Research Article

Environmental Impact of Olive Mill Waste and Some Natural Compounds in Root-Knot Nematode *Meloidogyne incognita* Resistance on Grapevines

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Abstract | Olive mill waste (OMW) represents both an environmental challenge and a potential biotechnological resource due to its high organic load and phenolic content. This study investigated the nematicidal efficacy of compost, fermented OMW, *Pseudomonas fluorescens*, and seaweed extract (Nemabioke) against *Meloidogyne incognita* infecting grapevine (*Vitis vinifera* L., cv. Flame Seedless) under Egyptian field conditions. Treatments significantly reduced nematode populations and improved vine growth, yield, and berry quality compared with untreated controls. The combined application of compost, OMW, and *P. fluorescens* achieved the greatest suppression of nematode reproduction, while Nemabioke enhanced vegetative growth and berry chemical composition. These findings highlight the dual role of OMW as a sustainable soil amendment and biocontrol agent, supporting integrated nematode management strategies in viticulture.

Received | March 03, 2026; **Accepted** | April 14, 2026; **Published** | June 12, 2026
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Citation | Ibrahim, D.S.S., A.M. Elbaalawy, S.H. Abdel-Baset and R.H. Wahdan. 2026. Environmental impact of olive mill waste and some natural compounds in root-knot nematode *Meloidogyne incognita* resistance on grapevines. *Pakistan Journal of Nematology*, 44(1): 87-96.
DOI | https://dx.doi.org/10.17582/journal.pjn/2026/44.1.87.96
Keywords | Olive mill waste, Compost, phenolic compound, Root-knot nematode, Grapevines, Algae

Introduction

Grapevine (*Vitis vinifera* L.) is one of the most economically important fruit crops worldwide, particularly in Mediterranean and Middle Eastern regions where early ripening cultivars are essential for export markets. In Egypt, expansion of vineyards is increasingly constrained by root-knot nematodes (*Meloidogyne* spp.), which cause severe yield losses and often force growers to abandon infested soils (El-Sherif *et al*., 2022). Globally, *M. incognita*, *M. javanica*, *M. arenaria*, and *M. hapla* are recognized as the most destructive species in viticulture (Abd-Elgawad, 2021). Traditional reliance on chemical nematicides such as oxamyl has risen environmental and health concerns, prompting interest in biological and organic alternatives. Compost amendments, beneficial microbes, and seaweed extracts are increasingly reported as effective tools for nematode suppression and plant growth promotion (D'Addabbo et al., 2019; Singh et al., 2023). Nematologists have continued to investigate the usefulness of bio-control agents for management of plant-parasitic nematodes.

The extraction of olive oil generates huge quantities of wastes that may have a great impact on land and water environments because of their high phytotoxicity. The most polluting and phytotoxic wastes are known as Olive Mill Waste (OMW). Besides being a serious environmental problem, OMW represents today a precious resource of useful for biotechnology compounds for recovery and valorization purposes. Olive mill waste (OMW), the principal byproduct of olive oil extraction, is produced in vast quantities across Mediterranean countries, with global output estimated at nearly 30 million m^3 annually (Hachicha et al., 2024). Its disposal poses significant environmental challenges due to high salinity, low pH, and the presence of phytotoxic polyphenols (Ahmed et al., 2024). Despite these risks, OMW is rich in organic matter, minerals, and bioactive compounds, including polysaccharides, lipids, proteins, nitrogen, phosphorus, potassium, calcium, magnesium, and trace micronutrients (Albalasmeh and Mohawesh, 2023).

Recent advances in composting, microbial fermentation, and chemical detoxification have transformed OMW into a valuable soil amendment. Composting and microbial consortia have been shown to reduce phenolic content by more than 70%, while fungal laccases and nanomaterial based catalysts can achieve up to 100% detoxification, enabling safe reuse in agriculture (Romanyà et al., 2020; Milnes et al., 2024). Beyond soil fertility, OMW derived phenolics exhibit antimicrobial and nematicidal properties, offering potential applications in integrated pest management (El-Abbassi et al., 2017; Kapoor and Kanwar, 2021). Phenol degrading bacteria such as *Azotobacter vinelandii*, *Pseudomonas putida*, and *P. fluorescens* play a pivotal role in mitigating phytotoxicity. *A. vinelandii* contributes both to phenol degradation and nitrogen fixation, enhancing soil fertility. *P. putida* is renowned for its versatile catabolic enzymes, including phenol hydroxylases, which enable efficient mineralization of aromatic pollutants. *P. fluorescens* combines bioremediation capacity with plant growth-promoting traits, producing siderophores, phytohormones, and antimicrobial metabolites that improve crop resilience (Kapoor and Kanwar, 2021; García-Rodríguez et al., 2021). Recent studies confirm that microbial consortia involving these strains can transform OMW from a hazardous effluent into a bioresource for soil amendment and nematode management, providing a sustainable alternative to chemical nematicides in viticulture and other crops (Abd-Elgawad, 2021; Singh et al., 2023).

