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Nematicidal Potential of Biogenic Volatile Organic Compounds (BVOCs)

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Abstract

Plant-Parasitic Nematodes (PPNs) cost global agriculture US $173 billion in economic losses in (Elling, 2013). According to Kumar et al. (2020), they are said to result in yearly agricultural losses of 21.3% in India, or Rs. 102,039.79 million (1.58 billion USD). Biogenic VOCs have been known to exhibit nematicidal effects. *Pseudomonas putida*-produced VOCs such as 2-octanone, 2-undecanone, dimethyl disulfide, and 2-nonanone have demonstrated contact nematicidal activity. The majority of biogenic VOCs induce oxidative stress in nematode cells, which leads to aberrant nematode function [1]. According to Cheng et al. (2017) [2], VOCs such 2-nonanone and 2-decanone impair the integrity of the gut and pharynx of *M. incognita*, causing mortality rates of 87.6% and 82.6% *in vitro* and greenhouse, respectively. According to Silva *et al.* (2019) [3], gall reduction through soil bio-fumigation with *Cymbopogon nardus* and *Dysphania ambrosioides* was 19% and 37%, respectively. Solid-phase microextraction is used to sample VOCs, and chromatography is used to detect them based on the partition coefficient. As a result, a potential approach for managing PPN that might be investigated is the use of biogenic VOCs to create analogues for nematicidal effects in soil.

Background: Plant-parasitic nematodes are notorious agricultural pests that cause substantial economic losses by attacking crops and disrupting root systems. Conventional nematode control methods often rely on chemical pesticides, which have raised concerns about environmental safety and the development of resistance. In recent years, there has been growing interest in exploring alternative strategies for nematode management that are effective, environmentally friendly, and sustainable. Biogenic Volatile Organic Compounds (BVOCs), emitted by various plant and microbial sources, have emerged as promising candidates for controlling plant-parasitic nematodes due to their diverse chemical compositions and potential for affecting nematode behaviour and physiology.

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Introduction

One of the most intricate and numerous organisms in the universe are nematodes, which are small worms. When it comes to nematodes that live in soil, some play important ecological roles in the soil food web by controlling carbon and recycling nutrients (such nitrogen, which increases the availability of the nutrient to plants), while others are viewed as phytosanitary risks. Plant-Parasitic Nematodes (PPNs) are nematode species that harm cultivated plant species, and they are challenging to control. They parasitize numerous crops all throughout the world, resulting in significant yearly economic losses. Some of the most economically detrimental PPNs on horticultural and field crops are the Root-Knot Nematodes (RKNs), *Meloidogyne spp*., which can damage practically all cultivated plant species. *Meloidogyne incognita*, one of the most prevalent RKNs in the world, may infect a wide range of plant species. The successful establishment and spread of PPN depend on its rapid reproductive rate and polyphagous lifestyle. It is challenging to locate nematodes for treatment because of their non-uniform, patchy horizontal and vertical distributions, which depend on the depth of the plant's root system and the availability of moisture. *Meloidogyne spp*., *Globodera spp*., *Heterodera spp*., *Pratylenchus spp*., the burrowing nematode (*R. similis*), and the stem and bulb nematode (*D. dipsaci*) are typically the nematodes that cause the most damage to agricultural crops. Unquestionably, *B. xylophilus* is the most harmful organism in forestry systems.

Nematicides made by chemicals have a significant role in control measures. They do, however, also come with some serious drawbacks, including as harm to the environment and human health. The use of many synthetic pesticides has been restricted due to growing public concern over the toxicity of chemical pesticides and ecological protection. Our scientist has recently developed an interest in working on environmentally friendly management solutions to control PPNs below the economic threshold level. According to Campos *et al.* (2010), Volatile Organic Compounds (VOCs), which are found in microbes and plants, have the potential to be used in the creation of commercial nematicides. According to Xu *et al*. (2015), VOCs include a wide variety of alcohols, aldehydes, ketones, esters, ethers, phenols, alkenes, alkynes, and alkyl compounds. Biogenic VOCs (BVOCs) are the aggregate term for the VOCs that are released into the environment by plants and microorganisms. Here, we've highlighted the value of BVOCs as a tool for managing PPNs in agroecosystems and achieving crop cultivation sustainability to secure future generations' access to food.

