



Study Title\

Study on the Effectiveness of Fertilization with Harmal Plant in Killing or Inhibiting the Movement of Nematodes in Plants



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Abstract:

The research aims to study the effectiveness of using *Peganum harmala* (Harmal) plant fertilization to kill or inhibit the movement of nematodes in plants. This study was conducted in the Al Buraimi Governorate, where a decline in tree growth accompanied by yellowing leaves and gradual plant death was observed. The Globe Environmental Team sought to investigate the cause and find



solutions to address the problem in collaboration with relevant authorities and the STEM team. We developed a timeline for the project and began with field visits to the study site. We analyzed the soil properties at the study location and collected another soil sample from a site 3 meters away from the study area. We examined the pH, soil color, conductivity, salinity, and other soil characteristics using a soil protocol.

The proposed solution involved preparing a *Peganum harmala* extract by drying the plant's leaves and applying them to plants infected with nematodes. We studied the effect of adding *Peganum harmala* leaves on the number of leaves and plant height in two soil samples: a control sample (soil from the study site) and an experimental sample (soil treated with *Peganum harmala* leaves). Additionally, we collaborated with Sultan Qaboos University to analyze the *Peganum harmala* leaves using HPLC (High–Performance Liquid Chromatography) to measure the chemical compounds in the extract and determine the optimal concentration of the extract to achieve the best results in combating nematodes.

The results were as follows: The findings confirm that *Peganum harmala* (Harmal) extract has a strong effect on nematodes, making it a promising candidate for use as a natural pesticide in agricultural pest control. The Harmal extract demonstrated effectiveness in killing or inhibiting the movement of nematodes, particularly at higher concentrations. Additionally, the results showed that plants treated with Harmal extract exhibited improved growth compared to untreated plants. The efficacy of Harmal in combating nematodes can be attributed to the presence of natural chemical compounds such as alkaloids (harmaline and harmine) and flavonoids like quercetin and kaempferol, which possess anti–parasitic properties.

This study provides preliminary insights into the potential use of *Peganum harmala* as a natural alternative for nematode control in agriculture. If the results prove the extract's effectiveness, Harmal could become a promising option for farmers seeking to reduce their reliance on chemical pesticides. Key recommendations from the study include:

- 1. Conducting further research to determine the precise mechanism of Harmal's effectiveness against nematodes.
- 2. Testing the impact of Harmal on a wider range of crops and soil types.

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- 3. Evaluating the economic feasibility of using Harmal as a natural pesticide in agriculture.
- 4. Producing natural extracts from the plant and distributing them to those interested in sustainable agriculture.
- 5. Conducting additional studies to assess the extract's effect on other types of nematodes and under different environmental conditions.

This study could contribute to the development of sustainable and environmentally friendly methods for agricultural pest control, enhancing productivity and reducing dependence on harmful chemicals.

Key Terms:

- 1. Fertilization: A substance or plant added to the soil to help plants grow or eliminate specific pests.
- 2. **Nematodes:** Round, unsegmented, elongated, colorless worms, usually microscopic, that live in the soil and feed on plant roots, causing significant damage to crops.
- 3. **Harmal** (*Peganum harmala*): A perennial herbaceous plant with dense, spiky leaves and a distinctive smell, belonging to the Zygophyllaceae family. It is used in traditional medicine for its therapeutic properties.
- 4. HPLC (High–Performance Liquid Chromatography): An analytical technique used to separate, identify, and measure chemical compounds in plant extracts, such as Harmal leaf extract.

Research Questions:

- 1. What is the effectiveness of using Harmal in fertilizing plants infected with nematodes?
- 2. What are the appropriate concentrations of Harmal needed to achieve an antinematode effect?

3. Can commercial products be developed from Harmal to replace chemical fertilizers, and what challenges might this development face?

Introduction:

Agricultural crops are among the most important foundations of human life, contributing to economic and social development, especially in developing countries, where they play a vital role in poverty alleviation and sustainable rural development. The GLOBE environmental team at Um Dhar Al Ghafari School in Al Buraimi Governorate applies GLOBE environmental protocols (air, soil, land cover, and water) in several locations within the governorate. Recently, a decline in the growth of newly planted trees was observed in one of the sites, accompanied by yellowing leaves and gradual plant death. The GLOBE team sought to investigate the cause and find solutions to the problem. It was found that the plants were infected with root–knot .nematodes

Most agricultural crops are affected by various pests, including nematodes. Nematodes are microscopic organisms that live in the soil and cause significant damage to crops by attacking roots, hindering water and nutrient absorption, and potentially causing plant damage. Nematodes feed on the outer layer of the host plant's roots, and with continued feeding, cells die, tissues die, and the plants become stunted, with yellowing leaves and slow growth (Al Rashid, 2017).Controlling nematodes is a significant challenge for farmers, especially with increasing restrictions on the use of chemical pesticides due to their negative effects on the environment and human health. Therefore, natural and effective alternatives for controlling these pests have been sought, including the use of medicinal plants such as Peganum harmala (Harmal).

