts slightly.

CAMPHOR;



Fig 7; camphor

Camphor is derived from the wood of camphor laurel and other related trees of laurel family. Camphor is bicyclic mono terpenoid. It is a white crystalline substance with strong odor and pangent tast. It is a waxy flammable substance obtained from steam distillation, purification and sublimation of wood, twings and bark of the tree.

Uses;

- Topical analgesic
- ✤ Antiseptic
- Muscle relaxant
- Anti- inflammatory
- ✤ Contraceptive
- Cough suppressant

BEES WAX;



Fig; 8 bees wax

Bees wax obtained from the honey comb of the bees Apis mellifera and other species of apis belonging to the family Apidae. Order Hymenptera. It is also known as yellow wax, cera alba. It is yellow to yellowish – brown in colour. Insoluble in water and soluble in alcohol, ether, chloroform, carbon etc.

Uses;

- In preparation of ointments, plasters and polishes.
- Also used in the manufacturing of candles, moulds in dental and electronic industries, cosmetics, lip sticks.
- Is an ingredient of paraffin ointment.

METHODOLOGY;

EXTRACTION OF COCONUT OIL: HOT PROCESSING OF COCONUT MILK: 1.SETTLING OF COCONUT MILK:

The extracted coconut milk is allowed to stand for maximum three hours in the refrigerator or ice box.

2.SEPARATION:

Separate the cream from the coco skin milk, coco skin milk is the used for the preparation of beverage.

3.HEATING OF COCONUT MILK:

Milk is slow heated in double walled boiler about 2 to 2.5 hours.

4.SEPARATION:

Separate the class A oil from proteinaceous residue (kalkkam). Remaining kalkkam can be re heated to recover more oil.

5.DRYING:

Placing the extracted oil in double walled boiler at 50°C.

6.FILTRATION:

Filtered in muslin cloth or sterilized cotton wool or filter paper or fabricated pressure filter.



Fig;9A. extraction of coconut oil



B. filtration of coconut oil



C. coconut oil

EXTRACTION OF EUCALYPTUS OIL;

The leaves of the eucalyptus tree are a popular health remedy all over the world. These leaves have antibacterial and anti- fungal properties. When distilled in to oil, eucalyptus creates an effective inhalant or a chest rub. A few drops of eucalyptus oil in a bath soothes aches and pains.

1.FIND FRESH EUCALYPTUS;

Eucalyptus trees grow wild in warmer climates. In colder climates, eucalyptus is sold by nurseries as a potted plant or shrub. You'll need a good handful -approximately 1/4 cup of leaves for every cup of oil intend to make.

- Eucalyptus is found at most florists, as it is a favourite addition
- ✤ to many floral displays.
- In warmer climates, you might find eucalyptus for sale at farmers market or gardening stores.
- You may find eucalyptus for sale in online. Although it's technically a tree or shrub, it's also labelled as an herb for its.
- The best time of day to cut eucalyptus is early in the morning, when its leaves contain a high concentration of oil.

2.WASH THE LEAVES IN THE SINK WITH WATER;

Rinse well, and set aside to dry. You may also choose to dry the leaves with a clean, dry cloth or towel.

- This step is particularly important when buying eucalyptus from a florist, as the leaves may be sprayed with preservative.
- Get the leaves as dry as you can, but if there's a little water left it will evaporate.

3.MEASURE ONE CUP OF OIL;

The best oil will be coconut oil. A strong scent is unnecessary because you want the eucalyptus scent to dominate the oil.

- If you'd like to make less than 8oz of eucalyptus oil, use less oil and fewer leaves. For example, if you'd like to make 4oz (1/2cup) of oil, measure 40z of oil, and use approximately 1/8 cup eucalyptus leaves.
- If you want to make more, just keep the propotions the same: 4parts oil to 1 part leaves.

4.STRIP EUCALYPTUS LEAVES FROM STEM, AND GENTLY CRUSH WITH YOUR HANDS;

This will start to extract the oil, and your hands will smell of the leaves.

- You can also chop the leaves with a sharp knife. If there are bits of stem and twig in the mix, that's okay.
- If you want to use mixed herbs to create your oil, you should add them at this time.

