GRAFTING ON *Solanum sisymbriifolium* ROOTSTOCK TO MANAGE ROOT-KNOT OF TOMATO IN PLASTIC TUNNEL

S. Baidya*, RD Timila*, Ram Bahadur KC**, HK Manandhar*, C Manandhar* and CK Timalsina***
*Plant Pathology Division, Khumaltar, Lalitpur
**Horticulture Research Station, Malepatan, Kaski
***Regional Agricultural Research Station, Lumle, Kaski

ABSTRACT

Rroot knot nematode (Meloidogyne spp.) is difficult to manage once established in the field because of its wide host range and soil-borne nature. For the management of root knot nematode, grafted plant with resistant rootstock of wild brinjal (Solanum sisymbriifolium) was tested under farmers' field conditions at Hemja of Kaski district during 2008-10. Grafted and seedling planting (non-grafted) materials were grown in root-knot nematode free soil. Around three week-old grafted and non-grafted tomato plants were transplanted in four different plastic tunnels where the nematodes had been reported previously. The plants were planted in diagonal position to each other as a pair plot in 80 × 60 cm spacing in an average of 20 × 7 m plastic tunnels. Galling index was recorded three times in five randomly selected plants in each plot at 60 days intervals. The first observation was recorded two months after transplanting. Total fruit yield was recorded from the same plants. In the observation, the root system of grafted plants was totally free from gall whereas on an average of 7.5 galling index (GI) in 0-10 scale was recorded in the seedlings . Fruits were harvested from time to time and cumulated after final harvest to calculate the total fruit yield. It was estimated that on an average tomato fruit yield was significantly increased by 37 percent in the grafted plants compared with the seedlings non-grafted plants. Grafting technology could be used effectively for cultivation of commonly grown varieties, which are susceptible to root-knot nematode in disease prone areas. This can be used as an alternative technology for reducing the use of hazardous pesticides for enhancing commercial organic tomato production.

Key words: Grafting, root-knot nematode (*Meloidogyne* spp.), tomato, wild brinjal (*Solanum sisymbriifolium*)

INTRODUCTION

Plant parasitic nematode is microscopic un-segmented worms, commonly described as filliform or thread-like multicellular invertebrates. Every cultivated plant species has at least one nematode species reported to parasitize it (Luc *et. al.*, 2005). Among the plant parasitic nematode, root-knot nematode (*Meloidogyne* spp.) is most commonly prevalence parasitic nematode in the country. Once high populations of *Meloidogyne* have developed in a field, it is virtually impossible to suppress and maintain populations at sufficiently low level regardless of control method practiced. Root-knot nematode is common and destructive plant parasitic nematode which has wide host range and can significant yield loss in many crops. It is polyphagous and reported in many crops such as tomato, brinjal, okra, potato, chilly, radish, carrot, onion, cabbage, cauliflower, lettuce, chickpea, broad bean, cowpea, cucumber, pumpkin and various gourds from plains to high hills of the country (Amatya and Shrestha, 1969, and Annual report, 2009). More than 90 species of *Meloidogyne* have been reported from different part of world but only four species such as *M. javanica*, *M. arenaria*, *M. incognita* and *M. hapla* have been reported in Nepal (Mathur *et.al.*, 1998, and Luc *et.al.*, 2005).

In tomato, root-knot nematode is a major biotic factor which is responsible for low yield. It has also been reported that in an average of 30 percent tomato fruits was reduced due to nematodes tomato growing in plastic tunnel at Hemja of Kaski district (Annual report, 2008). The problem of nematode has been increasing like a forest fire especially in year round plastic tunnel tomato cultivation. The intensive cropping of tomato in the plastic tunnels due to its high value is one of the barriers for the control of the nematode through crop rotation. Continuous mono cropping is inevitable in vegetable production in indoor areas and this reduces the yield and quality of produce due to damage caused by nematodes and other soil borne diseases (Augustin *et. al.*, 2002; Poffley, 2003). Although nematodes are difficult to eradicate from a field, it is essential to reduce them below the economic damage threshold to help avert a reduction in the productivity. Preventing the development of nematodes problem may be more economical than managing the nematodes once the problem develops. In many developed countries, chemical control is more widely used in nematode management. However, increasing concerns of the environmental risk posed by nematicide and development of number of viable alternatives, the concept of chemical control is not only inefficient and uneconomic but also biologically unsound and unacceptable to communities. In an alternative to chemical pesticides, various strategies have been employed by researchers as well as farmers for the management of the nematodes around the world. Among them, root-knot nematodes can also be effectively minimized by using resistant rootstock grafting with susceptible cultivars (Bersi, 2002, and Luc *et.al.*, 2005). Hence, this research study was conducted as grafted plants using resistant rootstock of wild brinjal (*Solanum sisymbriifolium*) against the root-knot nematode is the most viable alternative practice to cope the existing farmer's problem especially in the case of plastic tunnel tomato cultivation.

