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## **INTERACTIVE EFFECT OF *MELOIDOGYNE JAVANICA* AND OZONE ON LEAF PIGMENT, NITROGEN AND PROTEIN CONTENT OF PEA**

by  
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**Summary.** The effect of *Meloidogyne javanica* and ozone (0.1 and 0.2 ppm) on leaf pigment and nitrogen and protein content of seeds in pea was studied under conditions of artificial exposure in a glasshouse. *M. javanica* or ozone alone caused significant reductions in all the considered parameters. However, together they exhibited a synergistic interaction which resulted in greater quantitative reduction in chlorophyll, carotenoid and nitrogen content of leaves and protein content of seeds.

Ozone (O<sub>3</sub>), an important phytotoxic air pollutant, causes significant loss in plant growth and yield (Blum *et al.*, 1982). It accumulates in the palisade layers of the leaves and causes bleaching or discolouration through destruction of chlorophyll (Sakaki *et al.*, 1985). The quantitative decrease of leaf pigments and protein content of seeds caused by plant parasitic nematodes are reported to be increased in several plants in artificial exposures to O<sub>3</sub> (Khan, 1989, Singh, 1989).

In the present study an attempt was made to assess the interactive effect of *Meloidogyne javanica* (Treub) Chitw. and O<sub>3</sub> on quantitative changes in leaf pigments, nitrogen and seed protein content in pea (*Pisum sativum* L., cv. Rachna) in artificial exposures in a glasshouse.

### **Materials and methods**

Surface sterilized seeds of pea obtained from IARI, Seed Centre, New Delhi, were sown in clay pots (30 cm diam.) containing autoclaved

sandy loam field soil. Prior to sowing, some seeds were treated with commercially available root nodule bacteria, *Rhizobium leguminosarum* of pea strain. Three-week-old seedlings (one seedling/pot) were inoculated with 2000 freshly hatched second stage juveniles (J<sub>2</sub>) of *M. javanica*. The combinations of *Rhizobium* and nematode are given in Table I.

The test plants were exposed to O<sub>3</sub> in a dynamic state exposure chamber (Khan and Khan 1993). Ozone was generated by subjecting dry oxygen to the action of silent electric discharge in an ozoniser i.e. UV generator. A polyvinyl chloride tube from the ozoniser was connected to the inlet of the blower assembly of the exposure chamber.

Exposure of seedlings to O<sub>3</sub> started immediately after nematode inoculation. The pots were placed in the exposure chamber and exposed to O<sub>3</sub> for 3 h on every alternate day throughout the period of the experiment (70 days). The concentrations of O<sub>3</sub> used for the exposures were 0.1 and 0.2 ppm which were predetermined by sampling the gas inside the chamber

by a Handy Air Sampler (Kimoto Electricals, Japan) and analysed in the laboratory by employing the potassium iodide method (Anonymous, 1986).

Chlorophyll (a, b, and total), carotenoids and nitrogen content of leaves were determined by the methods of MacKinney (1941), Machlachlan and Zalik (1968) and Linder (1944), respectively. Protein (soluble, insoluble and total protein) estimation was done by the method of Lowry *et al.* (1951).

Each treatment was replicated five times and the pots were arranged in a completely randomised block design. The data were subjected to Analysis of Variance (ANOVA) to determine the significant effects.

## Results and conclusions

The data in Table I indicate that plants not exposed to O<sub>3</sub> but inoculated with *R. leguminosarum* and without *M. javanica* had higher levels of chlorophylls (a, b and total), carotenoids, nitrogen content of leaves and protein content of seeds, than plants with nematodes and exposed to O<sub>3</sub> (Tables I and II). However, the quantitative decrease in these parameters caused by *M. javanica* alone was comparatively greater in the absence of *R. leguminosarum*.

Pea plants exposed to 0.1 and 0.2 ppm of O<sub>3</sub> had decreased amount of chlorophylls and carotenoid in the leaves, the percent reduction being greater with 0.2 ppm O<sub>3</sub> (Table I). Pea

TABLE I - Effect of O<sub>3</sub> and *Meloidogyne javanica* on chlorophyll and carotenoid contents in pea.

Treatments	O <sub>3</sub> (ppm)			O <sub>3</sub> (ppm)			O <sub>3</sub> (ppm)			O <sub>3</sub> (ppm)		
	0	0.1	0.2	0	0.1	0.2	0	0.1	0.2	0	0.1	0.2
	Chlorophyll a (mg/g)			Chlorophyll b (mg/g)			Total chlorophyll (mg/g)			Carotenoid (mg/g)		
Pea	0.59	0.47	0.37	0.53	0.41	0.31	1.35	1.00	0.76	0.38	0.34	0.31
Pea + <i>Rhizobium</i>	0.89	0.58	0.45	0.78	0.53	0.40	1.90	1.23	0.94	0.47	0.47	0.39
Pea + Nematode	0.50	0.46	0.35	0.44	0.40	0.29	1.10	0.06	0.74	0.35	0.32	0.30
Pea + <i>Rhizobium</i> + Nematode	0.69	0.57	0.41	0.54	0.44	0.36	1.52	1.19	0.86	0.41	0.37	0.35
Interaction CD	0.004			0.005			0.006			0.005		

TABLE II - Effect of O<sub>3</sub> and *M. Javanica* on seed protein and nitrogen contents in pea.

Treatments	O <sub>3</sub> (ppm)			O <sub>3</sub> (ppm)			O <sub>3</sub> (ppm)			O <sub>3</sub> (ppm)		
	0	0.1	0.2	0	0.1	0.2	0	0.1	0.2	0	0.1	0.2
	Soluble protein %			Insoluble protein %			Total protein %			Nitrogen content %		
Pea	8.70	7.88	7.38	10.61	9.84	9.37	19.31	17.64	16.75	3.74	3.32	3.10
Pea + <i>Rhizobium</i>	9.23	8.63	8.32	12.22	11.13	10.54	21.45	19.76	18.86	5.47	5.27	4.88
Pea + Nematode	8.58	7.58	7.13	10.34	9.66	9.20	18.92	17.24	16.33	3.38	3.15	2.83
Pea + <i>Rhizobium</i> + Nematode	9.07	8.10	7.60	11.80	10.52	9.78	20.85	18.66	17.38	4.40	4.09	3.92
Interaction CD	0.07			0.05			0.10			0.06		

plants inoculated with root-knot nematode and root-nodule bacteria jointly but not exposed to O<sub>3</sub>, had more pigments than plants inoculated only with root-knot nematode but less than root-nodule inoculated plants.

Likely both the concentrations of O<sub>3</sub> inhibited the formation of proteins in the seeds, the effect being greater with exposures to 0.2 ppm O<sub>3</sub> than to 0.1 ppm. This pattern of inhibition was same in all the treatment irrespective of the presence or not of nematode and bacteria. However, protein reduction was less in plants inoculated with root-nodule bacteria only than in plants with root-knot nematode + root-nodule inoculated plants or in plants inoculated with root-knot nematode.

The results show that O<sub>3</sub> acted synergistically with root-knot nematode to cause significant loss of pigments and proteins in pea plants. The effect was less in the presence of root-nodule bacteria.

**Acknowledgements.** The senior author is grateful to CSIR for providing Financial Assistance as Research Associate.

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