Immune-Modifying and Antimicrobial Effects of Eucalyptus Oil and Simple Inhalation Devices

Angela E. Sadlon, ND, and Davis W. Lamson, MS, ND

Abstract
Eucalyptus oil (EO) and its major component, 1,8-cineole, have antimicrobial effects against many bacteria, including *Mycobacterium tuberculosis* and methicillin-resistant *Staphylococcus aureus* (MRSA), viruses, and fungi (including Candida). Surprisingly for an antimicrobial substance, there are also immune-stimulatory, anti-inflammatory, antioxidant, analgesic, and spasmolytic effects. Of the white blood cells, monocytes and macrophages are most affected, especially with increased phagocytic activity. Application by either vapor inhalation or oral route provides benefit for both purulent and non-purulent respiratory problems, such as bronchitis, asthma, and chronic obstructive pulmonary disease (COPD). There is a long history of folk usage with a good safety record. More recently, the biochemical details behind these effects have been clarified. Although other plant oils may be more microbiologically active, the safety of moderate doses of EO and its broad-spectrum antimicrobial action make it an attractive alternative to pharmaceuticals. EO has also been shown to offset the myelotoxicity of one chemotherapy agent. Whether this is a general attribute that does not decrease the benefit of chemotherapy remains to be determined. This article also provides instruction on how to assemble inexpensive devices for vapor inhalation.
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Angela Sadlon, ND – Holder of the Thorne Post-Doctoral Fellowship 2009-2010; Research associate to Dr. Davis Lamson
Correspondence address: dr.a.sadlon@gmail.com

Davis Lamson, ND, MS – Adjunct faculty in oncology, Bastyr University, Kenmore, WA; private practice, Tahoma Clinic, Renton, WA; product consultant, Thorne Research; contributing editor, *Alternative Medicine Review*

Introduction

Eucalyptus oil (EO) has antibacterial, antiviral, and antifungal components and a long history of use against the effects of colds, influenza, other respiratory infections, rhinitis, and sinusitis.

Inhalation of the vapor is safe; historical usage employed the method of breathing the vapor over a bowl of hot water containing a few drops of EO with a towel-tent over the head. Readers are likely to have experience with eucalyptus oil via their parents, who may have applied Vicks VapoRub® to the chest area and even to the nose for respiratory difficulty or infection. It seems that Vicks VapoRub not only works through inhalation, but also through absorption into the tissues of the chest.[1,2]

Devices are now sold in pharmacies that allow one to inhale vapor, foregoing the bowl, towel, stove, and risk of a par-boiled face. These devices perhaps provide greater convenience than the bowl of hot water, but the rather expensive per-use pads required for the device contain little EO.

This article reviews the published data on the medicinal attributes, safety, and efficacy of eucalyptus oil. The article also describes a simple, low-cost home delivery system for the vapor and an easily constructed pocket inhaler. Included is limited information on activity of other plant oils with similar components.

Most of the references cited have been obtained from a PubMed search of eucalyptus oil cross-referenced with other subjects. Some of the references cited were not found in PubMed, but in bibliographies of other publications. A PubMed search for 1,8-cineole (Figure 1), the major component of most EO species and present in tea tree, rosemary, and other plant oils, lists 635 publications. References on EO as an antiparasitic or antiprotozoan, bug repellent, or insecticide are not covered in this discussion; use of EO to increase penetration of other agents through the skin is also not included.

Components of Eucalyptus Oil

The percentage of components varies with species, plant part, and batch. Table 1 lists the major components of EO from five species and tea

tree oil (TTO). The species selected were randomly chosen except for *E. globulus* (Figure 2),[3] which is listed twice to demonstrate the variability of constituents within the same species, likely due to location of tree growth or time of year harvested. One publication claimed 43 components in the oil of *E. tereticornis* from fresh leaves as analyzed by gas chromatography.[4] Oil yield from leaves ranging from 3.57-10.6 mg/g volatile components was demonstrated when various species were steam-distilled. *E. globulus* produced 5.25 mg oil/g of fresh leaf.[5] Research interpretation of microbial action is complicated by the use of different EO species with varying percentages of individual components. No formal correlation between the amount of major constituents and the antibacterial activity has been determined.[6]

Immunomodulatory/Anti-inflammatory Effects *In vitro*

E. globulus oil dose-dependently stimulated phagocytosis (according to several parameters) by human monocyte-derived macrophages (MDMs) *in vitro*, without producing pro-inflammatory effects (Table 2). Phagocytic activity was not increased when the MDMs were exposed to TTO or lavender oil. Eucalyptus oil's phagocytic ability was halted when EO-stimulated cells were treated with a microtubule-destabilizing chemical, suggesting EO's phagocytic ability is dependent on the microtubule network.[7]

To determine the effect on inflammation, a cytokine profile was performed on MDMs treated with EO only, lipopolysaccharide (LPS) only, and with EO pretreatment, then LPS. EO produced no difference in cytokine profile from the control group. LPS significantly elevated interleukins (IL-4, IL-6) and tumor necrosis factor-alpha (TNF-α). EO reduced the inflammatory effect of LPS. Neither LPS nor EO had influence on IL-2, IL-10, or interferon-gamma (IFN-γ).[7]

1,8-cineole inhibited the stimulated cytokine production from human lymphocytes and monocytes. At 1.5 mcg/mL (10^{-5} M), 1,8-cineole significantly inhibited cytokine production in lymphocytes of TNF-α > IL-1β > IL-4 > IL-5 by 92-, 84-, 70-, and 65 percent, respectively. Cytokine production in monocytes was also inhibited: TNF-α > IL-1β > IL-6 > IL-8 by 99-, 84-, 76-, and 65 percent, respectively. At 10-fold lower concentration, 1,8-cineole demonstrated dose dependency with smaller declines of cytokines. At 0.15 mcg/mL (10^{-6} M), production of TNF-α and IL-1β was significantly inhibited in lymphocytes by 16- and 36 percent, respectively, while in monocytes, TNF-α and IL-1β were inhibited by 77- and 61 percent, respectively. Thus, 1,8-cineole at 10^{-6} M had a larger impact on TNF-α and IL-1β production in monocytes compared to lymphocytes, but similar effects at 10^{-5} M. The authors characterized 1,8-cineole as a strong inhibitor of TNF-α and IL-1β, with smaller effects on chemotactic cytokines. These authors suggest that since 1,8-cineole can control airway mucus hypersecretion, it might reduce exacerbations in asthma, sinusitis, and chronic obstructive pulmonary disease (COPD).[8]