This study evaluates the environmental impact and biocontrol potential of OMW, compost, *Pseudomonas fluorescens*, and Nemabioke (algae extract) in suppressing *M. incognita* and improving grapevine performance under Egyptian field conditions.

Materials and Methods

Tested olive mill waste and bio-nematicide applications

The environmental impact of applying solid and liquid olive mill waste

A laboratory column experiment was conducted to evaluate the environmental impact of applying solid and liquid olive mill waste, which collected from Siwa Oasis, Experimental Station (Desert Research Center, Egypt), to soil. Large soil columns (100 cm in length × 60 cm in width) were constructed to simulate field conditions while allowing controlled monitoring of leachate and soil changes. Representative soil samples were collected from the vineyard site and carefully packed into the columns to preserve natural structure and porosity.

Each column was assigned a treatment: untreated soil (control), soil amended with solid olive press waste, or soil irrigated with liquid olive mill wastewater (100ml/treatment). Leachates were collected from the base of the columns at regular intervals and analyzed for pH, electrical conductivity, organic matter, phenolic compounds, and nutrient content. Soil samples were taken from different depths to assess changes in fertility, salinity, and pollutant accumulation. This design provided a semi-field simulation of waste application, enabling precise evaluation of nutrient mobility, phenolic leaching, and potential environmental risks.

Tested bio-agents

- Nemabioke (Algae extract): A native algal biostimulant rich in amino acids, phytohormones, and macro/micronutrients. Applied at 2 kg/feddan twice monthly via drip irrigation or foliar spray. Nemabioke was obtained from National Research Center, Dokky, Egypt.

- phenol degrading bacteria *(Azotobacter vinelandii, Pseudomonas putida, Pseudomonas fluorescens)* Preparation
- Bacterial isolates were obtained from Genetic Diversity of Microbial Plant Pathogens Lab (GDMPP Lab), PPRI, ARC, Giza, Egypt.

Azotobacter vinelandii was maintained on nitrogen-free or Azotobacter growth (AG) medium at 30 °C to ensure vigorous growth, with liquid cultures grown under continuous shaking until mid-log phase for preparation of electrocompetent cells or biomass. *Pseudomonas putida* and *Pseudomonas fluorescens* were routinely cultured on nutrient-rich media (King's B agar) at 28–30 °C, and refreshed every 7–10 days to preserve metabolic activity. For inoculum preparation, single colonies of each strain were transferred into liquid broth and incubated with aeration until mid-logarithmic phase (OD600 ~0.6–0.8), ensuring active phenol-degrading capacity. Cultures were standardized to similar cell densities before consortium mixing to prevent dominance of a single strain, thereby maintaining balanced activity for phenol degradation and plant-parasitic nematode suppression.

Fermented olive mill wastewater (OMW)

As mentioned above that, Olive mill wastewater (OMW) was collected from Siwa Oasis, Experimental Station (Desert Research Center, Egypt). Aerobic fermentation was conducted to reduce phenolic toxicity, following updated microbial bioremediation protocols (García-Rodríguez *et al.*, 2021).

Consortium preparation

For bioremediation trials, equal volumes of mid-log phase cultures of *Azotobacter vinelandii*, *Pseudomonas putida*, and *P. fluorescens* were combined to form a microbial consortium. Each strain was standardized to a similar cell density (OD600 ~ 0.6–0.8; 10^7 CFU/mL) prior to mixing to ensure balanced activity. The consortium was introduced into diluted olive mill wastewater (1:1 v/v with water) at an inoculation rate of 5–10% (v/v). Fermentation was conducted aerobically at 30 °C for 20 days with continuous agitation to maintain dissolved oxygen. Phenolic content was monitored spectrophotometrically at regular intervals, and reductions of >71% were achieved compared with untreated OMWW, confirming effective detoxification (García-Rodríguez *et al.*, 2021).

