PIRIFORMOSPORA INDICA: A BIO-CONTROL MARVEL IN MORPHOLOGICAL GROWTH UNDER COMBINED STRESS OF ROOT KNOT NEMATODE AND SALINITY IN TOMATO

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ABSTRACT

Abiotic and biotic stresses, including salinity and root-knot nematode infestation, significantly undermine tomato productivity globally. Root-knot nematodes (Meloidogyne spp.) are substantial pests that inflict extensive damage on crops, necessitating the development of sustainable control strategies. Elevated salt concentrations in soil or irrigation water impair plant-water relations, ion homeostasis, and metabolic functions, resulting in substantial reductions in growth and yield. Combined stress of salinity and root knot nematode markedly diminished plant height, root and shoot biomass, leaf count, and overall canopy growth. Increased salinity resulted in observable symptoms including chlorosis, leaf curling, and wilting. Piriformospora indica, an endophytic fungus, has garnered significant interest due to its effects on promoting plant growth and enhancing stress resistance in various host plants. To date, there have been no reports regarding its application in root knot nematode. This study presents the colonization capacity of P. indica in the roots of two tomato cultivars: Pusa ruby and Pusa early dwarf in the presence of salt and RKN. The findings indicated that, at 2 months post-inoculation (mpi), the P. indica-colonized seedlings demonstrated enhanced total root- shoot length, root -shoot dry and fresh weight. Tomato seeds were inoculated with 2% P. indica mycilium suspension and nematode juveniles (100J2/seedling) under different concentration of NaCl. Following a 60-days period, nematode infestation in saline soil diminished seedling growth, reduced root and shoot biomass, and a decline in photosynthetic pigments.

Keywords: Salinity, Root Knot Nematodes, Piriformospora indica, Root Gall, Egg Mass

INTRODUCTION

Within ecosystems, plants engage with diverse biotic and abiotic factors. Plants in both natural and agricultural environments are consistently subjected to various biotic and abiotic stresses that significantly impair their growth, development, and productivity. Abiotic stresses, including salinity, drought, and extreme temperatures, disrupt critical physiological processes such as photosynthesis, nutrient absorption, and water regulation (Umar *et al.*, 2021; Nawaz *et al.*, 2023; Mazumder *et al.*, 2025). Salinity is a significant constraint on agricultural productivity in arid and semi-arid regions, impacting more than 20% of irrigated land worldwide (Munns & Tester, 2008; El Sabagh *et al.*, 2020; Negacz *et al.*, 2022). In plants subjected to salt stress, elevated levels of sodium (Na⁺) and chloride (Cl⁻) ions induce osmotic stress and ionic toxicity, resulting in diminished cell expansion, oxidative damage, and nutrient imbalance (Parida & Das, 2005; Khare *et al.*, 2015; Ahmad *et al.*, 2019; Arif *et al.*, 2020). Simultaneously, biotic stresses induced by pathogens and pest such as bacteria, fungi, viruses, and nematodes—diminish plant health and productivity. Root-knot nematodes (*Meloidogyne spp.*) are notably deleterious, causing gall formation, impairing nutrient and water transport, and rendering plants susceptible to secondary infections (Sasser & Freekman, 1987; Ibrahim *et al.*, 2019; Azlay *et al.*, 2023;). The harm inflicted by root-knot nematodes is frequently intensified under abiotic stress conditions, resulting in a complex interaction that

amplifies the adverse impacts on plant performance (Atkinson & Urwin, 2012; Shah and Smith 2020; Habteweld *et al.*, 2024).

