

# Research Journal of Pharmaceutical, Biological and Chemical Sciences

## Assesment of some genetic parameters in diallel crosses to resistant of roots knot nematode *Meloidogyne spp* in the food processing tomato.

Inad. Dhafer. Abood Al-mafragi\*, and R.WA. abd ALrahman.

Dept. of plant protection —college of Agric.

### ABSTRACT

Five pure lines were selected for most phenotypic traits, but they differed in their resistance to nematode root disease, and they were crossed in one direction For obtianed 10 hybrids have been tested and with their parents to this disease and that by exposing them to the second juveniles stage two of nematode 5000 J2 / kg soil to assessment of someGenetics parameters in these variaties , found that the resistance character was influence by dominant genes action and that the additive genes action was low, It was found that the linear relationship between the severity of the disease and the production of the genotypes was high and positive and reached 91.7%. The lines 1, 2 and 3 were sensitive to the disease while 4, 5 were resistant according to the pathological index of the number of nodes, and the crosses 2\*3 ,1\*3, 1\*4and sensitive genetic lines and hybrids formed at their roots more than 30 – 100 galls within 60 days from the beginning of exposure to the j2 in the case and the disease severity from 75-100% after 90 days in the field and accordingly decreased production rate of 35.1-58.3% .while the presistante purelins and their hybrids was formed galls attheirs roots 1-10 gall/plant in pots treatment and disease severity in the field traite respectirily.

**Keywords:** root knot nematode, Tomato processing, Meloidgyne spp, Genetic parameters

\*Corresponding author

## INTRODUCTION

*Tomato Lycopersicon esculentum*. Mill belonged to solanaceae has a population of 48 species and more than 3611 plant species. It is one of the most diverse plant families (Foolad 2007), one of the most important vegetable crops in Iraq in terms of area cultivated annually in production and consumption (Central Bureau of Statistics, 2010). This crop has received considerable attention from plant breeders, for the purpose of fresh consumption or for industrialization due to its high adaptability and suitability to its various environments and their high productivity and suitability for use both fresh and processed (Shankar et al., 2013) and is grown globally on an area of 4.4 million hectares and total production 160 million tonnes (FAO, 2014). Therefore, most of the Tomato breeding programs have now focused on the development of high yielding hybrids and their quality to start cultivating local varieties which replaced of imported varieties (Al-Kamar 2000 and Ibraheem 2001). The aim of genetic improvement is to produce varieties Distinctive In order to achieve superior genetic improvement, parents which differ geographically and genetically are selected, thus increasing the likelihood of obtaining superior hybrids in their properties (Shankar et al., 2013). The use of the hybrid vigor phenomenon in the production of varieties and the acquisition of hybrids for manufacturing was considered The top of the practical application of the foundations of the science of plant genetic improvement, which depends primarily on the selection of the appropriate inheritance parents, which can be accessed through the best combinations of the most important agricultural traits (Hannan et al., 2007). Therefore, the exploitation of this phenomenon has played a major role in the production of commercial hybrids, not only to increase the quantity but to improve the quality of production. The efforts and the many studies focused on the crop of tomato within this The results of the studies indicate that the cultured embryo hybrids show a hybrid vigor for the characteristics of syphilis and vegetative growth and the quality of production and surpass the commercial cultivars, and many scientists have conducted several experiments related to the production of hybrids (2002 BBalli) *Meloidogyn spp* is an important pest that causes high economic losses in a wide variety of vegetables and all over the world (Castagnone-sereno, 2006). The estimated amount in the United States of America for controlling root-knot nematodes is \$ 500 million (Keren- Zur et al., 2000). Globally, it is one of the Aggressive pests on crops, causing high economic losses of \$ 157 billion (Abad et al., 2008). It is difficult to control because its life cycle is shorter than 6 to 8 weeks and live between roots. In spite of the obvious qualitative differences in resistance to diseases that can be observed when comparing diseases between genetic plant lines which have different resistance genes. Factors that affect the effectiveness of newly introduced resistance species are the choice of pathotype or pureline Which are able to support the resistance. However, methods can be deployed to take advantage of reduce the collapse of resistance. Several specieses of crops have resistance against root-knot nematodes, but new sources of resistance to some of these species are needed to improve the resistance level of root-knot nematodes. To date, no resistant genetic has been identified in a number of other crop species. The resistance was transferred to highly acceptable commercial varieties to be able to find genetic material from adaptive species or from independent breeding lines (Hussey and Janssen, 2001). The plants resistance to for nematodes comes from wild plant species or from plant breeding such as genetic segregation Is an important source of genes for resistance to the disease of the complexity of the roots, and there are great possibilities to identify additional genes. For example efforts are concentrated to identify additional genes resistant to the root knot nematode in the wild tomato of the origin of gene Mi has revealed the existence of at least 8 additional genes in the *Lycopersicon peruvianum* tomato is likely to discover genes Mi (Foolad and Panthee, 2012). The study aimed to Test 5 different sensitivity, resistance and production pureline to get hybrids with high production and produce good resistance