Figure 1. Structure of 1,8-Cineole

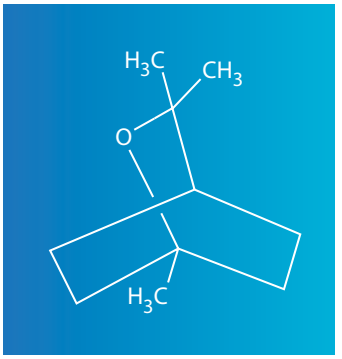

Figure 2. Photo of *Eucalyptus globulus*

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Stimulation of a human monocyte cell line with LPS increased early growth response factor-1 (Erg-1) in the nucleus and the whole cell. (Erg-1 is a transcription factor implemented in regulation of cell proliferation and apoptosis.) Pretreatment of the cells with 1,8-cineole (1-100 mg/L) decreased expression of Egr-1 in the nucleus and whole cell in a concentration-dependent manner. There was no change in the expression of nuclear factor kappa B

Key words: eucalyptus, tea tree, antimicrobial, cineole, MRSA

Table 1. Eucalyptus and Tea Tree Oil Constituents

Constituent	Boiling points°C	*E. globulus*[3]	*E. globulus*[6]	*E. citriodora*[3]	*E. nicholii*[3]	*E. terticornis*[6]	*E. camaldulensis*[19]	Tea tree oil[52]
cineole	176	46.8%	44.3%	-----	83.63%	6.2%	33.8%	5.1%
α-Pinene	156	28.9	9.3	0.14	2.93	8.3	4.20	2.6
d-Limonene	178	4.9	5.1	-----	4.50	-----	-----	1.0
β- Pinene	165	0.6	2.7	0.36	0.03	2.5	16.70	-----
Myrcene	167	0.2	-----	-----	0.03	1.3	0.71 (β)	-----
α-Phellandrene	136	-----	-----	-----	0.17	1.6	1.97	-----
Camphene	158	-----	23.1	-----	-----	-----	-----	-----
p-Cymene	178	1.4	1.6	-----	1.00	28.6	-----	2.9
Terpinene	175	0.1 (γ)	-----	-----	0.09 (γ)	0.8(γ)	0.51 (α)	23.0 (γ) 10.4 (α)
α-Terpinolene	136	-----	-----	-----	0.04	-----	0.68	3.1
Linalol	154	-----	0.3	0.66	-----	-----	-----	-----
Isopulegol	154	-----	-----	3.41	-----	-----	-----	-----
Citronellal	154	-----	-----	80.10	-----	-----	-----	-----
Iso-isopulgeol	-----	-----	-----	8.51	-----	-----	-----	-----
4-Terpineol	-----	-----	0.2	-----	0.13	1.7	5.15	40.1
α-Terpineol	175	1.8	0.3	-----	1.91	5.6	0.80	2.4
Citronellol	156	-----	0.1	4.18	-----	-----	-----	-----
α-Terpinyl acetate	196	0.2	1.2	-----	-----	0.2	-----	-----
Citronellyl acetate	-----	-----	-----	0.02	-----	-----	-----	-----
Geraniol	154	-----	0.2	-----	-----	-----	-----	-----
Caryophyllene	204	-----	-----	0.39	0.17	0.6 (β)	-----	-----
Aromadendrene	-----	2.9	1.3	-----	0.08	-----	-----	1.5
Bicyclogermacrene	-----	-----	-----	-----	0.08	-----	0.70	-----
Oxy-sesquiterpenes	-----	3.2	-----	0.10	0.48	-----	16.77	-----
Myrtenal	-----	-----	-----	-----	-----	-----	2.10	-----
α-Thujene	136	-----	-----	-----	-----	-----	2.15	-----
trans-Pinocarveol	-----	-----	-----	-----	-----	-----	1.47	-----
Cryptone	-----	-----	1.3	-----	-----	17.8	5.85	-----
Borneol	154	-----	-----	-----	-----	0.3	-----	-----
Cuminaldehyde	-----	-----	-----	-----	-----	6.5	-----	-----
Globulol	-----	-----	7.3	-----	-----	0.5	-----	0.2
Spathulenol	-----	-----	-----	-----	-----	1.8	-----	-----
δ-Cadinene	-----	-----	-----	-----	-----	-----	-----	1.3
Sabinene	-----	-----	-----	-----	-----	-----	-----	0.2
Viridiflorol	-----	-----	-----	-----	-----	-----	-----	0.1

EO and TTO constituents as percentages determined by GC-MS. Note with EO the differences between species and the variability within the same species. Out of 800+ species of Eucalyptus, five were chosen to list their constituents. The dashes represent either unfound boiling points or undetectable constituents.

(NF-κB).[9] In a similar experiment using α-pinene (a lesser component of EO than 1,8-cineole), nuclear translocation of NF-kB was reduced, inhibiting NF-κB activity.[10] Thus, EO (at least from *E. globulus*) as a whole has NF-κB reducing activity.[11]

An *in vitro* study of 1,8-cineole (10 mcg/mL concentration) on LPS-stimulated human monocytes showed a dose-dependent and significant inhibition of TNF-α (99%), IL-1β (74%), leukotriene (LT) B4 (47%), and thromboxane B2 (91%) after 20 hours. IL-1β-stimulated MDMs showed 98-percent reduction in TNF-α with the same concentration of 1,8-cineole.[12]

Animals Studies

Implementation of innate cell-mediated immune response was also observed *in vivo* after EO administration. In rat peripheral blood, after 15 days of oral EO treatment, a significant increase in monocytes occurred, without effect on granulocytes or lymphocytes. There was also a significant increase of CD44 and CD25 monocyte surface markers, but not on granulocytes or lymphocytes. This effect continued for five days after cessation of EO and was interpreted as monocyte activation and extravasation.[7]

