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USE OF ESSENTIAL OILS AS GREEN BIOPESTICIDES

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ABSTRACT

In recent years, there has been a growing interest in the application of herbal products, especially essential oils (EOs) in agriculture, as an alternative to the use of synthetic pesticides, for healthy crops and environmentally sustainable production systems. This was the main reason why the properties of EOs as natural pesticides for their promotion and use in organic farming were evaluated. EPA (Environmental Protection Agency) defines biopesticide as a safer product, with a lower persistence in the environment, biodegradable, less toxic to non-target organisms, less likely to contaminate soil and groundwater. Many EOs obtained from medicinal plants have a wide range of activities against harmful insects, bacteria and phytopathogenic fungi, weeds. Their action varies from insecticidal activities, repellents, antifeedants, growth regulation, etc. This review presents the most effective biopesticides based on EOs obtained from medicinal plants and the future development prospects of this industry.

INTRODUCTION

The use of chemicals in agriculture helps to increase crop production yields to meet the food needs of the growing population. However, their irrational application causes chronic human health problems and can destroy the environment and biodiversity *(Elahi, E., Weijun, C., et. al., 2019; Durán-Lara, E. F., et. al. 2020)*. Depending on their chemical structure, some products persist in the environment and can accumulate in animal meat, vegetables and fruits, which are then consumed by humans *(Yao, S., Zhao, Z., et. al., 2021*). In this context, numerous global initiatives have emerged, which aim to reduce the use of agrochemicals in agriculture.

In 2015, the Ministry of Agriculture and the Central Government of China launched the "Action Plan for the Zero Increase of Fertilizer Use – APZIFU", which includes actions to increase the zero use of chemical fertilizers and pesticides in the following period *(Shuqin, J., Fang, Z., 2018)*. In Europe there is a plan that aims to reduce the use of agrochemicals by about 50% by 2025 *(Lee, R., den Uyl, R., Runhaar, H. 2019)*.

Directive 2009/128/EC "Establishing framework for Community action to achieve the sustainable use of pesticides", which aims to reduce the risk and adverse impact of pesticides on human health and the environment. This involves the development of alternative agricultural techniques, including integrated pest management. Therefore, alternative or complementary plant protection practices are needed in order to reduce the use of pesticides, to obtain healthy crops, ecologically sustainable production, but also to protect the health of agricultural workers and consumers *(Sturchio, E., Donnarumma, L., Annesi T., et. al., 2014)*.

Regulation (EC) No. 1107/2009 introduced a new term: *"Basic substances (BS)"*. This Regulation provides for criteria for the approval of BS in Article 23, with specific provisions to ensure that such active substances can be used legally in the EU, insofar as they do not have an immediate or delayed adverse effect on human, animal health or on the environment. However, this regulation may not apply to the use of essential oils (EOs) as the concept behind the regulation was that various food additives could be authorized as BS without further formulation adaptations (e.g. emulsifiers or additives). Therefore, the approval process of some active substances (e.g. EOs) could be more complicated because the use of emulsifiers is required for their application. Any product that deviates from the definition of Article 23 of Regulation (EC) No. 1107/2009, including those containing an already approved BS, but also a co-formulant, will then be considered a plant protection product and will no longer be qualified as BS *(Pavela, R. 2014; Pavela, R., Benelli, G. 2016)*. These strict criteria have been established to protect human health from hazardous pollutants, but also due to the identification of high levels of food contamination with pesticide residues in EU countries*)*. In general, a natural substance proposed for pest management in agriculture must meet the following requirements: be effective against the target organism, safety and biological selectivity, composition and formulation be standardized and immediately available. Natural compounds generally have a lower persistence and toxicity than synthetic compounds, reducing the negative impact on the environment. EOs are complex mixtures composed of several chemical compounds (terpenes, alcohols, aldehydes and phenols), which together have potential herbicidal and antimicrobial properties *(Tworkoski T., 2002; Zanellato, M., Masciarelli, E., et. al., 2009)*. EOs are synthesized by 17,500 plant species mostly belonging to several families (*Lamiaceae, Asteraceae, Myrtaceae*, etc.). They have a role in the direct and indirect protection of plants against herbivores, pathogens, in reproduction processes of by attracting pollinators, seed dispersal and in adapting plants to stress conditions. The synthesis and accumulation of EOs is performed with the help of specific, secretory structures: glandular trichomes (plants from the family *Lamiaceae*), secretory cavities (plants from the family *Myrtaceae, Rutaceae*), resins, etc. Many EOs are obtained from plants whose cultivation is expensive or disadvantageous because of yields. Even the plants that are currently grown for commercial production of EOs cannot be easily grown. One reason would be that the physiological expression of the secondary metabolism of plants can vary depending on the stages of development, and the percentage of monoterpenes accumulated depends on temperature, circadian rhythm and varies depending on the phenological phase of the plant *(Hansted, L., et. al.,1994)*. Also, soil moisture, photoperiod, temperature, directly affect the secondary metabolism of the plant and the composition of an EO. Therefore, obtaining a standardized final product is a challenge, but new technologies for plant cultivation have been suggested, in order to increase production and standardize the qualitative and quantitative parameters of EOs *(Mahmoud, S.S., Croteau, R.B. 2002; Pavela, R., et.al., 2016*). Currently, EOs are extracted from plants using conventional, classical methods (standard distillation of plant material). Investments in new technologies (e.g. ultrasound, microwaves) in recent decades have led to the emergence of more efficient extraction processes, which have led to reduced extraction time and energy consumption, increased extraction efficiency, improved quality of EOs obtained *(Asbahani, A., et. al., 2015)*. These new trends in medicinal and aromatic plant research, together with the choice of suitable varieties and cultivars with high yields and better biological efficiency, will open new perspectives in the sustainable production of EOs.

