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Red Light-Induced Systemic Resistance to Root knot Nematodes in Tomato

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Abstract
This study was conducted to evaluate efficiency of red light to induce systemic resistance in tomato plants grown in 1kg plastic pots under green house conditions against *Meloidogyne* spp. Results indicated that tomato seedlings exposed to red light for 2 and 3wks were significantly (p=0.05) more resistant to the root knot nematodes than seedlings exposed to white light (control seedlings). This was manifested by the less number of galls on the roots of treated compared with control plants 30d after inoculation. Root gall index(gi) was 2.77 for red light treated and 5 for control plants. Root fresh and dry weight was 5.00 and 0.39 g for red light treatment compared with 6.31 and 0.64 g for white light control treatment respectively. Significantly (p=0.05) less (55) second stage juveniles (J2) enter the roots of red light treated plants compared to 123.66 J2 in control.
Keywords: Induced resistance, *Meloidogyne* spp, Red light, Tomato.

1. Introduction

Tomato, *Solanum lycopersicum* is one of the most important vegetable plants in the world. It is the second most important vegetable crop after potato with annual world production of about 152.9 million ton in 2009 (Anonymus, 2009). Annual tomato production in Iraq was estimated at 830.000 tons in 2008 (FAOSTAT, 2008). Tomato plants are attacked by many important plant pathogens. The root knot nematodes, *Meloidogyne* spp are considered as the most important nematode species worldwide and in Iraq. Many effective control measures were used to manage these pathogens such as resistant cultivars, soil solarization, chemical and biological control. Recently a new and renovated control, the acquired induce systemic resistance was also tried to control different plant pathogens including the root knot nematodes. Chemical, physical and biological inducers to control the root knot nematodes were used (Sahebani et al., 2011). Since light affect all the biological aspects of plants especially photosynthesis, the ultimate source of energy, it is therefore affect disease development in plants (Hong Wong et al., 2010). Light of different wave length plays an important role in the defense system of the plant host (Honda and Yunoki, 1976; Tan, 1978; Kumagai,1988) . Numerous research work have been accumulated in the literature reporting successful induced resistance against many plant pathogens such as fungi (Khanam, 2005; Ismail et al., 1998,2002,2004;Rahman et al.,2003). Recently, Ismail et al., (2008) showed that red light can induced resistance to *M. javanica and Pseudomonas syringae* in *Arabidopsis thaliana*.
This work was done to evaluate the effectiveness of red light in inducing acquired systemic resistance to *Meloidogyne* spp in tomato.

2.Materials and Methods

The experiments were performed in a greenhouse (27±5 C) at the Department of Plant Protection, College of Agriculture, Baghdad University and tomato, *Solanum lycopersicum* L. "Supper Regina" highly susceptible to *Meloidogyne* spp plants were used. Plants were grown and maintained in1kg plastic pots throughout the experiments.

2.1. Tomato plants

Tomato, *Solanum lycopersicum* L. cv. "Super Regina" (Genetics International Inc, Modesto, California, USA) susceptible to *Meloidogyne* spp was used.

2.2. Nematode inoculums

Soil from cucumber grown plastic house heavily infested with *Meloidogyne* spp (predominantly *M. javanica*) was collected and stored at 4C in polyethylene pages until use to inoculate tomato seedlings. The nematode infested soil was mixed with peat moss in 1:1 ratio and used as nematode inoculums.

2.3 Physical Treatment (Exposure to Red Light)

Tomato seedlings containing 4-5 true leaves were exposed to red light from 6 lamps (50-60 HZ, 220-240 V, and German Technology) and to regular white light 12h daily. Plants exposed to white light only for the same duration are considers as control treatment. The duration of exposure to red light was 1, 2, and 3wks. Seedlings then were transplanted in plastic pots containing nematodes infested soil and peat moss at 1:1 ratio and maintained in a greenhouse. Each treatment was replicated 4 times and each pot contained one plant.