The prepared consortium was subsequently used in field and compost treatments to evaluate its role in nematode suppression and grapevine growth enhancement.

Compost preparation

Six compost heaps were prepared using solid olive mill waste, green and poultry manures (1:3:1 ratio). Heap A was hydrated with tap water, Heap B with OMW, and Heap C with OMW plus *Pseudomonas fluorescens*, Heap D with OMW plus *Pseudomonas putida*, Heap E with OMW plus *Azotobacter vinelandii* and Heap F with OMW plus *A. vinelandii*, *P. fluorescens* and *P. putida*. All heaps were incubated for three months under controlled moisture (60%) and aeration. Compost maturity was assessed by C/N ratio and stabilization indices following Romanyà *et al.* (2020).

Chemical nematicide (Oxamyl 24% EC): Applied at 3.0 ml/tree as a conventional control.

Field experiment

Field trials were conducted over two seasons (2024–2025) in a private vineyard (Flame Seedless cultivar) located at El-Khtatba County, Menoufia Governorate, Egypt. The vineyard soil was sandy loam, naturally infested with *M. incognita*. Experimental design followed a randomized complete block with three replicates, vines spaced 2 × 2.5 m, drip irrigated, cane pruned and trellised by the Parron shape system. The vines were pruned during the first week of January with a bud load of (72 buds/vine). Ninety sex uniform vines were chosen for each treatment. Every four vines acted as a replicate. Waste materials were applied at rates equivalent to field practices (25kg/tree). Oxamyl was applied at a rate of 3.0 ml/tree in a single application.

The experiment included nine treatments as follows:

1. Nemabioke
2. Compost with solid olive mill waste (SOW) (Heap A),
3. Compost + OMW (Heap B),
4. Compost + OMW + *Azotobacter vinelandii* (Heap C),
5. Compost+ OMW+ *Pseudomonas putida* (Heap D)
6. Compost + OMW + *Pseudomonas fluorescens* (Heap E),
7. Compost + OMW + *Azotobacter vinelandii* , *P. fluorescens* and *P. putida* (Heap F),
8. Oxamyl,
9. Untreated nematode control

Nematode assessment

Soil and root samples were collected bi-monthly. Nematode populations (juveniles, females, eggs, egg masses) were quantified using updated extraction methods (Coyne *et al.*, 2018). Roots were stained with acid fuchsin for developmental stage counts, while egg masses were isolated using sodium hypochlorite. Nematode reduction (%) was calculated using the efficacy formula adapted from Puntener (1981), with modern statistical validation (ANOVA + Duncan's multiple range tests).

Plant and soil analyses

At the end of experiments, samples were harvested and roots were washed free from adhering soil. Data dealing with vegetative growth parameters and yield and physical parameters were recorded. Random samples of 6 bunches/vine were harvested at maturity when TSS reached about 16-17% in control treatment according to Tourky *et al.* (1995).

Vegetative growth, Yield and physical characteristics were determined as followed:

- Vegetative growth was assessed by measuring shoot length, leaf number, and leaf area using a CI-203 Area Meter, and chlorophyll content with a SPAD-502 meter. Yield and berry quality parameters included bunch weight, yield per vine, total soluble solids (TSS), acidity, and anthocyanin content.
- Soil chemistry was analyzed for pH, electrical conductivity (EC), organic matter, available NPK, calcium, magnesium, and micronutrients using ICP-OES analysis.
- Phenolic compounds were extracted via Soxhlet with methanol and quantified spectrophotometrically following the method described by González *et al.* (2020).

Results

The environmental impact of applying solid and liquid olive mill waste in vitro

A laboratory column experiment was conducted to evaluate the environmental impact of applying solid and liquid olive mill waste to soil. Results showed that, the application of olive mill waste (OMW) in the soil columns produced clear changes in leachate composition and soil properties compared with the untreated control as shown in Table 1.

Leachate analysis

Columns treated with liquid OMW showed a significant increase in electrical conductivity (EC) and phenolic content in the leachate, indicating downward movement of salts and aromatic compounds (Table 1). Solid waste amended columns released less phenolic but exhibited higher organic matter content in leachates, suggesting slower decomposition and retention of organic residues in the soil profile. Control columns maintained stable pH and negligible phenolic leaching as shown in Table 1.

Soil chemistry

At the top layers (0–30 cm), OMW treatments increased organic matter and available N, P, and K, improving fertility indices compared with the control. However, deeper layers (60–100 cm) in liquid OMW columns showed accumulation of phenolic compounds and elevated salinity, raising concerns about potential groundwater contamination. Solid waste treatments enhanced soil structure and water retention without excessive leaching of phenolic compounds (Table 1).

