

# Nematicidal Activity of Essential Oils

Subjects: Plant Sciences

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Essential oils (EOs) can be a large source of new food-safe and healthy nematicidal products, due to their strong activity on crop pathogens and pests, including phytoparasitic nematodes, as well as to their low environmental persistence.

Keywords: aromatic plants ; bionematicides ; essential oils ; fumigation ; phytoparasitic nematodes ; sustainable control ; terpenes

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## 1. Introduction

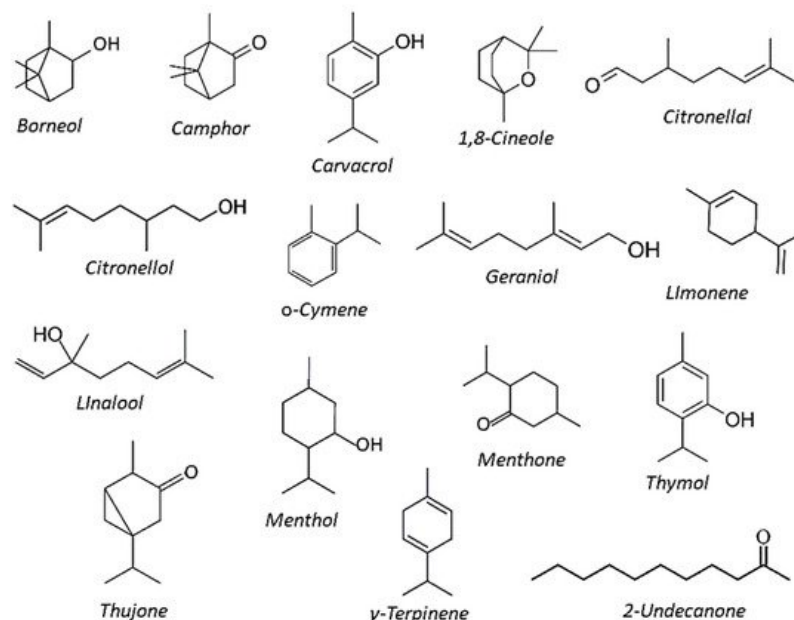
Food safety and human health preservation require severe restrictions on the use of synthetic pesticides traditionally applied for the control of crop pathogens and pests, due to their hazardous effects on soil, animals, and humans <sup>[1][2]</sup>. Phytochemicals from a large variety of plants have received an increasing interest as an alternative strategy for phytoparasitic nematode management <sup>[3][4][5][6]</sup>. Within plant-derived nematicidal compounds, a major role, as both a research topic and a source of new nematicides, has been increasingly provided to essential oils (EOs) from a wide range of aromatic and medicinal plants <sup>[7][8][9]</sup>. EOs' activity has been extensively documented on root-knot nematodes of the genus *Meloidogyne*, as the phytonematode species economically most damaging and prone to worldwide spread <sup>[10][11][12]</sup>, as well as on the pinewood nematode *Bursaphelenchus xylophilus* (Steiner and Buhrer) Nickle, due to the serious phytosanitary problems raised by this nematode in the pine forests of South Korea and Portugal <sup>[13][14][15]</sup>. Adversely, a minor attention was provided to EOs' activity on other potentially harmful phytonematodes, such as the cyst-forming (*Heterodera* spp. and *Globodera* spp.) and root-lesion (*Pratylenchus* spp.) species or the stem nematode *Ditylenchus dipsaci* (Kühn) Filipjev <sup>[16][17][18][19]</sup>. The 10-year studies of our research group were addressed to characterize the chemical features and nematicidal properties of 16 EOs from different botanical and geographic origins, as well as their structure–activity relationship. Main findings from these studies are reviewed and commented in this work in comparison with the related literature data.

## 2. Essential Oils

EOs are mixtures of volatile lipophilic constituents generally produced by the specialized metabolism of aromatic and medicinal plants from a wide range of botanical families (Lamiaceae, Myrtaceae, Rutaceae, Apiaceae, and others) as responsible for their distinctive odor, flavor or scent, though also present in non-vascular plants such as some liverworts <sup>[20]</sup>. EOs are stored in plant secretory epithelial or parenchymal cells, forming structures of various kinds such as glandular trichomes or excretory idioblasts. The ecological role of EOs is still not clearly defined, though plant secretory products have been suggested to provide adaptive benefits, including plant protection against phytopathogens and parasites.