The purpose of this analysis is to present studies conducted in recent years on the biological effectiveness of biopesticides obtained from EOs against phytopathogenic bacteria and fungi, insects and weeds, reported in the scientific literature, marketed biopesticides and development prospects of this industry.

MATERIAL AND METHOD

As an alternative to synthetic pesticides, the use of EOs obtained from medicinal plants has increased recently. There are several compounds identified in EOs that have demonstrated biopesticidal activity (Table 1).

Table 1

Compounds extracted from plants and used as biopesticides

Compound name	LD_{50} for rats (mg kg^{-1} body weight)	Plant (genus)	Reference
Allicin	60	Allium	FAO
Allyl sulfide	2980	Allium	FAO
Carvacrol	810	Carum, Cinnamomum, Mentha, Ocimum, Origanum, Thymus	FAO
Cinnamaldehyde	1160	Cassia, Cinnamomum, Lavendula, Pogostemon	FAO
Citronellal	5000	Citrus, Corymbia, Cymbopogon	Opdyke D.L.J.
Citral Geranial+Neral	4960	Citrus, Cymbopogon, Eucalyptus, Lavendula, Ocinum, Piper, Thymus,	Isman M.B., Machial C.M.
Eugenol	2680	Acorus, Ageratum, Cinnamomum, Citrus, Cymbopogon, Lantana, Laurus, Lavendula, Nicotiana, Ocimum, Pimpinella, Piper, Pogostemon, Syzygium	Isman M.B., Machial C.M.

Eucalyptol (1,8-Cineole)	2480	Alpina, Artemisia, Blumea, Cinnamomum, Curcuma, Eucalyptus, Eugenia, Laurus, Lavendula, Mentha, Ocinum, Piper, Rosmarinus, Salvia, Syzygium, Zingiber	FAO Isman M.B., Machial C.M.
Limonene	4600	Anethum, Apium, Carum, Chenopodium, Cinnamomum, Citrus, Coriandrum, Croton, Cuminum, Cymbopogon, Eucalyptus, Lavendula, Mentha, Myristica, Nicotiana, Ocimum, Origanum, Pimpinella, Piper, Rosmarinus, Salvia, Valeriana	FAO
Linalool	2790	Artemisia, Cinnamomum, Citrus, Coriandrum, Cymbopogon, Eucalyptus, Laurus, Lavendula, Mentha, Ocimum, Origanum, Rosmarinus, Salvia, Syzygium, Thymus, Zingiber	FAO
Menthol	3180	Mentha, Thymus	FAO
Nicotine	1	Nicotiana	FAO
Pulegone	150	Mentha, Origanum	FAO

Thymol	980	Carum, Lavendula, Ocimum, Origanum, Thymus	FAO Isman M.B., Machial C.M.
Zingiberene	5000 (Ginger oil, with 29% α-Zingiberene)	Zingiber	Koul O

*takeover from *Durán-Lara, E. F., et. al. 2020*

ANTIBACTERIAL ACTIVITY

În recent years, a greater number of studies have been published on the antibacterial properties of EOs against plant pathogens, with a growing interest in biocontrol methods. However, the number of studies reporting EOs as antibacterial agents from the perspective of the plant pathogen is still limited, while most studies on the antibacterial properties of EOs focus on the preservation and storage of food or health. A study conducted by Salamci, E., Kordali, S., *et.al*., 2007, showed a low success of EO obtained from the *Tanacetum* species, ineffective against the bacteria *Erwinia amylovora or Xanthomonas sp*. *Badawy, M.E.I.,* and *Abdelgaleil, S. A. M., 2013*, present in their paper, the results of tests obtained from the use of EOs from 18 plants of Egyptian origin. Isolated EOs were tested against phytopathogenic bacteria *Agrobacterium tumefaciens* and *Erwinia carotovora var. carotovora*. The tested oils showed a variable degree of antibacterial activity, and based on the values of the minimum inhibitory concentration (MIC), EOs were more effective against *E. carotovora var. carotovora* than against *A. tumefaciens*. *Thuya occidentalis* EO showed the best antibacterial activity of all EOs tested for both bacteria.

