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RESEARCH ARTICLE

THE ESSENTIAL OILS AS BIOPESTICIDES AND SOME MECHANISMS OF ACTION

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ABSTRACT

Pests such as weeds, pathogens and insects represent a problem for agricultural crops since they significantly reduce crop production. To protect the agricultural crops use significant amounts of synthetic pesticides around the world. However, these chemical compounds have been toxic into the living systems. Thus, new natural pesticides are being investigated which are friendlier to the environment and few effects on human health.

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INTRODUCTION

Pesticides are chemical substances which are used to kill harmful. Pesticides have been grouped in diferent classes based on their uses and handling, for example, bactericides, fungicides, nematicides, algicides, herbicides, rodenticides, insecticides. They are composed of organophosphates, pyrethroids, organochlorines, carbamates and chlorines. Some pesticides are soluble in water while others are soluble in organic solvents. They usually touch the nervous system of the pests and destroy them (Dad *et al*., 2022). Due to use of pesticides in a variety of sectors like food, forestry, agriculture and aquaculture, these chemical compounds have been toxic into the living systems. The harmful effects of them have been observed on plants affecting the growth, metabolism, producing genotypic and phenotypic changes; the soil, aquatic environments and human health have also been affected. In this last, it has been reported that pesticides produce genetic alteration, cancer, allergies, and asthma (Pathak *et al*., 2022). As a result of the above, new natural pesticides are being investigated that are more friendly to the environment and few effects on human health (Batish *et al*., 2008; Flores-Encarnación *et al*., 2023). In this context, aromatic plants and their essential oils have been used since antiquity as flavorings (condiments or spices), as well as in fragrances, medicines, as antimicrobial agents (Daferera *et al*., 2003; Flores-Encarnación *et al*., 2016).

Essential oils have also been reported to be good insect repellents or insecticides and have served to protect stored products against pests. These organic sources have been proposed as a valuable alternative to synthetic pesticides, since they do not produce adverse effects on the environment and cause less harm to human, animal health and to habitats and the ecosystem. Currently, novel, highly selective and easily degradable chemical products have been investigated. These natural biopesticides can be crude extracts or essential oils obtained from any part of the plant and are called "green pesticides" (Bakkali *et al*., 2008; Dorman and Deans, 2000; Isman and Machial, 2006; Nollet and Rathore, 2019). Therefore, in this work some novel mechanisms of action attributed to biopesticides are shown, especially related to essential oils.

ESSENTIAL OILS AND THEIR BIOLOGICAL POTENTIAL

Essential oils have been used for thousands of years in various cultures for medicinal and health purposes. They are concentrated hydrophobic liquids containing volatile aromatic compounds in great quantities. Because of their antidepressant, stimulating, detoxifying, and calming properties, they are recently gaining popularity as a natural, safe and cost-effective therapy for a number of health concerns. Essential oils are extracted from different parts of plants for example: leaves, barks, seeds, flowers, fruit peel. They were obtained by extraction using steam distillation and hydro distillation. Essential oils possesses antibacterial and antiviral properties and have been screened as a potential sources of novel antimicrobial compounds, alternatives to hazardous chemical preservatives and agents promoting food preservation (Burt, 2004; Fometu *et al.,* 2019; Mahato *et al*., 2019; Solórzano-Santos *et al*., 2012; Tajkarimi *et al.* 2010; Tongnuanchan and Benjakul, 2014). It has been reported that terpenes, terpenoids, aldehydes, alcohols, phenols, esters, low molecular weights aromatic and aliphatic substances are the main compounds in essential oils. Typically, the major components of essential oils are the main components responsible for their biological properties. However, minor compounds may also play an important role in bioactivity, either by potentiating the action of major components or through antagonistic or additive effects (Bassolé and Juliani, 2012). In general, essential oils are composed of approximately 20–60 components at different concentrations, but some of them may contain more than 300 different substances. However, two or three components are usually present in large proportions (20–70%) compared to other constituents present in small concentrations. For example, 1,8-cineole or eucalyptol (70–90%) is the major component of *Eucalyptus globulus* Labill essential oil while cinnamaldehyde (60–90%) is the major component of *Cinnamomum zeylanicum* Blume essential oil (Behbahani*et al.*, 2020; Boukhatem *et al*., 2020; Chouhan *et al*., 2017; de Sousa *et al*., 2023). Other example is the essential oil of *Thymus vulgaris* (thyme) which contains monoterpenes as thymol (49%), ρ- cimene (18%), carvacrol (6%), γ- terpinene (9%), linalool (3%), car-3-eno (2%), β- mirceno (2%), α- pinene (1%), limonene (1%) and camphane (0.5%) (Ben *et al*., 2019; Flores-Encarnación*et al*., 2022; López, 2004; Sakkas and Papadopoulou, 2007). The phenol monoterpene derivatives (thymol and carvacrol) are the major ingredients in thyme essential oil with antimicrobial and pharmacological properties such as anti-metastatic activity, anti-oxidative, and anti-inflammatory (Kianersi *et al*., 2021; Powers *et al*., 2018). In relation to antibacterial properties, Erkana *et al*., (2012) demonstrated the action of a group of terpenoids extracted from leaves of *Murraya koenigii* (curry) versus Gram-negative bacteria. Prabuseenivasan *et al*., (2006) reported that 19 essential oils showed antibacterial activity.