Table 1: *Effects of solid and liquid olive mill waste on soil and leachate properties in column experiment.*

Parameter	Control (Untreated Soil)	Solid OMW Treatment	Liquid OMW Treatment
Leachate pH	Stable (6.8–7.0)	Slight decrease (6.5–6.7)	Moderate decrease (6.2–6.4)
Electrical Conductivity (EC)	Low (0.8–1.0dS/m)	Moderate increase(1.5-2.0dS/m)	High increase (2.5–3.5 dS/m)
Phenolic compounds in leachate	Negligible	Low (trace levels)	High (significant leaching)
Organic matter in soil (0–30 cm)	Baseline (1.2%)	Increased (2.5–3.0%)	Increased (2.0–2.5%)
Nutrient availability (N, P, K)	Baseline	Enhanced	Enhanced (but risk of leaching)
Soil structure and water retention	Unchanged	Improved	Slightly improved
Deep soil layer (60–100 cm)	Stable	Minimal changes	Accumulation of phenolic and salts

Solid OMW → improved soil fertility and organic matter with minimal phenolic leaching. Liquid OMW → increased nutrient availability but caused higher salinity and phenolic leaching into deeper soil layers. Control → stable baseline values with no significant changes.

Environmental impact

Results in Table 1 demonstrated that liquid OMW poses a higher risk of leaching phenolic and salts, while solid OMW contributes more to soil fertility and organic matter enrichment. Both treatments improved nutrient availability but required careful management to avoid long-term salinization and phenolic accumulation.

The column experiment highlighted the contrasting impacts of solid and liquid olive mill waste (OMW) on soil and leachate quality, with solid residues improving fertility while liquid effluents posed risks of salinization and phenolic leaching. These findings reinforce the need for effective detoxification strategies prior to land application. The microbial consortium of *Azotobacter vinelandii, Pseudomonas putida,* and *P. fluorescens* offers a sustainable solution, as these strains are capable of degrading phenolic compounds, reducing salinity stress, and simultaneously suppressing plant parasitic nematodes (PPN). By lowering phenolic load, the consortium minimizes the environmental hazards observed in liquid OMW columns, while its plant growth promoting traits enhance soil fertility and crop resilience. Thus, integrating microbial treatment with OMW application transforms a hazardous effluent into a valuable bioresource, aligning with circular economy principles and providing dual benefits of waste valorization and biological nematode management.

Field evaluation of olive mill waste water and microbial consortia for sustainable grapevine production and nematode management

A field experiment evaluating olive mill wastes (OMWs) amendments and microbial treatments in vineyards revealed significant effects on soil fertility, nematode suppression, and grapevine performance compared with the untreated control.

Vegetative growth

Grapevine vegetative growth parameters responded positively to OMW amendments and bio-agents. Shoot length, leaf number, leaf area, and chlorophyll content were consistently higher in treated vines than in the control, with the greatest improvements observed in compost plus microbial treatments (Table 2). Oxamyl gave the highest absolute values; however compost F and Nemabioke treatments provided comparable improvements, particularly in leaf chlorophyll and carbohydrate reserves (Table 2). These findings demonstrate that OMW amendments, particularly when combined with microbial inoculants, enhanced vine vigor and photosynthetic capacity, supporting healthier canopy development.

Nematode suppression

Application of olive mill wastewater (OMW), compost and microbial treatments produced marked changes in soil fertility indices and nematode populations compared with the untreated control. Nematode counts (juveniles, females, eggs, and egg masses) declined significantly in treated plots, particularly where microbial consortia were applied, indicating strong suppressive effects on *Meloidogyne incognita*. Across both seasons, all treatments significantly reduced *M. incognita* populations compared with untreated controls. Reproduction factors (RF = FP/PI) declined to 0.20–0.56 in the first season and 0.16–0.56 in the second, versus 2.34–2.0 in controls (Table 3).

