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Association of *Meloidogyne incognita* (Kofoid and White) Chitwood with wilt complex disease of betelvine (*Piper betle* L.)

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Abstract
An intensive roving survey was conducted during 2015-16 and 2016-17 to record the incidence of betelvine wilt complex disease in northern parts of Karnataka. Percent disease incidence ranged from 10.6 to 28.4, typical galling symptom of root knot nematode was observed in wilt affected betelvine plants. Isolation and identification of pathogens was confirmed by following Koch's postulates, nematode inoculated plants showed the typical symptoms as recorded in the field. Root knot nematode was more frequently associated with *Sclerotium rolfsii*, *Rhizoctonia bataticola* and *Fusarium* spp. resulting in maximum wilt disease incidence.

Keywords: Betelvine, wilt complex, root knot, identification, *Meloidogyne incognita*

1. Introduction
Betelvine (*Piper betle* L.) regarded as natural green gold, is an important horticultural crop of commercial values and belongs to the family Piperaceae [1]. The plant is economically, medicinally and traditionally important in the whole world. Successful cultivation of betelvine suffers due to several biotic and abiotic factors including root and aerial diseases, among these wilt/ root rot caused by many fungal pathogens like *Phytophthora* spp. [1], *Rhizoctonia solani* and *R. bataticola* [2], *Fusarium* spp. [3], *Pythium* spp. and *Sclerotium rolfsii* [4]. All these wilt pathogens cause severe crop losses alone or when they are associated with root knot nematode *Meloidogyne incognita* [5]. The losses from wilt disease vary from 2 to 100 percent [6].
The association of plant parasitic nematodes with other microorganisms like fungi and bacteria often results in disease complexes where damage expression on crop may be additive, neutral or synergistic. The nematodes have been found important and vital in the development of wilt and root rot diseases caused by fungi and bacteria. Sitaramaiah and Devi [7] observed positive correlation between the populations of *M. incognita* with wilt disease incidence in infested gardens. By considering above facts, present investigations were undertaken to study the association of *Meloidogyne incognita* with other pathogens inciting wilt complex disease of betelvine.

2. Materials and methods
An intensive roving survey was conducted in major betelvine growing districts *viz.*, Bagalkot, Belagavi, Davanagere and Haveri districts of northern Karnataka, during 2015-16 and 2016-17.
Samples of infected and healthy plants with root and stem along with adhering rhizosphere soil from each orchard were collected. These samples were brought to laboratory for isolation of pathogens. The disease incidence in the orchards was assessed with the following formula give by Maiti and Sen [8].

$$\text{Percent disease incidence} = \frac{\text{Number of plants infected}}{\text{Total number of plants examined}} \times 100$$

2.1 Extraction of nematode
Cobb's sieving and decanting technique [9] was followed, for which 200 cc of the sample was taken in a container and mixed thoroughly with water.

Hard particles and stones, if any, were removed by stirring the suspension and then passed through set of sieves of 100, 250, 300 and 460 mesh sizes. The sievates from 300 and 460 mesh sizes were collected on a tissue paper spread over a coarse mesh, which was then placed in a Petri dish containing enough water so as to keep the tissue paper just wet.

The nematode suspension collected in the Petri dish was examined using research stereo binocular microscope. The root knot nematode present in the suspension was identified by observing different morphological characters.