Microorganisms, particularly plant-parasitic nematodes are critical biotic stressors due to their reciprocal interactions with plants (such as carrot, potato, tomato, brinjal, guava etc.) and significant impact on agricultural crop productivity (Phani et al., 2021; Kashyap et al., 2024). One of the most destructive groups of plant-parasitic nematodes is the root-knot nematode, which belongs to the genus Meloidogyne (Tapia-Vázquez et al., 2022). Root-knot nematodes (RKNs), are pivotal in both economic and scientific domains due to their intricate host plant interactions, broad host range and destructive capabilities facilitated by their stylet (Rutter et al., 2022). Meloidogyne spp. are among the most harmful plant parasites which penetrates root epidermal tissue and inducing gall formation. During the infective second juvenile (J2) stage, they secrete hydrolytic enzymes to invade roots, disrupting water and nutrient uptake, nitrogen fixation and mineral absorption. Infestation leads to stunted growth, reduced photosynthesis and oxidative stress due to reactive oxygen species (ROS) accumulation (Khan et al., 2023). ROS play a crucial role in host-pathogen interactions, influencing signaling pathways and plant defense responses. Due to the rising occurrence of environmental extremes and the enduring presence of soil-borne pathogens, there is an urgent necessity for integrated strategies that bolster crop resilience (Oadir et al., 2024). Investigating plant-microbe interactions, stress physiology, and genetic resistance presents promising strategies for alleviating the detrimental impacts of biotic and abiotic stresses. Tomato (Solanum lycopersicum), a widely cultivated and nutritionally rich (contains various vitamins and minerals, carbohydrates, flavonoids and antioxidants) crop in India is highly consumed. Economically, its production is severely affected by *Meloidogyne spp.* which pose a significant threat to its morphology, physiology and yield. Their infestations accounts for 30-50% of yield losses in India, contributing to an estimated global economic loss of \$358 billion. Such infestations pose a significant challenge to farming, particularly in organic systems. While synthetic nematicides were once the primary control method, concerns over their environmental and health risks have led to their phased withdrawal. This shift has intensified the search for cost-effective sustainable alternatives. Globally it comprises of 19% of vegetable consumption. So, addressing RKN-induced losses is critical for sustainable agriculture (Vashisth et al., 2024; Sharma and Ohri 2025). Biological agents, including microbes have emerged as promising eco-friendly solutions, offering both nematocidal effects and plant growth benefits in modern agriculture. Tomato a significant horticultural crop is particularly vulnerable to salinity and nematode infestation. Combine stress of salt and RKN diminish morphological parameters, including plant height and biomass, while also compromising fruit quality and market value (Cuartero & Fernández-Muñoz, 1999; Kaur et al., 2017; Prasad et al., 2024). The synergistic impact of biotic and abiotic stressors frequently leads to more significant damage than either stressor independently, underscoring the necessity to comprehend the fundamental mechanisms of stress interaction and plant response (Jing et al., 2024). The impact of saline soil on the infection caused by root knot nematodes is contingent upon various growth parameters, including the morphological, physiological and biochemical in tomato (El-sheikh and Osman, 2002; Li et al., 2020; Cui et al., 2022; Alsantely et al., 2023). The impact of fungal infection under salinity stress varies depending on the severity of salt stress, with either enhancement or suppression observed (Wiese et al., 2004; DiLeo et al., 2010; Dikilitas M, and Karakas S. 2014; Camacho al., 2023). The increased vulnerability was ascribed to the buildup of ABA in roots generated by salt, which inhibited the SA-mediated defense mechanism in tomato (Ali M, et al., 2023). Kissoudis et al., (2016) studied on tomato, when exposed to low concentrations of salt, S. lycopersicum exhibited a suppression of its defense mechanisms against Oidium neolycopersi, which is the causal agent of powdery mildew. Conversely, when subjected to high concentrations of salt, the plant exhibited resistance to the pathogen. The growth of fungi is hindered by a significant concentration of salt, which can be related to the enhanced resistance acquired at higher salt concentrations. The fungi appear to exhibit tolerance towards low levels of salt, which can trigger the signaling of abscisic acid (Haller E. et al., 2020). In recent years, plant-microbe interactions have become a promising method to improve plant resilience to abiotic

stresses. The endophytic fungus *Piriformospora indica*, now reclassified as Serendipita indica, has garnered significant interest among advantageous root-associated microbes due to its extensive host range and capacity to impart various stress tolerances (Varma *et al.*, 1999; Abd-Elsalam and Mohamed 2023). Numerous studies have shown that *P. indica* can markedly mitigate the adverse effects of salt stress in various plant species. *P. indica* enhances salt tolerance in rice and barley by elevating antioxidant enzyme activities, preserving ion homeostasis, and facilitating osmolyte accumulation, including proline (Jogawat *et al.*, 2013; Gill *et al.*, 2016; Kundu *et al.*, 2022). *P. indica* has been linked to enhanced morphological characteristics in tomato, such as increased plant height, root biomass, and leaf area under salinity stress, as well as improved photosynthetic efficiency (Zarea *et al.*, 2012; L Li and Hao 2023; Subhash *et al.*, 2025).