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Harmal is a perennial herbaceous plant with dense, spiky leaves and a distinctive smell, belonging to the Zygophyllaceae family. It has been used in traditional medicine for centuries due to its therapeutic properties. It grows in arid and semiarid regions, especially in the Middle East, North Africa, and Central Asia. Studies have shown that Harmal extract is effective as a natural fungicide against soil-borne fungi due to its content of compounds such as harmine and harmaline, which inhibit the growth of pathogenic fungi (Duke, 2002). Based on field observations and the problems faced by farmers, and after reviewing previous studies, the GLOBE team decided to study the problem and answer the following questions: What is the effectiveness of using Harmal in fertilizing plants infected with nematodes? What are the appropriate concentrations of Harmal needed to achieve an anti-nematode effect? Can commercial products be developed from Harmal to replace chemical fertilizers, and what challenges might this development face? This requires the application of GLOBE protocols and collaboration with relevant authorities, given the importance of this research in understanding and solving the problem for the benefit of the environment and its inhabitants.

Variables of the Study:

- Control Sample: Soil from the study site.
- Experimental Sample: Soil treated with Harmal leaves.
- Dependent Variable: Soil properties, plant growth.
- Constant Variables: Water and soil quality, irrigation amount and duration, light.

Research Plan:

1. **Identifying the Problem**: Through observing weak growth of newly planted trees, yellowing leaves, and gradual plant death.

- 2. Selecting the Research Problem: Defined through discussions with the GLOBE environmental team.
- 3. **Determining Study Tools:** GPS, soil catalog, pH meter, conductivity, and salinity meters to apply atmospheric and soil protocols.
- 4. **Formal Communication**: Contacting the Ministry of Agriculture and Sultan Qaboos University to present the project idea and collaborate on finding solutions and analyzing samples.
- 5. **Conducting Meetings**: With agricultural specialists from the Ministry of Agriculture and Sultan Qaboos University to discuss proposed solutions and how to reduce the problem.
- 6. Determining Study Locations Using GPS.
- 7. **Applying Protocols**: Soil and atmospheric protocols and entering data into the GLOBE website.
- 8. Collecting, Analyzing, and Converting Data into Charts.
- 9. Extracting, Interpreting, and Comparing Results with Other Research and Writing Recommendations.

Notes	Responsible	Plan	Month
Contacting relevant authorities through the school's communication system	Student: Salima Al Rashidi Student: Al Yazia Al Rashidi In collaboration with the GLOBE team	 Identifying the problem Contacting the Ministry of Agriculture Contacting Sultan Qaboos University Providing project tools (water and soil protocols) 	September
Providing project tools	Student: Salima Al Rashidi Student: Al Yazia Al Rashidi In collaboration with the GLOBE team and the	 Applying soil protocols at specified sites Analyzing infected soil samples 	October

Timeline for Research Preparation:

	Ministry of Agriculture		
Data entry Meetings with the team	Student: Salima Al Rashidi Student: Al Yazia Al Rashidi In collaboration with the GLOBE team and Sultan Qaboos University	 Entering data into the GLOBE website Collaborating with relevant authorities to draft recommendations and begin writing the research 	November
Student: Al Yazia Al Rashidi: Abstract, introduction, and research problem (joint application) Student: Salima Al Rashidi: Results, analysis, conclusion, and research formatting	Student: Salima Al Rashidi Student: Al Yazia Al Rashidi Supervised by the GLOBE program supervisor	- Writing the research and preparing for participation in the competition - Creating an electronic poster	December

Study Location:

(Sultanate of Oman, Al Buraimi Governorate), Ard Jaw area (*Latitude 24.24696*), (*Longitude 55.82804*), (*Elevation 313 m*). Soil and vegetation cover protocols were applied.









Data Collection and Analysis:

Data related to this research were collected through the international information network (Internet), in addition to conducting interviews with specialists from the Ministry of Agriculture in Al Buraimi Governorate and specialists at the Central Research Center at Sultan Qaboos University. The GLOBE environmental protocols for agricultural soil and atmospheric samples were also applied to the study area to determine the extent of improvement in the growth of plants tested in this research. Soil samples were taken from two sites at different distances for each sample and examined with the assistance of the Plant Protection Research Center at the Directorate General of Agricultural and Animal Research. Harmal extract was analyzed to identify the compounds present in the extract that contribute to biological control, with the assistance of Sultan Qaboos University.









Additionally, a sample of infected plant roots was taken, and the roots infected with nematodes were exposed to different concentrations of Harmal extract, with the assistance of the Central Laboratory at the College of Agricultural and Marine Sciences at Sultan Qaboos University. Experimental conditions (such as temperature, humidity, and the type of nematodes used) were controlled to ensure the accuracy of the results.



