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Comparative analysis and management of plant parasitic nematode population associated with walnut *(Juglans regia* L.) In Hazara Division, KP, Pakistan

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Abstract

ABSTRACT

Nematodes are round small worms found almost in every type of environment. Plant parasitic nematodes cause great damage to crops and trees and as a result loss in yield and plant heath occurred. Plant parasitic nematode infestation goes undetected and untreated because of less awareness. A survey was conducted to estimate the ratio of plant parasitic nematodes associated with Walnut (*Juglans regia* L.) in different areas of Hazara Division, KP, Pakistan. District Abbottabad, Mansehra and Kohistan were surveyed. Soil samples from these districts were collected and processed through Bearmann funnel technique. Nematodes were quantitatively analyzed. Ratio of plant parasitic nematodes and saprophytic nematodes was analyzed and calculated. Results showed that samples from Abbottabad and Mansehra had plant parasitic nematodes in greater ratio than saprophytic nematodes while Kohistan samples were found saprophytic. District Abbottabad showed 40% while district Mansehra sowed 60% plant parasitic nematodes.*Helicotylenchus* sp. were found in abundance from Abbottabad and Mansehra. Data was plotted in histograms and ANOVA was done which showed that P value was significant. P value for the effect of Carbofuran in Mansehra and Abbottabad was 0.0001 and 0.0001 while for poultry manure it was 0.0001 and 0.0009 respectively. Carbofuran and Poultry manure were used for the management of plant parasitic nematodes. Carbofuran totally eradicate the nematode population while poultry manure reduced the population to a remarkable point. Therefore both were found effective but poultry manure s more environment friendly in nature.

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Introduction

Walnut (*Juglans regia* L.) is one of the most widespread nut tree found worldwide. There is a great variety of walnuts varies in chemical and physical nut type, productivity, and forestry. The seed of the walnut is of nutritional importance. Pakistan, officially called the Islamic Republic of Pakistan, is located in the Middle East near the Arabian Sea and the Gulf of Oman. In Pakistan it is widely domesticated in the areas such as Galliyat, Abbottabad, Swat, Dir, Shangla, Buner, Chitral and Bajaur agency. They are found in many localities in Hazara Division, KPK, Pakistan. Some are under domestic conditions while some are in wild habitat. Pakistan is an important country regarding flora and fauna. Hazara Division includes Havelian, Abbottabad, Mansehra, Kohistan.

The climatic conditions of Abbottabad, Mansehra and kohistan are suitable for the growth of walnut trees. Plant parasitic nematodes alongwith diseases caused by pests and insects affect plant growth and yield. Plant parasitic nematodes are of great importance because of the damage caused by them .Plant parasitic nematodes are destructive to plants and agricultural crops, and may results in countable yield losses (Bang *et al*, 2016). Plant parasitic nematodes include several groups resulting in severe crop losses (Hugot *et al.*, 2001). A spadeful of soil can contain more than million nematodes because nematodes are so numerous and occur in so many habitats Many workers identified many plant parasitic nematode species and encountered plant parasitic nematode population associated with walnut (*Juglans regia* L.) throughout the world. A number of workers have reported nematodes associated with walnut causing yield losses from various parts of the world (Askary *et al.*, 2012; Britton *et al.*, 2009; Buzo *et al.*, 2009; Ciancio *et al.*,1996; Khan *et al.*, 1993; Khan *et al.*, 2002; Kaul *et al.*, 1989; Leila *et al.*, 2009; Lorrain, 2000; Liskova *et al.*, 1998; Mangan, 2015; Mazdosh *et al.*, 2005; McKenry and Roberts, 1985; Taheri *et al.*, 2013; Tarihi, 2001; Tokobaev *et al.*, 1986; Westerdahl *et al.*, 2006; Zaki and Mantoo, 2003; Zari *et al.*, 2011; Zarina *et al.*, 2010).

Management of the plant parasitic nematodes is very important economically as well as from plant health and yield point of view. Organic and inorganic amendments have been used for this purpose throughout the world and some are found promising. Many farmers are unaware of these plant parasitic nematodes and their management which results in yield loss and poor plant growth. In this study carbofuran (a nematicide, inorganic amendment) and poultry manure (Organic amendment) was used against plant parasitic nematodes in Mansehra and Abbottabad.