Terpenoids are the most important group of specialized components of plant EOs and are principally represented by mono- and sesquiterpene compounds, sometimes associated with low molecular weight phenylpropanoids. Plant terpenoids are synthesized via two different metabolic pathways: the mevalonate (MVA) pathway in cytosol leads to the formation of sesquiterpenoids, while the methylerythritol phosphate (MEP) pathway occurring in chloroplasts results in the synthesis of monoterpenoids. Phenylpropanoids derive instead from the shikimate pathway mainly occurring in chloroplasts .

Plant EOs can be also characterized by phytochemical polymorphism, as individual plants from the same species can produce several chemotypes with different EOs' compositional profiles, possibly resulting from diversification of EOs biosynthetic pathways under different environmental conditions <sup>[20]</sup>. This phenomenon was clearly evidenced within the Lamiaceae plants, in which several chemical types were identified in *Origanum*, *Lippia*, *Mentha*, *Lavandula*, and *Ocimum* species, though chemotypes were detected also within the Asteraceae, such as among *Matricaria*, *Tagetes*, and *Achillea* species. A summary of some of the most common components in plant essential oils is depicted in **Figure 1** .



**Figure 1.** Chemical structures of selected EOs' components.

The GC and GC-MS analysis of these EOs highlighted largely differentiated compositional profiles, mainly consisting of high concentrations of oxygenated monoterpenes (**Figure 1**). Carvacrol was present in the EOs from *Monarda didyma*, *M. fistulosa* and *Ruta graveolens* (14, 24 and 15%, respectively), whereas its isomer thymol was among the monoterpene components of EOs from *Thymus satureioides*, *M. didyma*, and *M. fistulosa* (12%, 6%, and 8%, respectively). However, EOs from *M. didyma* and *M. fistulosa* were also characterized by relevant amounts of γ-terpinene (22% and 25%, respectively). Large amounts of 1,8-cineole (syn eucalyptol) were found in the EOs of *R. officinalis* and *Eucalyptus globulus* (47% and 92%, respectively) and in the EO of *Cinnamomum camphora* (22%). Camphor was detected in the EOs of *Artemisia herba-alba* (26%) and *Rosmarinus officinalis* (12%), whereas main constituents of *Pelargonium asperum* EO were citronellol (35%) geraniol (22%) and linalool (13%). The phenylpropanoid *E*-cinnamaldehyde (85%) prevailed in the EO of *C. verum*, while limonene was almost the unique constituent in *Citrus aurantium* and *C. sinensis* and citronellal was abundant (84%) in *E. citriodora*.

### 3. Phytoparasitic Nematodes

Plant-parasitic nematodes are among the most serious constraints to world agriculture, globally causing damages estimated at USD 80 billion per year, most of which is due to root-knot species of the genus *Meloidogyne*. Moreover, these losses are presumably underestimated due to nonspecific symptoms and difficult recognition of nematode attacks.

A worldwide distribution on a large number of host crops is also presented by root lesion nematodes of genus *Pratylenchus*, quite rightly included among the most devastating nematode pests. In particular, *P. vulnus* Allen et Jensen is a severe parasite of fruit trees widespread in commercial orchards and nurseries of the Mediterranean region and United States.

The dagger nematode *Xiphinema index* Thorne et Allen is an ectoparasite species distributed throughout the world which feeds on grapevine root tips and directly causes root swelling and gall formation with a consequent reduction of plant growth. However, the economic impact of this species is mainly related to its vehiculation of dangerous grapevine viruses, such as the grapevine fanleaf virus.