The objective of this study is to investigate the migration patterns, penetration mechanisms, and gall formation characteristics of *Meloidogyne spp*in tomato cultivars. We intend to identify variations in susceptibility among cultivar of Pusa ruby and Pusa early dwarf. This will ultimately result in an increase in the sustainability of tomato production and a reduction in the economic losses that are caused by root-knot nematodes. The present study aimed to mitigate the disease progression of M. incognita in S. lycopersicum seedlings by colonizing *P. indica* under different concentration of salinity. This study distinctly investigates nematicidal potential of *P. indica*. The experiment conducted with *Piriformospora indica* under combined salinity and root-knot nematode stress demonstrated its effectiveness in enhancing the morphological growth of tomato plants.

MATERIALS AND METHODS

Plant Material and Root Knot Nematode

Freshly harvested seeds of the tomato variety Pusa Ruby were obtained from the Department of Horticulture, IARI, New Delhi. Culture of *Meloidogyne spp* was obtained from Dr. Zakaullah Khan at the Division of Plant Quarantine, National Bureau of Plant Genetic Resources (NBPGR), New Delhi. Root-knot nematode (*Meloidogyne spp*) infection in tomato

Establishment of root-knot nematode (Meloidogyne spp.) culture in tomato

The autoclave (121°C at 16 psi) sterilized sandy loam soil for 60 minutes. After sterilization, soil was stored safely and used to raise RKN nurseries. In 9-inch earthen pots, sterilized sandy loam soil was filled and transplanted tomato plants of susceptible varieties were carefully placed on the soil surface and covered with 1 cm of soil. For pure root-knot nematode culture, tomato seedlings were transplanted at 3-5 leaf stages (14 days). One egg mass from root-knot nematode, M. incognita-infected tomato plant roots was kept in sterilised water in a Petri dish for hatching. One healthy seedling per pot was kept at the centre after 14 days. A small pipe (2cm long, 5 cm bore) was inserted into the soil near each seedling's root zone for RKN inoculation holes. Inoculating J2, after 35 days of *M. incognita* infection, plants were carefully harvested for pure culture multiplication.

P. indica colonization in the root of tomato

Healthy and uniform-sized seeds of tomato are surface-sterilized in a sodium hypochlorite solution (1%) placed for germination into petri plates. After seven days of germination in petri plate seeding transferred into seed germinating paper for inoculation of *P. indica* as shown in Table 3.7. Germinating paper left for 14 days after transplanting of seedling and then at 14^{th} days experiment check for colonization of *P. indica*. Roots stain with the 0.02 and 0.05% trypan blue with 12 hrs of KOH treatment separately. Stain of 0.02% trypan blue for 12 hour of KOH treatment showed the clear colonization than stained with 0.05% trypan blue.

Harvest treated tomato cultivars Pusa Ruby and Pusa early dwarf

At 60th days of tomato cultivars (Pusa ruby and Pusa early dwarf) inoculated with P.indica and M. incognita under saline condition are harvested for the analysis of morphological growth aspects i.e., Root-shoot length, root-shoot dry weight and fresh weight.

STATISTICAL ANALYSIS

Statistical analyses were performed with statistical software's Graph Pad Prism (version 7.04) and XLSTAT (version 2023.3.1).