Moghaddam, M., Alymanesh, M.R., et. al., 2014, demonstrated that the effect of EO obtained from Basil on various bacteria, induced various responses in terms of inhibiting a wide range of pathogens. It was proved to be particularly effective against *Pseudomonas tolaasii*, while *Brenneria nigrifluens* was barely affected. In addition, *Xanthomonas citri* and *Rhodococcus fascians* were also inhibited, but at higher concentrations of EO compared to *P. tolaasii*.

Gakuubi, M.M., Wagacha, J.M. et. al., 2016 experienced the antibacterial activity of EO obtained from *Tagetes minuta* against phytopathogenic bacteria *Pseudomonas savastanoi pv. phaseolicola*, *Xanthomonas axonopodis pv. phaseoli* and *X. axonopodis pv.*

manihotis, which are responsible for causing diseases in beans. The results obtained confirmed the biopesticidal nature of EO and its use as a potential cheap, safe and effective alternative to chemicals used in crop protection.

Tests performed by *Stan (Tudora), C., et. al.* in *2019*, highlighted the rather bacteriostatic and not bactericidal activity of EO and less of the floral water, obtained from a new variety of Hyssop (*Hyssopus officinalis L*.) of Romanian origin ('*Cătălin'* variety), on the bacterium with phytopathogenic potential *Pseudomonas marginalis*.

ANTIFUNGAL ACTIVITY

The EO obtained from the tea tree showed promising results in the occurrence and severity of the disease induced by *Cercospore beticola* in beets and *Alternari solani* in potatoes. Also, the same EO was effective against the onset of *Fusarium sp.* on wheat, barley and oat leaves *(Terzi V., et.al., 2007)*. The application of oil in concentrations of 2.0% led to the inhibition of mycelial growths for *Botrytis oryzae, Alternaria brassicicola, Fusarium moniliforme, Aspergillus flavus, Fusarium proliferatum (Thobunluepop et. al., 2009).* The study conducted by *Carović-Stankoa, K., Frukb, G., et. al., 2013*, presents the results obtained after testing EOs obtained from 4 Basil taxa, preliminary screening of their antifungal activity against the phytopathogenic fungus *Monilinia laxa*. The best efficacy was obtained by EOs obtained from *Ocimum basilicum var*. *purpurascens* and *Ocimum tenuiflorum*, whose antifungal activity was sustained throughout the 23-day testing period. The paper of *Badawy, M. E.I., and Abdelgaleil, S. A. M., 2013,* present the results of tests obtained from the use of EOs obtained from 18 plants of Egyptian origin by hydrodistillation (*Artemisia judaica, A. monosperma, Callistemon viminalis, Citrus aurantifolia, C. lemon, C. paradisi, C. sinensis, Cupressus macrocarpa, C. sempervirens, Myrtus communis, Origanum vulgare, Pelargonium graveolens, Rosmarinus officinalis, Syzygium cumini, Schinus molle, S. terebinthifolius, Thuja occidentalis* and *Vitex agnus-castus).* These EOs were tested on phytopathogenic fungi: *Alternaria alternata, Botrytis cinerea, Fusarium oxysporum* and *Fusarium solani.* Inhibition of mycelial growths for most EOs was pronounced, and EO of *A. Monosperma* had the strongest inhibitory effect for *A. alternata, B. cinerea, F. oxysporum* and *F. solani*. On the other hand, EOs also caused a strong reduction in the germination of fungal spores compared to the control. EOs of *A. judaica* and *A. monosperma* caused the greatest inhibition of spore germination in *F. oxysporum*.

Of all the fungi tested, *F. Oxysporum* was the most sensitive to all EOs tested, except for *S. molle* EO.