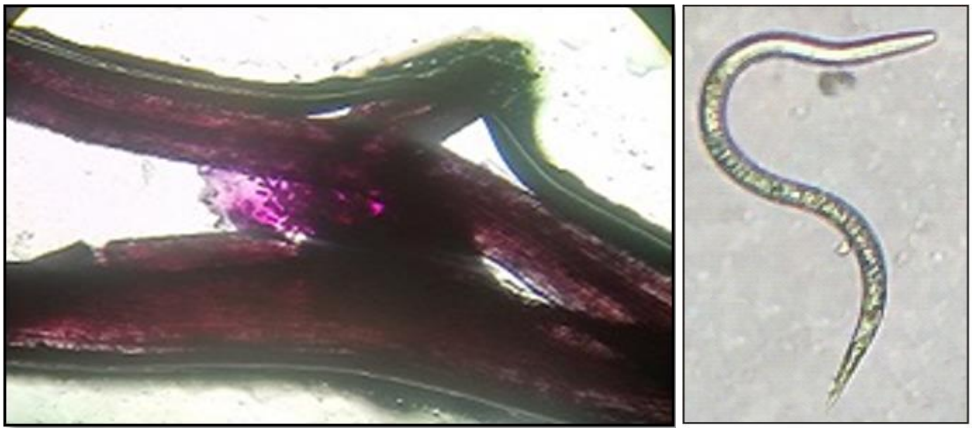
2.2 Proving the pathogenicity

The betelvine plants were raised in earthen pots containing sterilized soil, sand and farm yard manure in 3:1:1 ratio. The pots were inoculated with *Meloidogyne incognita* and a control treatment was maintained without inoculum. The egg masses of *M. incognita* from stock culture were transferred carefully to a wire gauze sieve containing two layers of facial tissue paper trimmed down to edge of wire gauze and kept in a Petridish holding sufficient water to remain in contact with the bottom of Petridish. After 24 hr, the contents of a Petridish were emptied into a beaker, diluted to a suitable volume and population counts were made with the help of Fenwick's multi chamber counting slide. Based on the requirement, the suspension was diluted with sterile water and inoculated to 90 days old betelvine plants.

The pots were maintained at 25 percent water holding capacity and the moisture loss was maintained by adding water on weight basis. Observation was made every day regarding development of wilt symptoms. Plants showing wilt symptoms were carefully uprooted and the associated organisms were re isolated by following standard procedure. The culture thus obtained was compared with original culture to confirm the identity of the fungus and subsequent confirmation of Koch's postulates.

3. Results and Discussion

The *Meloidogyne incognita* was identified based on the morphological characters. The observed characters were compared and confirmed with the description given by Eisenback *et al*. [10] (Plate 1).



A. B.

Plate1: Identification of *Meloidogyne incognita:* A) Female of *Meloidogyne incognita* inside the root and B) Juvenile of *Meloidogyne incognita*

The infected plants exhibited symptoms of yellowing, reduced leaf size and stunted growth, yellow coloured small sized leaves with each vine bearing a few leaves. Root system of the infected plants showed prominent galls of varying sizes. Microscopic examination of the galls revealed the presence of eggs, juveniles, males and females of nematodes in the vascular bundle of roots (Plate 2). Similar findings were reported by earlier workers Parameshwari [11] Ammajamma [12] and Meena *et al*. [13] the root knot nematode infected plants exhibited symptoms of stunted growth, yellowing of leaves and reduction in leaf size with each vine bearing a few leaves. Prominent galls of varying sizes were observed in the root system of infected plants. Microscopic examination of the galls revealed the presence of eggs, juveniles, males and females of nematodes in the vascular bundle of roots.

A. B.

Plate 2: Proving of Pathogenicity: A) Galls on root system and B) Infected plant

3.1 Association of *Meloidogyne incognita* with other pathogens

The data presented in Table 1 revealed that, in Bagalkot district out of nine orchards surveyed one orchard showed the presence of root knot nematode (*M. incognita*) along with fungal pathogens (*S. rofsii*, *R. bataticola* and *Fusarium* spp.) with the mean disease incidence of 11.40 percent. In Belagavi district twenty orchards were surveyed, three orchards recorded the association of root knot nematode with one or more fungal pathogens. In Davanagere district out of twelve orchards surveyed, three orchards recorded the presence of *Meloidogyne incognita* with fungal pathogens. Haveri district twenty five orchards were surveyed *M. incognita* alone was found in one orchard and four orchards recorded the association of *M. incognita* with other fungi. The data also revealed that maximum disease incidence was recorded in the orchards where root knot nematode was found associated with the fungal pathogens. The nematode punctures on the root system provide entry points for colonization of saprophytic fungi. The results are in conformity with the findings of earlier workers, Acharya *et al.* [14] studies the association of *Meloidogyne incognita* with *Sclerotium rofsii* and *Xanthomonas betlicola* on betelvine. Jonathan *et al.* [15] reported the association of root knot nematode with betelvine. Brahmankar *et al.* [16] also reported the association of root knot nematode with *Fusarium* spp., *Rhizoctonia*, *Botrydiplodia*, *Phytophthora* and *Pythium* spp. causing wilt of betelvine in Vidarbha region of Maharashtra.