RESULTS AND DISCUSSION

Morphological Growth aspects under combine stress of salt and RKN in presence of P. indica Root Length

Root length decreased significantly with increasing salt concentration in both varieties. At 0 mM salt, the Pusa Ruby variety had a root length of 26.733 ± 0.643 cm, which was significantly longer(p<0.01) than the Pusa Early Dwarf variety (24.333 ± 0.577 cm). Under the highest salt stress (150 mM), the root length of Pusa Ruby (13.567 ± 0.351 cm) was also significantly greater (p<0.01) than Pusa Early Dwarf (11.133 ± 1.026 cm). The presence of *P. indica* helped maintain significantly longer roots (p<0.01) under salt stress in both varieties, while *M. incognita* infection led to the shortest roots, especially at the higher salt concentrations. The combination of *M. incognita* and salt stress had a significant synergistic negative effect on root length (p<0.001).

Table:1 Effect of different salinity concentrations (0 mM, 50 mM, 100 mM, and 150 mM) on Root Length in Pusa Ruby and Pusa Early Dwarf under non-inoculated, *P.indica*-inoculated, *M. incognita*-inoculated and *P.indica* + *M. incognita*-inoculated treatment.

Variety	Treatment	0 mM	50mM	100mM	150mM
	Control	26.733 ± 0.643	19.867 ± 0.569	16.4 ± 0.436	13.567 ± 0.351
Pusa	P. indica	31.1 ± 1.253	28.533 ± 1.361	24.6 ± 0.529	21.267 ± 0.643
	M. incognita	24.533 ± 0.551	16.433 ± 0.404	14.7 ± 0.608	9.7 ± 0.361
Ruby					
	P. indica +	27.067 ± 0.777	22.7 ± 0.755	19.3 ± 0.3	16.3 ± 0.436
	M. incognita				
	Control	24.333 ± 0.577	18.433 ± 0.404	15.033 ± 0.751	11.133 ± 1.026
Pusa	P. indica	28.667 ± 0.577	21.333 ± 0.814	19.333 ± 0.493	17.5 ± 0.5
Early					
	M. incognita	21.333 ± 1.528	15.533 ± 0.351	13.4 ± 1.217	8.1 ± 0.656
Dwarf					
	P. indica +	24 ± 1	18.867 ± 0.643	16.7 ± 0.5	12.9 ± 1.308
	M. incognita				

Shoot Length

Shoot length decreased significantly with increasing salt concentration in both varieties. At 0 mMsalt, the Pusa Ruby variety had a shoot length of 64.667 ± 0.577 cm, which was significantly longer (p<0.01) than the Pusa Early Dwarf variety (64.533 ± 0.503 cm). Under the highest salt stress (150mM), the shoot length of Pusa Ruby (51.267 ± 0.252 cm) was also significantly greater (p<0.01) than Pusa Early Dwarf (37.233 ± 1.537 cm). The presence of *P. indica* helped maintain significantly longer shoots (p<0.01) under salt stress in both varieties, while *M. incognita* infection led to the shortest shoots, especially at the higher salt concentrations. The combination of *M. incognita* and salt stress had a significant synergistic negative effect on shoot length (p<0.001).

Table 2: Effect of different salinity concentrations (0 mM, 50 mM, 100 mM, and 150 mM) on Shoot Lenght in Pusa Ruby and Pusa Early Dwarf under non-inoculated, *P.indica*-inoculated, *M. incognita*-inoculated and *P.indica* + *M. incognita*-inoculated treatments

Variety	Treatment	0 mM	50mM	100mM	150mM
	Control	64.667 ± 0.577	63.5 ± 0.5	54.333 ± 0.577	51.267 ± 0.252
Pusa	P. indica	69.033 ± 0.351	63.967 ± 0.451	62.1 ± 0.854	59.167 ± 0.289
	M. incognita	57.2 ± 0.265	52.267 ± 0.379	49.233 ± 0.252	34.4 ± 0.361
Ruby	_				
	P. indica + M. incognita	59.267 ± 0.379	57.067 ± 0.208	54.533 ± 0.252	52.267 ± 0.379
	Control	64.533 ± 0.503	54.167 ± 0.764	48.9 ± 0.819	37.233 ± 1.537
Pusa Early	P. indica	67.367 ± 0.473	62.167 ± 0.643	58.833 ± 0.902	54.767 ± 0.252
	M. incognita	54.967 ± 0.551	50 ± 0.608	45.433 ± 1.32	30.567 ± 0.513
Dwarf	-				
	P. indica +	57.333 ± 0.306	54.467 ± 0.351	52 ± 0.3	50.7 ± 0.608
	M. incognita				