Muscalu, A., Tudora, C., et.al., 2018, in the preliminary tests performed, highlighted the antifungal activity, strongly inhibiting of the EO obtained from Tagetes (*Tagetes patula L.),* against two entomopathogenic fungal strains, *Beauveria brongniartii* and *Beauveria bassiana*. *Stan (Tudora), C., et. al. in 2019,* tested 4 EOs obtained from new varieties of medicinal plants (2 varieties of Basil; Marigold and Hyssop), on the entomopathogenic fungus *Beauveria brongniartii* (BbgMm1a / 09) and a stored-product insect (*Sitophilus granarius*). The results obtained showed that, at different concentrations, Red Basil EO ('*Serafim'* variety) showed a strong inhibitory effect, compared to the other oils tested. Regarding the insecticidal activity, the preliminary tests performed with EOs obtained from new varieties of medicinal plants had no effect against the tested insect. In vitro tests performed by *Sumalan, R. M., Alexa, E. et. al. 2019,* on the growth of the phytopathogenic fungus *Fusarium graminearum* with Coriander (*Coriandrum sativum L.*) EO added in different concentrations, reveal its high antifungal potential, while the minimum concentration with fungistatic effect was 0.4% and the minimum fungicidal concentration was 0.6%. An increase in the antifungal effect was observed *in vivo* experiments with *F. graminearum*, when used in a mixture with an EO obtained from Thyme *(Satureja hortensis L.)*, and this effect is attributed to the synergistic effect between the two EOs used. Therefore, Thyme EO used alone or in mixtures with other EOs could be a useful alternative in organic farming.

INSECTICIDAL ACTIVITY

Recent studies have aimed to find alternatives to synthetic insecticides used to control the Colorado potato beetle (*Leptinotarsa decemlineata Say*) and the environmental benefits of EOs (*Göldel, B., Lemic, D., Bažok, R. 2020*). The various EOs obtained from *Eugenia caryophyllus* (Sprengel), *Mentha spicata L., Myrtus communis L., Ocimum basilicum L.*, *Satureja khuzistanica Jamzad* and *Thymus daenensis Celak* were tested for nutritional indications and efficacy against adults and larvae of generation IV. All the tested oils showed a discouraging effect, the most effective oil being *S. khuzistanica* (*Saroukolai, A.T., et.al., 2014).* Pyola, a natural compound found in canola and pyrethrins rapeseed oil, is applied not only against the Colorado potato beetle, but also against several harmful insects. As a large part of the commercially available rapeseed oil comes from genetically modified plants, this product may not be in line with organic farming rules, despite its success in pest management.

The study conducted by *Chrysargyris, A., Koutsoumpeli, E. et.al., 2021* shows that some compounds in the EO of *Mentha spicata* L., may have insecticidal or repellent effects on the insect *Lobesia botrana*. Also, several studies reported that *M. spicata* EO has a fumigant toxic effect against several species of stored-product insects of the order Coleoptera (*Kedia, A., Prakash, B., et. al., 2014*), but it also has repellent, larvicidal and ovicidal activity on insects of the order Diptera, families Culicidae and Drosophila *(Koliopoulos, G., Pitarokili, D., et. al., 2010)*. The compounds contained in this oil: limonene, pulegone, menthone, menthol and eucalyptol are known for their insecticidal and repellent activity.

BIOHERBICIDAL ACTIVITY

There is a growing interest in the use of EOs due to the compounds contained, which have bioherbicidal potential. Some of them have already been marketed and successfully launched in organic farming. They destroy the cuticle and contribute to dehydration or burning of young tissue *(Soltys, D., et.al. 2013)*. An example in this respect is the bioherbicide commercially available under the name "*GreenMatch EX*", which includes lemongrass oil *(Cymbopogon sp.)*, "Interceptor ™" with 10% pine oil (*Pinus sylvestris L*.) *(Dayan, F. E, et.al., 2009). Batish, D. R et. al, 2008*, showed that EO extracted from eucalyptus leaves (*Eucalipus sp.*) has phytotoxic potential on common weeds (*Cassia occidentalis* L. and *Echinochloa sp*.), sprayed in different concentrations (from 5%-10% v/v with 0.05% v/v Tween-80). The phytotoxicity of eucalyptus EO is due to the compounds: 1,8-cineole, citronellal, citronellol, citronellyl acetate, p-cymene, eucamalol, limonene, linalool, α-pinene, γ-terpinene, α-terpineol, etc. There are several natural compounds contained in EOs obtained from plants in the spontaneous flora, with potential for use in weed control: *Descurainia sophia L. (Li et.al., 2011), Lippia menosides* (*Marco et.al., 2012*) etc., which have the main compounds, thymol and carvacrol.

Kordali, S., et al., 2008, in studies performed with EO of *Origanum acutidens*, associate the presence of carvacrol and thymol compounds with inhibition of seed germination and seedling growth in weed species *A. retroflexus, Chenopodium album* and *Rumex crispus.*

Pinheiro, P.E., et. al., 2015, evaluated the bioerbicidal potential and the effect of *Plectranthus amboinicus* EO, of the compounds contained (carvacrol and thymol). They concluded that EOs obtained from its chemotypes delayed or inhibited germination and reduced root and aerial growth in monocotyledonous (*Sorghum bicolor*) and dicotyledonous (*Lactuca sativa*) species used in the tests. The compounds also caused changes in meristematic cells in *L. sativa* species, with chromosomal changes.