Effects on immune-suppression in rats were evaluated with a 5-fluorouracil (5-FU)/EO combination. EO inhibited 5-FU-induced myelotoxicity and raised the phagocytic activity of the granulocytic/monocytic system. It was suggested that EO might be useful as a cell-mediated immuno-regulatory agent in immune-suppressive pathologies, infectious diseases, or after cancer chemotherapy.[7]

E. globulus oil (most effective dose, 300 mg/kg) resulted in significant reduction of bronchitis symptom severity, lower infiltration of inflammatory cells, and decreased airway mucins in LPS-induced chronic bronchitis in rats.[13]

Human Clinical Trials

Asthma

LTB4 and prostaglandin E2 (PGE2), both produced in the pathway of arachidonic acid metabolism, were measured in stimulated monocytes from 10 patients with bronchial asthma and 12 healthy controls after treatment with 1,8-cineole (200 mg three times daily) for three days. Forced expiratory volume in one second (FEV_1) and airway resistance (RAW) were measured the day before treatment, during treatment, and after discontinuing treatment. Significant inhibition of LTB4 and PGE2 was observed in both groups. The FEV_1 increased by 23.7 percent and RAW decreased by 26.1 percent after three days of cineole treatment. When lung function was checked four days after the end of treatment, FEV_1 (28.7%) and RAW (-17.6%) were still significantly improved compared to before treatment.[14]

The anti-inflammatory effect of oral 1,8-cineole (200 mg three times daily) was demonstrated in a double-blind, placebo-controlled, 12-week trial of asthma patients. The required oral glucocorticoid dosage was decreased by a mean of 3.75 mg in the cineole group compared to 0.91 mg in the control group. Prednisolone dose prior to treatment was 5-24 mg daily with average of 11 mg. Of note, the use of the rescue medication salbutamol (albuterol) was increased almost two-fold in the control group when prednisolone was lowered by 2.5 mg; whereas, there was no significant increase in rescue puffs in the cineole group, even with a decrease of 5 mg prednisolone. Some participants dropped out at that reduction (four in the cineole group, 11 in the placebo group). The cineole group maintained lung function capacity (peak expiratory flow rate, FEV_1, and RAW) four times longer than placebo, even at a lower prednisolone dosage.[15]

Rhinosinusitis

A study showed that cineole is effective for the discomforts of non-purulent rhinosinusitis. Individuals (n=150) with subjective findings of

Table 2. Immunomodulatory Effects of Eucalyptus Oil and 1,8-Cineole

Eucalyptus Oil

- Increases phagocytic activity and number of monocytes/macrophages[7]
- Decreases significantly or inhibits IL-4, IL-6, TNF-α and NF-κB when inflammation present[7]
- No influence on IL-2, IL-10, INF-γ[7]
- Significantly lowers inflammatory cell infiltrates such as neutrophils[8,17]
- Decreases airway mucin secretion of tracheal and bronchiole epithelium[8]
- EO/5-FU combination inhibited myelotoxicity and increased phagocytic activity of granulocytes[7]

1,8-Cineole

- Inhibits or reduces TNF-α, IL-1β, IL-4, IL-5, IL-6, IL-8, LTB4, PGE2, TxB2[9,10,13]
- Affects monocytes/macrophages more than other leukocytes[13]
- Decreases localization of Egr-1[14]
- Had no effect on NF-κB[14]

TNF-α = tumor necrosis factor-alpha; **IL** = interleukin; **LTB4** = leukotriene B4; **TxB2** = thromboxane B2; **NF-κB** = nuclear factor-kappaB; **INF-γ** = interferon-gamma; **5-FU** = 5-fluorouracil

headache with or without bending, tenderness to pressure points of the trigeminal nerve, impairment of general condition, nasal obstruction, and nasal secretions (rated by quantity and viscosity) were randomized in a double-blind, placebo-controlled trial. Subjects in the treatment group (n=75) showed over 80-percent improvement after seven days of oral 1,8-cineole (200 mg three times daily) compared to less than 50-percent improvement in the placebo group. Improvement was determined by symptoms-sum-score. Ultrasonography at the end of the study showed that sinus shadowing remained in 37 patients in the placebo group and four patients in the cineole group. All patients also inhaled 100 mcg of the decongestant xylometazoline three times daily to relieve nasal congestion.[16]

COPD

The efficacy of oral cineole (200 mg three times daily) was demonstrated in 242 COPD patients in a double-blind, placebo-controlled, six-month trial. Cineole demonstrated a significant decrease in the exacerbation frequency compared to placebo (occurrences in six months, 0.4 versus 0.9), severity (by subjective scoring), and duration (average 4.0 versus 5.7 days). Patient medications at the start of the study were not altered during the study. Lung function tests showed no significant differences between treatment and control. Other symptoms of COPD, such as "trouble breathing" or dyspnea, showed improvement scoring in both groups, with no statistically significant differences between groups.[17]

Analgesic Effect

Three species of EO (*E. citriodora*, *E. tereticornis*, and *E. globulus*) showed dose-dependent and time-dependent peripheral and central acting analgesic properties in rodents compared to morphine, using standard experimental test models. *E. tereticornis* had the greatest anti-inflammatory action in a model of rat paw edema. The anti-inflammatory action of EO compared to dexamethasone showed an average of 75-percent versus 97-percent inhibition, respectively, of neutrophil migration into the rat peritoneal cavity. A test for vascular permeability effect showed reduction, but widely varied by species and permeability agent.[18]

When rats or mice were injected with pro-inflammatory substances, 1,8-cineole was shown to prevent pain sensation. The opioid antagonist, naloxone, did not reverse the analgesic effect of 1,8-cineole, implying that 1,8-cineole does not work through the mu-opioid receptors in the body.[19]

Using rats and mice in pain evaluation methods, 1,8-cineole was found comparable to morphine in analgesic effect on the central and peripheral nervous system. A synergistic effect was observed between cineole and morphine, with naloxone failing to antagonize the effect of cineole. EO with morphine can allow the same strength of analgesia with lower morphine dose. β-pinene had an anti-nociceptive supraspinal effect in rats; but in contrast to cineole, showed opioid antagonist activity on morphine comparable to naloxone.[20]