The main body of the review

Biogenic volatile organic compounds (BVOCs)

Low boiling point substances known as "biogenic volatile organic compounds" (BVOCs) are frequently produced by secondary metabolic pathways in plants. According to Loreto and Schnitzler (2010), BVOCs can be released into the atmosphere by several vascular plants. One of the main sources of BVOC emissions, the forest accounts for around 70% of all vegetation-related BVOC emissions (Guenther et al., 2012). According to Loreto and Schnitzler (2010), the MEP pathway in plants is where isoprene and monoterpenes are synthesised, and both play a significant role in the overall biogenic VOC emission globally. BVOCs typically develop constitutively or as a result of stress induction. These elements can increase a plant's ability to withstand biotic and abiotic stressors such high temperatures, oxidative stress, and pest infestations (e.g., competing plants and microorganisms) (Loreto and Schnitzler 2010; Filella et al. 2013). The following table shows different BVOCs from bacteria, fungi, and plant species.

Nematicidal Effects of BVOCs

Table 1: The nematicidal potential of different biogenic volatile organic compounds (BVOCs) produced by various bacteria and fungi.

Microorganisms	VOCs	Nematodes	Reference
Pseudomonas putida 1A00316	Dimethyl disulfide, 2-nonanone 2-octanone (Z)-hexen-1-ol acetate 2-undecanone	*Meloidogyne incognita*	[1]
Pseudomonas koreensis T3GI1	3-methoxy-2,5-dimethyl pyrazine 1-undecene Dimethyl disulfide		[2]
Pseudomonas soli T13GI4	1-undecene Dimethyl disulfide		[2]
Paenibacillus polymyxa KM2501-1	furfural acetone 2-decanol 2-undecanone 2-undecano 2-decanone	*Meloidogyne incognita*	[2]
Pseudomonas koreensis T3GI1	3-methoxy-2,5-dimethyl pyrazine, 1-undecene, dimethyl disulfide	*Meloidogyne javanica*	[4]
Variovorax paradoxus T1GI1	dimethyl disulfide		[4]
Comamonas sediminis T13GI2	1-undecene, dimethyl disulfide		[4]

Pseudomonas monteilii T8GH4	1-undecene, dimethyl disulfide, 2-undecanone		[4]
Pseudomonas soli T13GI4	1-undecene, dimethyl disulfide		[4]
Daldinia concentrica	3-methyl-1-butanol 2-methyl-1-butanol 4-heptanone Isoamyl acetate		[5]
Metarhizium brunneum	1-octen-3-ol 3-octanone	*Meloidogyne hapla*	[6]
Pseudoalteromons marina H-42	Dimethyl disulphide, Dimethyl trisulphide	*Bursaphelenchus xylophilus*	Yu et al., (2015)
Vibrio atlanticus	dimethyl disulphid, dimethyl trisulphide, benzaldehyde, tert-butylamine		
Annulohypoxylon sp. FPYF3050	1,8-cineole		[7]
Trichoderma sp. YMF 1.00416	6-pentyl-2H-pyran-2-one		[8]

Table 2: The nematicidal potential of different biogenic volatile organic compounds (BVOCs) produced by various plant species.

Nematode	Plant Sources	Family	VOCs	References
M. incognita	*Agastache rugosa*	Lamiaceae	Methyleugenol, estragole, eugenol	[7]
	Rosmarinus officinalis		1,8-cineole, camphor, -pinene	[10]
	Thymus satureioides		Borneol, thymol	[10]
	Mentha spicata		Carvone	[11]
	Mentha pulegium		Menthofuran, Pulegone, Trans-anethole, Carvacrol	[11]
	Origanum vulgare		Carvacrol, Thymol, Terpinen-4-ol	[12]
M. javanica	*Origanum dictamnus*		Tarvacrol, Thymol, Terpinen-4-ol	[12]
Busaphelenchus xylophilus	*Origanum vulgare*	Lamiaceae	carvacrol, -terpinene, p-cymene	[3]
M. incognita	*Helianthus annuus*	Asteraceae	2,3-butanediol, sabinene, eucalyptol, limonene,	[3]
M. incognita *M. javanica*	*Tagetes minuta*	Asteraceae	dihydrotagetone, (Z)-ocimene, (E)-ocimenone	[12]
M. incognita	*Eucalyptus meliodora*	Myrtaceae	Trans-anethole, benzaldehyde	[13]
M. incognita	*Syzygium aromaticum*	Myrtaceae	eugenol	[14]
M. incognita	*Brassica juncea*	Brassicaceae	isothiocyanate	[15]
M. incognita	*Cymbopogon nardus*	Poaceae	Citronellal	[16]
B. xylophilus	*Cymbopogon citratus*	Poaceae	Geranial, Neral, -Myrcene	[17]