2.4. Parameters of Evaluation of Red Light Induced Resistance
2.4.1. Rate of Root Gall Index of *Meloidogyne* spp Infected Tomato
The rate of infestation of the root knot nematodes was determined using 5 level scale (Dube and Smart, 1987): 1= no galls on roots, 2= galls on 25% or the root, 3= galls on 50% or the root, 4= galls on 75% of the root, and 5, galls on 100% of the root.
2.4.2. Nematode Penetration
To determine the effect red light treatment on nematode penetration of tomato roots 1wk after nematode inoculation, roots were stained with acid fuchsine (Byrd et al., 1984), washed with water to remove excess stain and 1g of each root was individual examined under a compound microscope to count nematodes inside the roots.
2.4.3. Fresh and Dry Weight of Root and Shoot Systems
Plants were carefully uprooted and roots were washed under tap water to remove adhering soil. To determine shoots and roots dry and wet weight, shoots and roots were separately weight and then dried at 70C for 48h or until weight fixed.

3. Statistical Analysis
The data were subjected to analysis of variance and means were separated by the least significant method at (p=0.05) using SAS, 2004.

4. Results
4.1. Effect of Red Light on Rate of Root Gall Index (GI)
Red light treatment induced resistant to *Meloidogyne* spp in tomato plants. Red light significantly (p=0.05) reduced the GI of treated plants compared with white light- treated control plants (Table 1).While red light treated plants recorded the lowest average GI of 3.50, untreated plants recorded the highest average GI of 5, 30d after nematode inoculation .Two and 3 wks exposure to red light render tomato plants significantly more resistant to the nematode than 1wk exposure to red light or white light exposed control plants. This was manifested by the low GI, 2 and 2.33 for 2 and 3wks compared with the high GI of 4 and 5 in 1wk exposure to red light and white light exposed control plants respectively.
4.2. Effect of Red Light on Nematode Penetration
Red light treatment caused significant (p=0.05) reduction in the number of J2 penetrating tomato roots (Table 2). The average number of J2 in red light exposed plants was 55.00 compared to 123 J2 in white light treated plants. Significantly, the lowest J2, 26.66 was detected in roots of plants treated with red light for 2wks compared with 93.66, 44.66 and 123.66 J2 in 1wk, and 3wks red light treated plants and white light treated controls respectevely.
4.3. Effect of Red light on Root Weight
Red light treatments significantly (p=0,05) reduced root fresh and dry weight (Rfw and Rdw) 30d after nematode inoculation (Table 3).The average Rfw and Rdw of red light treated plants scored 5.00 and 0.39 compared with 6.31 and 0.61g for white light treated control plants respectively. No significant differences, however, were detected in root weight of plants exposed to 1, 2 and 3wks of red light. Results also indicated that 2wk exposure to red light caused significantly the highest Rfw and Rdw reduction, 4.00 and 0.63 compared to7.94 and 0.79g in control plants.
4.4. Effect of Red light on Shoot Weight
Red light treatments caused significant (p=0.05) increase in shoot fresh and dry weight (Sfw and Sdw) of tomato plants compared white light control treatments (Table 4). The average Sfw and Sdw were 26.12 and 1.70 in red light treated plants compared with 16.89 and 1.01g in white light control plants respectively. Plants exposed to red light for 2wks recorded the highest Sfw and Sdw,39.64 and 3.25 compared with 17.31, 0.35, 21..42, 1.55g in 1wk and 3wks treated plants respectively.

5. Discussion
A variety of biotic and a biotic agents have been reported to induce systemic acquired resistance to plant pathogens including plant parasitic nematodes (Sahebani et al., 2011). Results of this work demonstrated that exposure of tomato plants to red light significantly enhance their resistance to *Meloidogyne* spp and confirmed the only published report on red light-induced plant nematode resistance in which red light induced resistance to *M. javanica* in *Arabidopsis thaliana* (Islam et al., 2008). The mechanism of red light-induced resistance is not yet fully understood, however, result of this study suggest that red light treatment render roots of tomato plants less attractive to J2 of *Meloigogune* spp. light of different wave lengths play important roles in plant defense against various plant pathogens (Honda and Yunoki, 1976; Tan, 1978; Kumagai,1988). Photosynthesis, the ultimate energy source process for green plants, is one of the most important factors which affect the development of plant diseases (Hong Wong et al., 2010). Light was reported to inactivate toxins produced in