Rat superior cervical ganglion was used to measure the nerve excitability (via intracellular recording) when exposed to 1,8-cineole at concentrations of 0.1, 1.0, 3.0, and 6.0 mM injected intracellularly. Inhibition of excitability was seen at 1.0, 3.0, and 6.0 mM. The 6.0 mM concentration showed a significant decrease in excitability, causing a complete action potential block in all the tested neurons. The authors state that one mechanism of action is indirectly due to depolarization of the neuronal cytoplasmic membrane.[21]

Efficacy of EO against Microorganisms

Antibacterial Action

The antibacterial activity of several essential oils was evaluated against *S. aureus*. The oil activity order on growth inhibition was TTO (13 mm) > chamomile (12.5 mm) > *E. globulus* (12 mm) using 4 mm discs. Injection of the essential oils into nutrient broth inhibited cell growth. A 100-percent inhibition resulted from 100 µL of EO. Chamomile oil suppressed growth above 50 µL and TTO gave significant inhibition at only 10 µL. With the alginate-bead method, about three times the amount of EO as TTO was necessary for 100-percent inhibition. The affinity of cell binding was about double for TTO as the other oils, leading to speculation that TTO's superior antibacterial activity is due to its strong cell adhesion.[22]

An *in vitro* study tested antibacterial and anti-adhesion effects of five essential oils: manuka, tea tree, *Eucalyptus radiata*, lavender, and rosemary. On periodontopathic bacterial strains, manuka, tea tree, and eucalyptus oils showed the lowest concentration needed for bacteriostatic, bacteriocidal, and bacterial adhesion inhibition. *Porphyromonas gingivalis, Actinobacillus actinomycetemcomitans, Fusobacterium nucleatum,*

Streptococcus mutans, and *Streptococcus sobrinus* were completely killed by exposure to manuka, tea tree, or eucalyptus for 30 seconds at concentrations between 0.03-1.0 percent.[23]

An *in vitro* study examined dental biofilm production of *Streptococcus mutans*, the primary cause of dental caries, with 1-, 2-, and 4-mg/mL concentrations of *Mentha spicata* oil, *Eucalyptus camaldulensis* oil, and chlorhexidine. At 1 mg/mL, chlorhexidine showed the greatest potency. Regarding specific biofilm formation, *E. camaldulensis* versus chlorhexidine scored 6.3 (at 2 mg/mL) and 1.9 (at 4 mg/mL) versus 13.0 (at 2 mg/mL) and 4.0 (at 4 mg/mL), respectively, demonstrating a superior ability of the EO.[24]

An *in vivo*, four-week study of 100 volunteers was conducted on plaque formation, with different concentrations of *M. spicata* oil, *E. camaldulensis* oil, and chlorhexidine added to standard toothpaste. EO at all concentrations had significantly more inhibition compared to *M. spicata* and chlorhexidine.[24]

The oils from different plant parts (leaf, stem, and flower) of two Eucalyptus species (*E. sideroxylon* and *E. torquata*) were tested for antimicrobial activity against nine bacterial species. All four gram-positive bacteria (*Staphylococcus aureus*, *Staphylococcus epidermidis*, *Enterococcus faecalis*, and *Bacillus subtilis*) showed high sensitivity to EO; whereas, only two of the five gram-negative bacteria (*Klebsiella pneumonia* and *Proteus mirabilis*) showed mild susceptibility; *Escherichia coli*, *Pseudomonas aeruginosa*, and *Salmonella typhi* showed little-to-no susceptibility.[25]

The antimicrobial effect of *E. globulus* oil was studied on bacteria from human specimen samples. Included were 120 isolates of *Streptococcus pyogenes*, 20 isolates of *Streptococcus pneumoniae*, 40 isolates of *Streptococcus agalactiae*, 20 isolates of *Staphylococcus aureus*, 40 isolates of *Haemophilus influenza*, 30 isolates of *Haemophilus parainfluenzae*, 10 isolates of *Klebsiella pneumonia*, and 10 isolates of *Stenotrophomonas maltophilia*. The most sensitive bacteria were *H. influenza*, *H. parainfluenza*, and *S. maltophilia*. Some sensitivity was shown against *S. pneumoniae* and *S. agalactiae*, while there was little antibacterial effect against *S. pyogenes* or *S. aureus*. EO showed no effectiveness against *K. pneumoniae*.[26] (Note: some results disagree with this report, which may be due to the difference in species of EO.)

In a trial of 21 plant essential oils at several dilutions against six bacterial species (*E. coli*, *K. pneumoniae*, *P. aeruginosa*, *P. vulgaris*, *B. subtilis*, and *S. aureus*), 19 oils showed antibacterial activity against at least one species. *E. globulus* oil showed the least activity against the bacteria.[26]

Antibacterial Action of Vapor

An *in vitro* study examined the effect of vaporized *E. radiata* and other essential oils against six strains of bacteria (*H. influenza*, *S. pyogenes*, penicillin-susceptible *S. pneumoniae*, penicillin-resistant *S. pneumoniae*, *S. aureus*, and *E. coli*). The oils were from cinnamon bark, lemongrass, perilla, thyme, peppermint, tea tree, coriander, lavender, rosemary, eucalyptus, and citron. *E. coli* showed least susceptibility. For most of the essential oils, *H. influenza* was most susceptible, followed by *S. pneumoniae* and *S. pyogenes*, then *S. aureus*. While all oils had a degree of effectiveness, EO was one of the least. Individual oil constituents were also examined.[27]

Tuberculosis (TB)

In a striking case, a 28-year-old female (diagnosed with TB by sputum culture and chest x-ray), who had refused conventional treatment, employed *E. globulus* oil inhalation (3 mL EO to 500 mL boiling water) three times daily for three weeks. After 10 days the malaise reduced, appetite improved, cough subsided, and weight was gained. Objectively, the temperature normalized and sputum cultures were negative, although erythrocyte sedimentation rate (ESR) remained high at 110 (normal range 0-15 mm/hr for males and 0-20 mm/hr for females) and there was no change in the chest x-ray. After three weeks, the patient was given conventional treatment anyway.[28] The present authors were unable to locate any report on the *in vitro* effect of EO on *Mycobacterium tuberculosis*. (Note: In a private communication to the authors, a radiographics expert stated the chest x-ray would not be expected to change significantly in 10 days for a patient with a positive film for tuberculosis.)

