

Free phenol and the improved tolerance in four *Meloidogyne* (root-knot nematode) susceptible hosts

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Abstract. One hundred ppm of phenol in 2% NPK fertilizer treatments of 4 *Meloidogyne incognita* susceptible hosts, reduced the pathogenic impact and improved their tolerance.

Keywords. Host tolerance; susceptibility ratings; environmental resistance factor; metabolic compatibility.

1. Introduction

Plant metabolism and the complex wood chemistry imply the role of phenols in the plant's structure and function (Cowling and Horsfall 1980). The role of phenols in plant pathogenesis and disease resistance are well documented (Acedo and Rhode 1971; Cowling and Horsfall 1980; Gibel 1974, 1982; Goodman *et al* 1967). Feldman and Hanks (1968, 1971) opened up the exciting field of improving the plant tolerances with phenolics. Soil amendments (Singh and Seetha Ramiah 1967, 1971) indicated the possible nematicidal roles of phenols through organic amendments.

Free phenol when introduced into the plant (Ribereau-Gayon 1972) is actively absorbed and metabolized due to the highly reactive -OH group in its ring. Phenols condense with the plant's metabolites (glycosides, uronides, proteins etc.), thereby alter the substrate relations of the nematodes (Feldman and Hanks 1971; McIntyre 1980), creating a physical barrier for the nematodes, making the roots unsuitable for the nematodes. They also inhibit the free enterprise of the nematode's enzymes (Wallace 1973) leading to starvation of nematodes, with consequent lesser infectivity of the pathogen (Van Gundy *et al* 1967).

The above features reveal the metabolic compatibility of phenol with regard to the host's metabolites and the pathogen's enzymes, involving its bidirectional roles on the host as well as the pathogen's enzymes.

In pilot experiments it was found that free phenol in NPK solutions inhibited the egg hatchings appreciably. Taking advantage of the above facts, 100 ppm of phenol in 2% NPK fertilizer solution was employed to assess the effect on 4 *Meloidogyne* (root-knot nematode) viable hosts.

2. Methodology

Seven day old seedlings (from surface sterilized seeds) of *Hibiscus canabinus*, *Phaseolus mungo*, *Hibiscus esculentus* and *Solanum melongena* maintained in 10 cm dia. plastic pots (1 kg sterile sandy loam-2:1) were inoculated with 10^3 *Meloidogyne incognita* juveniles. For each host, 12 pots were treated with 2% NPK solution and another set of 12, treated with 100 ppm phenol in 2% NPK solution. Moisture was

kept at 60% levels with the irrigants applied, thrice weekly. Forty five days after the inoculation, the experiments were closed and the plants assayed for the pathogen population (table 1) and redox enzyme activities (table 2).

The environmental resistance factor (ERF) as a measure of the resistance encountered by the pathogen was assessed as follows:

$$\text{ERF} = \text{RR}/\text{RPI}$$

When RR = Rate of pathogen's reproduction,
RPI = Rate of the population increase of pathogen.

$$\text{RR} = \frac{(P_f \div \text{g root wt}) \div \text{♀ per g root}}{1 \times 100}$$

Table 1. Population dynamics of *M. incognita* in 4 susceptible hosts under the influence of phenol.

	<i>H. cannabinus</i>		<i>P. mungo</i>		<i>H. esculentus</i>		<i>S. melongena</i>	
	UT	PT	UT	PT	UT	PT	UT	PT
Egg mass	35	24	29	21	23	16	20	15
Eggs/egg mass	530.2	382.6	435.4	370.2	325.6	240.8	275.8	188.3
Total eggs (10 ³)	18.56	9.18	12.63	7.77	7.49	3.85	5.52	2.83
Population (10 ³):								
Root	0.53	0.42	0.47	0.36	0.33	0.26	0.22	0.15
Soil	6.41	4.65	4.27	3.30	3.12	2.20	2.12	1.48
Total	25.50	14.25	17.37	11.43	10.94	6.31	7.86	4.46
RR	5.45	3.79	4.38	3.72	3.26	2.41	2.76	1.88
RPI	0.54	0.29	0.36	0.23	0.22	0.12	0.15	0.08
ERF total	10.09	13.07	12.17	16.17	14.82	20.08	18.4	23.5
ERF plant (P)	7.55	8.81	9.18	11.51	10.59	13.09	13.43	15.71
ERF soil (S)	2.54	4.26	2.99	4.66	4.23	6.99	4.97	7.79
ERF P/S	2.97	2.07	3.07	2.47	2.50	1.87	2.70	2.02

Values are average of 6 replicates (± 0.01 to ± 0.002).