Mode of actions of BVOCs on PPN

Fumigant Action

Fumigants are volatile compounds that, upon application to the soil, transition into a gaseous phase. These vapours diffuse through the soil's pore space, spreading evenly. Certain compounds known as biogenic volatile organic compounds (BVOCs) have the ability to enter the nematode's body through natural openings like the excretory pore, anus, and mouth, as well as permeate through the nematode's cuticle. For the normal growth and development of nematodes, a series of enzymatic reactions are essential for proper cellular-level metabolic functions. Enzymes possess nucleophilic sites that readily share lone pairs of electrons, while BVOCs exhibit electrophilic sites with a high affinity for accepting electrons. Consequently, when enzymes and BVOCs come into contact, complex formations occur as the enzymes and BVOCs interact. This interaction leads to the inactivation of enzymes by BVOCs, thereby affecting biochemical pathways and protein synthesis in nematodes. As a result of enzyme inactivation, affected nematodes experience a disruption in their respiratory system, leading to hyperactivity, paralysis, and ultimately death. The detrimental effects of BVOCs on nematodes are manifested through the interruption of crucial biological processes, ultimately compromising their overall survival and well-being (Cheng et al., 2017) [2].

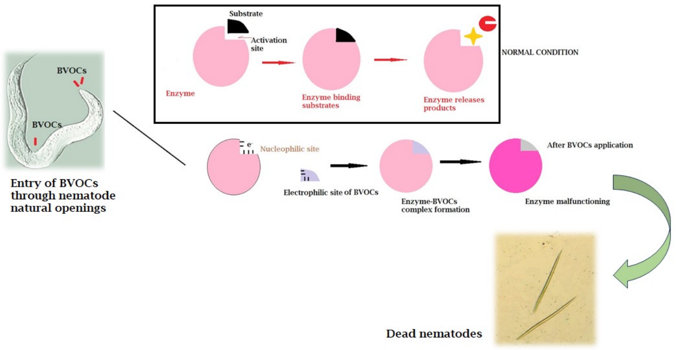

Figure 1: Fumigant action of BVOCs on nematodes in the soil.

Oxidative stress: Reactive Oxygen Species (ROS) are produced inside the worm's body during biochemical processes under normal circumstances without the presence of BVOCs. The right concentration of ROS is controlled by a variety of enzymes, including the primary endogenous enzymatic defense mechanisms of all aerobic Cells Are Catalase (CAT), Glutathione Reductase (GR), Glutathione Peroxidase (GPx), Superoxide Dismutase (SOD), and GPx. By superoxide radicals and hydrogen peroxide and converting them to less reactive species, they provide protection (Figure 2A). When the nematode is exposed to BVOCs, the electrophilic sites of the BVOCs bind to the nucleophilic sites of the enzymes, forming a complex with the enzymes. Endogenous enzymes were unable to scavenge radicals into less reactive species as a result of enzyme inactivation (Deng *et al*., 2022) [18]. Hence, the concentration of ROS inside the nematode gradually raises (referred to as oxidative stress), ultimately causing cell damage, shortened lifespan, and death of the nematode (Figure 2B).

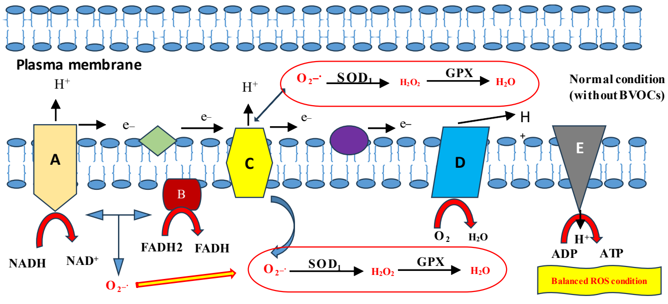

Figure 2A: Balanced ROS condition in the nematode's plasma membrane in the absence of BVOCs.