5.COMBINE OIL AND LEAVES IN A CORK POT SET ON LOW FLAME;

Make sure the lid is on your cork pot. There should be about ¹/₄ cup of oil resting above your leaves.

- Allow the mixture to steep for at least 6 hours. The longer it steeps, the stronger your eucalyptus oil will be.
- The scent of the steaming oil will fairly strong throughout your home. Make sure you're making your eucalyptus oil at a time you can enjoy it.

6.POUR THE EUCALYPTUS OIL THROUGH A FINE MESH STRAINER WHEN THE OIL IS COOL;

Catch the oil in a jar. Ideally, this will be a dark glass jar, but any jar will serve so long as it's kept in a dark location in your home.

- It's important to wait until the oil has cooled before pouring, so the so the glass does'nt crack from the sudden heat.
- Use clean, glass jars with tight fitting lids. Make sure they are dry as well. Any water or moisture in the jars can cause mould.



Fig 10;A. extraction of Eucalyptus oil



B. filtration of eucalyptus oil



C. Eucalyptus oil EXTRACTION OF VITEX NEGUNDO OIL:

- The vitex negundo plants were collected and washed thoroughly and cut into small parts.
- The respective plant parts were allowed to shade dried and then boiled in about 5 times of volume of water\ solvent extract was filtered and boiled with the suitable base (coconut oil) until all the water\ solvent is evaporated.
- The oil obtained was filtered for any residual particles if seen and was stored in suitable container as amber color container.



Fig11;A.extraction of vitex Negundo oil



B.filtration of vitex negundo oil



C. vitex negundo oil

METHOD OF PREPARATION OF HERBAL PAIN RELIEF BALM:

STANDARDIZATION OF HERBAL FORMULATION:

Herbal medicine is not a simple task since many factors influence the biological efficacy and reproducible therapeutic effect. Standardized herbal products of consistent quality and containing well defined constituents are required for reliable clinical trials and to provide consistent beneficial therapeutic effects. Pharmacological properties of an herbal formulation depend on phytochemical constituents present there in. Development of authentic analytical methods which can be reliably profile the phytochemical composition, including quantitative analysis of market\ bioactive compounds and other major constituents, is a major challenge to scientists.

An overview covering various techniques employed in extraction and characterization of herbal medicines as well as herbal nanomedicines standardization is reported. Standardization of herbal formulation is essential in order to asses of quality drugs based on the concentration of their active principles physical, chemical, phytochemical standardization and invitro, in vivo parameters.

The quality assessment of herbal formulations is of paramount importance in order to justify their acceptability in modern system of medicine. One of the major problems faced by the herbal industry is the unavailability of rigid quality control profiles for herbal materials and their formulations.

In India, the development of Ayush government of India launched a central scheme to develop standard operating procedures for the manufacturing process to develop pharmacopeial standards for ayurvedic preparations. The subject of herbal drug standardization is massively wide and deep. There is so much to know and so many seemingly contradictory theories on the subject of herbal medicines and their relationship with human physiology and mental function.

India needs to explore the medicinally important plants. This can be achieved only if the herbal products are evaluated and analysed using sophisticated modern techniques of standardization.

World health organization (WHO) encourages, recommends and promotes traditional\ herbal medicines in natural health care programmes because these drugs are easily available at low cost, safe and people have faith in them. The WHO assembly in number of resolutions has emphasized to need to ensure quality control of medicinal plant product by using modern techniques and applying suitable standards.

WHO guidelines for quality standardized herbal formulations

- a. quality control of crude drugs material, plant preparations and finished product.
- b. Stability assessment and shelf life.
- c. Safety assessment documentation of safety based on experience are toxicological studies.
- d. Assessment of efficacy by ethnomedical information and biological activity evaluations.

Quality control of herbal formulation

Quality control for efficacy and safety of herbal products is of paramount importance. Quality can be defined as the status of formulation that is determined by identity, purity, content and other physical, chemical and biological properties are by manufacturing process. Quality is a term that refers to processes involved in maintaining the quality and validity of the manufactured product.

Stability assessment and shelf life

The past decade has seen a significant increase in the use of herbal formulations as a result of WHO promotion of traditional formulation or medicine. Countries have been seeking the assistance of the organization in identifying safe and effective herbal formulations for using natural health care systems.