Methicillin-Resistant *Staphylococcus aureus* (MRSA)

An *in vitro* comparison of *E. globulus oil* and thyme oil was conducted on 14 isolates of MRSA and other gram-positive and -negative bacteria. Although EO was effective against MRSA, it was

out-performed by thyme oil, which performed comparably to vancomycin or chloramphenicol. Both oils were out-performed by antibiotics (imipenem, ciprofloxacin, tetracycline, cotrimoxazole, gentamicin, norfloxacin, and sulfamethoxazole) against *B. cereus, E. coli,* and *K. pneumoniae*.[29]

In two human cases of MRSA following traumatic injury, Polytoxinol™ (eucalyptus, tea tree, thyme, clove, and lemongrass extracts in ethanol) was used topically for daily treatment. Both cases resulted in complete clearance of MRSA, one after five days of application, the other after three weeks. No antibiotics were used during the herbal combination therapy.[30] Polytoxinol was also used to treat chronic MRSA osteomyelitis, which became progressively worse and unresponsive to antibiotics over two years. The essential oil combination was administered for two days at the site of the infection (via Osteoset Pellets soaked with 3 mL of the herbal antiseptic) and 1 mL/day funneled to the beads in the wound cavity for an additional two days. By three months the wound healed with complete resolution of symptoms. Although the ESR was slightly elevated at 14, the C-reactive protein (CRP) decreased by almost 50 percent (to 12) and wound cultures were negative for infection. X-ray showed resolution of infective process with incorporation of the bone graft.[31]

Table 3 summarizes the sensitivity of microbes to various species of eucalyptus.

Antiviral

An *in vitro* study showed *E. globulus* oil has a mild but noticeable plaque reduction effect for mumps virus, but no effect on adenovirus.[32] The essential oils of eucalyptus (species not given), tea tree, and thyme were tested *in vitro* for antiviral activity against *Herpes simplex* virus type 1 (HSV-1); individual components of the oils were also tested. While all oils greatly reduced viral count, dilution revealed that TTO was more active than thyme, which was more active than EO. Viral replication was reduced 96 percent when the whole essential oil (TTO, thyme, or EO) was used, and all oil dilutions were well below the cytotoxic level. Although individual monoterpenes were effective, 1,8-cineole demonstrated the weakest effect. Antiviral effects were demonstrated by essential oils and monoterpenes, whether added to host cells prior to infection or after entry of HSV-1 into cells. The high anti-HSV-1 activity occurred by direct inactivation of free virus particles. All materials tested interacted in a dose-dependent manner. It was stated that monoterpene

Table 3. Microbial Sensitivities to Eucalyptus

Microbe	Sensitivity	Ref.
S. aureus	Moderate – *E. globulus* High – *E. sideroxylon, E. torquata* None – *E. globulus* Little to none – *E. globulus*	[22] [25] [26] [27]
S. epidermidis	High – *E. sideroxylon, E. torquata*	[25]
E. faecalis	High – *E. sideroxylon, E. torquata*	[25]
B. subtilis	High – *E. sideroxylon, E. torquata* Little to none – *E. globulus*	[25] [27]
Haemophilus influenza	High – *E. globulus*	[26]
H. parainfluenza	High – *E. globulus*	[26]
Stenotrophomonas maltophilia	High – *E. globulus*	[26]
Mycobacterium tuberculosis	High – improved symptoms and negative sputum culture reported; no studies found on the bacterial strain directly – *E. globulus*	[29]
Porphyromonas gingivalis	High – *E. radiata*	[23]
Actinobacillus actinomycetemcomitans	High – *E. radiata*	[23]
Fusobacterium nucleatum	High – *E. radiata*	[23]
Streptococcus mutans	High – *E. radiata*	[23]
Streptococcus sobrinus	High – *E. radiata*	[23]
C. albicans	High – *E. sideroxylon, E. torquata* High – *E. globulus*	[25] [34]
Aspergillus flavas	High – *E. sideroxylon, E. torquata*	[25]
A. niger	High – *E. sideroxylon, E. torquata*	[25]
Tinea corporis	High – *E. pauciflora*	[35]
Tinea cruris	High – *E. pauciflora*	[35]
Tinea pedis	High – *E. pauciflora*	[35]
K. pneumonia	Mild – *E. sideroxylon, E. torquata* None – *E. globulus* Little to none – *E. globulus*	[25] [26] [27]
P. mirabilis	Mild – *E. sideroxylon, E. torquata*	[25]
S. pneumoniae	Mild – *E. globulus*	[26]
S. agalactiae	Mild – *E. globulus*	[26]
Mumps virus	Mild – *E. globulus*	[26]
S. pyogenes	Little – *E. globulus*	[26]
P. vulgaris	Little to none – *E. globulus*	[27]
E. coli	Little to none – *E. sideroxylon, E. torquata* Little to none – *E. globulus*	[25] [27]
P. aeruginosa	Little to none – *E. sideroxylon, E. torquata* Little to none – *E. globulus*	[25] [27]
S. typhi	Little to none – *E. sideroxylon, E. torquata*	[25]
Adenovirus	None – *E. globulus*	[26]

Microbial susceptibility to EO *in vitro*. The exceptions are Tinea and TB, which were done *in vivo*. Note: the data from the study of gaseous EO exposure on bacteria is omitted from the table because of experimental difference. In the gaseous experiment, EO behaved comparably to TTO on *H. influenza, S. pyogenes*, and *S. pneumoniae*, but considerably less on penicillin-resistant *S. pneumoniae, S. aureus*, and *E. coli*.

hydrocarbons were slightly superior to monoterpene alcohols in antiviral activity, with α-pinene and α-terpineol demonstrating the greatest effect. The Selectivity Index (Total Cytotoxicity$_{50}$/Inhibitory Concentration$_{50}$), the best indicator of effectiveness, was 5.3 for EO, 60.0 for TTO, and 6.4 for thyme oil.[33]