Root Fresh Weight

Root fresh weight decreased significantly with increasing salt concentration in both varieties. At 0 mM salt, the Pusa Ruby variety had a root fresh weight of 2.237 ± 0.025 g, which was significantly higher (p<0.01) than the Pusa Early Dwarf variety (2.16 ± 0.026 g). Under the highest salt stress (150 mM), the root fresh weight of Pusa Ruby (1.188 ± 0.011 g) was also significantly greater (p<0.01) than Pusa Early Dwarf (1.157 ± 0.031 g). The presence of *P. indica* helped maintain significantly higher root fresh weights (p<0.01) under salt stress in both varieties, while *M. incognita* infection led to the lowest root fresh weights, especially at the higher salt concentrations. The combination of *M. incognita* and salt stress had a significant synergistic negative effect on root fresh weight (p<0.001).

Table 3: Effect of different salinity concentrations (0 mM, 50 mM, 100 mM, and 150 mM) on Root Fresh Weight in Pusa Ruby and Pusa Early Dwarf under non-inoculated, *P.indica*-inoculated, *M. incognita*-inoculated and *P.indica* + *M. incognita*-inoculated treatments

Variety	Treatment	0 mM	50mM	100mM	150mM
	Control	2.237 ± 0.025	2.097 ± 0.095	1.95 ± 0.173	1.188 ± 0.011
Pusa	P. indica	3.743 ± 0.047	3.467 ± 0.159	3.45 ± 0.02	2.981 ± 0.127
	M. incognita	3.173 ± 0.029	2.012 ± 0.075	1.767 ± 0.015	1.734 ± 0.031
Ruby					
	P. indica +	3.143 ± 0.042	2.823 ± 0.025	2.473 ± 0.038	2.21 ± 0.009
	M. incognita				
	Control	2.16 ± 0.026	1.767 ± 0.015	1.54 ± 0.026	1.157 ± 0.031
Pusa	P. indica	3.493 ± 0.155	3.35 ± 0.02	3.202 ± 0.066	2.912 ± 0.075
Early					
	M. incognita	2.133 ± 0.049	1.912 ± 0.075	1.583 ± 0.079	1.549 ± 0.052
Dwarf	_				
	P. indica +	3.097 ± 0.036	2.757 ± 0.067	2.504 ± 0.091	2.005 ± 0.094
	M. incognita				

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Shoot Fresh Weight

Shoot fresh weight decreased significantly with increasing salt concentration in both varieties. At 0 mM salt, the Pusa Ruby variety had a shoot fresh weight of 12.901 ± 0.051 g, which was significantly higher (p<0.01) than the Pusa Early Dwarf variety (13.93 ± 0.113 g). Under the highest salt stress (150 mM), the shoot fresh weight of Pusa Ruby (11.217 ± 0.06 g) was also significantly greater (p<0.01) than Pusa Early Dwarf (7.907 ± 0.096 g). The presence of *P. indica* helped maintain significantly higher shoot fresh weights (p<0.01) under salt stress in both varieties, while *M. incognita* infection led to the lowest shoot fresh weights, especially at the higher salt concentrations. The combination of *M. incognita* and salt stress had a significant synergistic negative effect on shoot fresh weight (p<0.001).

Table 4: Effect of different salinity concentrations (0 mM, 50 mM, 100 mM, and 150 mM) on Shoot fresh weight in Pusa Ruby and Pusa Early Dwarf under non-inoculated, *P.indica*-inoculated, *M. incognita*-inoculated and *P.indica* + *M. incognita*-inoculated treatments