## MATERIALS AND METHODS OF WORK

The genotypes of purelines and 10 hybrid crosses were taken from I.D.Abood The nematode inoculum was obtained from the infected roots of plants (Tomato, eggplant) Extraction and calculation of eggs and juveniles from Root nematodes: using the Hussey and Barker method, roots after cutting the roots into small pieces with scissors. Put a quantity of roots in a bowl and add 0.5% NaOCl The roots are taken with 200 µm and 500 µm sieves and washed with Tap water several times to obtain the clear water containing the eggs and J2. The number of eggs is then calculated using the counting slide under the compound optical microscope, or Take the roots and add the Naocl concentration of 1% (Hemming and Whitehead, 1965) and take the cut roots and the surrounding soil (75, 50, 25) µm under tap water currents. The starter is taken from the last sieve and placed in a flask. After a procedure to extract several times, the number of eggs and J2 is calculated by slide counting and count its under the microscope compound light. To calculate of different levels of nematode inoculum for sensitive variety (supermarimande) was inoculation by 1000, 2000, 3000, and

5000 J2 /Kg soil and after two month the plants were taken out and washing the roots carefully and calculate the No.of according of .(sasser and Freckman , 1987) scales.

**Screening of tomato varieties for root knot nematode : -**

Inoculation of tomato seedling by (5000 eggs, J2 / kg soil) After two weeks of planting at 3/3/2017, three holes were made in a triangle around the root of the plant and put the inoculum (Eggs and J2), while keeping 3 replicates for each variety for comparison (without any inoculation). The pots were arranged in full random order and kept in the plastic house and were placed and irrigated every two days in sufficient quantities of water without getting water outside to keep the inoculum from washing and after Three months were taken out of plants on (25/6/2017) and the number of gills was calculated according to the scale of number node (sasser and Freckman , 1987) ( 0= no gill ,1= 1-2 gill , 2= 3-10 gill , 3=11-30 gill , 4= 31-100 gill , 5 = more than 100 gill ) , after washing the roots with water running quietly to maintain the complete of roots Determination of genetic for root. Quietly to complete of root .Determination of genetic sensitivity for root knot nematode in the field: transferred the seedlings to the pot and after week the seedling were inoculation with 1000 J2 / per pot/ 200 g of soil. And the seedling were transfer to the field after month from date of inoculation , the experiment was design as complete random block with three replicates, then transferred inoculum seedlings to the field on 2/4/2017 was taken care of watering and extracting fertilization for three month and then calculate the disease severity according to scale of ( 0= no gill of total root ,1= 1-10% no gill of total root , 2= 11-25% no gill of total root , 3=26-50% no gill of total root , 4= 51-75 No gill of total root , 5 = 76-100% No. gill of total root (Taylor and sasser,1978) and McKinney equation are used to calculate of disease severity ( McKinney , 1, 23 ) at (17/7/2017 ) and took the following data: - Measurement of percentage of Total TSS: Ten similar fruits were taken at approximately maturity of each treatment (genetic variety ) in each replicater at random Block 3 drops of juice from each fruits were placed on glasses of man refractometer was reading data as .ratio of total solved solids and the mean of the data was calculate for each genotype and each replicates.