Antifungal

Two Eucalyptus species (*E. sideroxylon* and *E. torquata*) and their different plant parts (leaf, stem, and flower) were tested for antifungal activity. The EO showed (in order) strong antifungal activity against *Candida albicans*, *Aspergillus flavus*, and *A. niger*, compared to a fluconazole control (with no action from oil of *E. sideroxylon* flower).[25]

E. globulus oil and 29 other essential oils (including TTO) were evaluated against biofilm-forming *C. albicans* strains, both sensitive and resistant to fluconazole. Eighteen oils showed anti-Candida activities with the strongest being EO, peppermint, ginger grass, and clove, with respective percentage inhibitions of 81-, 74-, 40-, and 29 percent. Fluconazole showed 78-percent inhibition, which was less than EO. Note that TTO was not among the highest inhibitors.[34]

An herbal preparation with the active principal one-percent oil of *E. pauciflora* was applied twice daily for three weeks to 50 patients with a skin infection by any of three species of Tinea. After two weeks, all patients were KOH-negative and after three weeks, 60 percent of patients completely recovered and 40 percent showed improvement; none of the recovered patients had relapsed at two months. A five-percent EO concentration produced no adverse skin effects during three weeks of application.[35]

Antioxidant Activity of Eucalyptus Oil

EO can react biologically as an antioxidant. The free radical scavenging capability of *E. tereticornis* oil from fresh or decaying leaves and separate oil constituents was studied against superoxide anion and hydroxyl radical. Both oils showed strong antioxidant ability either comparable or exceeding the standard antioxidants ascorbic acid and t-butylhydroxytoluene, respectively. Interestingly, the sum of the major individual constituents of the oil did not match the exceptional results of the whole oil, suggesting a synergistic effect when in combination.[4]

The oils of three eucalyptus species (*E. polyanthemos, E. globulus*, and *E. perriniana*) were assessed for antioxidant ability. *E. polyanthemos* showed an antioxidant effect comparable to α-tocopherol, as it inhibited hexanal from oxidizing to hexanoic acid for at least 30 days. At 500 µg/mL, *E. polyanthemos, E. globulus,* and *E. perriniana* inhibited hexanal oxidation by 99-, 55-, and 16 percent, respectively. In comparison, 50 µg/mL α-tocopherol inhibited hexanal oxidation by 98 percent.[5]

Additional research demonstrates the antioxidant ability of EO by the same experimental method, although the species was not given.[36] However, when *E. globulus* was compared with 10 other essential oils for free radical scavenging ability, it performed poorly, and the authors of the study attributed it to the high amount of monoterpenes. In regard to lipid oxidation, *E. globulus* demonstrated moderate inhibition (48.6%).[37]

Nasal Ciliary Beat Frequency (CBF)

EO can affect the cilia in the respiratory tract. Ciliated nasal cells from healthy volunteers were treated with EO, pine needle oil, or a combination of menthol, EO, and pine needle oil at concentrations from 0.2 to 11 g/m^3. Nasal cells experienced a dose-dependent decrease in the CBF starting with the smallest concentration of the oils tested.

For EO, at 0.2 g/m^3 the relative decrease was 14.6 percent compared to the control cells. At 7.5 g/m^3, the relative decrease in CBF was 32.5 percent. This appears to imply that, in the case of vapor inhalation of oils for respiratory health, multiple exposures at lower concentration could be more beneficial than fewer exposures at higher concentration.[38]

In another CBF study, the ciliated epithelial brushings of the inferior nasal turbinate were examined with oils of sesame, soy, peanut, thyme, lavender, eucalyptus, and menthol (all diluted with Miglyol 840, a neutral, low-viscosity carrier oil). Brushings were placed on slides and exposed to test solutions for 2, 5, 10, and 20 minutes. Most of the oils (less for thyme oil and none for Miglyol 840) produced an increase in the CBF. The beat frequency increased by 20 percent at 10 minutes with EO (0.2%) and remained elevated at 20 minutes. EO (2%) increased the ciliary beat frequency 11.8 percent at five minutes, with a continual decrease seen at 10 minutes and longer.[39]

Antispasmodic Effect

Eucalyptus tereticornis Sm. oil and 1,8-cineole were evaluated on chemically- and electrically-induced muscle contraction of guinea pig tracheal smooth muscle. EO inhibited potassium-induced smooth muscle contraction at 200-1,000 µg/mL, with 50-percent contraction inhibition at 248 µg/mL. EO (200-400 µg/mL) enhanced the contractions when induced by acetylcholine, but caused relaxation at 800-1,000 µg/mL. Cineole inhibited potassium-induced, smooth muscle contraction at 600-1,000 µg/mL with 50-percent contraction inhibition at 446 µg/mL. Cineole significantly enhanced the acetylcholine-induced contractions at all concentrations (10-1,000 µg/mL).[40]

Cineole applied to guinea pig tracheal smooth muscle resulted in a statistically significant decrease in contraction. Cineole also significantly relaxed smooth muscle when combined with ovalbumin stimulation (in previously sensitized animals), but no effect occurred with muscarinic-induced contractions, demonstrating the effect is from the sympathetic branch of the nervous system.[41]

Another study demonstrated that 1,8-cineole vapor had little to no effect on citric acid-induced cough in guinea pigs. However, the authors noted a possible dose-dependent, antitussive effect on the respiratory tract, but failed to increase the concentration to verify. It was noted that different species of EO might produce better or worse results depending on the cumulative effect of their respective constituents.[42] Other studies using α- and β-pinene show spasmolytic effects on ileum preparations.[43,44]

In Vitro Malignancy Study

Two cancer cell lines – MCF7 breast cancer cells and HEPG2 liver cancer cells – were exposed to EO. *E. torquata* oil had a notable cytotoxic effect on MCF7 cells and the oil of *E. sideroxylon* had a mild effect; neither had a substantial effect on the HEPG2 cell line.[25]

Safety of EO

The safety information on EO is based on oral and topical administration. Although no articles were located specifically assessing inhalation toxicity, there is a reference reporting toxicity when the inhalation solution was ingested by young children.[45] There is one case report of vocal cord dysfunction with EO exposure; a 46-year-old female who had daily exposure to a Eucalyptus plant experienced chest tightness, sore throat, dyspnea, cough, and wheezing. During an inhalation trial the patient was blinded to multiple smells while determining the trigger. No change was seen with exposure to water, ammonia, or pine oil. When EO was combined with ammonia, the vocal chords adducted, recreating her symptoms. The patient, who had a past medical history of allergic rhinitis and had three children with asthma, was instructed in breathing techniques that stopped the symptoms.[46]

Natural Medicines Comprehensive Database states that foods in the United States containing eucalyptus oil are safe. Ingesting 3.5 mL or more of the undiluted oil at one time has been reported to be fatal,[47] although other reports show ingestion of more than 3.5 mL to be non-fatal (see below).