They demostrated a significant inhibitory effect by cinnamon, clove, geranium, lemon, lime, orange and rosemary essential oils. Cinnamon essential oil showed the highest activity at low concentrations; aniseed, eucalyptus and camphor essential oils showed the lowest antibacterial activity against the tested bacteria (*Klebsiella pneumoniae*, *Proteus vulgaris*, *Bacillus subtilis*, *E. coli*, *P. aeruginosa*, and *S. aureus*). It has been reported also the potent inhibitory effect of essential oil of oregano, thyme and basil on the growth of *E. coli* and *P. aeruginosa* (Al-Bayati, 2008; Flores-Encarnación *et al*., 2016; Gracia-Valenzuela *et al*., 2012; Paredes-Aguilar *et al*., 2007). It has been reported also that the essential oil of *Thymus vulgaris* had a negative effect on growth and biofilm formation of uropathogenic *E. coli* (Flores-Encarnación *et al*., 2018). On the other hand, it has been reported that essential oils inhibit or slow the growth of yeasts and molds. Maness and Zubov (2019) reported that essential oils of *Rosmarinus officinalis*, *Cinnamomum verum* and *Citrus paradisi* inhibited the growth of *Trichophyton mentagrophytes*, *Microsporum gypseum* and *Rhizopus stolonifer*. Gucwa *et al*., (2018) reported that *T. vulgaris*, *Citrus limonum*, *Pelargonium graveolens*, *Cinnamomum cassia*, *Ocimum basilicum*, and *Eugenia caryophyllus* essential oils showed both fungistatic and fungicidal activity toward *Candida albicans* and *C. glabrata* isolates, resulting that *C. cassia* essential oil have the highest activity. The study also showed that *T. vulgaris* and *C. limonum* affected the cell membranes and *T. vulgaris* produced a potassium ion efflux. In addition, it was observed that all of the tested oils showed the ability to inhibit the transition of yeast to mycelium form. Candidiasis (frequently caused by *C. albicans*, *C. glabrata*, *C. tropicalis*, *C. krusei*, or *C. parapsilosis*) is associated with the formation of biofilms on the surface of medical devices and tissues (Feyaerts *et al*., 2018; Flores-Encarnación*et al*., 2022; Ramage *et al*., 2006). Rajkowska *et al*., (2019) reported that clove and thyme essential oils can be efficiently used preventing the formation of biofilm in abiotic surfaces (glass, polyethylene terephthalate, polypropylene) by *Candida* sp.; clove and thyme essential oils showed anti-biofilm activity. In relation to the effect of essential oils in viruses, it has been reported in vitro that *Lippia alba* and *Lippia citriodora* essential oils inhibited the replication of dengue virus serotypes. The use of the *Lippia* sp. essential oils could be a potential resource for treatment of tropical disease like dengue, especially in developing countries. The *L. citriodora* infusions have been used for the treatment of colds, flu, bronchitis, coughs, asthma and others (Flores-Encarnación *et al*., 2020*b*; Ocazionez *et al*., 2010; Pascual *et al*., 2001). It has been reported that *L. alba* essential oil contains as major components: carvone (40-51%), limonene (30-33%) and bicyclesesquiphellandrene (7-9%). Viral inhibitory effect was not observed by addition of the essential oil after virus adsorption. It seems that inhibitory effect of *Lippia* sp. essential oil is attributed to direct inactivation of virus before adsorption on the host cell (da Silva *et al*., 2020). It is speculated that direct inactivation of virus by the essential oils can be due to disruption of lipid viral envelope by action of terpenes and terpenoids (Meneses *et al*., 2009). Other authors have reported that these compounds have showed anti-HIV activity inhibiting the virus adsorption to target cell and causing inactivation of virus reverse transcriptase. In case of yellow fever virus, the antiviral effect of *L. alba* and *L. citriodora* essential oils could be explained by the presence of lipid compounds as carvone, limonene, geranial, neral, and others (Meneses *et al*., 2009; Sun *et al*., 1996). Some mechanisms of action by antibacterial activity of essential oils have been reported. One of them is the the ability to alter and to penetrate the lipid membrane of bacteria and fungi or affecting the lipid envelope of viruses, making them more permeable and causing leaking ions and cytoplasm (bacterial and fungi lysis and death). There are few reports about the bactericidal mechanism of the essential oil of *T. vulgaris*. It is believed that this essential oil alters the permeability of membrane as do other oils (Hussein *et al*., 2018; O'Bryan *et al*., 2015). It has been reported that carvacrol is a hydrophobic compound that influences cell membranes by altering the composition of fatty acids, which then affects the membrane fluidity and permeability (Rudramurthy *et al*., 2016). However, its exact mechanism of action is still unclear. It was reported that carvacrol significantly depleted the internal ATP pool of bacterial cells and induced the leakage and loss of ATP from bacterial cells (Rudramurthy *et al*., 2016; Swamy *et al*., 2016; Ultee *et al*., 2002). In a previous study, it was reported in uropathogenic *E. coli* a new possible mode of action of essential oil of *T. vulgaris*, which consists of the blockage of the respiratory chain of the bacterium and that could explain the loss of ATP from bacterial cells reported by other authors (Flores-Encarnación *et al.*, 2020*a*).