UT, No phenol treatment; PT, phenol treated.

Table 2. Redox metabolism in 4 *M. incognita* susceptible hosts expressed as μg of TTC reduced per mg wt of tissues.

Enzyme activities	<i>H. cannabinus</i>		<i>P. mungo</i>		<i>H. esculentus</i>		<i>S. melongena</i>	
	UT	PT	UT	PT	UT	PT	UT	PT
TDH Root	375.8	310.4	298.1	256.8	238.2	202.6	198.4	162.4
Shoot	198.4	175.8	162.7	142.4	129.8	102.4	98.2	78.9
Total	574.2	486.2	460.8	399.2	368.0	305.0	296.6	241.3
Per mg Wt	287.1	243.1	230.4	199.6	184.0	152.5	148.3	120.65
TER Root	275.8	236.8	214.1	190.4	169.9	152.4	137.1	124.1
Shoot	152.4	128.4	108.5	97.1	80.2	60.4	59.0	40.0
Total	428.2	365.2	322.6	287.5	250.1	212.8	196.1	164.1
Per mg Wt	214.1	182.6	161.3	143.75	125.05	106.4	98.05	82.05
Rate of synthesis (%)	74.57	75.11	70.01	72.02	67.96	69.77	66.12	68.01
TER TDH $\times 100$								

Values are average ($\pm 0.001 - 0.01$) of 6 replicates.

UT, No phenol; PT, phenol treated.

$$R_{Pi} = \frac{P_f - P_i}{P_i \times \text{post inoculation period (45 days)}}$$

P_f is the final pathogen population and P_i is the initial inoculum applied.

Triphenyl tetrazolium chloride (TTC) was employed as the artificial electron acceptor for determining the redox enzyme activities. The rates of synthesis in the plants were then computed as a measure of relations between the total dehydrogenases (TDH) and the total endogenous reductases activities (TER) (table 2) (Kannan 1967, 1968).

3. Observations

Table 1 shows that for the initial inoculum (P_i) of 10^3 juveniles, the 4 viable genomes exhibit varied amount of final population (P_f) at the end of 45 days, indicating their susceptibility ratings (SR). The SR in them runs in a decreasing order as follows: *H. canabinus* > *P. mungo* > *H. esculentus* > *S. melongena*, indicating the high susceptibility of *H. canabinus* and low susceptibility of *S. melongena* under NPK influence.

The introduction of phenol resulted in a smaller size of the final population, reflecting the alleviating effect of phenol by reducing the pathogen impact, though of course in accordance with the susceptibility ratings of those hosts. The final population load in the 4 viable hosts under NPK and under the influence of phenol, indicate the host responses in this screening test.

In contrast to the hosts' susceptibility ratings, the total ERF-(table 1) encountered by the pathogen in its two environs, viz. inside the plant and outside the soil, is observed to operate in the reverse direction of the hosts' susceptibility ratings, as given here: $ERF = S. melongena > H. esculentus > P. mungo > H. canabinus$. Thus a host with high SR as *H. canabinus* is associated with low ERF and *S. melongena* with low SR, is associated with high ERF, both under NPK and under phenol influences.

Host pathogen relations (HPR) is the resultant of 2 forces viz. plant and the pathogen, both of them subjected to the soil influences. It can be observed that success of the pathogen is the result of a low resistance encountered by it in the plant environment and also the soil, which is finally exhibited as high or low pathogen impact on the host, which is now graded genetically for its SR or resistance as the case may be. Therefore it is clear that the final interpretations of the host-pathogen relations are based on the interactions of the 2 forces viz. SR and ERF. Hence HPR can be signified as $SR \leftrightarrow ERF$ or $HPR = SR/ERF$ indicating the role of the host and pathogenic factors.