Materials and methods

Different localities of Hazara Division were surveyed for walnut trees both in domestic and wild conditions. Soil samples were collected from different localities of Abbottabad, Mansehra and Kohistan from the walnut trees. Soil around tree trunk was dig with the help of spade up to 2 to 3 feet deep. A handful of soil sample was taken and stored in polythene bags. It is important to airtight them properly to avoid drying of a sample. Name of Locality, host and date was mentioned. Soil samples were stored in a cool place as optimum temperature and moisture is required for nematode population. Soil samples were processed using Bearmann funnel technique (Southey, 1970). After 24 hours, each sample was studied under Watson Barnet stereoscope at 4X power per 200ml of soil sample for plant parasitic nematode population. Quantitative analysis was done by counting nematode population under stereoscope microscope with the help of needle in different localities of Abbottabad, Mansehra and Kohistan. Recorded data was analyzed statistically by subjected to one way ANOVA and histogram graphs.

For the management of plant parasitic nematodes, nematicide Carbofuran and Poultry Manure was applied around walnut trees affected by nematodes. Carbofuran was applied at the rate of 250 gram/ tree and poultry manure 8kg/ tree. Soil around the tree trunk was dug with the help of spade and applies the amendments around it. Normal watering was done and avoids any other pesticide or agricultural sprays.

Soil samples with subsamples were collected after 3 Month, 6 Month and 12 Month of application and processed through Bearmann funnel technique. Nematode population was quantitatively analyzed under stereoscope microscope. Untreated trees were kept for the comparison with the treated ones.

Statistical analysis

The recorded data was subjected to statistical analysis. One way ANOVA and histogram graphs were used to analyze the recorded data. Statistical software PRISM 5 was used for statistical analysis.

Results and discussion

Quantitative analysis showed that nematode population comprised both parasitic and saprophytic species as shown in figure 1. Some localities were completely saprophytic while some showed both plant parasitic nematodes and saprophytic nematodes. Parasitic nematodes comprised of different species.

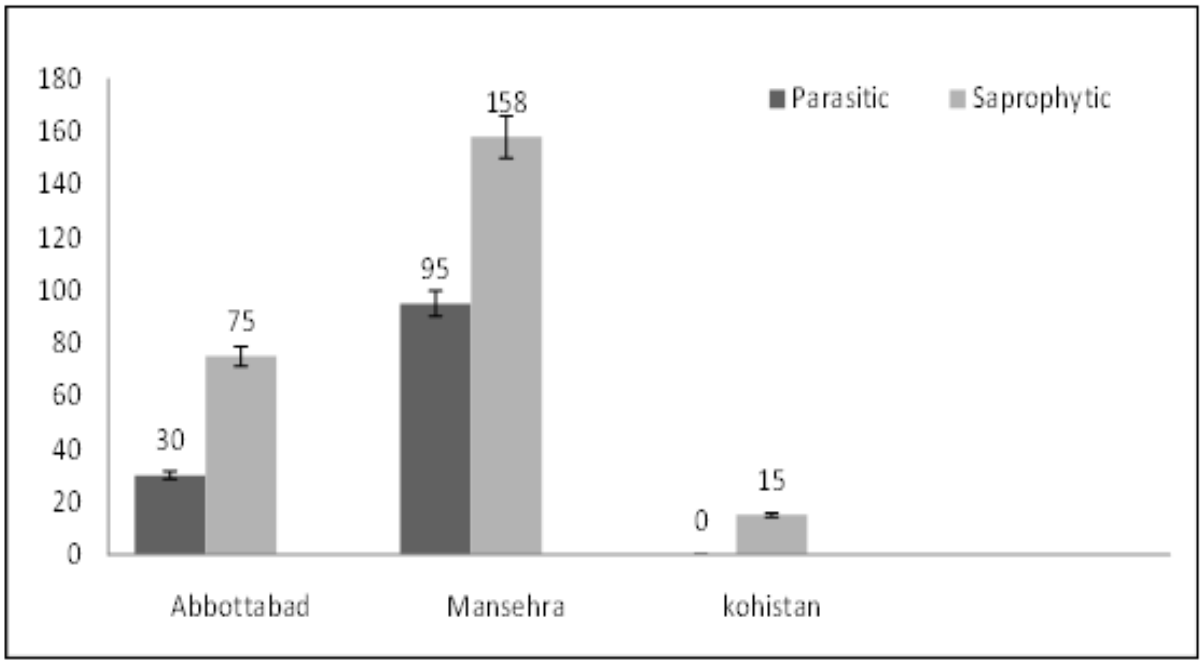

Fig. 1. Histogram showing the parasitic and saprophytic nematode population in different localities.