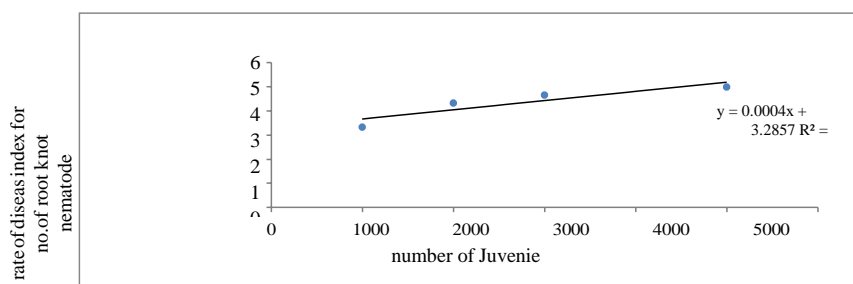
**Test designs:** In the plastic house experiments, use the full CRD design with three replicates. In the field, use the Random complete block designs (RCBD) with three segments.

**Statistical analysis:** The analysis of data collected by Genstat statistical was used with the least significant difference of LSD (5%) to compare the differences

**RESULTS AND DISCUSSION**

**Effect of different levels of nematode inoculum in rate of disease index for number of nodes**

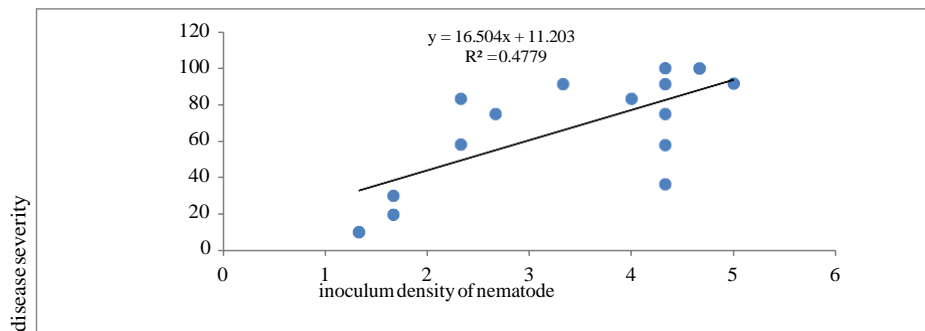
The result of the relationship of the rate disease index for the nodes number with the inoculum density of nematodes showed that there was clear effect by increasing inoculum density lead to increasing of rate disease index , and the positive relationship was reach to 0.816 and the reason for the high proportion to this relationship is because the low levels of the inoculum caused to large numbers of root knot in the sensitive variety and high levels of inoculum density was lead to increased number of j2 which infected the sensitive roote and happen outbreak of nematode and lead to highest number of gills read to disease index 5 at the density of 5000 eggs& j2 and lowest rate of number node 3.3 at the inoculum density 1000 eggs & j2 and the disease index not decrease at inoculum density 2000,3000 eggs and j2 reach 4.3 ,4.6 respectivity this result nearly same of the result Durban et.al , 2003 and moslihi etal . ,2010.



