



EUROPEAN MEDICINES AGENCY
SCIENCE MEDICINES HEALTH

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Committee on Herbal Medicinal Products (HMPC)

Assessment report on *Thymus vulgaris* L., *Thymus zygis* Loefl. ex. L., aetheroleum

Final

<Based on Article 10a of Directive 2001/83/EC as amended (well-established use)>

<Based on Article 16d(1), Article 16f and Article 16h of Directive 2001/83/EC as amended (traditional use)>

Herbal substance(s) (binomial scientific name of the plant, including plant part)	not applicable
Herbal preparation(s)	<i>Thymus vulgaris</i> L., <i>Thymus zygis</i> Loefl. ex. L., aetheroleum ¹
Pharmaceutical forms	In liquid dosage forms for oral use and in liquid or semi-solid dosage forms for cutaneous use and use as a bath additive
Rapporteur	Reinhard Länger
Assessor(s)	

¹ The liquid herbal substance complies with the Ph. Eur. monograph (ref. 01/2008:1374)



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1. Introduction

1.1. Description of the herbal substance(s), herbal preparation(s) or combinations thereof

- Herbal substance(s)

Not applicable.

- Herbal preparation(s)

Thyme essential oil (= thyme oil).

Definition

Essential oil obtained by steam distillation from the fresh flowering aerial parts of *Thymus vulgaris* L., *T. zygis* Loefl. ex. L. or a mixture of both species (Pharm. Eur. monograph 01/2008:1374).

T. zygis Loefl. ex. L. not in IPNI, only *T. zygis* L.

Pharm. Eur. monograph on Thymi herba: *T. zygis* L.

Appearance

Clear, yellow or very dark reddish-brown, mobile liquid with characteristic, aromatic, spicy odour, reminiscent of thymol (Ph. Eur.).

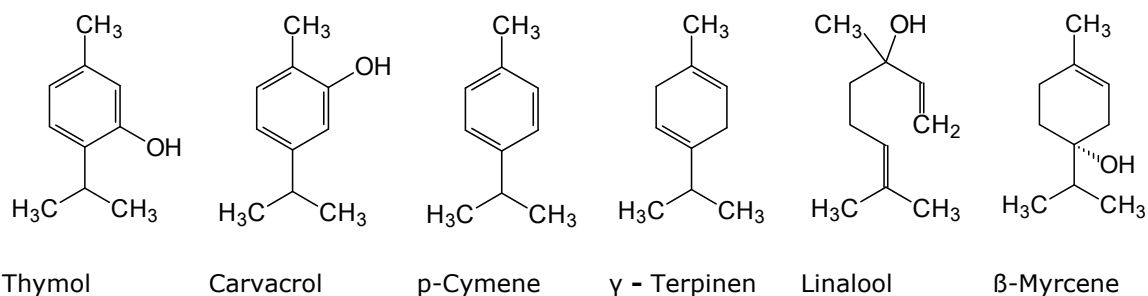
Commercially the crude thyme oil is called "red thyme oil" because of its deep colour. After redistillation "white thyme oil", a light yellow oil, which smells similarly but sweeter and less pungent is obtained (Böhme et al. 2008).

Composition:

Essential oil: there are at least 6 chemotypes of *Thymus vulgaris* (Thompson et al. 2003) with different compositions of the essential oil; only the 'thymol'-type with thymol as predominant compound complies with the definition in the European Pharmacopoeia. The dried herbal substance contains up to 2.5% essential oil; the main components are thymol, carvacrol, p-cymene, γ -terpinene, linalool, β -myrcene, terpinen-4-ol. Some compounds occur partly as glycosides (e.g. p-cymene-9-ol (Takeuchi et al. 2004, Kitajima et al. 2004, Stahl 1991)).

Composition according to Pharm. Eur.:

- β -Myrcene: 1.0 per cent to 3.0 per cent,
- γ -Terpinene: 5.0 per cent to 10.0 per cent,
- p-Cymene: 15.0 per cent to 28.0 per cent,
- Linalol: 4.0 per cent to 6.5 per cent,
- Terpinen-4-ol: 0.2 per cent to 2.5 per cent,
- Thymol: 36.0 per cent to 55.0 per cent,
- Carvacrol: 1.0 per cent to 4.0 per cent.