Topically, the undiluted oil potentially causes neurotoxicity when used for a prolonged period of time or administered in large amounts over the body.[47] An example of toxicity was observed in a six-year-old female with full body pruritic urticaria, who had received some benefit from application of bandages soaked with a mixture of vinegar, olive oil, ethanol, and EO containing 80- to 85-percent cineole. The solution was applied over two days, culminating in a final one-time application of 50 mL of the EO in the form of the solution. The patient developed slurred speech and unsteady gait, and lost consciousness within 30 minutes. Upon arousal, the patient had nausea, vomiting, inability to walk, decreased deep tendon reflexes, hypotonia, hypotension, and proteinuria. The patient made a full recovery after six hours following removal of EO from the skin. The initial body-wide pruritic urticaria resolved within 48 hours with no further treatment and no further recurrence.[48]

The most inclusive review on EO poisoning in children (109 children, mean age two years) was conducted retrospectively over a 12-year period. Various extremes of EO poisoning were reported, with no notable effects occurring when 1.7 mL or less of pure EO was ingested. Minor poisoning occurred with doses around 2.0 mL, including ataxia, vomiting, abdominal pain, and/or miosis. Moderate poisoning occurred after a mean of 2.5 mL, which included a decrease in the level of consciousness or a Glasgow coma scale of 8-14. Major poisoning occurred with ingestion of at least 7.5 mL and included unconsciousness, unresponsiveness to verbal command, absent eye opening, abnormal extensor/flexor responses to noxious stimuli, and/or a Glasgow coma scale score of 3-7

for any duration without hypoventilation. Life-threatening poisoning is considered unconsciousness with hypoventilation. Caution is to be exerted with a vomiting patient, since aspiration of pure EO can cause pneumonitis.[49]

For pregnant or lactating women, small oral amounts of EO found as flavoring in food are considered safe. Use of EO by pregnant or lactating women for medicinal purposes should be avoided because of insufficient toxicity information.[47]

A product of encapsulated EO for intestinal release is available and anecdotal reports on its use for respiratory problems are highly favorable. A side effect can be constipation, which might be due to imbalance of gut flora or relaxation of smooth muscle contraction, similar to antispasmodic effects discussed above.

Inhalation Pharmacokinetics

Study of the pharmacokinetics of inhaled 1,8-cineole in humans show peak plasma concentration after 18 minutes. Elimination from blood is biphasic, with mean distribution of 6.7 minutes and an elimination half-life of 104.6 minutes.[50] No studies were found showing the inhaled pharmacokinetics of whole EO. Several studies show the pharmacokinetics of skin permeation by EO.

Inhalation Delivery Devices

Commercial Devices for Vapor Inhalation

Devices for vapor inhalation range from those that enrich the air with vapor to those that allow direct inhalation into the nose or mouth, with a range of accessories. While these devices are not expensive, there are even less expensive self-assembly devices made from commonly available parts. Two delivery methods are demonstrated for eucalyptus vapor. The first method is a heated cup with a few drops of eucalyptus oil. The second is a pocket inhaler that uses body heat to vaporize the oil.

Self-Assembly of Electrical Inhalation Device

Obtain a cup heater and a mug that is slightly wider than the heater plate, providing a spacer to slow heating (Patient Handout 1). After the heater and cup have warmed for about 10 minutes, add three drops of EO to the cup and inhale through the mouth and nose for a few minutes. The amount of vapor inhaled can be varied by the distance from the cup and the number of drops. Keep eyes closed while inhaling and avoid wearing glasses as the oil can affect plastic lenses.

Self-Constructed Pocket Inhaler

A suitable-sized piece of stockinet or loosely woven fabric can be rolled on a small wooden skewer and twisted into a two-dram glass vial (Patient Handout 2). The fabric easily accepts 70 drops of EO without saturation. Carrying the vial inhaler in a pocket close to the body furnishes heat for satisfactory vaporization. Hold the vial close to or touching the lips or nostrils during inhalation. The amount of vapor intake is regulated by the distance from the vial and the speed of breathing – the closer and slower, the more vapor. Best results seem to be obtained by alternating mouth and nasal inhalation during sessions of 3-5 minutes several times daily.

Tea Tree Oil

A discussion of the activity of eucalyptus oil would be incomplete without discussing tea tree oil as the two oils share most of the same terpenoid molecules as constituents. The major difference is the predominance of 1,8-cineole (about 45%) in EO with little or no terpinen-4-ol, compared to terpinen-4-ol of 30 percent or more in TTO and cineole of 15 percent or less.

Terpenoid molecules are structured from multiples of the five-carbon isoprene unit, and many are interconvertible under *in vitro* acid catalysis to equilibrium mixtures. Whether this occurs in animals or humans is an interesting speculation that could account for some curiosities that arise from examination of research data. There is a report of the balance of TTO constituents changing in the bottle.[51] Under phytochemical conditions particular enzymes likely dictate the balance of constituents.

The medicinal properties of TTO and several of its constituents were reviewed in 2006, listing the antimicrobial effects on 27 bacterial strains and 24 fungal strains, along with viral and protozoal data; anti-inflammatory activity is also elucidated.[52] There is evidence that antimicrobial activity of TTO may exceed EO in some cases, but little data on oral use of TTO exists. Its activity is thought due chiefly to terpinen-4-ol and α-terpineol; the former is the major component of TTO, but both are minor components of EO.