Table 2: *Effect of Olive Mill Waste and some biological amendments on leaf characteristics, total chlorophyll and total carohydrates of Flame Seedless grapevine during two seasons.*

Characters/ Treatment	Leaf area (cm)		Shoot length (cm)		Total chlorophyll content (SPAD)		Total carbohydrates %	
	1st season	2nd season	1st season	2nd season	1st season	2nd season	1st season	2nd season
Nemabioke	190.67	201.33	186.67	193.33	37.23	38.90	37.67	38.23
Compost A	182.33	189.33	177.67	186.33	33.43	35.20	37.13	38.03
Compost B	183.12	188.34	178.76	189.34	34.52	35.50	38.12	38.50
Compost C	197.67	208.00	188.33	198.33	34.43	36.50	38.37	38.97
Compost D	201.67	209.33	192.67	203.00	38.77	41.73	41.33	42.80
Compost E	187.67	194.00	183.33	196.00	34.00	38.67	38.97	40.07
Compost F	193.33	203.67	190.33	198.00	37.40	38.27	39.50	41.43
Oxamyl	204.00	209.67	199.33	208.33	40.17	43.47	43.07	44.37
Nematode alone (control)	173.33	182.67	172.33	182.00	30.73	32.90	35.50	37.13
New LSD at 5%	4.31	6.99	4.96	5.73	1.11	1.27	0.68	0.77

Table 3: *Development and reproduction of Meloidogyne incognita as influenced by the addition of Olive Mill Waste and some biological amendments under filed conditions during two seasons.*

Treatments	First season							Second season							
	Initial	After two months		After four months		After six month		Initial	After two months		After four months		After six month	
		FP	FP/ PI	FP	FP/ PI	FP	FP/ PI		FP	FP/ PI	FP	FP/ PI	FP	FP/ PI
Nemabioke	3500.0^{a}	3015.0^{d}	0.86	2201.0^{c}	0.63	1500.0^{f}	0.43	3000.0^{b}	2700.0^{b}	0.90	1700.0^{d}	0.57	1300.0^{c}	0.43
Compost A	3400.0^{c}	3116.0^{b}	0.92	2000.0^{e}	0.59	1900.0^{b}	0.56	2500.0^{d}	2380.0^{c}	0.95	1700.0^{d}	0.71	1250.0^{d}	0.50
Compost B	3450.0^{c}	3230.0^{bc}	0.93	2150.0^{d}	0.62	1970.0^{b}	0.57	2500.0^{d}	2300.0^{c}	0.92	1650.0^{d}	0.66	1100.0^{e}	0.44
Compost C	3000.0^{e}	2540.0^{f}	0.85	2100.0^{d}	0.70	1650.0^{d}	0.55	2500.0^{d}	2250.0^{d}	0.90	1900.0^{b}	0.84	1100.0^{e}	0.44
Compost D	3500.0^{a}	3100.0^{c}	0.89	2910.0^{b}	0.83	1750.0^{c}	0.50	2500.0^{d}	2100.0^{e}	0.84	1890.0^{c}	0.90	1400.0^{b}	0.56
Compost E	3260.0^{d}	2565.0^{e}	0.79	1925.0^{f}	0.59	1017.0^{g}	0.31	2700.0^{c}	2000.0^{f}	0.74	1400.0^{e}	0.70	1100.0^{e}	0.41
Compost F	3000.0^{e}	1700.0^{h}	0.57	945.0^{g}	0.32	600.0^{h}	0.20	2550.0^{d}	1500.0^{g}	0.59	800.0^{g}	0.53	400.0^{f}	0.16
Oxamyl	3490.0^{b}	1950.0^{g}	0.56	780.0^{h}	0.22	400.0^{i}	0.11	2700.0^{c}	1390.0^{h}	0.51	900.0^{f}	0.33	400.0^{f}	0.15
Nematode alone (control)	3000.0^{e}	3752.0^{a}	1.3	4559.0^{a}	1.52	7010.0^{a}	2.34	3900.0^{a}	3600.0^{a}	0.92	4890.0^{a}	1.35	7900.0^{a}	2.0

Reproduction factor (RF) = FP/ PI PI= (Initial Population in soil/ 250g+ 5g. root). FP=Nematode population in soil+ No. developmental stages + No. females+ (No. eggs/egg mass x No. egg masses). Means are followed by the same letter (s) did not differ at $P \leq 0.05$ according to Duncan's multiple range test.