1.2. Information about products on the market in the Member States

Regulatory status overview

Member State	Regulatory Status				Comments (not mandatory field)
Austria	<input type="checkbox"/> MA	<input type="checkbox"/> TRAD	<input type="checkbox"/> Other TRAD	<input type="checkbox"/> Other Specify:	Combinations only
Belgium	<input type="checkbox"/> MA	<input type="checkbox"/> TRAD	<input type="checkbox"/> Other TRAD	<input type="checkbox"/> Other Specify:	Combinations only
Bulgaria	<input type="checkbox"/> MA	<input type="checkbox"/> TRAD	<input type="checkbox"/> Other TRAD	<input type="checkbox"/> Other Specify:	
Cyprus	<input type="checkbox"/> MA	<input type="checkbox"/> TRAD	<input type="checkbox"/> Other TRAD	<input type="checkbox"/> Other Specify:	
Czech Republic	<input type="checkbox"/> MA	<input type="checkbox"/> TRAD	<input type="checkbox"/> Other TRAD	<input type="checkbox"/> Other Specify:	Combinations only
Denmark	<input type="checkbox"/> MA	<input type="checkbox"/> TRAD	<input type="checkbox"/> Other TRAD	<input type="checkbox"/> Other Specify:	Combinations only
Estonia	<input type="checkbox"/> MA	<input type="checkbox"/> TRAD	<input type="checkbox"/> Other TRAD	<input type="checkbox"/> Other Specify:	Combination only
Finland	<input type="checkbox"/> MA	<input type="checkbox"/> TRAD	<input type="checkbox"/> Other TRAD	<input type="checkbox"/> Other Specify:	
France	<input type="checkbox"/> MA	<input type="checkbox"/> TRAD	<input type="checkbox"/> Other TRAD	<input type="checkbox"/> Other Specify:	
Germany	<input checked="" type="checkbox"/> MA	<input type="checkbox"/> TRAD	<input type="checkbox"/> Other TRAD	<input type="checkbox"/> Other Specify:	
Greece	<input type="checkbox"/> MA	<input type="checkbox"/> TRAD	<input type="checkbox"/> Other TRAD	<input type="checkbox"/> Other Specify:	
Hungary	<input type="checkbox"/> MA	<input type="checkbox"/> TRAD	<input type="checkbox"/> Other TRAD	<input type="checkbox"/> Other Specify:	
Iceland	<input type="checkbox"/> MA	<input type="checkbox"/> TRAD	<input type="checkbox"/> Other TRAD	<input type="checkbox"/> Other Specify:	
Ireland	<input type="checkbox"/> MA	<input type="checkbox"/> TRAD	<input type="checkbox"/> Other TRAD	<input type="checkbox"/> Other Specify:	No products
Italy	<input type="checkbox"/> MA	<input type="checkbox"/> TRAD	<input type="checkbox"/> Other TRAD	<input type="checkbox"/> Other Specify:	Combination only
Latvia	<input type="checkbox"/> MA	<input type="checkbox"/> TRAD	<input type="checkbox"/> Other TRAD	<input type="checkbox"/> Other Specify:	
Liechtenstein	<input type="checkbox"/> MA	<input type="checkbox"/> TRAD	<input type="checkbox"/> Other TRAD	<input type="checkbox"/> Other Specify:	
Lithuania	<input type="checkbox"/> MA	<input type="checkbox"/> TRAD	<input type="checkbox"/> Other TRAD	<input type="checkbox"/> Other Specify:	
Luxemburg	<input type="checkbox"/> MA	<input type="checkbox"/> TRAD	<input type="checkbox"/> Other TRAD	<input type="checkbox"/> Other Specify:	
Malta	<input type="checkbox"/> MA	<input type="checkbox"/> TRAD	<input type="checkbox"/> Other TRAD	<input type="checkbox"/> Other Specify:	
The Netherlands	<input type="checkbox"/> MA	<input type="checkbox"/> TRAD	<input type="checkbox"/> Other TRAD	<input type="checkbox"/> Other Specify:	No products
Norway	<input type="checkbox"/> MA	<input type="checkbox"/> TRAD	<input type="checkbox"/> Other TRAD	<input type="checkbox"/> Other Specify:	
Poland	<input type="checkbox"/> MA	<input type="checkbox"/> TRAD	<input type="checkbox"/> Other TRAD	<input type="checkbox"/> Other Specify:	No products
Portugal	<input type="checkbox"/> MA	<input type="checkbox"/> TRAD	<input type="checkbox"/> Other TRAD	<input type="checkbox"/> Other Specify:	
Romania	<input type="checkbox"/> MA	<input type="checkbox"/> TRAD	<input type="checkbox"/> Other TRAD	<input type="checkbox"/> Other Specify:	
Slovak Republic	<input type="checkbox"/> MA	<input type="checkbox"/> TRAD	<input type="checkbox"/> Other TRAD	<input type="checkbox"/> Other Specify:	Combination only
Slovenia	<input type="checkbox"/> MA	<input type="checkbox"/> TRAD	<input type="checkbox"/> Other TRAD	<input type="checkbox"/> Other Specify:	

Spain	<input type="checkbox"/> MA	<input type="checkbox"/> TRAD	<input type="checkbox"/> Other TRAD	<input type="checkbox"/> Other Specify:	No products
Sweden	<input type="checkbox"/> MA	<input type="checkbox"/> TRAD	<input type="checkbox"/> Other TRAD	<input type="checkbox"/> Other Specify:	No products
United Kingdom	<input type="checkbox"/> MA	<input type="checkbox"/> TRAD	<input type="checkbox"/> Other TRAD	<input type="checkbox"/> Other Specify:	

MA: Marketing Authorisation

TRAD: Traditional Use Registration

Other TRAD: Other national Traditional systems of registration

Other: If known, it should be specified or otherwise add 'Not Known'

This regulatory overview is not legally binding and does not necessarily reflect the legal status of the products in the MSs concerned.

2. Historical data on medicinal use

2.1. Information on period of medicinal use in the Community

Evidence of the period of traditional use:

The medicinal use of thyme oil is documented at least since 1589 (Dispensatorium Noricum, cited in Gildemeister et al. 1961). The essential oil of *Thymus vulgaris* is published in pharmacopoeias and standard text books of phytotherapy since many decades (e.g. British Pharmaceutical Compendium 1949, Tschirch 1917, Stahl 1962, Martindale 1972).

In Germany products containing thyme oil as the only active ingredient are on the market at least since 1976. Therefore, for thyme essential oil, a period of at least 30 years in medical use, as required by Directive 2004/24 EC for qualification as a traditional herbal medicinal product, is fulfilled.

The medicinal use of thyme oil in the specified indications is a European tradition.