**Figure 1: Nematode density relationship with rate of disease index for number of nodes**

**Relationship of inoculum density with disease severity :**

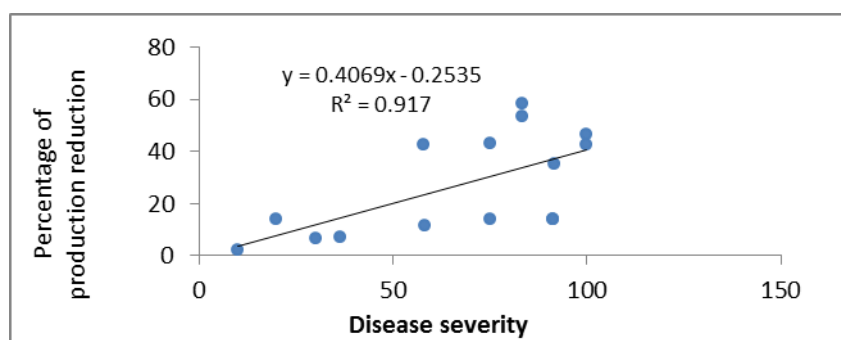
From the observation of Figure (2), the correlation between the inoculum density of nematodes and the percentage of the disease severity (physiological destruction of the root), the higher of inoculum density of the nematodes in the soil increased the severity of infection in the roots of different genotypes. In the resistant varieties, the plant response to infection by nematode is low compared with sensitive variety. whatever increasing of inoculum density, and the relationship of numerical density of the members of the nematodes in the soil a positive relationship accompanied by an increase of disease severity and that the correlation between them up to 0.4779. (Gharabadiy et al. 2013)



**Figure 2: The relationship of the inoculum density of nematodes in the soil with the physiological destruction of the root system (disease severity)**

**Correlation between diseases severity and reduce of production:**

(Figure 3). The disease severity at the root of the genotypes is related to the reduction of production. However, this relationship controls by disease of resistance of each genotyp or its sensitivity. If the genotype is resistant, the amount of reduction is low, or if the physiological damage to the roots increases due to the increased susceptibility In response to a nematode infection, the reduction in production was increased. The value of this relationship, although the response to nematode was varied, was estimated at 91.7%, the root destruction of pureline 2 and the 2 × 1 hybrid were 100% that the reduction of production reach 46.6, And 42.5 respectively while pureline 4 and hybrid 8H × 2 subversion ratio of destruction reach to 20 ,10 respectively that the reduction in production is low 13.8, 2.01 respectively, this result. Compatible with what is mentioned (Abrol and Shankar 2012).



**Figure (3)the relationship between diseases severity caused by nematodes *meloidgyne* spp and percentage of reduce of production reduce**

It is noted from the results in this table that the two purelines are 2, 1 are very sensitive because more than 100 gill on its roots within 60 days while the pureline (3) was formed at its roots 11-30 knots during the same period, another pureline 5,4 were resistant because are forming at its root 1-9 gill, noting that all pureline were inoculum with 1000 j2 /200 g soil, whereas 2 × 5 crossbreeding hybrid, was high resistance more than high its parents. The other hybrids were sensitive and the striking results were that the 4 × 5 hybrid was also resistant to the root knot nematode and did not give high resistance to Although his parents were

resistant, he indicated that the genetic sites of both parents were not pure towards the resistance. Therefore, when these sites converged during the hybridization process, individuals were placed with respite sites that were allergic to the disease. Some of the community had recessive genes at several sites. Which plays a key role in the resistance, and this confirms that hybridization should occur between pureline of genetic purity recessive with purline carrying the genetic purity for dominant gene for all genetic sites, so that each site has obtained two allese one of them are recessive and other are dominant gene to be a gene action is great when compared with its highest resistant parnets as hybrid 2 × 5.