Most common plant parasitic nematode found were *Helicotylenchus* species which are fungal feeders in their mode of life. Nematode population of different localities of district Abbottabad and Mansehra were compared and concluded that 40% of nematode population of Abbottabad and 60% of Mansehra were parasitic. Data was plotted statistically in histogram graphs showing the nematode population of both parasitic and saprophytic in localities of Abbottabad, Mansehra and Kohistan as can be seen in figure 1. One way AVOVA of the data showed that P value was not significant (P=0.0746).

Management data plotted in histogram graphs clearly showed that application of both nematicide Carbofuran and Poultry manure (as an organic amendment) for the control and management of plant parasitic nematodes associated with walnut trees showed positive results as shown in figures. Both Carbofuran and poultry manure reduced the nematode population associated with walnut trees in Mansehra and Abbottabad as can be seen by histogram graph bars. Treated trees data was compared with the data of untreated ones in the histogram graphs.

Carbofuran is inorganic in nature and is a chemical which is commercially made with nematicide nature for the control of nematodes associated with different plants and crops. Commercially it is available on agricultural shops and centers and can be purchased on need with an ease. Histogram graphs showed that

nematodes were reduced to a greater extend by the application of carbofuran. Comparison was clear in histogram graphs in both Mansehra and Abbottabad. Histogram graph was plotted against data collected from walnut trees treated with poultry manure in Mansehra and Abbottabad as in figure 4 and 5. Poultry manure is organic in nature and is a natural waste of poultry. It is in use for different crops and trees as a fertilizer from old times. It is in the approach of every person and easy to use and handle. Data showed that nematode population was reduced to a noticeable extent with the application of poultry manure. Therefore it controlled the nematode population in Mansehra and Abbottabad as well.

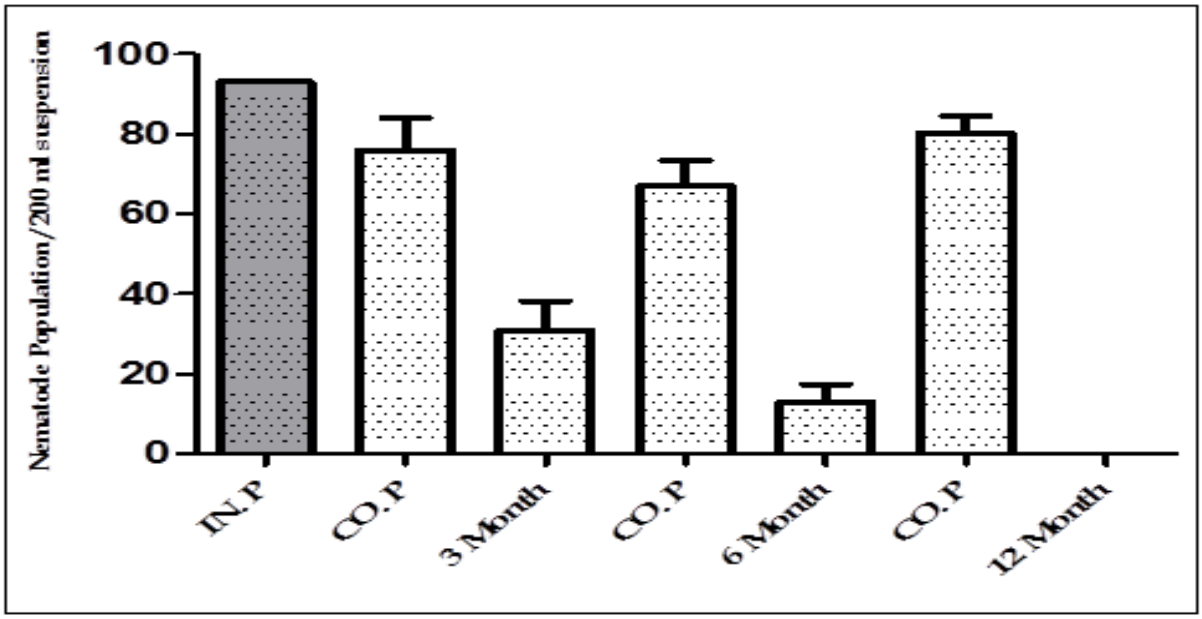

Fig. 2. Histogram Graph showing the effect of Carbofuran on Plant parasitic Nematode in Mansehra.
IN.P= Initial Population, CO. P= Controlled Population.