ESSENTIAL OILS AS BIOPESTICIDES: Pests such as weeds, pathogens and insects represent a problem for agricultural crops since they significantly reduce crop production (from 25 to 50%). So, to protect the agricultural crops use significant amounts of synthetic pesticides around the world. However, the excessive use of synthetic pesticides in crop fields, urban environment and water bodies (to eliminate harmful pests) has resulted in an increased risk of pesticide contamination, in addition to the emergence of resistant pests and the toxicological consequences for human health, including environmental contamination. Currently, natural pesticides are being investigated that are friendlier to the environment and few effects on human health. In this context, aromatic plants and their essential oils have been used since antiquity as flavorings (condiments or spices), as well as in fragrances, medicines, etc (Batish*et al.,* 2008; Daferera *et al*., 2003; Flores-Encarnación *et al*., 2016; Flores-Encarnación *et al.*, 2023). Essential oils have been explored for repellent, fumigant, larvicidal and adulticidal properties for pest control. They can be inhaled, ingested or skin absorbed by insects. Plants from the *Myrtaceae*, *Lamiaceae*, *Asteraceae*, *Apiaceae*and *Rutaceae* families are selected for their anti-insect activities, such as Lepidoptera, Coleoptera, Diptera, Isoptera, and hemiptera (Kumar*et al*., 2022; Tripathi *et al*., 2009). For example, it has been reported the toxicity of oil-in-water formulations of pine, lemongrass, geranium, eucaliptus, palmarosa and citral essential oils, against *Bemisia tabaci* (polyphagus whitefly pest) (Nebapure *et al*., 2022). Palmarosa essential oil was found to possess highest contact toxicity. Other authors reported that essential oil of *Ruta chalepensis* (*Rutaceae*) was a larvicidal effect against larvae of *Orgyia trigotephras* (a phytophagous insect) (Akkari *et al*., 2015; Fierascu *et al.,* 2020; Flores-Encarnación *et al.*, 2023). Below, some mechanisms of action of essential oils due to their pesticide activity will be shown.