These factors are applicable to both the NPK and phenol treatments as indicated by the varying pathogen loads (P_f) sustained in the 4 viable hosts, indicative of the SR and the varied ERF observed in the plant and in the soil (table 1). The influence of the environmental resistance over the pathogen, inside the plant and outside the soil, is well observed in the varied pathogen contents in these 2 environs and the specific influence of phenol is well observed (table 1) in the increased ERF in both the environs as a result of which the final pathogen load is depressed in these 2 environs, resulting now in altered HPR relations in the 4 viable genomes, which are now presumed to exhibit improved vigour through phenol amendments of the soil.

It is well known (Van Gundy and Stolzy 1961; Van Gundy *et al* 1964; Kirkpatrick

et al 1964; Dropkin 1969; Wallace 1969; Nardicci and Barket 1979), that, extremes of soil environment (soil heat, porosity, moisture, pH and O₂ relations) which impede the plant growth also impede the pathogen's success and that, a well nourished plant rallies round the infection by virtue of its nutrition. Thus the successful pathogenesis has the environmental bearing also, in addition to the host's genetic viability or otherwise.

It is therefore clear that breeding for resistant hosts, involving gene shuffle and soil amendments, are ultimately meant to pose altered environs to the pathogen, to contain its impact, if not totally eradicate that. The utility of soil amendments is centred around the environmental impacts on the pathogen, more possible and easily, by varied amendments with different agents as phenols, phenolics etc. since in genetic reshuffle it is not possible to dictate the polygenic interplay, while at the same time the evolution of resistant biotypes of pathogens cannot also be ignored, for a given genetic effort.

The source size, strength-sink size and strength (Cowling and Horsfall 1980) show the bidirectional transit of raw and finished products through the metabolic continuum in the plant. It is also known (Cowling and Horsfall 1980) that during crisis (dietary and pathogenic) the host spends the minimal energy to meet the crisis demands, conserving the rest for repair through synthesis of proteins, lipids, phenols etc. through metabolic pathways involving redox enzymes (Goodman *et al* 1967). It is needless to point out such synthesis during nematosis as extensively documented in nematology.

The quiescence of the above is reflected in table 2, indicating the redox metabolism during the pathogenic crisis under NPK and phenol influences. It is observed that the dehydrogenases are more active in a highly susceptible host as *H. cannabinus*, than in *S. melongena* in conjunction with their SR ratings, under NPK treatments. Their recoveries through endogenous reductases also run in the same fashion. With the introduction of phenol, which resulted in improved ERF values and hence lowered pathogen population, the dehydrogenase activities also fall down, indicating reduced catalysis due to reduced pathogenic impact. Correspondingly the synthetic rates as expressed by endogenous reductases, are also depressed, suiting the need of the demands.

The metabolic continuum in the plant is known (Cowling and Horsfall 1980) to compensate for the ill effects of crisis (dietary-pathogenic) by reallocation of resources, triggering reactions at all regions, which exhibit different responses of growth and metabolism, which ultimately are involved in compensations through growth and metabolism. Such compensations are also observed in the present studies in the differential redox metabolic responses of the root and shoot under NPK and phenol influences. A significant feature in the present studies is, the significant recovery gains effected ranging from 66–75% under various nutritional impacts as observed in table 2, indicating the efforts put up by the viable genomes to tide over the crisis. In other words, such viable genomes with genetically ingrained susceptibility, exhibit functional resistance, brought to play during the crisis, which is now interpreted in terms of improved vigour of those susceptible hosts.

Such an improved situation is due to phenol's high compatibility with the host's factors (metabolites) and its repressive effects on the pathogen's enzyme. Posed with these two hazards, viz. altered internal milieu of the plant and enzyme repression, this obligate endopathogen is faced with impeded nutrition with consequent impact

on its progeny and hence reduced pathogenic impact on the host, which is interpreted in terms of improved vigour of the viable host.

Therefore, agents like phenol exhibiting metabolic compatibility with host and pathogen factors, can be advantageously exploited for improving plant vigour for better agricultural gains. Such agents, if with extensive side radicals of high reactive nature, permitting high dilutions as phenol in the present case, require exploration since the high dilution factor will help reduce the cost of control measures.

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