Evidence regarding the indication in traditional use:

Oral use:

Catarrh of the upper respiratory tract, bronchial catarrh, symptoms of bronchitis, cough with spasms	Martindale 1972, Böhme et al. 2008, Leung 1980	Posology: Oral use: 4 - 5 drops (for example on a piece of sugar or in honey) 3-5 x daily (Hager CD-ROM 2008). Czech Pharmacopoeia (Český lékopis, 2007): single dose for oral use - 0.1 g
Anthelmintic	Tschirch 1917, Stahl 1962	no posology available

Oromucosal use:

antiseptic mouthwash, tooth pastes	Gildemeister et al. 1961	no data on the strength available; a concentration of 0.005% thymol is proposed (equivalent to 0.009 - 0.014% essential oil) Haffner et al. (1984)
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Cutaneous use:

Pruritus associated with dermatoses	German commission B (thyme oil as bath additive) Leung 1980	for bath additives see below
Bruises, sprains	Leung 1980	no posology available
Supportive treatment of acute and chronic diseases of the airways	German commission B (thyme oil as bath additive) Leung 1980	for bath additives see below; in unctions in a strength of 10%
Rubefacient, counter-irritation	Martindale 1972, Stahl 1962	no posology available

Inhalation:

Supportive treatment of acute and chronic diseases of the airways	German commission B (thyme oil as bath additive)	see below
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Madaus 1938: only reference to the use of thymol, not of the essential oil.

Stahl-Biskup E et al. in Wichtl (2009): only reference to thyme herb or thymol, not to thyme oil.

Authorized products containing thyme essential oil as the only active ingredient with evidence of tradition:

Germany:

Several bath additives are on the market at least since 1976.

Concentration of thyme oil in bath additives: 5 – 15%.

Posology of thyme oil:

- Adolescents, adults: 0.7 – 2.5 g / 100 l water (= 0.007% - 0.025%)
- Children 6-12 years: 0.35 – 1.68 g / 100 l water (= 0.0035% - 0.0168%)
- Children 2-6 years: 0.175 – 0.82 g / 100 l water (= 0.00175% - 0.0082%)
- Children 6 months – 2 years: 0.7 g / 100 l (= 0.007%)

Once daily or 3-4 times weekly

Water temperature: 35-38°C

Duration of a single bath: 10-20 minutes

Indication: for the relief of symptoms in coughs associated with cold with viscous mucilage.

Plausibility of the effects and critical discussion of traditional indications:

- Cough and cold, oral use:

The expectorant activity of thyme herb is attributed to the content of essential oil. The efficacy of thyme oil is plausible when administered orally.

- Cough and cold, cutaneous use, bath additive:

It is commonly accepted in literature (e.g. Hänsel & Sticher 2007) that essential oils are absorbed through the skin because of their lipophilic nature. After cutaneous administration and during a bath containing thyme oil the constituents evaporate from the vehicle. Thyme oil will be inhaled and absorbed via the lung additionally. It is therefore plausible that thyme oil applied to the skin in semi-solid dosage forms or used as a bath additive will act in coughs and colds similar to the oral

application. Expectorants may be interpreted for oral use by the layman. Therefore a different wording for the indication for cutaneous use and use as a bath additive is proposed.

- Antiseptic gargle:

The use as an antiseptic gargle or mouthwash is plausible due to the antibiotic activity of the essential oil. However, there is very limited evidence of an actual traditional use in this indication. Clinical experience (see chapter 4.2) is based on products containing synthetic antiseptic agents too. Therefore this indication will not be proposed.

- Bruises, sprains:

Cutaneous application results in an increased blood flow in the skin, an effect which is often used traditionally for the supportive treatment of bruises and sprains. However, for thyme oil there was only a very recent reference citing this indication. There is no evidence for a traditional use in this indication.

- Pruritus:

The indication 'pruritus associated with dermatoses' is mentioned in the respective monograph of the German commission B. However, this indication does not seem to be plausible according to the data from pharmacological experiments. Therefore this indication will not be proposed for traditional herbal medicinal products.

- Anthelmintic:

The use as anthelmintic agent is obsolete.

2.2. Specified strength/posology/route of administration/duration of use for relevant preparations and indications

Proposals for indications for traditional use:

Different indications are proposed depending on the route of administration:

Oral use:

Traditional herbal medicinal product used as an expectorant in cough associated with cold.

Cutaneous use, use as bath additive:

Traditional herbal medicinal product for the relief of symptoms in coughs and colds.

Further indications found in literature are not supported by traditional use or by a traditional posology.

Posology:

Oral use: 4 - 5 drops (for example on a piece of sugar or in honey) 3 - 5 x daily (Hager CD-ROM 2008). 1 drop = app. 0.05 ml = app. 0.045 g.

Dosage recommended in the last version of the Czech Pharmacopoeia (Český lékopis 2007): single dose for oral use - 0.1 g (= app. 2-3 drops).

At least 0.004 g essential oil per litre for preparation of a full bath (= app. 1 drop per litre = 120 drops per full bath = app. 0.5 g) (German Commission B).

In ointments in concentrations up to 10% (Hager CD-ROM 2008).

Use in children and adolescents:

The only available data come from the authorized products in Germany (see above) and from confidential data provided from the German Authorities. See chapter 5.6.

Duration of use:

Duration of a bath: 10-20 minutes.

Although thyme oil is considered as safe the duration of use should be limited to 1 week, otherwise a doctor or a qualified health care practitioner should be consulted.

3. Non-Clinical Data

3.1. Overview of available pharmacological data regarding the herbal substance(s), herbal preparation(s) and relevant constituents thereof

Antibacterial activity

The essential oil exerts a strong antibacterial activity on Gram-positive and Gram-negative bacteria. The activity is mainly attributed to thymol and carvacrol (numerous articles published, e.g., Simeon de Buochberg et al. 1976, Janssen et al. 1986, Menghini et al. 1987, Patakova et al. 1974, Allegrini et al. 1972, Janssen 1989, Farag et al. 1986, Lens-Lisbonne et al. 1987, Vampa et al. 1988, Chalchat et al. 1991, Dorman et al. 2000, Hersch-Martinez et al. 2005).