Assessment of quality

All procedures should be accordance with good manufacturing practices.

Safety assessment

Assessment of the safety of herbal products therefore is the, first priority in herbal research. These are various approaches to the evaluation of safety of herbal formulations. The toxic effects of herbal preparation may be attributed mainly to the following: inherent toxicity of plant constituents and ingredients and manufacturing malpractice and contamination.

Conventional methods for standardization of herbal formulations

Standardization of herbal formulation requires implementation of good manufacturing practices (GMP). There are many polyherbal formulations which requires standardization as there are frequently used based only on their ethonobotanical use. Standardization minimizes batch to batch variations. Assure, safety, efficacy, quality and acceptability of the polyherbal formulations.

FORMULATION OF HERBAL PAIN RELIEF BALM;

Thake one container in that weigh and add 5gm of petroleum jelly, place the container in a hot plate and boil it until all the amount of petroleum jelly completely dissolved. Weigh 5ml of methyl salicylate and boil the solution in hot plate.

- In the dissolved petroleum jelly solution weigh and add 5gm of bees wax, stir it and boil until the bees wax added completely dissolve in the petroleum jelly.
- ✤ After that, weigh and add 5gm of menthol crystals to the above solution and boil it until the menthol completely dissolved.
- Weigh 10ml of vitex negundo oil, stir the solution and boil the solution.
- ♦ Weigh 10ml of eucalyptus oil, stir the solution and boil the solution.
- Weigh 5gm of sodium benzoate and add it to the solution, stir it well and boil the solution, for complete dissolution of the solution.
- When all the added ingredients were completely dissolved and turns in to the liquid form then take the solution out of the hot plate and keep the herbal balm solution for cooling. Finally the prepared solution cools down and turns into a semi solid herbal balm.

TABLE;1 MATERIALS USED IN FORMULATION OF HERBAL PAIN BALM

S.NO	INGREDIENTS	QUANTITY	MEDICINAL USES	
1	Coconut oil	50ml	Solvent	
2	Eucalyptus oil	10ml	Pain reliever	
3	Vitex negundo oil	10ml	Relieves arthritic pain	
4	Petroleum jelly	5gm	Relieves dry skin, healing	
5	Menthol	5gm	Counter irritant	
6	Camphor	5gm	Relives cough	
7	Methyl salicylate	5gm	Analgesic, skin absorbent	
8	Sodium benzoate	5gm	Preservative	



Fig 12;preparation of herbal pain relief balm

EVALUATION OF PREPARED HERBAL FORMULATION; PHYSICAL PARAMETERS;

Clarity and colour was checked by naked eyes against white background, the odour was smelled.

PH;

The PH of the prepared formulation was determined by using digital PH meter by dipping the glass electrode completely in to the gel system to cover the electrode. The measurement was carried out in triplicate and the average of the three readings was recorded.





Fig13;A.herbal pain balm PHASE SEPARATION:

B. marketed pain balm

The prepared balm was transferred in a suitable wide mouth container. Set aside for storage, the oil phase and aqueous phase separation were visualising after 24h.

VISCOSITY;

Viscosity of balm was determined using brook filled viscometer (S-62,model LVDV-E)at 25°C with a spindle speed of the viscometer rotated at 12rpm.

SPREDABILITY;

Two sets of glass slides of standard dimensions were taken. The herbal balm formulation was placed over one of the slides. The other slides was placed on the top of the gel, such that the gel was sandwiched between the two slides in an area occupied by a distance of 7.5cm along the slides. Hundred g weight of gel was placed on the upper slides so that the gel was between the two slides was pressed uniformly to form a thin layer. The weight was removed and the excess of gel adhering to the slides was scrapped off. The two slides in position were fixed to a stand with out slightest disturbance and in such a way that only upper slides to slip off freely by the force of weight tied on it. A20 g weight was tied to the upper slide carefully. The time taken for the upper slide to travel the distance of 7.5 cm and separated away from the lower slide under the influence of the weighed was noted. The experiment was repeated for three times and the mean time was taken for calculation.