Mixed nematode culture found in different localities of study area showed that nematodes are damaging the crops and trees. Plant parasitic nematodes associated with walnut trees were reported by Taheri *et al.* in 2013 which were damaging walnut trees in the same way. Plant parasitic nematode damage to trees was supported by Taheri and his coauthor's work.

They cause severe damage to the plants as a result of which plant health and yield was affected. In Pakistan Khan *et al*, in 1993 reported the damage caused by plant parasitic nematodes to walnut trees as reported in this study. Different species were reported associated with walnut trees in the studied area as shown in histogram graph in figure 1 confirm the presence of these damaging nematodes. It in turn affects the economic health of the host trees and area as well.

As they are unseen enemies therefore they got unnoticed and were not considered by the farmers and planters of the area.

These areas are habitat of walnut trees naturally but yield was decreased with the passage of time. One of the factors for the yield loss in these areas was nematode infestation which normally goes unseen and unaware. Mixed culture of plant parasitic nematodes in figure 1 showed that different species were present associated with walnut trees. In Mansehra, population was found high as compared to the Abbottabad. In Kohistan, only saprophytic species were found. Reason for the absence of plant parasitic nematodes from this area could be use of old farming techniques and extreme low and high temperature. Saprophytic nematode population observed was very limited one. Nematodes require optimum temperature for their growth and development.

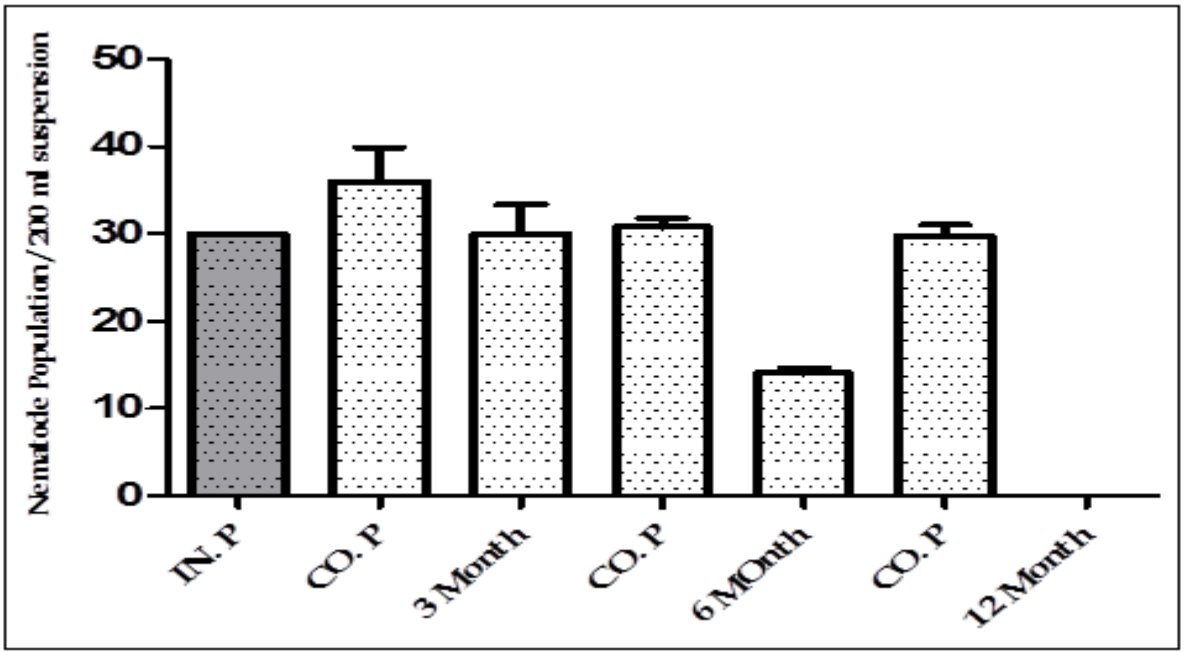

Fig. 3. Histogram Graph showing the effect of Carbofuran on Plant parasitic Nematode in Abbottabad.
IN.P= Initial Population, CO. P= Controlled Population.

In low temperatures they cannot survive and reproduce as mentioned by Lambert and Bekal in 2002 in their study showed that onset of winter and dry soil are harmful to nematode survival. Old farming techniques include the application organic wastes in the fields and trees. Organic wastes were found very helpful in the eradication and control of nematode population.