Oils with higher percentage of phenolic compounds show higher inhibitory activity (Penalver et al. 2005). Correlations between concentrations of thymol and MIC and minimal bactericidal concentration suggest that the formation of membrane perforations is the principal mode of action of thymol against oral bacteria (Shapiro et al. 1995).

Thyme essential oil had the lowest minimum inhibitory concentration (0.03% V/V) against *Escherichia coli* and *Candida* among 20 essential oils tested (Hammer et al. 1999).

The antibacterial activity of 14 essential oils and their major components was evaluated by agar-plate dilution assay under sealed conditions, with agar used as a stabilizer for homogeneous dispersion by Inouye et al. (2001). Of the selected strains of four major bacteria causing respiratory tract infection, *Haemophilus influenzae* was most susceptible to the essential oils, followed by *Streptococcus pneumoniae* and *Streptococcus pyogenes*. *Staphylococcus aureus* was less susceptible. No cross-resistance was observed between penicillin-sensitive and penicillin-resistant *S. pneumoniae*. *Escherichia coli*, used as a control bacterium, showed the lowest susceptibility. Essential oils containing aldehyde or phenol as a major component showed the highest antibacterial activity, followed by the essential oils containing terpene alcohols.

Antifungal activity

The essential oil is highly antifungal, when tested on fungi and yeasts, e.g. *Candida albicans*. This activity is mainly attributed to phenol compounds thymol and carvacrol. Thymol interferes with the formation and viability of hyphae and induces morphological alterations in the envelope of *Candida albicans* (Braga et al. 2007, 2007a).

Thyme oil inhibits the mycelial growth of *Aspergillus flavus* and *A. niger* (Paster et al. 1990) and at concentrations of ≤ 500 ppm completely inhibits dose-dependently fungal growth and mycotoxin production of *Aspergillus flavus*, *A. parasiticus*, *A. ochraceus* and *Fusarium moniliforme* (Soliman & Badeaa 2002).

The therapeutic efficacy of a 1% solution of thyme oil and thymol against *Trichophyton mentagrophytes*, *T. rubrum* and *T. tonsurans* was examined on 2-months old Wistar rats. During the 37-day observation period the oil - treated rats were cured (Sokovic et al. 2008). In dilution assays thyme oil showed much higher antifungal potency than the commercial fungicide bifonazole (Sokovic et al. 2009).

Thyme oil was antagonistic by vapour contact against an experimental tinea pedis in guinea pigs infected with *Trichophyton mentagrophytes*. Thyme oil killed the conidia, inhibited germination and hyphal elongation at concentrations of 1-4 µg/mL air (Inouye et al. 2001a).

Inouye et al. (2007) investigated in vitro the ability to treat tinea pedis with a combination of essential oils, heat and salt in a foot bath.

Agar blocks implanted with *Trichophyton mentagrophytes* were immersed in 0.1% aqueous agar containing two-fold dilutions of essential oils with or without sodium chloride at 27°C, 37°C and 42°C for 10 and 20 min. The fungicidal activity of essential oils was markedly enhanced by treating at 42°C for 20 min as compared with that at 27°C, showing 1/4 - 1/32-fold reduction of minimum fungicidal concentration (MFC to kill 99.99%). Thyme essential oil rich in thymol showed a conspicuous activity. MFCs were further reduced to 1/2 - 1/8 by the addition of 10% sodium chloride.

The in vitro activity of some essential oils (EO) (thyme red oil, fennel, clove, pine, sage, lemon balm and lavender) against clinical and environmental fungal strains was investigated. The minimal inhibitory concentrations were determined by a microdilution method in RPMI 1640 and by a vapour contact assay. The inhibiting effects of EO in vapour phase were generally higher than those in liquid state. According to both methods thyme red oil and clove were found to be the oils with the widest spectrum of activity against all fungi tested (Tullio et al. 2007).

Spasmolytic activity

Thyme oil shows a spasmolytic effect on the smooth muscle and a contracture involving a direct action on skeletal muscle by an unknown mechanism. The essential oils were diluted in methanol to give a final bath concentration of 5×10^{-5} and 2×10^{-4} g/ml for rat diaphragm in vitro with muscle stimulated directly or via phrenic nerve and 4×10^{-6} - 8×10^{-5} g/ml for field-stimulated guinea-pig ileum studies (Lis-Balchin et al. 1997).

Thymol has in vitro an agonistic effect on $\alpha 1$ -, $\alpha 2$ - and β -adrenoreceptors; the spasmolytic activity is detectable in concentrations $> 10^{-6}$ M. In a concentration of 10^{-4} M, thymol suppresses the spontaneous contractile activity of the non striated muscles of the stomach of the guinea-pig. In higher concentrations thymol exhibits a spasmolytic activity in the ratio of 1:10 compared to papaverine (Beer et al. 2007).

Thyme oil inhibits the phasic contractions of the ileal myenteric plexus-longitudinal muscle preparation of the guinea-pig. The ED₅₀ was found for thyme oil at 6.9 mg/L compared to papaverine which is 5 times more effective and isoprenaline (ED₅₀ 0.0044). On the tracheal guinea-pig preparation papaverine was 700-times more effective than thyme oil (Brandt 1988, Reiter & Brandt 1985).

Secretomotoric activity

Only historic reports are available from experiments on the secretomotoric activity.

Gordonoff (1931, 1932, and 1933) and Vollmer (1932) demonstrated secretomotoric and secretolytic properties of thymol and thyme preparations.

A stimulation of the ciliary movement in the pharynx mucosa of frogs treated with diluted solutions of thyme oil, thymol or carvacrol has been reported by Freytag (1933, cited in Hager CD-ROM 2008).