TRADING

Currently, there are fewer biopesticides registered in the EU than in the USA, India, Brazil, China, and the relatively low level of research in this field in the EU is related to the complexity of biopesticide regulations *(Damalas, C. A., Koutroubas, S., D. 2018).*

Globally, they comprise a small part of the total crop protection market, with a value of about $ 3 billion, representing only 5% of it *(Olson S., 2015).* There are over 200 products available on the US market, compared to 60 similar products on the European Union market. The size of the biopesticides market is expected to equal that of synthetic ones between the late 2040s and early 2050s, but there are uncertainties about absorption rates in areas such as Africa and South-East Asia *(Marrone, P.G., 2014; Damalas, C. A., Koutroubas, S., D. 2018).*There are few commercial biopesticides based on essential oils on the market, some of which are shown in Table 2.

Table 2

Commercial pesticides based on essential oils

Trade name	Active ingredients	Disease and pest control
Ant Out®	Clove oil (20%) Cottonseed oil (40%)	Ants, Spiders, Crickets
BioRepel™	Garlic oil (10%)	Aphids, Thrips, Flies
Bonide ® All Seasons	Mineral oil containing petroleum distillates	Bean aphids, beet thrips, almond mites, caterpillar eggs, coconut moth, apple weevil, greenhouse whitefly, mites, spiders, etc.

Bonide ®	Neem oil	Aphids, Flies, Mites, Rust, Powdery mildew, Black spot
Monterey Fruit Tree Spray Plus	Pyrethrin (0.25%), Neem oil (70%)	Powdery mildew, Rust, Leaf stain, fuzzy mould
Pest Out ®	Cottonseed oil (40%) Clove oil (20%) Garlic oil (10%)	Aphids, Mites, Thrips
QL Agri® 35	Quillaja Saponaria (35%)	Ectoparasitic and endoparasitic nematodes, Control of mites and insects from different crops: vines, citrus, tomato, apple, cherry, hazelnut.

*takeover from *Durán-Lara, E. F., et. al. 2020*

There are a number of issues regarding the trading of new products:

- deficiencies in the quantity of raw materials available;
- low persistence of treatment effects over time. EOs comprise lipophilic metabolites, highly volatile and with a molecular weight below 300. Terpenoids tend to be volatile, thermolabile and can be easily oxidised or hydrolysed depending on their structure. The chemical composition of an EO depends on the conditions during plant material storage, processing and its subsequent handling. Although the contact effect of EO is good, the rapid evaporation into the environment and the gradual biodegradation of the active substances, which take place after application to plants, leads to a low persistence of their effect.
- the best results of some EOs are obtained from plants, which are difficult to cultivate or which have low yields in obtaining essential oils. Therefore, it is necessary to streamline the production of these crops, to industrialize the production of essential oils, to ensure adequate control of the quality of these products on the market.
- there are strict registration laws for these products, which require toxicological testing and studies, especially for "non-target" organisms;

The authorization process is complex and expensive, as the authorization of any new product is. Authorizations in EU countries require safety documentation through appropriate toxicological studies. Their manufacture is often of local importance, on a small scale, because production is restricted by the limited availability of raw materials. Therefore, biopesticide manufacturers try to market such products outside the scope of the authorization process (e.g. as perfumes, secondary pesticide fertilizers, etc.). This practice is ineffective for many manufacturers because they cannot openly declare the efficacy against pests on the product label, which usually leads to low sales.

- there is no collaboration between companies that produce these products on a large scale and research centres, and studies are very limited *(Pavela, R., Benelli, G. 2016; Durán-Lara, E. F., et. al. 2020).*

The use of essential oils as biopesticides also has a number of clear *advantages:*

- high efficacy against a wide range of diseases and pests of agricultural importance;
- multiple mechanisms of action;
- due to the large number of active compounds in each mixture, the development of resistance is less likely;
- low toxicity to “non-target” organisms, including humans;
- low risk to health during applications due to low toxicity of residues.

A large number of EOs have been approved in Europe for use in agriculture. EOs obtained from plants of different origins have been recorded for specific uses, in particular for the biocidal effect, such as: *Mentha arvensis* and *Mentha spicata*, *Juniperus mexicana, Citrus x sinensis*, Piper nigrum, *Cinnamomum zeylanicum, Boswellia carterii*, *Cymbopogon flexuosus*, *Citrus aurantium*, etc. *(Raveau, R., et. al., 2020).*

Commercially available products: BIOXEDA-Clove-based EO, used against pathogens in fruit storage facilities (apple and pear), BIOX-M-*Mentha spicata*-based EO, used as a potato growth regulator, or LIMOCIDE-OROCIDE-PREV-AM and ESSEN'CIEL (orange EO against whitefly, thrips, powdery mildew, mould, etc. found in vegetable, fruit, ornamental, but also tobacco, vine crops). To these EOs can be added other aromatic plant extracts (either main compounds or purified EO obtained by specific extraction techniques, such as supercritical fluid extraction), from *Lavandula* *angustifolia, Artemisia alba, Citrus bergamia, Melaleuca leucadendron, Cinnamomum camphora, Elettaria cardamomum, Coriandrum sativum, Cupressus sempervirens, Eucalyptus globulus* and *Citrus paradisi (ECHA; Raveau, R., et. al., 2020).*

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CONCLUSIONS

Despite all the problems raised, the biopesticides market for organic farming has grown due to different formulations and producers, and the bioactivity of the new products is better than in the past. Cooperation between the public and private sectors is needed to facilitate the development, manufacture and sale of these products as environmentally friendly alternatives.