PATCH TEST;

Apply the product to a small patch of skin where a person is unlikely to accidentally wash or rub it away. Good areas may included the inside of the arm or bend of the elbow. Apply the product to a quarter-sized patch of skin. A person should apply the product as thickly as they would when using it regularly. Leave the product on the patch of skin for as long as it would normally be on the skin. If a person is testing a product that they would usually wash off, such as a cleanser, they should keep the patch on for 5min or long as the instructions advice. Repeat the patch test twice a day for between 7-10 days. A reaction may not happen immediately, so it is important to continue applying the product for this length of time. If a person skin react to the product, they should wash it off as soon as possible and stop using it. a person can use a cool compress or petroleum jelly to relieve the skin if needed.

ACCELERATED STABILITY STUDIES:

Accelerated stability testing of prepared herbal balm formulation was at room temperature studied for one week, at $50^{\circ}C \pm 1^{\circ}C$ for 3 months. The herbal balm formulation were kept both at room and elevated temperature and observed on 0th, 15th, 20th, 30th, 40th, 50th, 60th, 70th, 80th and 90th day for the following parameters.

RESULTS AND DISCUSSION;

The physicochemical parameters of the prepared balm were determined parameters such as colour, odour, appearance and PH were tested. The formulations exhibited good in appearance characteristic as well as PH was found in the range 7.0 which is the desired PH of the skin.

S.NO	ORGANOLEPTIC	HERBAL BALM	MARKETED BALM	
	CHARACTERS			
1	Formulation	Pain balm	Pain balm	
2	Colour	Light green	white	
3	Odour	Fragrant	Fragrant	
4	Appearance	Good	Smooth	
5	State	Semi solid	Semi solid	

TABLE2; PHYSICAL PARAMETERS OF HERBAL PAIN BALM

S.NO	PARAMETERS	RESULTS
1	РН	6.5
2	Spreadability	7.4g. cm/sec
3	Phase separation	No phase separation
4	Viscosity	39010 cps
5	Patch test	Non allergenic

TABLE3; EVALUATION RESULT OF HERBAL PAIN BALM

TABLE4; RESULT OF STABILITY STUDIES

FORMULATION	PERIOD	COLOUR	PH	ANY OTHER
	Room temperature	Light green	7.0	Nil
	$25^{\circ}C \pm 2^{\circ}C/60\%$	Light green	6.8	Nil
PAIN BALM	±5% RH			
	$30^{\circ}C \pm 2^{\circ}$ C/ $65\% \pm$	Light green	6.7	Nil
	5% RH			
	$30^{\circ} \text{ C} \pm 2^{\circ} \text{ C}/65\% \pm$	Light green	6.5	Nil
	5%			

SUMMARY AND CONCLUSION:

•Herbal balm was prepared by using Hot Processing Technique and were found to be without particles transparent components which are used in formulation are having good compatibility without any significant changes.

•The Eucalyptus leaves extracts have relieving pain property, vitex negundo leaves extracts used to relieve Arthritic pain, cures high fever and alleviates menstrual cramps.

•The prepared formulation showing good physical characteristics.

•Further evaluated by various evaluation parameters such as PH, Extrudability, Spreadability, Viscosity, Patch test and gives good result.

•Based on the study research it can be concluded that herbal components can be effectively formulated as in the form of balm by using Hot

•Processing Technique which having excellent pain-relieving property.

REFERENCES;

1. Acebo E, Raton JA, Sautua S, Eizaguirre X, Trébol I, Pérez JL. Allergic contact dermatitis from *Boswellia serrata* extract in a naturopathic cream. *Am J Contact Dermat.* 2004;51:91–2. [PubMed] [Google Scholar]

2. Aguilar JL, Rojas P, Marcelo A, Plaza A, Bauer R, Reininger E, et al. Anti-inflammatory activity of two different extracts of *Uncaria tomentosa* (Rubiaceae) *J Ethnopharmacol.* 2002;81:271–6. [PubMed] [Google Scholar]

3. Almekinders LC, Gilbert JA. Healing of experimental muscle strains and the effects of nonsteroidal anti-inflammatory medication. *Am J Sports Med.* 1986;14:303–8. [PubMed] [Google Scholar]

4. Almekinders LC. Anti-inflammatory treatment of muscular injuries in sport. An update of recent studies. *Sports Med.* 1999;28:383–8. [PubMed] [Google Scholar]