Antioxidant activity

Antioxidative effects of thyme oil have been determined in various test systems in vitro (e.g., Youdim et al. 1999a, Dorman et al. 2000, Kulisic et al. 2005, 2005a). Essential oils with high proportions of the phenolic components thymol and/or carvacrol showed the highest antioxidant activities (Jukic et al. 2005, Chizzola et al. 2008). The antioxidant activity of p-cymene-2.3-diol, a minor component of the essential oil, is considered as more potent than thymol or carvacrol which could be due to its dihydroxy structure (Schwarz et al. 1996).

Thyme essential oil exhibited a dose-dependent protective effect on the copper-induced LDL oxidation. The protective effect of essential oils is assigned to the presence of phenolic monoterpenes, thymol and carvacrol, which are identified as the dominant compounds (Kulisic et al. 2007).

Youdim et al. (1999, 1999a, 2000, 2002) investigated the influence of thyme oil and of thymol on the phospholipid polyunsaturated fatty acid composition, antioxidant enzyme activity and the phospholipid fatty acid composition in several rat tissues. The rats were fed with a diet containing thyme oil or thymol in an amount of 42.5 mg/kg BW/day. Beneficial effects could be found in different experimental settings. Thymol alone was not more effective compared to the entire essential oil.

Anti-inflammatory activity

Thyme oil inhibits prostaglandin biosynthesis, thymol was less active in the COX-inhibition test (Wagner et al. 1986).

Thymol was shown to inhibit dose-dependently the experimentally induced release of neutrophil elastase. The authors concluded that thymol may have a helpful effect in the control of inflammatory processes present in many infections (Braga et al. 2006).

Further activities

Effects on the alimentary canal

In the stomach thymol (<0.5 mM) suppressed the generation of action potential and slow potential changes without any marked change in membrane potential and membrane resistance. Increased concentrations of thymol (>0.5 mM) reduced the membrane potential and membrane resistance. In the ileum and rectum, thymol suppressed spike activity without any marked change in the membrane potential. Although the membrane was completely depolarized, thymol (>1 mM) suppressed the generation of phasic and tonic responses of the K-induced contracture evoked in the various regions of the alimentary canal. Thymol (0.5 mM) suppressed spontaneous mechanical responses in the various regions of the alimentary canal (Ito et al. 1974).

ACE-inhibition

Jukic et al. (2007) examined in vitro the inhibitory activity exerted by the main constituents of essential oil obtained from *Thymus vulgaris* on acetylcholinesterase (AChE). The total essential oil and selected compounds, specifically linalool and thymol, carvacrol and their derivatives thymoquinone and thymohydroquinone, were tested for AChE inhibition. Thymohydroquinone exhibited the strongest AChE inhibitory effect over the range of concentrations. The AChE inhibitory potential decreased in the following order: thymohydroquinone > carvacrol > thymoquinone > essential oil > thymol > linalool.

Wound healing

After topical treatment of burned rats with thyme oil (1:1 diluted with olive oil) an increase in the formation of new tissue could be observed (Dursun et al. 2003).

Bone Metabolism

Thyme oil and thymol have been demonstrated to be efficient inhibitors of bone absorption in rats. Thymol is a direct inhibitor in the osteoclast absorption pit assay (Mühlbauer et al. 2003).

Cardiovascular system

Thymol in concentrations of 1-10 mg/kg BW inhibited calcium channels in rats and lowered blood pressure (Aftab et al. 1995).

Magyar et al. (2002) achieved a similar inhibition of calcium and potassium channels in canine and human ventricular cardiomyocytes.

Szentandrassy et al. (2004) concluded from experiments on the Langendorff-perfused guinea pig heart that the negative inotropic action of thymol can be explained by reduction in calcium content of the sarcoplasmic reticulum due to the combination of the thymol-induced calcium release and inhibition of the calcium pump. The calcium-sensitizer effect, observed at lower thymol concentrations, indicates that thymol is likely to interact with the contractile machinery also.

Skeletal muscles

Thymol suppresses both Ca(2+) and K(+) currents in enzymatically isolated rat skeletal muscle fibers in a concentration-dependent manner (Szentandrassy et al. 2003).

Thymol and carvacrol were able to evoke Ca(2+) release with EC(50) values of 158 +/- 16 and 211 +/- 55 µM respectively in heavy sarcoplasmic reticulum vesicles isolated from skeletal muscle and actively loaded with calcium (Sarkozi et al. 2007).

Effects on the CNS

Lim et al. (2005) investigated the stimulating or sedative effects of inhaling thyme essential oil by using the forced swimming test (FST) with mice. The inhalation of thyme oil (p<0.05) resulted in 22.87% reduction of immobility. The same results were achieved when over-agitation was artificially induced in the mice by an intraperitoneal injection of caffeine.

Mohammadi et al. (2001) investigated several phenol derivatives with regard to their ability to activate directly the gamma-aminobutyric acid (GABA(A)) receptors in the absence of the natural agonist. This mechanism is supposed to contribute to its sedative-hypnotic actions. Only compounds with the phenolic hydroxyl attached directly to the benzene ring and with aliphatic substituents in ortho position to the phenolic hydroxyl activated chloride currents in the absence of GABA. The concentrations required for half-maximum effect were 200 µM for thymol, and 23 µM for the positive control propofol.

Insecticidal actions

Thyme oil is lethal against adult *Oryzaephilus surinamensis*, *Rhyzopertha dominica* and *Sitophilus oryzae* (Shaaya et al. 1991).