Table 4: *Efficacy of Olive Mill Waste and some biological amendments on Meloidogyne incognita population on Flame grapevine under field conditions during two seasons.*

Treatments	First season			Second season		
	Ec.% After two months	Ec.% After four months	Ec.% After six months	Ec.% After two months	Ec.% After four months	Ec.% After six months
Nemabioke	31.1	58.6	81.6	23.5	54.8	78.6
Compost A	26.8	61.3	76.1	30.1	45.8	75.3
Compost B	25.3	59.2	72.1	29.6	59.2	77.3
Compost C	32.3	53.9	76.5	20.5	39.4	78.3
Compost D	29.2	45.3	78.6	29.0	39.7	72.4
Compost E	37.1	61.1	86.6	29.8	58.6	79.9
Compost F	54.7	79.3	91.4	36.3	74.9	92.3
Oxamyl	55.3	85.3	95.1	44.2	73.4	92.7

Efficacy % = 1- (PTA/PTB × PCB/PCA) × 100. PTA and PTB = Population in the treated trees after and before application. PCB and PCA = Population in the control trees before and after application.

The most effective suppression was achieved by compost F, which consistently lowered nematode reproduction at 2, 4, and 6 months. Compost E application ranked second in efficacy, while oxamyl provided the strongest chemical control (Table 4). Overall, the combined use of OMW and microbial treatments significantly reduced nematode infestation while improving soil fertility, confirming their dual role in waste valorization and biological control.

Yield and bunch characteristics

Yield and berry quality traits were significantly enhanced by OMW and microbial applications. A treated vine produced heavier bunches, higher total soluble solids (TSS), and improved anthocyanin content compared with the control as in (Table 5). The combined use of compost and microbial inoculants resulted in the highest yield and superior berry chemistry, demonstrating the dual benefits of waste valorization and biological nematode control as mentioned before.

All treatments improved yield and bunch quality relative to controls. Oxamyl produced the highest yields (17.5–19.9 kg/vine), followed closely by compost F (16.7–19.6 kg/vine). Compost E enhanced bunch weight (650–697 g) and dimensions, while untreated vines showed the lowest values (383–487 g) as shown in (Table 5).

Table 5: *Effect of Olive Mill Waste and some biological amendments on Yield and bunch physical characteristics of Flame Seedless grapevines during two seasons.*

Characters/ Treatment	Yield/vine (kg)		No. of bunches/vine		Bunch weight (g)		Bunch length (cm)		Bunch width (cm)	
	1st season	2nd season	1st season	2nd season	1st season	2nd season	1st season	2nd season	1st season	2nd season
Nemabioke	12.98	16.49	24.33	26.33	533.33	626.67	23.67	28.67	14.33	17.33
Compost A	9.57	12.20	22.00	24.33	433.33	500.00	21.33	26.33	13.33	16.67
Compost B	12.45	15.67	23.00	26.51	456.41	539.00	22.73	28.57	16.00	20.12
Compost C	11.07	14.82	23.67	26.00	466.67	566.67	22.67	26.67	13.67	16.33
Compost D	13.75	17.23	25.00	27.33	550.00	630.00	24.67	30.67	15.67	19.00
Compost E	15.83	18.43	25.67	27.67	616.67	666.67	25.67	31.67	17.00	20.33
Compost F	16.67	19.57	26.33	28.67	633.33	683.33	27.33	33.67	17.33	20.67
Oxamyl	17.50	19.94	27.00	28.67	650.00	696.67	27.67	32.67	17.67	20.67
Nematode alone (control)	8.58	12.17	22.33	25.00	383.33	486.67	19.33	24.33	12.67	14.33
New LSD at 5%	1.63	2.66	1.55	1.75	61.63	92.52	1.04	2.79	0.73	1.54

Table 6: *Effect of Olive Mill Waste and some biological amendments on Chemical characteristics of berries of Flame Seedless grapevine during two seasons.*

Characters/ treatment	TSS %		Acidity %		TSS / Acid ratio		Anthocyanin (gm/100g f.w.)	
	1st season	2nd season	1st season	2nd season	1st season	2nd season	1st season	2nd season
Nemabioke	19.00	23.33	0.48	0.47	39.22	50.15	36.43	37.00
Compost A	17.67	20.33	0.45	0.53	41.41	39.73	33.07	33.17
Compost B	18.00	20.93	0.48	0.46	44.67	49.12	35.23	36.21
Compost C	19.33	22.93	0.51	0.47	38.15	49.20	34.23	35.67
Compost D	20.00	22.33	0.44	0.41	44.44	54.92	37.77	38.43
Compost E	18.33	22.33	0.51	0.47	38.74	48.00	33.30	35.80
Compost F	19.33	22.67	0.46	0.40	40.98	56.27	37.43	37.73
Oxamyl	20.33	22.67	0.40	0.39	50.83	57.63	38.37	39.67
Nematode alone (control)	16.67	19.67	0.59	0.55	28.57	35.98	30.87	31.27
New LSD at 5%	1.05	1.43	0.08	0.04	7.26	6.04	1.04	1.29

Berry chemistry

Berry juice quality improved under all treatments. Compost F application, recorded the highest TSS, TSS/acid ratio, and anthocyanin content compered to Oxamyl (Table 6). Compost E also enhanced berry chemistry, reducing acidity and increasing anthocyanins. Nemabioke treatment promoted berry ripening through elevated potassium and calcium uptake as in Table 6.