5. Andersohn F, Suissa S, Garbe E. Use of first- and second-generation cyclooxygenase-2-selective nonsteroidal antiinflammatory drugs and risk of acute myocardial infarction. *Circulation*. 2006;113:1950–7. [PubMed] [Google Scholar]

6. Boswellia serrata. Altern Med Rev. 1998;3:306–7. [PubMed] [Google Scholar]

7. Araujo CC, Leon LL. Biological activities of *Curcuma longa* L. *Mem Inst Osawaldo Cruz.* 2001;96:723–8. [PubMed] [Google Scholar]

8. Badria FA, El-Farahaty T, Shabana AA, Hawas SA, El-Batoty MF. Boswellia-curcumin preparation for treating knee osteoarthritis: A clinical evaluation. *Alt Complement Ther*. 2002;8:341–8. [Google Scholar]

9. Banerjee M, Tripathi LM, Srivastava VM, Puri A, Shukla R. Modulation of inflammatory mediators by ibuprofen and curcumin treatment during chronic inflammation in rat. *Immunopharmacol Immunotoxicol.* 2003;25:213–24. [PubMed] [Google Scholar]

10. Banno N, Akihisa T, Yasukawa K, Tokuda H, Tabata K, Nakamura Y, et al. Antiinflammatory activities of the triterpene acids from the resin of *Boswellia carteri*. *J Ethnopharmacol*. 2006;107:249–53. [PubMed] [Google Scholar]

11. Bengmark S. Curcumin, an atoxic antioxidant and natural NFk B, cyclooxygenase-2, lipooxygenase, and inducible nitric oxide synthase inhibitor: A shield against acute and chronic diseases. *JPEN J Parenter Enteral Nutr.* 2006;30:45–51. [PubMed] [Google Scholar]

12. Bernstein JE, Bickers DR, Dahl MV, Roshal JY. Treatment of chronic postherpetic neuralgia with topical capsaicin. A preliminary study. *J Am Acad Dermatol*. 1987;17:93–8. [PubMed] [Google Scholar]

13. Burton TM. Monsanto arthritis-pain drug, Celebrex, surpasses Viagra's early sales success. *The Wall Street Journal B: New York*. 1999 [Google Scholar]

14. Calder PC. N-3 Polyunsaturated fatty acids, inflammation, and inflammatory diseases. *Am J Clin Nutr.* 2006;83:1505S–19. [PubMed] [Google Scholar]

15. Caterina MJ, Julius D. The vanilloid receptor: A molecular gateway to the pain pathway. *Annu Rev Neurosci.* 2001;24:487–517. [PubMed] [Google Scholar]

16. Caterina MJ, Leffler A, Malmberg AB, Martin WJ, Trafton J, Petersen-Zeitz KR, et al. Impaired nociception and pain sensation in mice lacking the capsaicin receptor. *Science*. 2000;288:306–13. [PubMed] [Google Scholar]

17. Cho KJ, Yun CH, Yoon DY, Cho YS, Rimbach G, Packer L. Effects of bioflavonoids extracted from the bark of *Pinus maritime* on proinflammatory cytokine interleukin-1 production in lipopoysaccharide-stimulated RAW 264.7. *Toxicol Appl Pharmacol.* 2000;168:64–71. [PubMed] [Google Scholar]

18. Chrubasik S, Eisenberg E, Balan E, Weinberger T, Luzzati R, Conradt C. Treatment of low back pain exacerbations with willow bark extract: A randomized double blind study. *Am J Med.* 2000;9:9–14. [PubMed] [Google Scholar]

19. Chrubasik S, Künzel O, Model A, Conradt C, Black A. Treatment of low back pain with a herbal or synthetic anti-rheumatic: A randomized controlled study. Willow bark extract for low back pain. *Rheumatology*. 2001;40:1388–93. [PubMed] [Google Scholar]

20. Chung JM, Lee KH, Hori Y, Willis WD. Effects of capsaicin applied to a peripheral nerve on the responses of primate spinothalamic tract cells. *Brain Res.* 1985;329:27–38. [PubMed] [Google Scholar]