Good insecticidal activity (>90%) against larvae of *Lycoriella ingenua* was achieved with thyme oil at 30 x 10⁻³ mg/l air. Carvacrol and thymol were effective at 10 x 10⁻³ mg/l (Park et al. 2008). The LD50 against *Tyrophagus putrescentiae*, a stored food mite, is 10.2 µg/cm² in an impregnated fabric disk assay (Jeong et al. 2008).

Mosquito control

Thymol and carvacrol are potent repellents in concentrations of about 0.05% in topical treatment (Choi et al. 2002, Park et al. 2005).

Antihelmintic actions

Thyme oil in solutions of 1:2000 caused the death of ascarides in vitro. Non-phenolic constituents demonstrated less activity (Akacic & Petricic 1956).

Antiparasitic

Thyme oil is effective against *Trypanosoma cruzi*. Thymol may be the main component responsible for the trypanocidal activity (Santoro et al. 2007).

In an in vitro growth inhibition assay with bloodstream forms of *Trypanosoma brucei* the ED50 of thyme essential oil was found to be 0.4 µg/ml (Mikus et al. 2000).

Antiviral actions

The IC50 for thyme oil against herpes simplex virus type 2 was determined at 0.007% when the essential oil was added at different stages during the viral infection cycle (Koch et al. 2008).

3.1.1. Assessor's overall conclusions on pharmacology

Numerous pharmacological activities are reported for thyme oil or for the isolated compound thymol. Most experiments refer to the antimicrobial and to the antioxidant activity. These effects are primarily used in food industry, where thyme oil is an effective agent against spoiling. The spasmolytic activity and the few data on the secretomotoric activity make the use as expectorant in cough associated with cold plausible.

3.2. Overview of available pharmacokinetic data regarding the herbal substance(s), herbal preparation(s) and relevant constituents thereof

No non-clinical data on pharmacokinetics are available.

3.2.1. Assessor's overall conclusions on pharmacokinetics

No non-clinical data on pharmacokinetics are available.

3.3. Overview of available toxicological data regarding the herbal substance(s)/herbal preparation(s) and constituents thereof

Acute toxicity

The LD50 of the essential oil p.o. in rats was 2.84 g/kg body weight (Von Skramlik 1959).

The intraperitoneal LD50 of thymus zygis oil in mice was 600 mg/kg body weight (Jimenez et al. 1993).

During antimicrobial activity assessment of high doses of thyme oil (0.05% compared to 0.01% and 0.06 mM diluted in ethanol) and thymol (24% content in thyme oil) simultaneously strong cytotoxic effect on Caco-2 cells was stated (Fabian et al. 2006). The extent of damage in the cell population caused by enteroinvasive *E. coli* was also widened. In accordance with this, the presence of the evaluated substances at high concentrations in the digestive system could cause injury to intestinal cells.

Subchronic toxicity:

In commonly used doses (up to 20 drops per day; as culinary herb) no acute or chronic toxicity is reported for thyme oil (Mills & Bone 2000).

Chronic toxicity

No toxic effects were observed in rats after the addition of 1.0% of thymol to their diet for 19 weeks (Hagan et al. 1967).

Mutagenicity and genotoxicity

Essential oil:

Thyme essential oil had no mutagenic or DNA-damaging activity in either the Ames test (strains TA1535, TA1537, TA98, TA100, with and without metabolic activation) or *Bacillus subtilis* rec-Assay (Zani et al. 1991). Although the Ames test lacked one of the currently used strains the negative result could be regarded reliable, because the paper contains adequate description of findings and because the Ames test is supplemented by the rec-assay.

Thymol and other constituents:

Thymol did not show mutagenicity in *Salmonella typhimurium* strains TA97, TA98 and TA100, with and without S9 metabolic activation and 20 minutes standard preincubation time (Azizan et al. 1995).

Stammati et al. (1999) determined relative cytotoxicities of thymol and carvacrol and assessed their potential genotoxicity in short-term assays. Both substances inhibited the colony-forming ability of Hep-2 cells in dose-dependent manner. The results of an AMES-test in strains TA100 and TA98 were ambiguous. Both substances were marginally more toxic to repair-deficient strain than to its repair-proficient counterpart. The substances produced elevated revertant numbers in the strain TA100, but not to a level generally considered significant. Effects in the SOS chromotest are interpreted as signs of toxicity rather than real SOS induction. The authors conclude that the genotoxic potential of thymol and carvacrol is very weak.

Concentrations of thymol and γ -terpinene above 0.1 mM significantly induced DNA damage in human lymphocytes, however, below these concentration thymol and carvacrol significantly reduced the oxidative DNA damage induced by H₂O₂ (Aydin et al. 2005) or imidazolquinoline and mitomycin C (Aydin et al. 2005a).

Thymol and carvacrol reduced the level of DNA-lesions caused by H₂O₂ in HepG2 and colonic Caco-2 cells (Horvathova et al. 2006).

Thymol in concentrations up to 520 μ M did not increase the frequencies of chromosome aberrations in Syrian hamster embryo cells compared to the control cells (Hikiba et al. 2005).

Azirak & Rencuzogullari (2008) investigated the in vivo genotoxic effects of carvacrol and thymol in bone marrow cells of rats. Both carvacrol (10, 30, 50, and 70 mg/kg b.w. intraperitoneally) and thymol (40, 60, 80, and 100 mg/kg b.w. intraperitoneally) significantly induced the structural and total chromosome abnormalities (CA) in bone marrow cells for all treatment periods (6, 12, and 24 h) when compared with control. Both carvacrol and thymol showed similar effects with the positive control urethane on induction of the percentage of structural and total CA at the highest concentrations except the effects of carvacrol for 6 h treatment (70 mg/kg b.w. and 100 mg/kg b.w., respectively). In addition, carvacrol induced the numerical CA at all concentrations when compared to control and at two highest concentrations (50 and 70 mg/kg b.w.) when compared to solvent control. Thymol also induced the numerical CA especially at the highest concentration (100 mg/kg b.w.) for all treatment periods.