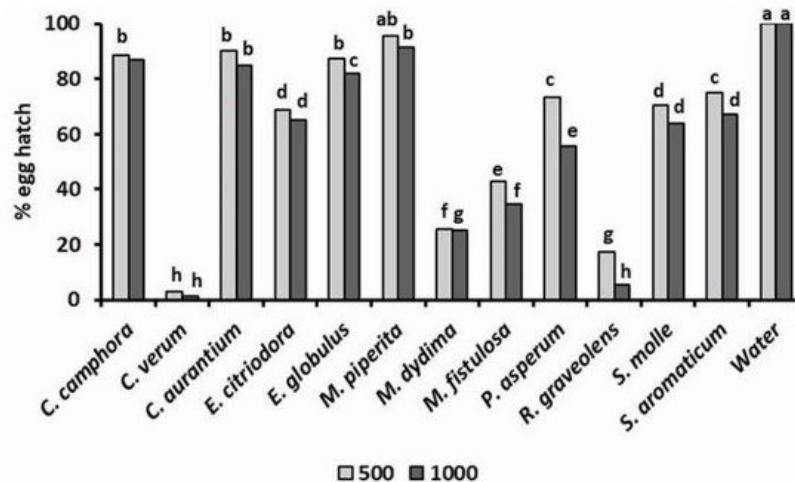
In our studies, *in vitro* toxicity bioassays were conducted on 2nd stage juveniles (J2) of *M. incognita*, mixed-age specimens of *P. vulnus* and *X. index* females, which were exposed for 4–96 h intervals to 2–100 µg mL<sup>-1</sup> concentrations of EOs or their single constituents. Egg hatchability bioassays were undertaken on *M. incognita* egg masses treated for 24–48 h with 500–1000 µg mL<sup>-1</sup> EO solutions. Finally, *in vivo* studies on tomato (*Solanum lycopersicum* L.) were carried out in soil infested by *M. incognita* (20 eggs and J2 mL<sup>-1</sup> soil) and treated with 50–200 µg kg soil<sup>-1</sup> doses of the different EOs, applied either in water solution or by fumigation.

## 4. Nematicidal Activity of Experimental EOs

The EOs from *A. herba-alba* and *R. officinalis* were highly active on *M. incognita* J2 and *X. index* females, as both resulted in an almost complete mortality after a 96 h exposure to only 2  $\mu\text{g mL}^{-1}$  solutions, but were less toxic to *P. vulnus* [1]. The lesion nematode *P. vulnus* was less sensitive than *M. incognita* and *X. index* also to the EO from *T. satureioides* and both *Monarda* EOs [1][2]. The two *Cinnamomum* EOs were differently active on *M. incognita* J2, as a similar 64% mortality occurred after a 24 h J2 exposure to 0.78 and 25  $\mu\text{g mL}^{-1}$  solutions of *C. verum* and *C. camphora*, respectively [3].

Analogously, toxicity to *M. incognita* J2 largely differed between the two Eucalyptus EOs, as only an 8 h exposure to a 12.5  $\mu\text{g mL}^{-1}$  solution of *E. citriodora* EO was enough to cause more than 90% J2 mortality, while similar rates were reached only after a 24 h immersion in a 100  $\mu\text{g mL}^{-1}$  solution of the *E. globulus* EO.

In addition to their toxicity to infective J2, the tested EOs also variously affected the hatchability (Figure 2) of root-knot nematode eggs. A 96 h exposure of *M. incognita* egg masses to a 500  $\mu\text{g mL}^{-1}$  solution of EOs of *C. verum* and *R. graveolens* reduced the percentage of egg hatch to only 1.2% and 7.0%, respectively. Analogously, egg hatchability was strongly limited by similar treatments with *M. didyma* and, at less instance, *M. fistulosa* EOs. A lower but significant egg hatch inhibition was also caused by the two *Eucalyptus* EOs, as well as by the EOs of *P. asperum*, *S. molle*, and *S. aromaticum*. Adversely, poor or nil effects on root-knot nematode egg viability were found for *C. aurantium* and *M. piperita* EOs.

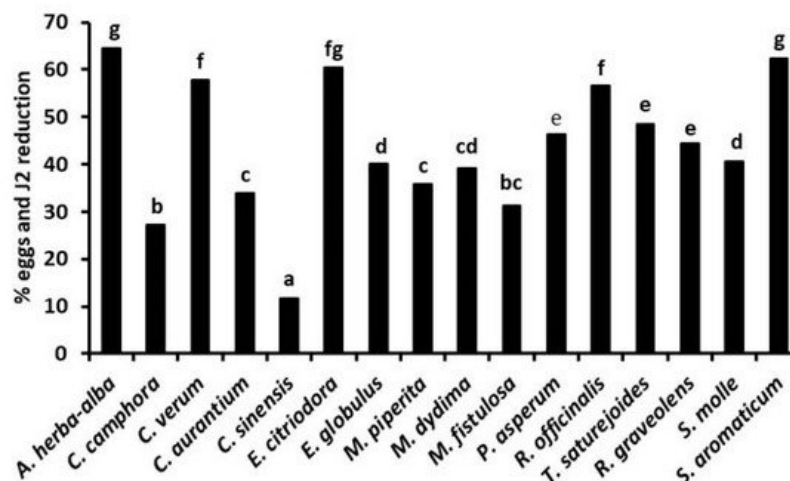


**Figure 2.** Relative hatchability of *M. incognita* eggs after a 48 h egg mass exposure to 500 or 1000  $\mu\text{g mL}^{-1}$  solutions of 12 different essential oils.