THE BIOPESTICIDE ACTIVITY OF ESSENTIAL OILS

Essential oils are synthesized through secondary metabolic pathways of plants as communication and defense molecules, and are responsible for the specific flavor and scent of aromatic plants (Dudareva*et al.*, 2006; Nagegowda, 2010; Pavela and Benelli, 2016). Essential oils play important roles in direct and indirect plant defenses against herbivores and pathogens, in plant reproduction processes through attraction of pollinators and seed disseminators, and in plant thermotolerance. Substances contained in essential oils are classified into two chemical groups according the metabolic pathway of their synthesis: terpenoids, which are mainly represented by monoterpenes and less commonly by sesquiterpenes, and phenylpropanoids with low molecular weight. The synthesis and accumulation of essential oils is associated with the presence of secretory structures such as glandular trichomes (*Lamiaceae*), secretory cavities (*Myrtaceae*, *Rutaceae*), and resin ducts (*Asteraceae*, *Apiaceae*), which can be found in various plant organs (Aharoni *et al*., 2005; Pavela and Benelli, 2016; Nagegowda, 2010; Regnault-Roger *et al*., 2012). As it has been reported, secondary metabolites obtained from plants (such as essential oils) have become popular as an alternative to combat mosquitoes, such as *Aedes aegypti*, *Culex quinquefasciatus*, *Anopheles stephensi*, as well as repellents in water-soluble formulations (Nuchuchua *et al*., 2009; Veerakumar *et al*., 2014; Vivekanandhan *et al.*, 2018; Vivekanandhan *et al.*, 2020; Vivekanandhan *et al.*, 2023). So, essential oils from eucalyptus leaves have multiple activities depending on insects. Recently vapour delivery of plant essential oils have been shown to alter pyrethroid efficacy and detoxification enzyme activity in mosquitoes. Further, studies have shown that the essential oils can delivered as a vapour for enhancement of deltamethrin efficacy in both pyrethroids susceptible and resistant strains of *A. aegypti* mosquitoes. The oil from other aromatic plants are also used to control other insect pests (O'Neal *et al.*, 2019; Vivekanandhan *et al.*, 2023). Natural plant metabolites attribute their toxicity to multiple mechanisms of action, which reduces the possibility of resistance development in target mosquitoes. Thus, the joint action of synthetic chemical pesticides and natural products (such as essential oils) increase the toxic efficacy in mosquitoes, affecting the early stages of mosquito, through the action of the substances contained in essential oils and affecting the nervous system central of the mosquito (inhibition of cholinesterase) by the action of the chemical insecticide (Hiramori and Nishigaki, 2001; Pavela and Benelli, 2016). Different mechanisms of action have been proposed regarding the bioinsecticidal action of essential oils and their components. So, he monoterpenes show a cytotoxic effect on insect tissues by decreasing cell membrane permeability reducing the number of intact Golgi bodies and mitochondria, and impairing cellular respiration. Since the nervous system is crucial to the functional integrity of insects, most essential oils find it to be an easy target of action. Numerous putative receptors for essential oil activity have been described in the literature, some of which include receptors for acetylcholinesterase (AChE), octopamine, adenosine triphosphatases (ATPases), gamma-aminobutyric acid (GABA)-gated chloride channels, butyrylcholinesterase (BuChE), and nicotinic acetylcholine. Acetylcholinesterase is a crucial enzyme involved in the breakdown of the neurotransmitter acetylcholine into choline and acetate, the termination of neurotransmission and synaptic signaling (Blenau *et al*., 2012; Cardenas-Ortega *et al*., 2015; Gaire *et al*., 2019; Gupta *et al.*, 2023; Kostyukovsky *et al*., 2002; Yeom *et al*., 2015). Other authors have reported that the acetylcholinesterase activity is inhibited by essential oils of *Cyclotrichium niveum* (Boiss.), *Thymus praecox* subsp. *caucasicus* (Willd. ex Ronniger) and *Anethum graveolens* L., causing neurotoxicity in living organisms (particularly arthropods). So, monoterpenoids like linalool act upon an insect's nervous system and affect the ion transport and release of the acetylcholinesterase enzyme. Suppression of acetylcholinesterase activity is a biomarker of essential oil-induced toxicity in insects (Blenau *et al.*, 2012; López and Pascual-Villalobos, 2010; Orhan *et al*., 2009; Orhan *et al*., 2013; Re *et al*., 2000; Seo *et al.,* 2014). On the other hand, the German cockroach (*Blattella germanica* L.) is an important domestic and industrial pest. Their feces and exuviae can cause allergic reactions in sensitive people and can be vectors of pathogenic microorganisms (viruses, bacteria, protozoa and helminths for humans) and wildlife. Additionally, cockroaches are disgusting to most people and indicate an unhealthy environment. German cockroaches have a short generation time and high fecundity, which increases their chances of developing resistance to insecticides used for population management (Barcay, 2004; Phillips and Appel, 2010). It has been reported that thymol, terpineol and linalool, main components of the essential oil of *Origanum majorana* showed fumigant toxicity against female German cockroaches. Similar effects were observed with *Mentha arvensis* L. essential oil, which contains menthol and menthone as main components. The *M. arvensis* L. essential oil had fumigant activity against the American cockroach (*Periplaneta americana* L.) and the German cockroach (Appel *et al*., 2001; Jang *et al*., 2005; Phillips and Appel, 2010). These are some examples about the mechanisms of action of essential oils as possible biopesticides. It is necessary to carry out more studies to learn more about its uses and its application in other areas of knowledge.

CONCLUSION

Biopesticides are a possible alternative to replace or complement the use of chemical pesticides currently used. Their ecological advantages make biopesticides more beneficial to the environment and humans. In this context, essential oils are compounds of natural origin that have been recognized for their multiple functions, including being biopesticide compounds. Therefore, it is important that more studies be carried out to learn about the biopesticidal properties of essential oils or their derivatives and their mechanisms of action.

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