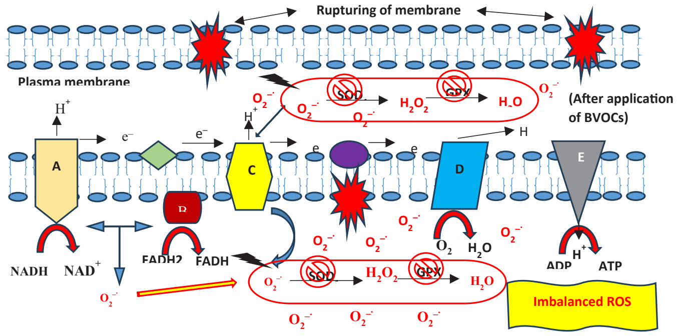

Figure 2B: Rupturing of the membrane due to excess ROS production after exposure of nematode to BVOCs.

Bio-fumigation

Volatile organic compounds having biocidal characteristics are produced as a result of the biological degradation of plant or animal tissues or byproducts. Brassicaceae plants are often used because they contain a lot of glucosinolates. Glucosinolates are converted during decomposition into volatile isothiocyanates, which are poisonous to soil organisms. more environmentally friendly and less hazardous than synthetic fumigants. Alternatively, sorghum can be employed, which results in the production of hydrogen cyanide with a similar outcome.

Mechanism of synthesis of biocidal isothiocyanates (ITCs)

On noxious soilborne pathogens, the Brassicaceae plant family is specifically responsible for the release of biocidal Isothiocyanates (ITCs) through the hydrolysis of glucosinolates (GSLs, thioglucosides) found in crop residues, which is catalysed by the isoenzyme myrosinase (MYR, thioglucoside glucohydrolase). The tissues of the plant sequester the hydrolysing enzyme MYR (stored in the cell wall or cytoplasm) and the secondary metabolite GSL (located in the cell vacuole). Sulfur-containing GSLs typically have low biological activity when present in their natural state, but when plant tissue is ruptured or lysed, GSL and MYR combine to form a number of biologically active volatile compounds, including ITCs (in high concentration), nitriles, epithionitriles, and thiocyanates (in low concentration) (Dutta *et al*., 2019) (Figure 1) [19].

Mode of action of Isothiocyanate (ITCs) The active sites of ITC or other volatiles react with the biological nucleophiles of the target nematodes, notably the thiol and amine groups of several enzymes, and become permanently alkylated. ITC could open a channel for DNA oxidative damage. According to in vitro studies, exposure to ITCs produced by brassica plants significantly decreases nematode motility, possibly as a result of the nematodes' reduced ability to find hosts. One study found that the exudates of brassica species may diminish the size of the dorsal pharyngeal gland nucleus in the potato cyst nematode (*Globodera rostochiensis*), potentially reducing the nematode parasitism of potatoes (Dutta *et al*., 2019) [19].

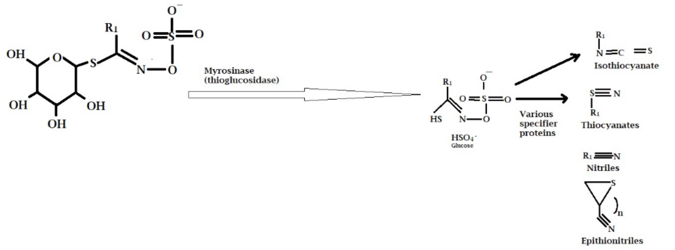

Figure 3: The biological breakdown of glucosinolates (Modified Dutta *et al*., 2019) [19].

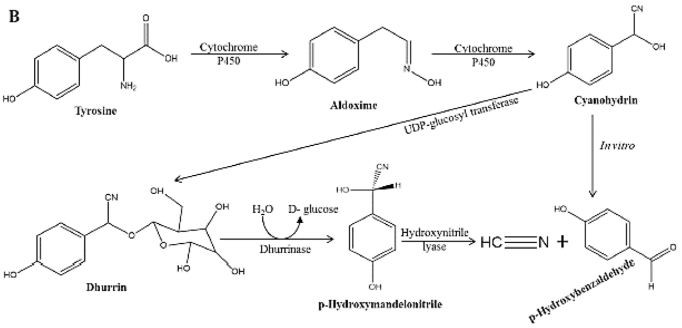

Figure 4: Dhurrin biosynthesis in sorghum and HCN production (Courtesy- Dutta *et al*.,2019) [19].