Further research is needed to integrate them into common production systems, to maintain low costs, a certain quality and availability of products for farmers. Regulations that promote the registration of low-risk products, the provision of incentives could facilitate the trading and availability of biopesticides on the market. While new products could serve as a promising option for use in pest control, more research is needed in the field to assess their effectiveness on pest-specific issues in different cropping systems.

Future research on the use of EOs as biopesticides for agriculture will focus on: the development of efficient stabilization processes (e.g. microencapsulation); simplification of legislation on biopesticide approval requirements; optimization of plant cultivation conditions and extraction processes, leading to obtaining EOs with homogeneous chemical composition.

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BIBLIOGRAPHY

1. Asbahani, A.E., Miladi, K., Badri, W., Sala, M., Aït Addi, E., Casabianca, H., El Mousadik, A., Hartmann, D., Jilale, A., Renaud, F. N. R., Elaissari, A., 2015 - *Essential oils: from extraction to encapsulation,* Int. J. Pharmacol. vol. 483, p. 220-243.
2. Badawy, M.E.I., Abdelgaleil, S.A.M., 2013 - *Composition and antimicrobial activity of essential oils isolated from Egyptian plants against plant pathogenic bacteria and fungi,* Industrial Crops and Products, vol. 52, p.776-782.
3. Batish, D.R., Singh, H.P., Kohli, R.K., Kaur, S., 2008 - *Eucalyptus essential oil as a natural pesticide,* Forest Ecology and Management, vol. 256(12), p. 2166-2174.
4. Carović-Stankoa, K., Frukb, G., Satovica, Z., Ivićc, D., Politeod, O., Severe, Z., Grdišaa, M., Strikićf, F., Jemrićbet, T., 2013 - *Effects of Ocimum spp. essential oil on Monilinia laxa in vitro,* The Journal of Essential Oil Research, vol.25(2), p.143-148.
5. Chrysargyris, A., Koutsoumpeli, E., Xylia, P., Fytrou, A., Konstantopoulou, M., Tzortzakis, N., 2021 - *Organic cultivation and deficit irrigation practices to improve chemical and biological activity of Mentha spicata,* Plants. Agronomy, vol.11 (3), p. 599.
6. Dayan, F.E., Cantrell, C.L., Duke, S.O., 2009 - *Natural products in crop protection,* Bioorganic & Medicinal Chemistry, vol.17(12), p. 4022-4034.
7. Damalas, C.A., Koutroubas, S.D., 2018 - *Current Status and Recent Developments in Biopesticide Use,* Agriculture, vol. 8(1) p.13.
8. Durán-Lara, E.F., Valderrama, A., Marican, A., 2020 - *Natural Organic Compounds for Application in Organic Farming,* Agriculture, vol. 10(2), p. 41.
9. Elahi, E., Weijun, C, Zhang, H., Nazeer, M., 2019 - *Agricultural intensification and damages to human health in relation to agrochemicals: Application of artificial intelligence*. Land Use Policy, vol. 83, p. 461-474.
10. Gakuubi, M.M., Wagacha, J.M., Dossaji, S.F., Wanzala, W., 2016 - *Chemical composition and antibacterial activity of essential oils of Tagetes minuta (Asteraceae) against selected plant pathogenic bacteria,* International Journal of Microbiology, article ID 7352509.
11. Göldel, B., Lemic, D., Bažok, R., 2020 - *Alternatives to Synthetic Insecticides in the Control of the Colorado Potato Beetle (Leptinotarsa decemlineata Say) and Their Environmental Benefits,* Agriculture, vol.10, p. 611.