21. Claeson P, Panthong A, Tuchinda P, Reutrakul V, Kanjanapothi D, Taylor WC, et al. Three non-phenolic diarylheptanoids with anti-inflammatory activity from *Curcuma xanthorrhiza*. *Planta Med*. 1993;59:451–4. [PubMed] [Google Scholar]

22. Clemett D, Goa KL. Celecoxib: A review of its use in osteoarthritis, rheumatoid arthritis and acute pain. *Drugs*. 2000;59:957–80. [PubMed] [Google Scholar]

23. Curtis CL, Harwood JL, Dent CM, Caterson B. Biological basis for the benefit of nutraceutical supplementation in arthritis. *Drug Discov Today*. 2004;9:165–72. [PubMed] [Google Scholar]

24. Curtis CL, Hughes CE, Flannery CR, Little CB, Harwood JL, Caterson B. N-3 fatty acids specifically modulate catabolic factors involved in articular cartilage degradation. *J Bio Chem.* 2000;275:721–4. [PubMed] [Google Scholar]

25. Curtis CL, Rees SG, Little CB, Flannery CR, Hughes CE, Wilson C, et al. Pathologic indicators of degradation and inflammation in human osteoarthritic cartilage are abrogated by exposure to n-3 fatty acids. *Arthritis Rheum.* 2002;46:1544–53. [PubMed] [Google Scholar]

26. Graham DJ. COX-2 Inhibitors, Other NSAIDs, and Cardiovascular Risk: The Seduction of Common Sense. *JAMA*. 2006;296:1653–6. [PubMed] [Google Scholar]

27. Daviglus ML, Stamler J, Orencia AJ, Dyer AR, Liu K, Greenland P, et al. Fish consumption and the 30-year risk of fatal myocardial infarction. *N Engl J Med.* 1997;336:1046–53. [PubMed] [Google Scholar]

28. Ehrich EW, Dallob A, De Lepeleire I, Van Hecken A, Riendeau D, Yuan W, et al. Characterization of rofecoxib as a cyclooxygenase- 2 isoform inhibitor and demonstration of analgesia in the dental pain model. *Clin Pharmacol Ther*. 1999;65:336–47. [PubMed] [Google Scholar]

29. Elmali N, Baysal O, Harma A, Esenkaya I, Mizrak B. Effects of resveratrol in inflammatory arthritis. *Inflammation*. 2007;30:1–6. [PubMed] [Google Scholar]

30. Ernst E. Adulteration of Chinese herbal medicines with synthetic drugs: A systematic review. *J Intern Med.* 2002;252:107–13. [PubMed] [Google Scholar]

31.ASHA, V.V.; PUSHPANGADAN, P. Anti-pyretic activity of Cardiospermum halicacabum. Indian J. Exp. Biol., v.37, n.4, p.411-414, 1999.

32.BABU, K.C.V.; KRISHNAKUMARI, S. Cardiospermum halicacabum suppresses the production of TNF- α and NO by human peripheral blood mononuclear cells. Afr. J. Biomed. Res., v.9, p.95-99, 2006.

33.BLONCO-FLONTE, H.; ANGUIANO-IGEA S.; OTERO-ESPINAR, F.J.; BLANCOMENDEZ, J. In-vitro bioadhesion of carbopol hydrogel. Int. J. Pharm., v.142, p.169-174, 1996.

34.CHOI, E.M.; LEE, Y.S. Luteolin suppresses IL-1b-induced cytokines and MMPs production via p38 MAPK, JNK, NF-kappaB and AP-1 activation in human synovial sarcoma cell line, SW982. Food Chem. Toxicol., v.48, n.10, p.2607-2611, 2010.

35.FELDMANN, M.; MAINI, S.R. Role of cytokines in rheumatoid arthritis: an education in pathophysiology and therapeutics. Immunol. Rev., v.223, p.7-19, 2008.

36.GHOSH, M.N. Fundamentals of experimental pharmacology. Kolkatta: Scientific Book Agency, 1984. p.156-157.

37.GIINTER, S.; IRMARGD, M.; UTE, W.; CHISTOPH, M.S. Anticarcinogenic effects of the flavonoid luteolin. Molecules., v.13, n.10, p.2628-2651, 2008.