Non-brassica bio-fumigation

Tyrosine (L-amino acid), a precursor for the dhurrin biosynthesis in sorghum, is hydroxylated and transformed into aldoxime and cyanohydrin (nitrile) by the catalytic actions of cytochrome P450 enzymes. UDP-glucosyl transferase catalyzes the glycosylation of hydroxy nitriles to the cyanogenic glycoside, dhurrin. HCN is produced as a result of dhurrin's hydrolysis. (Dutta *et al*.,2019) (Figure 2) [19].

Photo-activation of α-terthienyl in *Tagetes spp*.

The roots of marigolds (Tagetes spp., Asteraceae or Compositae family) contain large amounts of alpha-terthienyl (thiophene - polyacetylenic sulphur compound). Reactive oxygen species, which are known to be phytotoxic against insects and nematodes both *in vitro* and *in vivo*, are formed when the chemical alpha-terthienyl is photoactivated with near ultraviolet light (325-400 nm) (Figure 3). In contrast, irradiation of alpha-terthienyl, which is necessary for nematicidal activity, may not occur in the rhizosphere in the absence of light. The growth of *Tagetes spp.* exclusively affects endoparasitic nematodes, thus it was thought that the alpha-terthienyl is triggered within the live root system by means other than light (Dutta *et al*.,2019) [19].

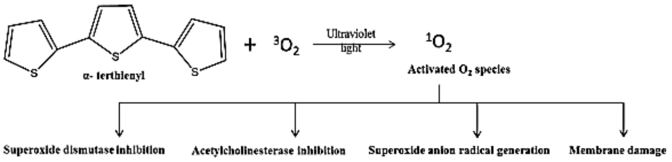

Figure 5: Photo-activation of α-terthienyl in Tagetes spp. And releases ROS molecules (Courtesy- Dutta *et al*.,2019) [19].

Analytical determination of VOCs

a. Sampling of VOCs

Solid phase microextraction

Principle: Using the SPME method, which is based on the partition mechanism theory, the analyte equilibrium is created between the sample matrix and the polymeric phase on the fibre.

The amount of analyte extracted by the coated surface is determined by the Nernst distribution law. The amount of analyte absorbed by the coating at equilibrium is directly proportional to the concentration of the analyte in the sample.

The two processes in the sample preparation process used by SPME are sample extraction and sample desorption. During the extraction stage, the analyte is adsorbed onto a coated silica fiber's surface. The fibre is retracted into the needle once balance has been attained, and the needle is released from the septum. Once the needle is inserted into the device's injection port, the target analytes are desorbed to the analytical instrument during the desorption process.

b. Desorption of the sample into a gas chromatogram

In analytical chemistry, Gas Chromatography (GC) is a popular method of chromatography used to separate and analyse substances that can be vaporised without decomposing.

Chromatogram

The chromatograph's output in graphic form. Different peaks or patterns on the chromatogram represent the various components of the separated mixture in the case of ideal separation. The concentrations of specific mixture components are shown by the area beneath the peak.

Future prospects of biogenic VOCs

Proper identification of biogenic plants that emit VOCs, mass production of VOCs-producing microbes and its commercial formulation making are the basis idea for future success as a management strategy to control the plant parasitic nematode. Efficient extraction techniques of VOCs are yet to be identified and a better understanding of the mode of action of VOCs on plant parasitic nematodes is necessary. Identifying nematode-specific VOCs is the future challenge and further residual toxicity of VOCs in the soil is mandatory to know the negative impacts on natural enemies and beneficial microorganisms. Above mentioned aspects are mainly focused to achieve the betterment of agriculture and sustainability in the agro-ecosystem.

Conclusion

VOCs offer a promising avenue for the development of environmentally friendly and sustainable approaches for controlling plant parasitic nematodes. The negative impact of biogenic VOCs on humans and animals is comparatively less when compared to the chemical nematicide alone. Therefore, biogenic VOCs could be the future eco-friendly managemental strategy to manage plant parasitic nematodes below the economic threshold level of damage.

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