**Effect general and specific combing ability:-**

**Table 1: Average number of nodes for purelines (diamlines values) and crossbreeding (values above diameter) of plants inoculume with nematodes 1000J2/200g Soil**

	P1	P2	P3	P4	P5
P1	4.33	4.33	4.33	5.00	2.67
P2		4.67	4.00	3.33	1.33
P3			2.33	4.33	2.33
P4				1.67	1.33
P5					1.67

LSD<sub>0.05</sub> = 1.055

The results indicated sensitive that the pureline has given the sensitivity to most of the hybrids formed by this purelines and positive values, its meaning, the sensitive hybrid was more than the highest parental sensitivity of the disease, while the negative values of the other hybrids had decreased sensitivity to the disease and approached of the resistance was more hybrid resistance was the hybrid 2 × 5, 4 × 5. The results were consistent with previous results in the interaction of resistance genes with the pathogenic genes of nematode roots. This is consistent with Naumkina et al. (2005)

Effect of Combining and specific combing ability

**Table (2) Effect of general combining ability of purelines (diameter values) and effect of specific ability (values above diameter)**

	P1	P2	P3	P4	P5
P1		7.14-	0.00	15.38	38.46
P2	3.70-		14.29-	28.57-	71.43
P3	30.00	14.29		85.71	0.00
P4	66.67	5.26	116.67		-18.40
P5	11.11-	57.89	16.67	18.40-	

Se HP = 12.90 Se MP% =15.20

The results in Table (3) we find that the negative values mean that the hybrid carries the resistance status and all the more hybrids be resistant is 2 × 5 if the value - 71.43, which means that is approached with the highest resistant parents but not the same degree of resistance may be. The hybrid had the hybrid vigor of - 100 it was close to the resistance at the highest its parents, while the 1×3, 3×5 hybrid carried 0.00 value in crossbreeding closed with the least sensitive parents and other parent are more sensitive while the hybrid 3×4 had positive approachd with the highest parents sensitive the hybrids that was compared to the average between parents the result showed that the hybrid also 2×5 was resistant than the average parent and negative value reach to -57.85. (Shankar et al .2013 , Figueiredo et al 2015)

**Table (3) Hybrid vigor estimated at the highest of parents (values above diameter) and mean between parents (values under the diameter)**

	P1	P2	P3	P4	P5
P1	0.846	0.157-	0.223	1.220	0.209-
P2		0.465	0.271	0.066-	1.161-
P3			0.084	1.315	0.220
P4				0.245-	0.424-
P5					1.150-

The results in Table (3) we find that the negative values mean that the hybrid carries the resistance status and all the more hybrids be resistant is  $2 \times 5$  if the value - 71.43, which means that is approached with the highest resistant parents but not the same degree of resistance may be The hybrid had the hybrid vigor of - 100 it was close to the resistance at the highest its parents, while the  $1 \times 3$ ,  $3 \times 5$  hybrid carried 0.00 value in crossbreeding closed with the least sensitive parents and other parent are more sensitive while the hybrid  $3 \times 4$  had positive approached with the highest parents sensitive the hybrids that was compared to the average between parents the result showed that the hybrid also  $2 \times 5$  was resistant than the average parent and negative value reach to -57.85. (Shankar et al .2013 , Figueiredo et al 2015)

Gene action and degree of inheritance

**Table 4: Some Genetic Values of the Interaction of R-Genes with *Meloidogyne spp***

$\sigma^2_p$	1.81
$\sigma^2_G$	1.63
$\sigma^2_E$	0.133
$\sigma^2_A$	0.696
$\sigma^2_D$	0.985
$\sigma^2_s$	0.696
$\sigma^2_g$	0.491
$\sigma^2_G/\sigma^2_s$	0.706
$h^2_{bs}$	92.67
$h^2_{ns}$	54.25

$\sigma^2_p$ =phenotypic variance  $\sigma^2_G$  =Genotypic variance  
 $\sigma^2_A$  =Additive genes effect ,  $\sigma^2_D$  =Dominant genes effect  
 $\sigma^2_s$  =specific combining ability effect ,  
 $\sigma^2_g$  =General of combining ability effect ,  
 $h^2_{bs}$  =heritability narrow sense .