Undeger et al. (2009) examined the genotoxicity of thymol and carvacrol using comet assay. V79 Chinese hamster lung fibroblast cells were treated with 1, 5, and 25 μ M thymol and carvacrol. The results of this study indicate a lack of clastogenic activity for thymol and carvacrol at biologically relevant concentrations, and a moderate antioxidant activity in vitro.

Assessor's comment:

The significance for risk assessment of the above findings with single components (thymol and others) is somewhat difficult to interpret, because thymol seems to have both genotoxic and antigenotoxic properties, depending on concentration and test system. The doses causing significant effects in the

study of Azirak and Rencuzugullari (2008) are at least 10 times higher than the estimated maximum human dose (25 drops of an assumed size of 25 µl, resulting in 0.6 g of essential oil and calculated doses of thymol and carvacrol as 4.7 and 0.34 mg/kg bw, respectively). Furthermore, the administration in the study of Azirak and Rencuzugullari (2008) was intraperitoneal, which most probably makes the difference between effective doses in rats and human posology even larger.

Reproduction toxicity:

Thyme essential oil consisting of 48% p-cymene and 24% thymol (0.25% essential oil in the feed over 2 weeks and during 4 days of pregnancy, n=15, number of embryos: 126) showed no influence on the growth and development of mouse embryos in vivo (Domaracky et al. 2006).

3.3.1. Assessor's overall conclusions on toxicology

Potential mutagenicity and genotoxicity of thyme essential oil and its main components, especially thymol, has been assessed in a number of studies on both prokaryotic and eukaryotic in vitro and in vivo experimental systems. Studies have many weaknesses, findings are often contradictory and reporting does not always contain sufficient details, which make interpretation difficult and conclusions equivocal. Thymol is also an antioxidant and prevents DNA damage in certain experimental conditions. It seems that in some studies thyme oil and its main components give weak indications towards genotoxicity, but at the best these indications are weak and debatable. Adequately performed mammalian cell studies are needed for the resolution of potential genotoxicity.

Although there is one of the strains of *S. typhimurium* lacking in the published AMES-test the data can be regarded as sufficient for the development of a list entry for the cutaneous use of thyme oil and the use as bath additive.

For a list entry for oral use data from an AMES-test fully complying with current guidelines are considered to be necessary.

Nevertheless, thyme oil can be regarded as safe when administered in the recommended posology.

4. Clinical Data

4.1. Clinical Pharmacology

No data available.

4.1.1. Overview of pharmacodynamic data regarding the herbal substance(s)/preparation(s) including data on relevant constituents

No data available.

4.1.2. Overview of pharmacokinetic data regarding the herbal substance(s)/preparation(s) including data on relevant constituents

Thymol:

After application of a single dose of thyme dry extract (corresponding to 1.08 mg thymol) only the sulfate could be detected in the human plasma, but not the free thymol nor the glucuronide. The sulphate could be detected 20 minutes after application; maximum plasma levels were reached after about 2 hours. Thymol can be detected in the plasma up to 38 hours; renal elimination was completed within 24 hours. Elimination half-life was determined as 10.2 hours (Kohlert et al. 2002).

4.2. Clinical Efficacy²

4.2.1. Dose response studies

No data available.

4.2.2. Clinical studies (case studies and clinical trials)

Indication: Cough associated with cold:

No data available from studies using thyme oil as the only active substance. Studies including combinations with synthetic drugs are considered as not relevant for the monograph on thyme oil.

Stomatitis:

A comparison of different mouthrinses (containing thymol, chlorhexidine, povidon + H₂O₂) showed no differences in the papillary bleeding score and in plaque index between the treatment with thymol and water (Maruniak et al. 1992).

The application of a combination of thymol, menthol, methyl salicylate and 1.8-cineol over 6 months did not show statistically significant differences between the vehicle and the essential oil group. Neither development of bacterial resistance nor emergence of opportunistic pathogens could be observed (Charles et al. 2000).

Many clinical trials are published which investigate the efficacy of combinations of chlorhexidine and thymol (e.g., Twetman et al. 1999). The contribution of thymol to the overall efficacy cannot be estimated.

4.2.3. Clinical studies in special populations (e.g. elderly and children)

No data available.

4.3. Overall conclusions on clinical pharmacology and efficacy

There are no clinical data available which would support the well-established use of thyme oil.

5. Clinical Safety/Pharmacovigilance

5.1. Overview of toxicological/safety data from clinical trials in humans

Case reports:

Germany has received 17 case reports for allergic reactions (urticaria, skin rashes, bronchospasm, asthma attack, anaphylactic shock) concerning the use of thyme herb.

Austria: In the pharmacovigilance database of the Austrian medicines agency AGES PharmMed no adverse events concerning *Thymus vulgaris* are reported (date 03.02.2009).

5.2. Patient exposure

No data available.

5.3. Adverse events

Adverse events related to the essential oil

² In case of traditional use the long-standing use and experience should be assessed.

In very rare cases allergic reactions may occur due to the content of thymol (Hänsel 1994).

Cross-sensitivity between plants belonging to the *Lamiaceae* family has been reported for the dust or extracts (Benito et al. 1996). No such data are available for the essential oil.

In concentrations higher than 8% in Vaseline irritation of the skin may occur. The daily application in gargles, mouthwashes and toothpastes over a longer period (no exact data available) may cause allergic reactions (Hager CD-ROM 2008).