certain plant- pathogen host interaction (Kohmoto et al., 1989). In this study red light treatment significantly (p=0.05) reduced gall development on treated plants and the number of J2 in roots compared with those in white light control plants (Tables 1&2). These results support previous report using red light to induce resistance to *M. javanica* (Islam et al., 2008). Red light treatment caused significant increase in root weight and reduction in shoot weight (Tables 3&4). *Meloigogune* spp infection is known to have negative effects on water and nutrient elements as well as photosynthesis (Melakeberhan et al., 2004). It was reported that *M.incognita* infection caused biomass accumulation in roots and this is controlled by the efficiency of the pathogen in capturing the light energy and directing it in favor of the pathogen or the infected host (Melakeberhan et al., 1988). Because of the relative large size of females of *Meloigogyne* spp and its ability to produce large number of eggs, it requires large amount of energy. Beside this energy requirement, these pathogens caused obvious distortion in xylem vessels, swellings of root cells and formation of giant feeding cells which alter root normal functions. It is also believed that induced resistance to root knot nematode is related to icresed levels of salicylic acid and pathogenesis related proteins in red light treated plants (Wang et al., 2010). Further research on this aspect of induced resistance to *Meloidogyme* spp is needed before a reliable and practical management of root knot nematodes by red light is achieved.

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Table 1. Effect of red light exposure duration on rate of root gall index of tomato, *Solanum lycopersicum* L. 30 days after inoculation with *Meloidogyne* spp

Exposure (wk)	Root Gall Index		Mean
	White	Red	
1	5	4	4.50
2	5	2	3.50
3	5	2.33	3.66
Mean	5	2.77	
LSD (P=0.05) Light = 0.24*, Treat. = 0.29*, Inter. = 0.41 *			

Each number is a mean of three replicates and two plant each.* indicate significant difference. Nematode inoculums (nematode infested soil + peat moss, 1:1) were added when plants were 4-5 true leaves. Gall index was according to 1-5 level scale : 1= no galls on the roots , 2= galls on 1- 25% of the root , 3= galls on 26- 50% of the root , 4= galls on 51- 75% of the root, and 5= galls on 76-100% of roots.

Table 2. Effect of red light exposure duration of tomato, *Solanum lycopersicum* L. on nematode penetration 1 wk after inoculation with *Meloidogyne* spp

Exposure (wk)	No. of J2/1g of root		Mean
	White	Red	
1	123.66	93.66	108.66
2	123.66	26.66	75.16
3	123.66	44.66	84.16
Mean	123.66	55.00	
LSD (P=0.05) Light = 3.27 * Treat. = 4.01*, Inter. = 5.67*			

Each number is a mean of three replicates and two plant each.* indicate significant difference. Nematode inoculums (nematode infested soil + peat moss, 1:1) were added when plants were 4-5 true leaves.

Table 3. Effect of red light exposure duration of tomato, *Solanum lycopersicum* L. on root weight 30 days after inoculation with *Meloidogyne* spp

Exposure (wk)	Root wt (g)				Mean	
	White		Red			
	Dry	Fresh	Dry	Fresh	Dry	Fresh
1	0.78	6.03	0.68	5.25	0.73	5.64
2	0.79	7.94	0.63	4.00	0.71	5.97
3	0.37	4.96	0.43	5.75	0.40	5.35
Mean	0.64	6.31	0.39	5.00		
LSD (P=0.05) Light = 1.21*, Treat.=1.48^{ns} , Inter.= 2.1* (fresh wt) LSD (P=0.05) Light = 0.1554*,Treat.= 0.1904*, Inter.= 0.2692* (dry wt)						

Each number is a mean of three replicates and two plant each.* indicate significant difference. Nematode inoculums (nematode infested soil+ peat moss, 1:1) were added when plants were 4-5 true leaves.

Table 4. Effect of red light exposure duration of tomato, *Solanum lycopersicum* L. on shoot weight 30 days after inoculation with *Meloidogyne* spp

Exposure (wk)	Shoot wt (g)				Mean	
	White		Red			
	Dry	Fresh	Dry	Fresh	Dry	Fresh
1	0.26	12.94	0.35	17.31	0.30	15.12
2	1.49	19.45	3.25	39.64	2.35	29.55
3	1.29	18.28	1.55	21.42	1.42	19.85
Mean	1.01	16.89	1.70	26.12		
LSD (P=0.05) Light= 2.45*,Treat.=3* , Inter.= 4.25 *(fresh wt) LSD (P=0.05) Light= 0.43*,Treat.=0.5313*, Inter.= 0.7514* (dry wt)						

Each number is a mean of three replicates and two plant each.* indicate significant deferens. Nematode inoculums (nematode infested soil + peat moss, 1:1) were added when plants were 4-5 true leaves.