38.GOPALAKRISHNAN, C.; DHANANJAYAN, R.; KAMESWARAN, L. Studies on the pharmacological actions of Cardiospermum halicacabum. Indian J. Physiol. Pharmacol., v.20, p.203-206, 1976.

39.GUPTA, M.; MAZUMDER, U.K.; BHAWAL, S.R. CNS activity of Vitex negundo Linn in mice. Indian J. Exp. Biol., v.37, n.2, p.143-146, 1999.

40.JAIN, S.; PADSALG, B.D.; PATEL, A.K.; MOALE, V. Formulation development and evaluation of fluconazole gel in various polymer bases. Asian J. Pharm., v.1, p.63-68, 2007.

41.JEYADEVI, R.; SIVASUDHA, T.; RAMESH KUMAR, A.; DINESH KUMAR, L. Antiarthritic activity of the Indian leafy vegetable Cardiospermum halicacabum in Wistar rats and UPLC-QTOF-MS/MS identification of the putative active phenolic components. Inflamm. Res., v.62, n.1, p.115-26, 2013.

42.KIM, J.Y.; SONG, J.Y.; LEE, E.J.; PARK, S.K. Rheological properties and microstructures of carbopol gel network system. Colloid Polym. Sci., v.281, n.7, p.614-623, 2003.

43.KUMAR, E.; MASTAN, S.K.; AMRANDER REDDY, G.; RAGUNANDAN, N.; SREEKANTH, N.; CHAITANYA, G. Anti-arthritic property of the ethanolic leaf extracts of Cardiospermum halicacabum Linn. Biomed. Pharmacol. J., v.1, p.2, 2008.

44.KUMARAN, A.; KARUNAKARAN, R.J. Antioxidant activities of the methanol extract of Cardiospermum halicacabum. Pharm. Biol., v.44, n.2, p.146-151, 2006.

45.LAIRD, J.M.A.; CARTER, A.J.; GRAUERT, M.; CERVERO F. Analgesic activity of a novel use-dependent sodium channel blocker, crobenetine, immuno-arthritic rats, Br. J. Pharmacol., v.134, n.8, p.1742-1748, 2001.

46.LOGANATHAN, V.; MANIMARAN, S.; JASWANTH, A.; SULAIMAN, A.; SHIVAPRASADHA, R.M.V.; SENTHIL KUMAR, B.; RAJASEKARAN, A. The effects of polymers and permeation enhancers on releases of flurbiprofen from gel formulations. Indian J. Pharm. Sci., v.63 n.3, p.200-204, 2001.

47.MARTIN, A. Physical pharmacy, kinetics. First Indian reprint. New Delhi: B.I Waverly, 1994.

48.MIZUSHIMA, Y.; TSUKADA, W.; AKIMOTO, T. A Modification of rat adjuvant arthritis for testing anti-rheumatic drugs. J. Pharm. Pharmacol., v.24, n.10, p.781-785, 1972.

49.MURPHY, C.T.; MCCARROLL S.A.; BARGMANN, C.I.; FRASER, A.; KAMATH, R.S.; Genes that act downstream of DAF-16 to influence the lifespan of Caenorhabditis elegans. Nature, v.424, p.277-283, 2003.

50.NAIR, A.M.; SARAF, M.N. Inhibition of antigen and compound 48/80 induced contraction of guinea pig trachea by ethanolic extract of the leaves of Vitex negundo linn. Indian J. Pharmacol., v.27, n.4, p.230-233, 1995.

51.NAIR, C.K.N.; MOHENAN, N. Medicinal plants in India with special reference to Ayurveda. Delhi, India: NAG Publisher, 1998.

52.NANDGUDE, T.; THUBE, R.; JAISWAL, N.; DESHMUKH, P.; CHATAP, V.; HIRE, N. Formulation and evaluation of pH induced in situ nasal gel of salbutamol sulphate. Int. J. Pharm. Sci. Nanotechnol., v.1, n.2, p.177-83, 2008.

53.NAPPINNAI, M.; PAKALAPATI, S.; ARIMILLI, R. Roficoxib gels-preparation and evaluation. Indian Drugs., v.43, p.513-15, 2006.