12. Hansted, L., Jakobsen, H.B., Olsen, C.E., 1994 - *Influence of temperature on the rhythmic emission of volatiles from Ribes nigrum flowers in situ,* Plant Cell Environ., vol. 17, p. 1069-1072.
13. Isman, M.B., Machial, C.M., 2006 - *Chapter 2: Pesticides based on plant essential oils: From traditional practice to commercialization. In Advances in Phytomedicine. Naturally Occurring Bioactive Compounds*, 1st ed.; Rai, M., Carpinella, M.C., Eds.; Elsevier: Amsterdam, The Netherlands.
14. Kedia, A., Prakash, B., Mishra, P.K., Chanotiya, C.S., Dubey, N.K., 2014 - *Antifungal, antiaflatoxigenic, and insecticidal efficacy of spearmint (Mentha spicata L.) essential oil,* Int. Biodeterior. Biodegrad., vol.89, p. 29-36.
15. Koliopoulos, G., Pitarokili, D., Kioulos, E., Michaelakis, A., Tzakou, O., 2010 - *Chemical composition and larvicidal evaluation of Mentha, Salvia, and Melissa essential oils against the West Nile virus mosquito Culex pipiens,* Parasitol. Res., vol.107, p. 327–335.
16. Kordali, S., Cakir, A., Ozer, H., Cakmakci, R., Kesdek, M., Mete, E., 2008 - *Antifungal, phytotoxic and insecticidal properties of essential oil isolated from Turkish Origanum acutidens and its three components, carvacrol, thymol and p-cymene,* Bioresource Technology, vol.99, p. 8788-8795.
17. Koul, O., 2016 - *The Handbook of Naturally Occurring Insecticidal Toxins*, 1st ed.; CABI: Wallingford, UK.
18. Lee, R., den Uyl, R., Runhaar, H., 2019 - *Assessment of policy instruments for pesticide use reduction in Europe; Learning from a systematic literature review,* Crop Protections*,* vol. 126, 104929.
19. Li, J., Liu, X., Dong, F., Xu, J., Li, Y., Shan, W., Zheng, Y., 2011 - *Potential allelopathic effects of volatile oils from Descurainia sophia (L.) Webb ex Prantl on wheat,* Biochemical Systematics and Ecology, vol.39, p. 56-63.
20. Marco, C.A., Teixeira, E., Simplício, A., Oliveira, C., Costa, J., Feitosa, J., 2012 - *Chemical composition and allelopathyc activity of essential oil of Lippia sidoides Cham.* Chilean Journal of Agricultural Research, vol. 72, p. 157-160.
21. Marrone, P.G., 2014 - *The market and potential for biopesticides. In Biopesticides: State of the Art and Future Opportunities;* Gross, A.D., Coats, J.R., Duke, S.O., Seiber, J.N., Eds.; American Chemical Society: Washington, DC, USA, p. 245–258.
22. Mahmoud, S.S., Croteau, R.B., 2002 - *Strategies for transgenic manipulation of monoterpene biosynthesis in plants,* Trends Plant Sci., vol. 7, p. 366-373.

23. Moghaddam, M., Alymanesh, M.R., Mehdizadeh, L., Mirzaei, H., Ghasemi P.A., 2014 - *Chemical composition and antibacterial activity of essential oil of Ocimum ciliatum, as a new source of methyl chavicol, against ten phytopathogens,* Ind. Crop. Prod., vol. 59, p.144-148.
24. Muscalu, A., Birsan, M., David, L., Grigore, I., Fătu, A. C., Tudora, C., 2018 - *Harvesting and processing of French marigold to obtain products with new uses,* Proceedings of the 46th International Symposium, Actual Tasks on Agricultural Engineering, 27 february - 1 March, Opatija, Croatia, p. 239-244.
25. Olson, S., 2015 - An analysis of the biopesticide market now and where is going, Outlooks Pest Manag. vol. 26(5), p. 203-206.
26. Opdyke, D.L.J., 1979 - *Monographs on Fragance Raw Materials. A Collection of Monographs Originally Appearing in Food and Cosmetics Toxicology,* 1st ed.; Pergamon Press Ltd.: Oxford, UK.
27. Pavela, R., 2014 - *Limitation of plant biopesticides. In Advances in Plant Biopesticides,* Ed., Springer, New Delhi
28. Pavela, R., Benelli, G., 2016 - *Essential oils as ecofriendly biopesticides? Challenges and constraints,* Trends Plant. Sci., vol. 21, p. 1000-1007.
29. Pavela, R., Žabka, M., Bednář, J., Tříska, J., Vrchotová, N., 2016 - *New knowledge for yield, composition and insecticidal activity of essential oils obtained from the aerial parts or seeds of fennel (Foeniculum vulgare Mill.),* Ind. Crops Prod. vol. 83, p. 275-282.
30. Pinheiro, P.E., Costa, A.V., Alves, T.A., Galter, I.N., Pinheiro, C.A., Pereira, A.F., Fontes, M.M.P., 2015 - *Phytotoxicity and cytotoxicity of essential oil from leaves of Plectranthus amboinicus, carvacrol, and thymol in plant bioassays,* Journal of Agricultural and Food Chemistry, vol. 63, p. 8981-8990.
31. Raveau, R., Fontaine, J., Hadj, I., Sahraoui, A. L., 2020 - *Essential Oils as Potential Alternative Biocontrol Products against Plant Pathogens and Weeds: A Review*, Foods, vol. 9, p. 365.
32. Yao, S., Zhao, Z., Lu, W., Dong, X., Hu, J., Liu, X., 2021 - *Evaluation of Dissipation Behaviour, Residues, and Dietary Risk Assessment of Fludioxonil in Cherry via QuEChERS Using HPLC-MS/MS Technique,* Molecules, vol. 26(11), p. 3344.
33. Salamci, E., Kordali, S., Kotan, R., Cakir, A., Kaya, Y., 2007 - *Chemical compositions, antimicrobial and herbicidal effects of essential oils isolated from Turkish Tanacetum aucheranum and Tanacetum chiliophyllum var. chiliophyllum,* Biochem. Syst. Ecol. vol. 35, p. 569-581.