( Griffen , 1956 c) The genetic valus in table 4 we find that the effect of additive genes ( $\sigma^2_A$ ) 0.696 less than the effect of Dominant gene(  $\sigma^2_D$  ) 0.985 and the  $\sigma^2_p$ : its meaning was consumed  $\sigma^2_G$  and  $\sigma^2_E$  , consumed  $\sigma^2_A$  and  $\sigma^2_D$  and  $\sigma^2_G$  meaning was Thus the effect of gene resistance on resistant of genotype for the *Meloidogyne spp* . are very high reach to 92.67 and the additive gene.action reach to 54.25 and dominant gene action reach to 38.42 these results mentioned to there many of resistant genes are cooperative with them and when compared with the dominant gene effect

**Relationship of nematode with reduce of production —**

**Table (5) Percentage of production reduce in purelines (diameter values) value and cross-breeding values(above-diameters values) of nematode- inoculum of plants which inoculum by nematode**

	P1	P2	P3	P4	P5
P1	42.63	40.53	43.33	35.10	13.87
P2		46.67	58.33	14.10	2.01
P3			53.33	14.23	11.47
P4				13.87	7.33
P5					6.44
LSD <sub>0.05</sub> = 22.024					

The infection of plants by nematode was effected on the metabolic events of the roots and reflected on the vegetative part of the plant and there for also on the reduction of production , the purelines was varied response to infection depended on different genes resistant and the pureline 5 was the lowest decrease in percentage of production reduced 6.44 % and pureline 3 was highest reduction of production 53.33 and the hybrids :2×5 , 4×5 , 3×5 , 1×5 which less reduction of production 2.01 , 7.33 , 11.47 , 13.87 respectively because the pureline (5) have more than gene of resistance and of hybrids be one of its parent which be resistance while the 2×3, 1×3 was highest production reduces because the pureline 3 very susceptible to infection by *Meloidogyne spp*

**Effect of nematodes on reduction of total soluble solids**

**Table 6: Effect of Nematodes on Total TSS Reduction of purelines (diameter Values) and cross-hybrids (above of diameter values)**

	P1	P2	P3	P4	P5
P1	9.1	15.0	14.2	25.4	11.7
P2		6.5	2.1	12.1	7.8
P3			4.2	13.9	14.5
P4				6.6	14.5
P5					20.3
LSD <sub>0.05</sub> = 1.016					

Table (6) The result mentioned to significant difference in the rate of total solids to the highest value of the 1×4 , and 1×2 hybrids : 25.4 , 15 respectively and the lowest value for pureline 1 and 2×3 hybrids and also lowest value with resistant character , No relationship between resistant genes with reduction of total soluble solids result , this result consistent with .

**REFERENCES**

Al-Kammer, Majid Khalif and Abdul-Jabbar Ismail. "Intercultural Hybridization and Analysis of the Coalition Capacity for Quantitative Properties in Tomato", Iraqi Agriculture Journal, Issue 3, 190-183.

Abad, P., J. Gouzy, J. Marc, J.M. Aury and P.C.Sereno. 2008. Genome sequence of the metazoan plant parasitic nematode *Meloidogyne incognita*. Nature Biotechnology, 26: 909-915.

AISH ,A.A. .2016. Induction of Systemic Resistance Against Root-Knot Nematode Using Beneficial Microorganisms. Agriculture Botany Department Faculty of Agriculture- Suez Canal University

Balliu A. and M. Hallidri.2002. "Combining Ability Test Between Some Tomato Genotypes" Acta Horticulturae (ISHS) 579:123-126 .

Central Statistical Organization. "Crop and Vegetable Production" Directorate of Agricultural Statistics. Planning Authority. Council of Ministers - Iraq, 2010.