54.NAYAK, S.H.; NAKHAT, P.D.; YEOLE, P.G. Development and evaluation of cosmeceutical hair styling gels of ketoconazole. Indian J. Pharm.Sci., v.52, p.231-33, 2005.

55.NIELEN, M.M.; SCHAARDENBURG, D.V.; REESINK, H.W.; TWISK, J.W.R.; VAN DE STADT, R.J.; VANDER HORST, B.I.E.; DE KONING, M.H.; HABIBUW, M.R.; DIJKMANS, B.A. Simultaneous development of acute phase response and auto antibodies in preclinical rheumatoid arthritis. Ann. Rheum Dis., v.65, n.4, p.535-537, 2006.

56.PANIGRAHI, L.; GHOSAL, S.K.; PATTNAIK, S.; MAHARANA, L.; BARIK, B.B. Effect of permeation enhancers on the release and permeation kinetics of lincomycin hydrochloride gel formulations through mouse skin. Indian J. Pharm. Sci., v.68, p.205-11, 2006.

57.PATIL, K.R.; PATIL, C.R.; JADHAV, R.B. Antiarthritic activity of bartogenic acid isolated from fruits of Barringtonia racemosa Roxb. (Lecythidaceae). Evid. Based Complim. Alternat. Med., p.1-7, 2009.

58.QUEIROZ, M.B.R.; MARCELINO, N.B.; RIBEIRO, M.V.; ESPINDOLA, L.S.; CUNHA, F.; SILVA, M.V. Development of gel with Matricaria recutita L. extract for topic application and evaluation of physical-chemical stability and toxicity. Lat. Am. J. Pharm., v.28, n.4, p.574-579, 2009.

59.RAJASEKARAN, A.; ARULKUMARAN, G.; ARIVUKKARASU, R. Acute and subacute toxicity study of methanol leaf extract of Cardiospermum halicacabum L and Vitex negundo L in rats. Pharmacog. Commun., v.5, n.1, p.39-45, 2015.

60.SHARIFIFAR, S.; DEHGHN-NUDEH, G.; MIRTAJALDINI, M. Major flavonoids with antioxidant activity from Teucrium polium L. Food Chem., v.112, n.4, p.885-888, 2009.

61.SHEEBA, M.S.; ASHA, V.V. Cardiospermum halicacabum ethanol extract inhibits LPS induced COX-2, TNF-alpha and iNOS expression, which is mediated by NF-kappa B regulation, in RAW264.7 cells. J. Ethnopharmacol., v.124, n.1, p.39-44, 2009.

62.SHEEBA, M.S.; ASHA, V.V. Effect of Cardiospermum halicacabum on ethanol induced gastric ulcers in rats. J. Ethnopharmacol., v.106, n.1, p.105-110, 2006.

63.SUBRAMANYAM, R.; NEWMASTER, S.G.; PALIYATH, G.; NEWMASTER, C.B. Exploring ethnobiological classifications for novel alternative medicine: a case study of Cardiospermum halicacabum L. (Modakathon, Balloon Vine) as a traditional herb for treating rheumatoid arthritis. Ethnobotany, v.19, p.1-18, 2007.

64.TAMHANKAR, C.P.; SARAF, M.N.; Anti-arthritic activity of Vitex negundo Linn. Indian J. Pharm. Sci., v.56, n.1, p.158-159, 1994.

65.TELANG, R.S.; CHATTERJEE, S.; VARSHNEYA, C. Studies on analgesic and antiinflammatory activities of Vitex negundo Linn. Indian J. Pharmacol., v.31, p.363-366, 1999.

66.TSAI, C.C.; LIN, C.C. Anti-inflammatory effects of Taiwan folk medicine 'Teng-Khaia-U' on carrageenan and adjuvant-induced paw edema in rats. J. Ethnopharmacol., v.64, n.1, p.85-89, 1999.

67.WAAKO, P.J.; GUMEDE, B.; SMITH, P.; FOLB, P.I. The in vitro and in vivo anti-malarial activity of Cardiospermum halicacabum L. and Momordica foetida Schumch. J Ethnopharmacol., v.99, p.137-143, 2005.

68.WALKER, R.B.; SMITH, E.W. The role of percutaneous penetration enhancers. Adv. Drug Deliv. Rev. v.18, n.3, p.295-301, 1996.