MATERIALS AND METHODS

Identification of root-knot nematode species from tomato roots

Galled roots were collected for identifying root-knot species from experimental plastic tunnels. Six different galled roots samples of tomato from three different plastic tunnels, two roots from each tunnel, were collected at Hemja of Kaski district. Five fresh adult females from each rooting system altogether 30 females were extracted and diagnosed the species of *Meloidogyne*. The galled roots were stained in cotton blue for removing body content of nematode easily. Stained females were dissected from root tissue by needle and kept in Petridisc containing water. The posterior end of dissected females was cut off with scalpel on the Petridisc cover and inner tissue was removed carefully by small soft brush. The cuticle was spread with outside uppermost that the cutting opening underneath and the perineal pattern uppermost on the slide having a drop of glycerol and covered with cover slide. The mounted perinial patterns were examined in microscope to identify the species with viewing on the different pattern of the posterior parts.

Estimation of galling index and total yield

Seedlings of wild brinjal (*solanum sysimbriifolium*) were grown for resistant rootstock in nursery at Horticulture Research Station (HRS), Malepatan of Kaski dristict. Around 40days-old wild brinjal plants were used as rootstock for grafting on the scion of the 21 day-old tomato plant (Thim-16) by cleft grafting technique. The grafted junction was covered with paraflim. Immediately, the grafted plants were kept inside the grafting house for 7-8 days at $25 \pm 2°C$ in more than 90 percent relative humidity (RH). The plants were transferred to the shade nursery house for hardening in open environment. After a week, the healthy grafted plants were selected for transplanting in plastic tunnel. Before transplanting, all the leaves grown below the graft union was removed. In the meantime, the seedlings of Thim-16 were also produced in the nematode free nursery bed. Both the grafted and seedling tomato plants were transplanted in four different plastic tunnels in an average of 20×7 m where the nematodes had been reported previously. The experiments were carried out in two years from 2008 to 2010 at Hemja of Kaski district. During planting, the graft union was kept above the ground to avoid root growth from the scion. In each tunnel, both grafted and seedling plants were transplanted in diagonally to each others in as pair plot in 80×60 cm spacing. In the tunnels, remaining all the required inputs (fertilizers, irrigation, staking, weeding, nutrition, and insecticides) were equally provided to all plants. Galling index was recorded three times in five randomly selected plants in each plot at 60 days intervals. Gall index was recorded in 0-10 scale as described by Bridge and Page, 1980. The first observation was recorded two months after transplanting. Total fruit yield was recorded from same plants. Fruits were harvested from time to time and cumulated the fruit yield after final harvest to calculate the total fruit yield. The total yields difference was calculated in between the grafted and seedling plants in T-test at 0.05 level in M-STAT.

Galling index system (0-10)	Percentage of total root system galled	Explanation of rating
0	0	Complete and healthy root system, no infection
1	10	Very few small galls can only be detected upon close examination
2	20	Small galls/ knot only but clearly visible main root clean
3	30	Some larger knot visible, main root clean
4	40	Larger knot predominate but main root clean
5	50	50% of root infested. Knotting on some main roots. Reduced root system
6	60	Knotting on main roots
7	70	Majority of main roots knotted
8	80	All main roots, including tap root, knotted. Few clean root visible
9	90	All roots severely knotted. Plant usually dying
10	100	All root severely knotted. No root system. Plant usually dead.