Although the 2006 review noted the therapeutic resistance associated with conventional antibiotics did not occur with TTO, two other publications conclude that TTO "habitual use" (defined as three days) at sub-lethal antibacterial doses does reduce efficacy of standard antibiotics against MRSA;

An Inexpensive Inhaler – Eucalyptus Oil Vapor for Respiratory Difficulty

Davis W. Lamson, MS, ND

Required Materials:
Eucalyptus oil
Coffee mug with base larger than heater, so doesn't touch heater
Electric cup heater

Eucalyptus oil is known to have antibacterial, antifungal, and antiviral actions. However, it does much more than that. Research shows eucalyptus oil has smooth-muscle relaxing effects, antioxidant activity, mucus thinning, immune stimulating, and anti-inflammatory actions. These actions work in unity to improve respiratory ailments ranging from bronchitis to asthma to sinusitis, etc.

The old-fashioned, but effective, method of administration was to breathe the vapor over a bowl of hot water containing a few drops of eucalyptus oil, with a towel over the head to make a tent. Be careful not to burn your face.

More modern equipment is available at your local drug store. There is a personal steam inhaler for about $40.00. The single-use pads are rather pricey and contain little eucalyptus oil. Applying a few drops of eucalyptus oil allows reuse of a pad.

A smaller dry device for about $20.00 may be more useful for children as it warms a pad containing eucalyptus and menthol to provide vapor while sleeping. Oil can be added to the pad as mentioned.

The picture above shows the least expensive electrical method

Instructions:
Obtain cup heater, mug with a base wider than the heater plate, and eucalyptus oil
Warm heater and mug for approximately 8 minutes
Add 2-4 drops of eucalyptus oil to the warmed mug
Close eyes when inhaling and do not wear glasses
Inhale through the mouth and nose for a few minutes
The amount of vapor inhaled can be varied by the distance from the mug

The authors authorize use of this handout, as long as author attribution remains.

Pocket Inhaler – Eucalyptus Oil Vapor for Respiratory Difficulty

Davis W. Lamson, MS, ND

Required Materials:

Eucalyptus oil
Screw cap glass vial, 2 dram, 17 x 60 mm (about 5/8" x 2.5")
Stockinet or loose fabric to prevent oil spillage

Eucalyptus oil is known to have antibacterial, antifungal, and antiviral actions. However, it does much more than that. Research shows eucalyptus oil has smooth-muscle relaxing effects, antioxidant activity, mucus thinning, immune stimulating, and anti-inflammatory actions. These actions work in unity to improve respiratory ailments ranging from bronchitis to asthma, to sinusitis, to COPD.

Equipment for Inhalation of Vapor

Another information page describes the equipment for vapor inhalation available in most drug stores. That page also includes a description for the use of an extremely low-cost electrical device for the vapor by assembly of a cup heater and cup.

The Pocket Inhaler and Use

A suitable-sized piece of stockinet or loosely woven fabric can be rolled on a small wooden skewer and twisted into the glass vial. In the pictured example, the fabric easily accepted 70 drops of eucalyptus oil without observable separate liquid.

Carrying the vial inhaler in a pocket close to the body furnishes the heat for satisfactory vaporization. Hold the vial close to the lips or nostrils during inhalation. The amount of vapor intake is regulated by the distance from the vial and the speed of breathing, the slower the more vapor. Best results are obtained by alternating mouth and nasal inhalation during sessions of about 3-5 minutes several times daily.

shorter application was not investigated.[53,54] This appears similar to the action of sub-lethal doses of standard antibiotics. Many other studies on the action of TTO on MRSA have been published since the TTO review; the reader is referred to PubMed citations for this information.

Clinical efficacy studies of TTO have been demonstrated topically for skin, intranasal, dental, and one case each for bacterial vaginosis and percutaneously into bone for infection.[52] There are few reports of ingested TTO, except for toxicity reports. In an anecdotal report to one of the authors, patients residing for extended periods in foreign countries believe that small oral doses of TTO acted as an anti-infective.

In two tuberculosis cases, 41- and 33-year-old females were unwell for 12 months and three weeks, respectively. Both patients had findings of elevated ESR, positive culture of antibiotic sensitive *M. tuberculosis*, and bilateral patchy consolidation on chest x-ray. The 33-year-old also had right pleural effusion. Before starting conventional TB medication, TTO was inhaled for 10 days (41-year-old) and five days (33-year-old). On the fourth and fifth day, respectively, the sputum cultures were negative for *M. tuberculosis* and physical symptoms subsided. In the 33-year-old woman, chest radiography showed the right pleural effusion had cleared. Both patients subsequently underwent conventional therapy.[55]

A 2000 study examined the susceptibility to TTO of cell wall-less bacteria, particularly the human pathogenic bacterium *Mycoplasma pneumoniae*. The minimum inhibitory concentration was determined to be 0.006% (v/v) of TTO for the wild type and 0.003% (v/v) for mutants of *M. pneumoniae* that lost the ability to adhere to host cells. The authors tentatively suggested using TTO for mouth washing and inhalation in the case of *Mycoplasma pneumonia* infection.[56]

In 2007, elucidation of the mechanism for the strong anti-inflammatory effect of inhaled TTO on the stimulated immune system was obtained in mice. It was demonstrated that the hypothalamic-pituitary-adrenal axis mediated the effect.[57]

Conclusion

Eucalyptus oil and its major component 1,8-cineole have a variety of antimicrobial, immune stimulatory, anti-inflammatory (decreasing certain inflammatory cytokines), antioxidant, and even analgesic and spasmolytic effects. Antimicrobial effects involve a range of bacteria, viruses, and fungi. However, Eucalyptus species or constituent composition determines the potency. Monocytes seem more affected than other white blood cells. Application by either the inhalation or oral route can provide benefit. There is a long history of folk usage with a good safety record; more recently, the biochemical details behind these effects have been clarified.

EO is reported useful in many circumstances, especially for purulent and non-purulent respiratory problems, including bronchitis, asthma, and COPD. Other plant oils may sometimes appear more microbiologically active; however, the combination of the safety of moderate doses of EO along with its broad-spectrum antimicrobial action (including against tuberculosis and MRSA) make it an attractive alternative to pharmaceuticals. It is unusual for an antimicrobial agent to also have anti-inflammatory and immune-stimulating properties. EO has also been shown to offset the myelotoxicity of one chemotherapy agent. Whether this is a general attribute or whether it affects chemotherapy benefit, remains to be determined.

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