Variety	Treatment	0 mM	50mM	100mM	150mM
	Control	12.901 ± 0.051	12.373 ± 0.177	12.123 ± 0.188	11.217 ± 0.06
Pusa	P. indica	18.783 ± 0.726	17.81 ± 0.165	17.023 ± 0.093	13.973 ± 0.031
	M. incognita	13.577 ± 0.311	10.354 ± 0.419	8.637 ± 0.146	$\boldsymbol{6.172 \pm 0.659}$
Ruby					
	P. indica + M. incognita	16.147 ± 0.162	12.244 ± 0.212	12.22 ± 0.211	11.45 ± 0.151
	Control	13.93 ± 0.113	11.537 ± 0.22	9.207 ± 0.067	7.907 ± 0.096
Pusa	P. indica	17.008 ± 0.943	16.32 ± 0.101	16.767 ± 0.204	12.582 ± 0.929
Early	M. incognita	12.388 ± 0.38	10.737 ± 0.574	8.854 ± 0.173	$\boldsymbol{6.472 \pm 0.511}$
Dwarf	P. indica + M. incognita	15.667 ± 0.243	12.174 ± 0.195	11.502 ± 0.501	10.418 ± 0.397

Root Dry Weight

Root dry weight followed a similar pattern to root fresh weight, decreasing significantly with increasing salt concentration in both varieties. At 0 mM salt, the Pusa Ruby variety had a root dryweight of 0.355 ± 0.008 g, which was significantly higher (p<0.01) than the Pusa Early Dwarf variety (0.297 ± 0.006 g). Under the highest salt stress (150 mM), the root dry weight of Pusa Ruby (0.236 ± 0.005 g) was also significantly greater (p<0.01) than Pusa Early Dwarf (0.173 ± 0.005 g). The presence of *P. indica* helped maintain significantly higher root dry weights (p<0.01) under salt stress in both varieties, while *M. incognita* infection led to the lowest root dry weights, particularly at the higher salt concentrations. The combination of *M. incognita* and salt stress had a significant synergistic negative effect on root dry weight (p<0.001).

Table 5: Effect of different salinity concentrations (0 mM, 50 mM, 100 mM, and 150 mM) on Root dry weight in Pusa Ruby and Pusa Early Dwarf under non-inoculated, *P.indica*-inoculated, *M. incognita*-inoculated and *P.indica* + M. *incognita*-inoculated treatments

Variety	Treatment	0 mM	50mM	100mM	150mM
	Control	0.355 ± 0.008	0.292 ± 0.01	0.256 ± 0.005	0.236 ± 0.005
Pusa	P. indica	0.425 ± 0.008	0.418 ± 0.02	0.411 ± 0.008	0.309 ± 0.005
	M. incognita	0.309 ± 0.024	0.29 ± 0.002	0.276 ± 0.003	0.243 ± 0.004
Ruby					
	P. indica + M. incognita	0.37 ± 0.007	0.361 ± 0.001	0.294 ± 0.004	0.292 ± 0.003
	Control	0.297 ± 0.006	0.216 ± 0.002	0.194 ± 0.006	0.173 ± 0.005
Pusa	P. indica	0.413 ± 0.006	0.394 ± 0.006	0.362 ± 0.027	0.292 ± 0.003
Early	M. incognita	0.286 ± 0.005	0.197 ± 0.001	0.091 ± 0.001	0.081 ± 0.001
Dwarf					
	P. indica +	0.322 ± 0.008	0.276 ± 0.004	0.229 ± 0.007	0.219 ± 0.008
	M. incognita				

Shoot Dry Weight

Shoot dry weight followed a similar pattern to shoot fresh weight, decreasing significantly with increasing salt concentration in both varieties. At 0 mM salt, the Pusa Ruby variety had a shoot dry weight of 3.122 ± 0.194 g, which was significantly higher (p<0.01) than the Pusa Early Dwarfvariety (2.297 ± 0.06 g). Under the highest salt stress (150 mM), the shoot dry weight of Pusa Ruby (1.868 ± 0.085 g) was also significantly greater (p<0.01) than Pusa Early Dwarf (1.33 ± 0.154 g). The presence of *P. indica* helped maintain significantly higher shoot dry weights (p<0.01) under salt stress in both varieties, while *M. incognita* infection led to the lowest shoot dry weights, particularly at the higher salt concentrations. The combination of *M. incognita* and salt stress had a significant synergistic negative effect on shoot dry weight (p<0.001).