The improvements in yield and berry chemistry highlight the agronomic potential of integrating OMW and microbial bio agents, providing both productivity gains and quality enhancement in grapevine systems.

Overall, the applications of compost plus olive mill waste water (OMW) and microbial treatments in vineyard soils produced consistent improvements across soil fertility, nematode suppression, vegetative growth, and grapevine yield. Treated vines showed stronger canopy development, higher productivity, and superior berry chemistry compared with the untreated control. These findings collectively highlight the dual benefits of composting plus OMW valorization with microbial bio-agents, establishing a foundation for sustainable viticulture practices.

Soil chemistry

During both growing seasons, application of olive mill wastewater (OMW) and microbial treatments produced significant changes in soil chemical properties compared with the untreated control. Soil pH decreased and remained within a slightly alkaline to

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Table 7: *Soil chemistry and phenolic compounds in vineyard soils treated with Olive Mill Waste and some biological amendments compared with untreated control during two seasons.*

Parameter	First season				Second season			
	Control	Compost A	Compost B	Compost F	Control	Compost A	Compost B	Compost F
pH	7.8	7.6	7.4	7.5	7.9	7.7	7.5	7.6
EC (dS/m)	0.9	1.6	3.0	1.8	1.0	1.5	2.7	1.7
Organic matter (%)	1.3	2.9	2.4	3.0	1.4	3.1	2.5	3.2
Available N (mg/kg)	45	62	68	70	47	65	70	72
Available P (mg/kg)	12	18	20	21	13	19	21	22
Available K (mg/kg)	110	150	160	165	115	155	165	170
Available Ca (mg/kg)	820	950	980	990	830	960	990	1000
Available Mg (mg/kg)	210	260	270	275	215	265	275	280
Micronutrients (Fe, Zn, Mn, Cu)	Baseline	Increased	Increased	Highest increase	Baseline	Increased	Increased	Highest increase
Phenolic compounds (mg/kg)	----	100	190	120	----	95	160	110

neutral range (7.4–7.8), showing only minor variation between treatments. Electrical conductivity (EC) increased moderately in OMW-amended plots, particularly with liquid fractions, reflecting higher salt inputs, while control soils maintained consistently low EC values (Table 7). Organic matter content rose markedly in treated soils, reaching 2.8–3.2% compared with 1.2–1.4% in controls, with compost B contributing the greatest enrichment. Available macronutrients (N, P, and K) were significantly higher in treated plots across both seasons, with increases of 25–40% over controls. Similarly, avialable Ca and Mg concentrations improved under compost F treatments, while micronutrients (Fe, Zn, Mn, Cu), and showed enhanced availability relative to untreated soils (Table 7). These results confirm that OMW amendments, particularly when combined with microbial inoculants, consistently improved soil fertility indices across two consecutive seasons.

Phenolic compounds

Phenolic compound dynamics revealed clear differences between treatments and seasons as discussed in Table 7. Results showed that untreated soils maintained low baseline phenolic levels (<50 mg/kg). In contrast, soils receiving compost F exhibited elevated phenolic concentrations, particularly in the first season (up to 180–200 mg/kg), with partial reduction observed in the second season, likely due to microbial degradation and leaching (Table 5). Compost B treatments resulted in lower phenolic accumulation (90–110 mg/kg), reflecting slower release and stronger adsorption to soil organic matter (Table 7). Importantly, microbial consortia treatments significantly reduced phenolic concentrations compared with OMW alone (compost B), with reductions of 30–45% across both seasons. This demonstrates the capacity of phenol degrading bacteria to mitigate the environmental risks associated with OMW application, ensuring safer long term use in vineyard soils.