Adverse events related to thymol

Thymol has been used orally in folk medicine as a vermifuge at therapeutic doses (0.3 – 0.6 g, max. 1 g). Thymol in these concentrations caused abdominal pain and transient collapse (Czygan et al. 2004).

Assessor's comment:

The symptoms described above may also be due to the worm infections. The doses of thymol correspond to 62 – 208 g herbal substance. These doses exceed the recommended daily doses by far. With the recommended amounts of thyme preparations only approximately 38 mg of thymol are administered. The proposed dosage of the pure essential oil (25 drops per day) corresponds to approximately 300 mg thymol. However, no adverse reactions from the oral use of the essential oil are published.

Thymol has caused dermatitis in dentists and, when used in toothpaste, cheilitis and glossitis (Hager CD-ROM 2008).

5.4. Serious adverse events and deaths

Case reports:

Germany has received 17 case reports for allergic reactions including serious adverse events (urticaria, skin rashes, bronchospasm, asthma attack, anaphylactic shock) concerning the use of thyme herb.

5.5. Laboratory findings

No data available.

5.6. Safety in special populations and situations

5.7. Intrinsic (including elderly and children) /extrinsic factors

The German National Authority provided confidential data on the use of thyme oil as bath additive in children. In an open multicenter post marketing surveillance study 491 children at an age from 1 month to 9 years suffering from acute or chronic infections of the upper airways were treated with baths containing app. 12 mg thyme oil per litre. Although the study design does not allow conclusions regarding the efficacy, the safety data might support the safe use of thyme oil in the paediatric population.

Patients: 61 patients <= 6 months of age; 130 patients 6 - 12 months; 175 patients 12 - 24 months, 69 patients 24 - 36 months; 27 patients 36-48 months; 13 patients 48 - 60 months; 15 patients older, 1 child no age given.

Adverse events: In 7 cases (1.4%) a causal relationship between AE (skin irritation, mostly exanthemas) and the study medication was rated as very likely. Unfortunately no data are available about the age of the concerned patients.

Skin irritation has to be rated as a common adverse event in the age group treated in this study. 92% of the patients in this study were below 4 years of age. Some products authorized in Germany allow the use in children up from 6 months of age.

Therefore the restriction to a minimum age of 3 years seems to be justified for the use of thyme oil as bath additive.

For the oral use and for the cutaneous use no safety data from the paediatric population are available. Additionally no data on the posology are published for children and adolescents. Therefore the oral and cutaneous uses should be limited to adults and elderly.

Like other essential oils thyme oil should not be applied to the face particularly in the nasal area of babies and infants under the age of two years because of the risk of a laryngospasm.

Full baths are contraindicated independent of the active substance in cases of large skin injuries, acute skin diseases, high fever, severe infections, severe circulatory disturbances and cardiac insufficiency.

5.8. Drug interactions

Topically applied thymol significantly enhanced the percutaneous absorption of 5-fluorouracil through porcine epidermis in comparison with control (Gao et al. 1997).

Thyme oil potentiates the antifungal action of amphotericin B. The strongest decrease (48%) of the MIC 80% was obtained with medium containing 0.2 µL/ mL of essential oil (Giordani et al. 2004).

Thyme oil increases the transdermal delivery of nitrendipine in two different skin models (Mittal et al. 2008).

5.9. Use in pregnancy and lactation

Thyme essential oil consisting of 48% p-cymene and 24% thymol (0.25% essential oil in the feed over 2 weeks and during 4 days of pregnancy, n=15, number of embryos: 126) showed no influence on the growth and development of mouse embryos in vivo (Domaracky et al. 2006).

In the absence of sufficient data the use during pregnancy and lactation is not recommended.

5.10. Overdose

Single reports of hyperthyroidism are mentioned after long term use of overdoses of thymol. No cases are reported for thyme oil.

5.11. Drug abuse

Not relevant.

5.12. Withdrawal and rebound

Not relevant.

5.13. Effects on ability to drive or operate machinery or impairment of mental ability

No studies on the effect on the ability to drive and use machines have been performed.

5.14. Overall conclusions on clinical safety

Thyme oil can be considered as safe when administered in the specified posology and the specified route of administrations.

6. Overall conclusions

Thyme oil is similarly used to thyme. Many beneficial effects have been attributed to thyme and its essential oil throughout history but just a few have scientific evidence. There is no specified clinical data available to support well-established use.

During the use of thyme oil spasmolytic, expectorant, antimicrobial, antioxidant, invigorating, appetizing, eupeptic and choleric properties have been experienced.

According to the traditional medicinal use the following indications are proposed:

Oral use:

Traditional herbal medicinal product used as an expectorant in cough associated with cold.

Cutaneous use, use as bath additive:

Traditional herbal medicinal product for the relief of symptoms in coughs and colds.

The lipophilic nature of the components of the essential oil makes absorption via the skin plausible. It is also plausible that absorption of volatiles occurs after inhalation of the essential oil when applied in semi-solid dosage forms, in a bath or in semi-solid dosage forms as embrocation.

Although there is one of the strains of *S. typhimurium* lacking in the published AMES-test the data can be regarded as sufficient for the development of a list entry for the cutaneous use of thyme oil and the use as bath additive.

For a list entry for oral use data from an AMES-test fully complying with current guidelines are considered to be necessary.

Nevertheless, thyme oil can be regarded as safe when administered in the recommended posology.

Many clinical trials are published which investigate the efficacy of gargles consisting of combinations of chlorhexidine and thymol. The contribution of thymol to the overall efficacy cannot be estimated. Since the clinical evidence is only poorly documented for isolated compounds of the thyme essential oil or for combinations with other essential oils and synthetic antiseptic drugs the traditional use of thyme oil as a gargle cannot be supported.

The use against pruritus associated with dermatoses is not plausible.

Annex

List of references