In the experiments in soil, the strongest suppression of gall formation and nematode egg density on tomato roots (Figure 3) occurred after soil treatments with the EOs from *E. citriodora*, *E. globulus*, *M. piperita*, *P. asperum*, and *R. graveolens*, applied at 50–200  $\mu\text{g kg}^{-1}$  soil doses either by fumigation or in an aqueous suspension. Nematode infestation on tomato roots was also strongly suppressed after soil fumigation with the same range of doses of *A. herba-alba*, *R. officinalis*, and *T. satureioides* EOs, as well as by soil irrigation with 50–200  $\mu\text{g kg}^{-1}$  soil doses of EOs from *S. aromaticum*, *C. verum*, and *E. citriodora* or *M. didyma* and *M. fistulosa* EOs (Figure 4). Adversely, the lowest suppressive performance was provided by the soil treatments with *C. aurantium* and *C. sinensis* EOs, in good agreement with the poor or limited in vitro activity of these two EOs.



**Figure 3.** Tomato roots from soil infested by *M. incognita*, non-treated and treated with EOs from *E. globulus* and *P. asperum* ( $100 \mu\text{g Kg}^{-1}$  soil), or nematicide fenamiphos.



**Figure 4.** Relative suppression (non-treated soil = 0) of *M. incognita* multiplication on tomato roots after soil treatment with a  $100 \mu\text{g Kg}^{-1}$  soil dose of 16 different essential oils.

No literature data were available on the nematicidal activity of *C. aurantium*, *S. molle*, and *Monarda* EOs, while only single *in vitro* assays stated a limited toxicity to root-knot nematode J2 and eggs of EOs from *C. sinensis* and *A. herba-alba* and, adversely, a strong toxicity of *T. satureioides* EO [4][5].

Previous reports on the nematoxicity of the other EOs investigated in our studies were mostly referred to *in vitro* assays on root-knot nematode species with EO samples not always chemically characterized, while toxicity to other phytonematode species was poorly or not at all documented. Most of these studies generally agreed with data from our experiments.

EOs from *Eucalyptus* species, also including *E. citriodora* and *E. globulus*, were always found to reduce J2 motility and viability and egg hatchability of different *Meloidogyne* species [6][7][8]. In agreement with our data, nematicidal properties of *Cinnamomum* EOs were described as consistently variable among the species, with a moderate suppressiveness of *C. camphora* EO to *M. incognita* on tomato and a strong *in vitro* toxicity of the EO of *C. verum* to the root-knot species *M. incognita* and *M. graminicola* Golden and Birchfield and the pinewood nematode *B. xylophilus* [9][10][11].

In accordance with the strong toxicity to *M. incognita* and *P. vulnus* observed in our studies, EOs of *R. graveolens* and *S. aromaticum* were described as highly active on J2 and eggs of *M. incognita*, *M. chitwoodi* Golden, O'Bannon, Santo and Finley, and *M. exigua* Goeldi [7][12][13], as well as on different stages of *B. xylophilus* [9][14]. Contrastingly, a greenhouse study of Meyer et al. [15] did not find any significant reduction of *M. incognita* population on various vegetable crops in soil treated with a *S. aromaticum* EO formulation.

The low activity of *M. piperita* EO in our experiments is also confirmed by literature data, which reported a poor in vitro toxicity of this EO to root-knot nematode J2 and eggs and its scarce effectiveness on the infestation of *M. arenaria* Chitwood on tomato in soil [16][6][17].

The EOs from *P. asperum* and *R. officinalis* resulted highly toxic to *M. incognita* in all our in vitro and in vivo experiments, but were generally documented as poorly active on root-knot nematode J2 and eggs by previous in vitro assays [4][6][16][18]. Contrastingly, experimental applications of *R. officinalis* EO to soil resulted in a strong reduction of the infestation of *M. incognita* on tomato [19] as well as of *M. javanica* and *P. brachyurus* (Godfrey) Filipjev and Schuurmans Stekhoven on soybean, *Glycine max* (L.) Merr. [20].

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