RESULT AND DISCUSSION

Study of posterior cuticular pattern so called perineal pattern of female root knot nematode is the one of the common practice to identify their species level. In our observation from the thirty fresh female root-knot nematodes, fourteen patterns were found the structure of *M.incognita* and twelve were *M. javanica.* Four patterns were not so clear to diagnose the species. These two species were found either together or alone in tomato growing plastic tunnels (Table 1). Rana and Ali (1992) have alsoreported the more than one species of *Meloidogyne* infested in the single species of plant. In addition, they reported from the field of winter vegetables crops in Chitwan district showed that *M. incognita* was most common species and 56 percent of crops were infested, followed by *M. arenaria* and *M. javanica* with 37 and 23 percents crop infestation respectively.

Table 1. Species identification of root knot nematode collected from the root samples of Hemza, Kaski district

Sample lot	No. of female	Diagnosed species	Number	Remarks
1	5	*Meloidogyne incognita*	5	-
2	5	*M. incognita*	2	1 *M. incognita* ?
		M. javanica	2	
		M. javanica	3	
3	5	*M. incognita*	2	-
4	5	*M. incognita*	3	2 *M. incognita* ?
5	5	*M. javanica*	3	1 *M. incognita* ?
		M. incognita	1	
6	5	*M. incognita*	1	-
		M. javanica	4	
Total	30		26	

Farmers have been generally cultivating more susceptible cultivar as monocropping for several years which enhance the increasing the inoculum density of nematodes and causing severe damage of crops. However, farmers are not ready or willing to substitute the cultivars; it's due to market demand on fruit quality and also lack of resistant cultivars. Management of the nematodes has become an essential regarding the substantial loss in the crop yield and quality. Grafting in many countries could prove to be an alternative management approach, especially where temperature does not affect the genes controlling resistance (Luc *et. al.*, 2005). Three species of *Solanum* i.e., *S. sisymbriifolium*, *S. torvum* and *S. toxicarium*. These species, when used as rootstocks, not only reduced galling on tomato, but also reduced bacterial wilt of aubergine caused by *Ralstonia solanacearum* (Mian *et.al.*, 1995). We also found that the used of root of wild brinjal (*S.sisymbriifolium*) as resistant rootstock against root-knot nematodes could be effectively managed the nematodes in tomato growing area. The result showed that the root system of grafted plants was totally free from galls of nematode infection whereas in an average of 7.5 galling index (GI) in 0-10 scale was recorded in non-grafted plants.

Due to healthy root system in grafted plants, the survivability of plants was also longer compared to seedloing plants. On an average of two months longer period of stand in the field by the grafted plants was reconded than that of seedlings (Figure 1). Normally, in non-grafted plants, the period of fruiting was ceased after seven months from the first date of fruit harvesting. When susceptible tomato was grafted on to rootstocks having resistance to *Meloidogyne* spp., yield increased significantly at the beginning and at end of the harvest compared with the non-grafted plants. The fruits at the end of the harvesting stage of grafted plants could also be obtained high price due to less quantity of tomato fruit supply in the markets because the production have already been terminated from most of the normal tomato plants due to the nematode infestation in the fields.