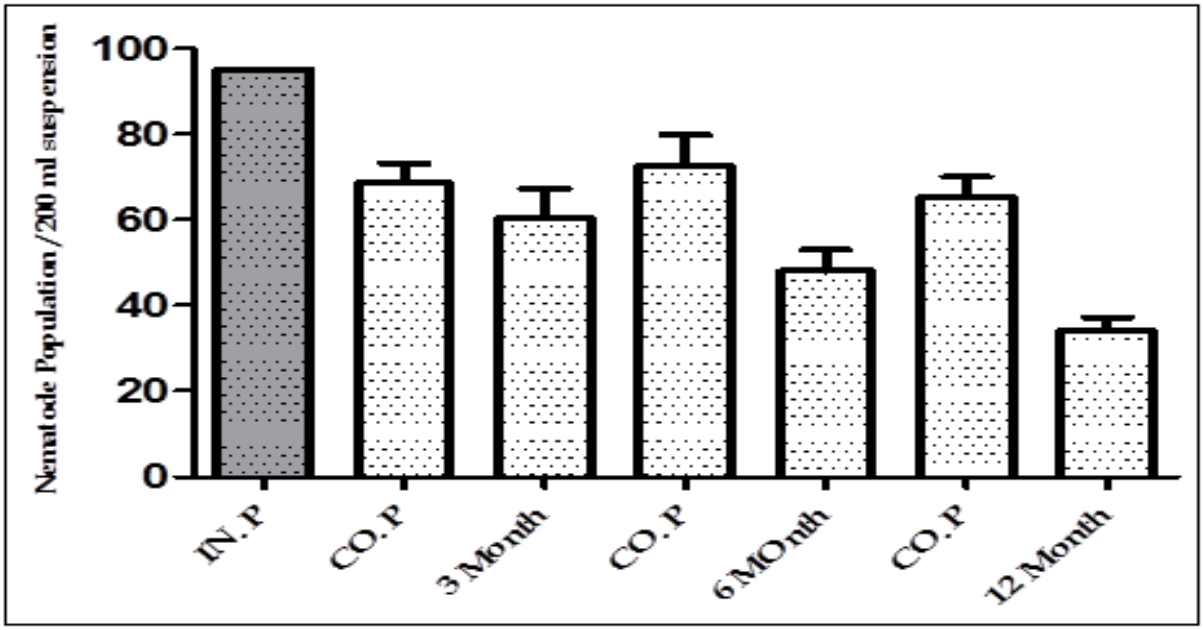

Fig. 4. Histogram Graph showing the effect of Poultry Manure on Plant parasitic Nematode in Mansehra.
IN.P= Initial Population, CO. P= Controlled Population.

Walnut trees treated with Carbofuran in Mansehra and Abbottabad showed a remarkable decline in nematode population as compared to the nematode population of the untreated or controlled ones as shown in figure 2 and 3. It showed that decline occurred just after the 3 months of application to the

affected tree in both Mansehra and Abbottabad. Further decline occurred after 6 months of application while after 12 month soil samples were found saprophytic. It showed that nematicide (Carbofuran) totally eradicate the plant parasitic nematode population associated with walnut trees. Carbofuran was found extremely effective against nematodes. Eradication of nematode also has positive impact on the plant growth and yield. While on the other hand, untreated trees showed high population of nematodes associated with walnut trees. Histogram graphs of effect of poultry manure on nematodes associated with walnut trees showed that nematode population was declined to a notice able level in both Mansehra and Abbottabad. Data of nematodes after 3 months of application showed the nematode population decline on comparison with the controlled ones.