34. Saroukolai, A.T., Nouri-Ganbalani, G., Hadian, J., Rafiee-Dastjerdi, H., 2014 - *Antifeedant activity and toxicity of some plant essential oils to Colorado potato beetle, Leptinotarsa decemlineata Say (Coleoptera: Chrysomelidae),* Plant Protect. Sci. vol. 50, p. 207-216.
35. Stan, (Tudora), C., Vlăduțoiu, L., Muscalu, A., Vlăduț, N.V., 2019 - *Harvest and quality of Hyssop (Hyssopus officinalis L.), Proceedings of the 47th International Symposium, Actual Tasks on Agricultural Engineering, 0*5 - 07 March 2019, Opatija, Croatia, p. 315-321.
36. Stan, (Tudora), C., Muscalu, A., Fătu, A.C., Dinu, M.M., Israel-Roming, F., 2019 - *Preliminary tests regarding fumigant effect of volatile oils obtained from medicinal plants,* Bulletin of USAMV Cluj-Napoca. Animal Science and Biotechnologies, vol. 76(1), p. 57.
37. Soltys, D., Krasuska, U., Bogatek, R., Gniazdowska, A., 2013 - *Chapter 20. Allelochemicals as Bioherbicides-Present and Perspectives,* p. 529-531.
38. Sturchio, E., Donnarumma, L., Annesi T., Millano, F., Cassori, L., Masciarelli, E., Zanellato, M., Meconi, C., Boccia, P., 2014 - *Essential oils: an alternative approach to management of powdery mildew diseases,* Phytopathologia Mediterranea, vol. 53(3), p. 385-395.
39. Sumalan, R. M., Alexa, E., Popescu, I., Negrea, M., Radulov, I., Obistioiu, D., Cocan, I., 2019 - *Exploring ecological alternatives for crop protection using Coriandrum sativum essential oil,* Molecules, vol. 24, 2040.
40. Shuqin, J., Fang, Z., 2018 - *Zero Growth of Chemical Fertilizer and Pesticide Use: China's Objectives, Progress and Challenges.* J. Resour., Ecol., vol. 9(1), p. 50-58.
41. Terzi, V., Morcia, M., Faccioli, P., Valè, G., Tacconi, G., Malnati, M., 2007 - *In vitro antifungal activity of the tea tree (Melaleuca alternifolia) essential oil and its major components against plant pathogens,* Letters in Applied Microbiology, vol. 44(6), p. 613-618.
42. Thobunluepop, P., Udomsilp, J., Piyo, A., Khaengkhan, P., 2009 - *Screening for the antifungal activity of essential oils from bergamot oil (Citrus hystrix DC.) and tea tree oil (Melaleuca alternifolia) against economically rice pathogenic fungi: a driving force of organic rice Cv. KDML 105 production,* Asian Journal of Food & Agro-Industry, Special Issue, S374-S380.
43. Tworkoski, T., 2002 - *Herbicide effects of essential oils,* Weed Science, vol. 50, p. 425-431.

44. Zanellato, M., Masciarelli, E., Casorri, L., Boccia, P., Sturchio, E., Pezzella, M., Cavalieri A., Caporali, F., 2009 -*The essential oils in agriculture as an alternative strategy to herbicides: a case study.* International Journal of Environment and Health, vol. 3(2), p. 198-213.
*** Food and Agriculture Organization of the United Nations. The Use of Spices and Medicinals as Bioactive Protectants for Grains; Agricultural Services Bulletin No. 137; FAO: Rome, Italy, 1999.
*** Directive 2009/128/EC of the European Parliament and of the Council of 21 October 2009 establishing a framework for Community action to achieve the sustainable use of pesticides, *Official Journal of the European Union* L 309/71.
*** Regulation (EC) No 1107/2009 of the European Parliament and of the Council of 21 October 2009 concerning the placing of plant protection products on the market.
ECHA (European Chemical Agency) - https://www.echa.europa.eu/