Table 1: Frequency of major pathogens associated with wilt complex disease of betelvine in major growing areas of northern Karnataka

				Pathogens														
District	**Taluk**	**PDI**	**No. of orchards surveyed**	**S**	**R**	**F**	**M**	**S+F**	**S+R**	**S+M**	**F+M**	**R+F**	**R+M**	**F+M**	**F+R+M**	**S+F+M**	**S+R+F**	**S+R+F+M**
Bagalkot	Badami	12.9	5					2				2						1
	Bilgi	10.9	2			1						1						
	Jamkhandi	10.6	2			1						1						
	Mean	11.4																
Belagavi	Chikkodi	13.0	6			1			1			2				1	1	
	Gokak	15.7	5			1		1		1		1					1	
	Hukkeri	16.1	5					1				1					2	1
	Raybag	15.0	4			2											2	
	Mean	14.9																
Davanagere	Channagiri	12.0	3			2						2						
	Harapanahalli	17.7	2					1							1			
	Harihar	25.1	4					1				1			1			1
	Honnali	17.1	3			1		1				1						
	Mean	18.0																
Haveri	Byadgi	11.9	3			2		1										
	Haveri	17.8	4			2						1					1	
	Hirekerur	20.7	4															
	Ranebennur	28.4	5				1	1									2	1
	Savanur	16.7	5	1	1							1					1	1
	Shiggaon	11.8	4			1			1			1			1			
	Mean	17.9																

4. Conclusion

The present investigation it was found that the root knot nematode (*Meloidogyne incognita*) was found to be associated with the fungal *pathogens viz., Sclerotium rolfsii, Rhizoctonia bataticola* and *Fusarium solani* that resulted in disease complex. The presence of nematode along with fungal pathogens also increases the wilt disease incidence.

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6. References

1. Gunther E. The Essential Oils. D. Van Nostrand & Co, Inc. New York. 1952; 5:160-161.
2. Marimuthu T. Fungal nematode wilt complex in betelvine (*Piper betle* L.). Plant Disease Researcher. 1991; 6(2):85-88.
3. Mc Rae W. Foot rot disease of *Piper betle* in Bengal. Indian Agricultural Science, 1934, 585-617.
4. Singh BP, Joshi LK. Studies on the diseases of *Piper betle* in Jabalpur (Madhya Pradesh). Four new world records of fusaria, Science and Culture. 1972; 38**:**285.
5. Dastur JF. Diseases of pan (*Piper betle* L.) in central provinces. Proceedings of Indian Academic Science. 1935; B.1(11):778-815.
6. Medhane NJ, Pawar AB. Proceedings of First National Workshop on Betelvine. 1982, 109-111.
7. Saksena S. *Phytophthora parasitica* the secourge of 'pan' (*Piper betle* L.). Indian Phytopathology. 1977; 30:1-16.
8. Sitaramaiah K, Devi GP. Influence of root knot nematode *Meloidogyne incognita* on sclerotium wilt of betelvine. Indian Journal of Nematology. 1990; 20:220-231.
9. Maiti S, Sen C. Fungal diseases of betelvine, PANS. 1979; 25:150-157.
10. Cobb NA. Estimating the nematode population of the soil. Agric. Tech. Circ. Bur. Pl. Ind. U.S. Dep. Agric. 1918, 1:48.
11. Eisenback JD, Hirschmann H, Sasser JN, Triantaphyllou AC. A guide to the four most common species of root knot nematodes (*Meloidogyne* spp.) with a pictorial key. A cooperative publication of the Department of Plant Pathology and Genetics. North Carolina State University and United States Agency for International Development, Releigh, North Carolina, 1981, 48.
12. Parameshwari B. Studies on wilt complex of betelvine (*Piper betle* Linn.), M.Sc. Thesis, University of Agricultural Sciences Dharwad, Karnataka (India). 2003, 56-57.
13. Ammajamma R. Studies on etiology, epidemiology and

management of wilt complex of *Coleus forskohlii* (Wild.) Briq. Ph. D. Thesis, Univ. Agric. Sci. Dharwad, Karnataka (India), 2010.
14. Meena SK, Ramyabharathi SA, Raguchander T, Jonathan EI. Interaction of *Meloidogyne incognita* and *Fusarium oxysporum* in carnation and physiological changes induced in plants due to the interaction. SAARC Journal of Agriculture. 2016; 14(1):59-69.
15. Acharya A, Dash SC, Padhi NN. Pathogenic association of *Meloidogyne incognita* with *Sclerotium rolfsii* and *Xanthomonas betlicola* on betelvine. Indian Journal of Nematology. 1987; 17(2):196-198.
16. Jonathan EL, Arulmozhiyan R, Muthswamy S, Manual WW. Field application of *Paecilomyces lilacinus* for the control of *Meloidogyne incognita* on betelvine, *Piper betle*. Nematologia Mediterranea. 2000; 28(2):131-133.
17. Brahmankar SB, Dange NR, Deepali GT. Organisms associated with wilted betelvine in Vidarbha. International Journal of Plant Protection. 2011; 4(1):236-238.