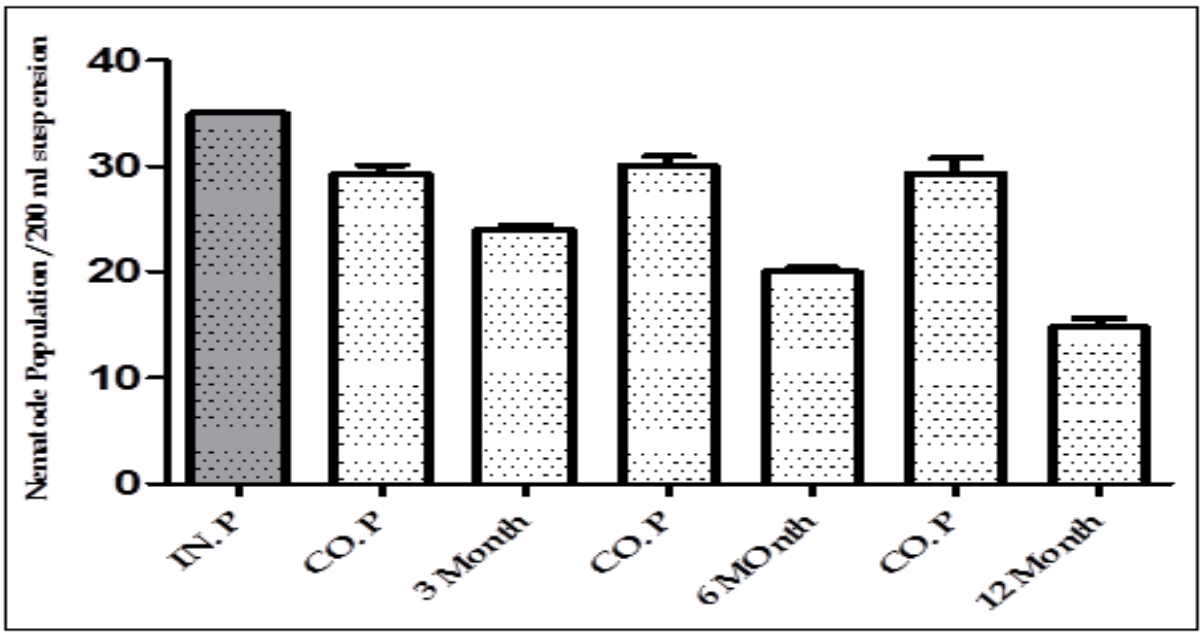

Fig. 5. Histogram Graph showing the effect of Poultry Manure on Plant parasitic Nematode in Abbottabad. IN.P= Initial Population, CO. P= Controlled Population.

In the same way further decline was noticed after 6 months and 12 months of application as shown in figure 4 and 5. Although they were not completely eradicated as in the case of Carbofuran but declined to a remarkable level from the initial population in both Mansehra and Abbottabad. It showed that poultry manure can be used for the management of plant parasitic nematodes associated with walnut and other crops in both Mansehra and Abbottabad. As poultry manure is organic in nature and environment friendly, therefore by keeping in view the atmospheric needs poultry manure is much recommended as compared to the nematicide which is a chemical in nature. Nematicide due to its chemical and poisonous nature, it is not good for health of human beings. Poultry manure decomposes and do not have any side effect on the human health. This is same to the study of Rehman *et al.*, from India in 2014 according to which Although it was studied that synthetic nematicide have noteworthy effect in limiting these plant parasitic nematodes but the toxic nature of these chemicals to the atmosphere and human beings required the research workers to go for safe and environment-friendly alternative for these chemicals. Statistical analysis was done on the recorded data of Carbofuran against plant parasitic nematodes associated with walnut trees in Mansehra and Abbottabad. ANOVA (Analysis of variance) was done which showed that P value was significant (P= 0.0001 for Mansehra and P=0.0001 for Abbottabad). Poultry manure was effective in reducing the nematode population associated with walnut trees. ANOVA results of effect of poultry manure in Mansehra and Abbottabad were also found significant as P value was

0.0001 and 0.0009 respectively. This is similar to the study of Osunlola and Fawole in 2015 whose studied data was statistically analyzed using ANOVA ($p \leq 0.05$).

Recorded data shows that all organic amendments and carbofuran were significant ($p \leq 0.05$) and reduced nematode reproduction and root damage.

Conclusion

After this survey it was concluded that plant parasitic nematode population was present in different localities of Hazara Division of KP, Pakistan. Both saprophytic and parasitic species were present. It was concluded that plant parasitic nematodes were causing damage to the walnut trees in the surveyed localities.

They go unnoticed and undetectable because of less awareness. They caused great damage to the plant health and fruit yield. Management is required to control plant parasitic nematodes. Inorganic amendment comprising of Carbofuran (Nematicide) and organic amendment including Poultry Manure reduced the plant parasitic nematode population. Carbofuran totally eradicate the population while Poultry Manure reduced to a noticeable extend. But as Poultry Manure is not toxic in nature therefore it is more suitable as compared to nematicide which is toxic in nature.

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