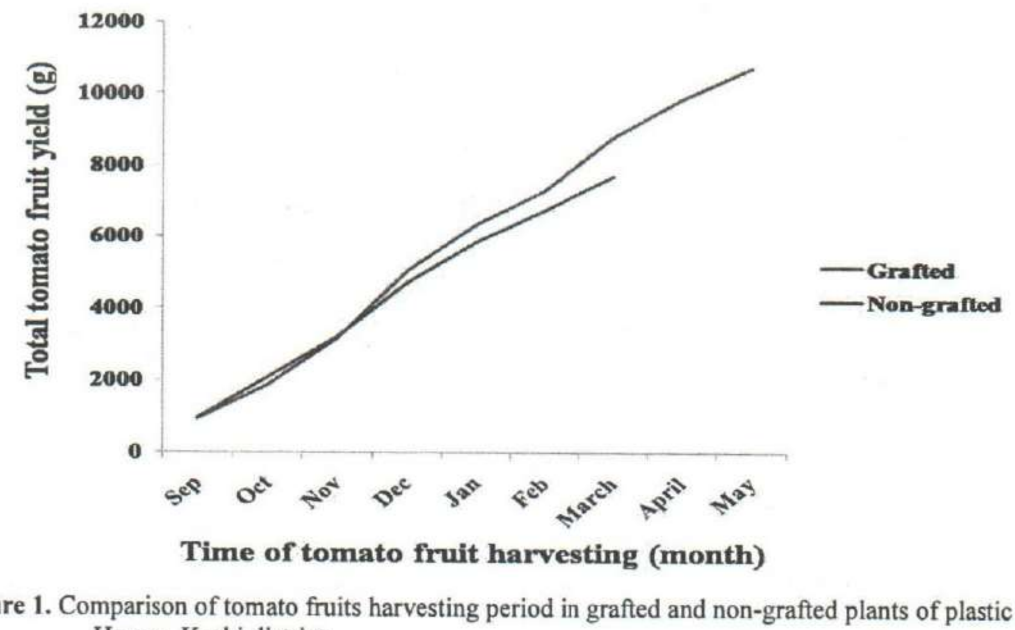

Figure 1. Comparison of tomato fruits harvesting period in grafted and non-grafted plants of plastic tunnel at Hemza, Kaski district

Fruit yield was also recorded after each harvesting from the selected plants and cumulated the total fruit yield after the final harvest. The total yield showed significantly increased fruit yield in grafted plants over seedlings. It was estimated that on an average of 37 percent fruit yield increased in grafted plants compare to seedlings (Table 2). Granges and Leger (1996) were also found that when susceptible tomato were grafted on to rootstocks having resistance to species of *Meloidogyne* and various root pathogens, yields increased 50 and 30 percent at the beginning and end of harvest when compared with the non-grafted plants, respectively. When increased the yield, the number of fruits were also increased significantly. It was recorded that around 28 and 58 percents total number of fruits increased in grafted tomato cultivars Baluroi and Prelane with compared to non-grafted plants respectively (Vuruskan *et.al.*, 1990). The production and productivity was increased due to suppression of nematode population in the field. In resistant rootstock, suppression of nematodes population could be caused by some factors including difference in penetration rate, death rate, and rate of multiplication. Generally, resistance is most frequently linked with biochemical events taking after nematode infestation or by particular morphological features differentiating the resistant plant/root from susceptible ones. According to Lee (1994), the increased yield of grafted plants is also believed to be due to enhanced water and mineral uptake. Useof the grafting plants not only increased the yield of crop in the given season, it is also reduced nematode population significantly in soil for coming cropping season. So, there are no need to cultivated grafted plants for many years, they can be substituted by susceptible cultivars when the initial population of nematodes became below the economic threshold level. Hence, host plant resistance has become an important strategy in the management of plant parasitic nematodes.

Table 2. Fruit yield and galling index estimated in grafted and non grafted tomato plant cultivated in RKN infested plastic tunnel at Hemza, Kaski district

Parameter	Yield (Kg)		GI (0-10)	
	Yr 2065-66	Yr 2066-67	Yr 2065-66	Yr 2066-67
Grafted[1]	12.064 (± 1.39)[5]	9.41 (± 1.39)[5]	0	0
Non grafted[1]	8.98 (± 0.68)[5]	6.78 (± 0.68)[5]	8	7
Yield increase over non grafted[2]	3.104	2.63		
% increase [3,4]	34.64**	38.79**		
Average	36.72		7.5	

Note:

1 Mean fruit yield calculated from four replications (Four plastic tunnels)
2 Mean fruit yield of grafted – mean fruit yield of non-grafted plant
3 % change (due to using grafting plant) = 100 x (mean of grafting plant/ mean of non grafting plant) – 100
4 Change was tested for statistical significance with t- test at ** P<0.05
5 STDEV value of mean fruit yield

CONCLUSION

Grafting is one of the most effective and innovative technique for the management of root-knot nematodes. Grafted plants not only control the root-knot nematodes, it helps to reduce the initial population of nematodes in the fields and also increased the production and productivity by lengthening the plant life than the non-grafted plants. The grafting technique for tomato production in plastic tunnel is economically benefited to the growers for long term low-input sustainable agriculture. It also gives increased soil borne diseases tolerance and vigour to plants. Hence, the grafting technology could be used effectively for cultivation of commonly grown varieties, which are susceptible to root-knot nematode in disease prone areas. It can also be used as an alternative technology for reducing the use of hazardous pesticides for enhancing commercial organic tomato production.

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