Castagnone-Sereno, P. 2006 . Genetic variability and adaptive evolution in parthenogenetic root-knot nematodes. Heredity. 96:282-289



- Darban, D.A., M.A., Pathan, M.M., Jiskani, K.H.R., and Wagan, . 2003. : Response of some tomato cultivars to root-knot nematode, *Meloidogyne incognita* (Kofoid and White) Chitwood. Pakistan Journal of Agriculture, Agricultural Engineering and Veterinary Sciences 19:36-38.
- FAO, .2014. Plant genetic resource for food and agriculture. Rome, Food and Agriculture Organisation of the United Nations
- Falconer, D.S. .1960. : Introduction to quantitative genetics. Printed in Great Britain for Oliver and Boyd, by Robert Mac Lehos and comp. London Glasgow : P 281-286.
- Foolad, M.R. .2007. Genome Mapping and Molecular Breeding of tomato. Hindawi Publishing Corporation International Journal of Plant Genomics Volume, Article ID : P 53-64.
- Foolad, M.R. and D.R. Panthee. 2012. Marker assisted selection in tomato breeding . Cr.T. Rev. Plant. Sci. 31: 93 – 123.
- Gharabadiyan Fatemeh, Salar Jamali and Hamid Reza Komeili .2013. Determining of root-knot nematode (*Meloidogyne javanica*) damage function for tomato cultivars. Journal of Agricultural Sciences 58:147-157.
- Hannan, M.M , M.K, Biswas, M.B., Ahmed and ,A. M. HOSS.2007. IN, M. Combining Ability Analysis of Yield and Yield Components in tomato (*Lycopersicon esculentum* Mill.) . Turk J Bot. 31:559-563
- Hussey, R. S . and G.J.W. Janssen, .2001. . In: Starr, J. L., Bridge, J. and Cook. R(2001) .( Resistance to Plant Parasitic Nematodes). PP:44-55.
- Hussey, R.S. and K.R. Barker. 1973. A comparison of methods of collecting inoculum of *Meloidogyne* spp., including a new technique plant disease. 57: 1025 – 8.
- Keren-Zur, M., J. Antonov, A., Bercovitz A., Feldman, G., Kerem, and N. Rebhum . 2000. *Bacillus firmus* formulation for the safe control of root knot nematodes. The BCPC conference. Pests and Disease, Brighton, UK. Pp 307-311.
- Moslehi, S.H.G.H., Niknam, S., Aharizad, .2010. : A study on the reaction of six tomato cultivars against root-knot nematode *Meloidogyne javanica* under greenhouse conditions. Iranian Journal of Plant Protection Science 41:19-27
- Mortazavi-Bak, A., and A.R. Ahmadi .2004. : An evaluation of resistance and tolerant tomato cultivars to root-knot nematode *Meloidogyne javanica* in Iran. Proceedings of the 16th Iranian Plant Protection Congress, p. 24
- Naumkina, Ti, V. Yakovlev. T., Titemok, T., Vasilchikov A. , Borisov, O. , Kolikova. .2005. : Pea breeding improve effectiveness of symbiotic nitrogen fixation. Russia : P88-102.
- Pattison, T. 2007. Tomato root knot nematode : Biology and control. Department of Primary Industry and Fisheries. Queensland, Australia Using nematodes as bioindicators for soil health in bananas. " Super oil". Proceedings of the International Soil Science Conference : P 4-9.
- Sasser, J. N. and D. W. Freckman. 1987. A world perspective on nematology: the role of the society. In vistas on Nematology. Edited by Veck, J. A., D W. Dickson and M. D. Hyattsville: society of Nematologists. 7 – 14.
- Shankar , A; R . REDDY, M.; Sujattha , M., ratap, M. .2013. Combining ability analysis to identify superior F1 hybrids for yield and quality improvement in Tomato (*Solanum lycopersicum* L.). Agrotechnolog : P2-3.
- Taylor A. L. and J. N. Sasser. 1978. Biology. Identification and control of root – knot nematodes (*Meloidogyne* spp.) Coop. Pub. Dep. Plant pathol. North Carolina state Univ., and U.S. Agency. In. T. Dev. Raleigh, N. C. PP. 111.
- Whitehead, A. G. and Hemming, J. R. 1965. A comparison of some quantitative methods of extracting small vermiform nematodes from soil. Annals of Applied Biology. 55:25:38.