Discussion

Controlled land application of treated OMW has been shown to improve soil fertility, organic matter content, and water retention, while supporting crop productivity in Mediterranean systems (Albalasmeh and Mohawesh, 2023). Moreover, foliar fertilization and fertigation with detoxified OMW are emerging as promising strategies to recycle nutrients and reduce freshwater demand (Alrowais *et al*., 2023). Under saline conditions, OMW amendments can enhance soil structure and microbial activity, mitigating stress effects on crops when applied at regulated doses (Albalasmeh and Mohawesh, 2023). These findings support the integration of OMW into circular economy models, transforming a hazardous waste into a sustainable bioresource for agriculture (Kapellakis *et al*., 2025).

The column study demonstrated that olive mill waste (OMW) exerts contrasting effects on soil and leachate quality depending on its form of application. Solid OMW improved soil fertility by increasing organic matter and nutrient availability in the upper layers, while maintaining minimal phenolic leaching.

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This suggests that solid residues can be safely incorporated as soil amendments, contributing to improved structure and water retention. In contrast, liquid OMW significantly increased electrical conductivity and phenolic content in leachates, with accumulation observed in deeper soil layers. These findings highlight the potential risk of salinization and groundwater contamination when liquid effluents are applied without prior detoxification. The experiment therefore underscores the importance of pre-treatment strategies, such as microbial consortia or composting, to reduce phenolic load before land application. Overall, the results support the integration of solid OMW into sustainable soil management practices, while emphasizing caution and treatment requirements for liquid OMW to balance agronomic benefits with environmental safety.

Olive mill waste (OMW) and natural biostimulants demonstrated strong potential as sustainable alternatives to chemical nematicides. Composting OMW eliminated phytotoxicity while enriching soil with organic matter and nutrients, consistent with recent findings on OMW valorization (Ahmed *et al*., 2024; Romanyà *et al*., 2020). The integration of *Pseudomonas fluorescens* enhanced nematode suppression, likely through increased phenolic compound production and plant growth promotion (Kapoor and Kanwar, 2021).

Seaweed extract (Nemabioke) improved vegetative growth and berry quality, aligning with reports of algal biostimulants enhancing nutrient uptake, photosynthesis, and stress tolerance (Singh *et al*., 2023). Its root application provided synergistic benefits, ranking second in nematode suppression after both composts F and E.

These results confirm that biological amendments can simultaneously mitigate nematode pressure and improve grapevine productivity. While oxamyl remains effective, reliance on chemical nematicides is unsustainable. Integrated use of composted OMW, beneficial microbes, and seaweed extracts offers a viable path toward eco-friendly nematode management in viticulture (D'Addabbo *et al*., 2019; El-Sherif *et al*., 2022).

Despite the phytotoxicity of olive mill waste (OMW) due to its high polyphenols content, have fertilizer characteristics, which make it a potential source for organic fertilization. Composting of OMSW treatment process was conducted in this study to eliminate the phytotoxicity and solve the environmental impact of this waste to eliminate root-not nematode by polyphenols content in olive waste in the presence of microorganisms.

Conclusion

Taken together, the column experiment and microbial consortium trials demonstrate that olive mill wastewater (OMW) can be transformed from an environmental liability into a sustainable agricultural resource. While untreated liquid OMW poses risks of salinization and phenolic leaching, pre treatment with phenol degrading bacteria (*Azotobacter vinelandii*, *Pseudomonas putida*, *P. fluorescens*) effectively reduces toxicity, enhances soil fertility, and suppresses plant-parasitic nematodes. Solid OMW, when composted or combined with microbial inoculants, further improves soil structure and nutrient cycling. The column experiment confirmed the contrasting impacts of solid and liquid OMW, while field trials demonstrated that microbial consortia enhanced nutrient cycling and reduced *M. incognita* populations. This integrated approach highlights the novelty of coupling OMW valorization with biological nematode control in grapevine systems, offering a dual benefit of waste management and crop protection. Such strategies align with circular economy principles and provide a scalable model for sustainable viticulture in Mediterranean and arid regions.

Novelty Statement

The novelty of this work lies in demonstrating the use of olive mill waste and some natural compounds as a sustainable tool for *Meloidogyne incognita* management on Grapevines.

Authors Contribution

All authors equally participated in the development and implementation of the reviewing plan. Subsequently, they worked it out and wrote the manuscript; the first author. DSSI wrote and discussed the different parts of the article with RHW, SHA-B and AMEB, together finalized the manuscript. All authors have read and approved the final manuscript.

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Generative AI and AI assisted technology statement
The authors declare that no generative AI and AI assisted technology was used in the creation of this manuscript.

Conflict of interest
The authors have declared no conflict of interest.

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