Table 6: Effect of different salinity concentrations (0 mM, 50 mM, 100 mM, and 150 mM) on Shoot dry weight in Pusa Ruby and Pusa Early Dwarf under non-inoculated, *P.indica*-inoculated, *M. incognita*-inoculated and *P.indica*+*M. incognita*-inoculated treatments

Variety	Treatment	0 mM	50mM	100mM	150mM
	Control	3.122 ± 0.194	3.12 ± 0.211	2.153 ± 0.038	1.868 ± 0.085
Pusa	P. indica	3.84 ± 0.044	3.489 ± 0.115	2.923 ± 0.032	2.847 ± 0.057
	M. incognita	2.82 ± 0.072	2.643 ± 0.042	1.887 ± 0.032	1.617 ± 0.031
Ruby					
	P. indica + M. incognita	3.367 ± 0.046	2.76 ± 0.026	2.637 ± 0.047	2.359 ± 0.031
	Control	2.297 ± 0.06	2.034 ± 0.058	1.827 ± 0.025	1.33 ± 0.154
Pusa	P. indica	3.101 ± 0.1	2.578 ± 0.115	2.433 ± 0.038	2.293 ± 0.086
Early					
	M. incognita	2.041 ± 0.07	1.373 ± 0.086	1.14 ± 0.026	0.897 ± 0.025
Dwarf					
	P. indica + M. incognita	2.441 ± 0.12	2.087 ± 0.076	1.853 ± 0.033	1.643 ± 0.006

DISCUSSION

According to the findings of the current study, the synergetic effect of root knot nematode and salinity poses a greater threat to tomato cultivars (Pusa Ruby and Pusa Early Dwarf). This study examines the influence of *Piriformospora indica* (*P. indica*) on the morphological resilience of tomato plants (Solanum lycopersicum) subjected to combined abiotic and biotic stress from salinity and *Meloidogyne incognita* (root-knot nematode). The results highlight the considerable potential of *P. indica* as a biocontrol agent and plant growth enhancer, particularly in stress-prone agro-ecological regions.

The morphological parameters of tomato plants, including shoot and root length, fresh and dry biomass, and overall plant vigor, exhibited significant enhancement in P. indica-treated groups relative to those experiencing only nematode or salinity stress. These findings align with previous studies indicating that P. indica provides tolerance to multiple abiotic stresses, such as salinity, drought, and heavy metal toxicity. P. indica's endophytic colonization likely influences these effects by regulating host plant hormone signaling, improving nutrient absorption, and enhancing antioxidative defense mechanisms. The synergistic stress of salinity and nematode infection significantly inhibited growth in untreated plants. Nevertheless, in P. indica-inoculated plants subjected to the same combined stress, the adverse effects were substantially alleviated. This dual protection can be ascribed to the fungus's capacity to modify root architecture, augment osmolyte accumulation, and activate the plant's innate immune system. Improved root systems not only enhance water and nutrient uptake in saline conditions but may also restrict nematode invasion and gall development, as evidenced by diminished gall indices in P. indica-treated plants. Moreover, the symbiotic association seems to elicit systemic resistance to M. incognita, probably engaging jasmonic acid (JA) and salicylic acid (SA) signaling pathways, which are recognized to be stimulated by advantageous endophytes. The enhanced plant tolerance to salinity stress may result from P. indica-induced upregulation of salt-responsive genes and improved ionic equilibrium via selective uptake of K⁺ over Na⁺, a mechanism extensively documented in prior research.

A notable aspect of the current findings is the prospective use of *P. indica* as a sustainable, eco-friendly substitute for chemical nematicides and synthetic stress mitigators. Its soil-borne characteristics, simplicity of cultivation, and wide-ranging effectiveness render it a promising option for integrated pest and stress management strategies in tomato cultivation and potentially other horticultural crops. Further studies are necessary to elucidate the precise molecular mechanisms underlying the synergistic alleviation of biotic and abiotic stress by *P. indica*. Field trials are crucial for validating greenhouse results and assessing the consistency of *P. indica*'s performance under varying environmental conditions.

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