Plant growth was also observed, including stem length (measured using a ruler), number of leaves, and leaf color. These data were entered into tables and analyzed graphically. Two samples of plants infected with nematodes were identified: the first was planted in agricultural soil without Harmal, and the second was planted with the addition of dried Harmal leaves. Both samples were irrigated with equal amounts of water daily, from the same source, and under the same environmental conditions (light and ventilation) to ensure fairness in testing all samples. A chemical analysis of the soil treated with Harmal was conducted to determine changes in soil properties.



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Results:

The physical and chemical properties of the Harmal plant were observed, and soil samples from the study site and another site 3 meters away were analyzed with the assistance of the Plant Protection Research Center at the Directorate General of Agricultural and Animal Research to confirm nematode infection in the plant roots and determine whether the infection was widespread in agricultural areas near the .study site

Moisture	pH	Density	Taste	Smell	Color
≤10%	5.5-7.5	0.9-1.2	Bitter	Strong,	Dark brown
		g/ml		aromatic,	to yellowish
				and	green
				bitter	

Table 1: Physical and Chemical Properties of Harmal Plant

Table 2: Results of Nematode Infection at the Study Site

Nematode Infection	Sample		No
	Туре	Sample	INU
Root Knot Nematode	Soil and	Soil from the study site	1
(Meloidogyne sp.)	roots	Soli nom the study site	1
No infection	Soil	Soil 50 meters from the	0
NO INIECION	301	study site	Z

GLOBE environmental protocols were applied to the soil, and data on plant growth manifestations were recorded and observed over a month as follows:

Table 3: Results of Applying GLOBE Protocols to Soil Samples Before Adding

	Harmal								
Temper	Soil					Textur	Consist	Structur	
ature	Color	Conductivity	Salinity	рН	Rocks	e	ency	е	Sample
	5Y					Sandy	Fragile		Infected
33.4	7/6	2	1.5 ppm	4.31	Few	loam	soil	Granular	soil
	5YR					Sandy	Fragile		Uninfec
36.2	6/8.7	5	3 ppm	6.72	Few	loam	soil	Granular	ted soil

 Table 4: Results of Applying GLOBE Protocols to Soil Samples After Adding

 Harmal

рН	Conductivity	Oxygen	Salinity	Temperature	Sample Type
	(μs)	(mg/l)	(ppm)	(°C)	
4.31	2	2	1.5	33.4	Control sample
					(soil only)
6.55	4	5	3	35.4	Experimental
					sample (soil +
					Harmal leaves

 Table 5: Effect of Adding Harmal Leaves on the Number of Plant Leaves in the

Study Sample

	Experimental Sample		Control Sample	
Leaf	Number of Leaves in	Leaf	Number of	Week
Color	Experimental Sample	Color	Leaves in	
	(Soil + Harmal)		Control Sample	
	8		8	Week 1 (1-
Light green		Light green		5/10/2024)
8 8	12	88	10	Week 2 (7-
				13/10/2024)
	15		9	Week 3 (14-
	15		5	20/10/2024)
Dark groop	18	Yellow	5	Week 4 (21-
Dark green	18	renow	5	27/10/2024)
	21		3	Week 5 (28-
	21		5	3/11/2024)

Figure 1: Chart Showing the Effect of Adding Harmal Leaves on the Number of Plant

Leaves in the Study Sample

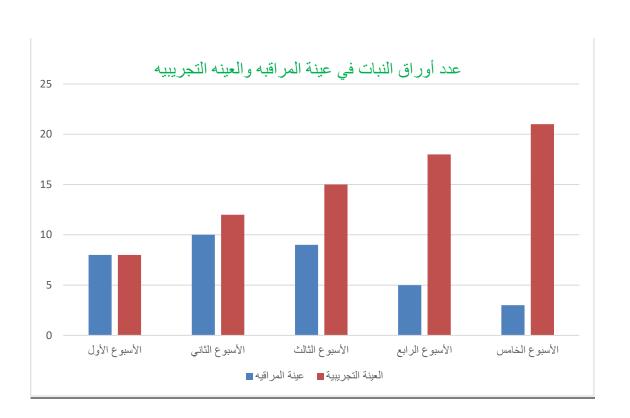


Table 6: Effect of Adding Harmal Leaves on Plant Height in the Study Sample (Using

a Ruler)

Ex	Experimental Sample		Control Sample	
Leaf Color	Plant Height in	Leaf Color	Plant Height in	Week
	Experimental		Control Sample	
	Sample (Soil +		(cm)	
	Harmal) (cm))			
	7		7	Week 1 (1-5/10/2024)
Light green	11	Light green	11	Week 2 (7-
				13/10/2024)

	45		44.5	Week 3 (14-
	15		11.5	20/10/2024)
Daulaanaan	10	Vallassi	11 5	Week 4 (21-
Dark green	19	Yellow	11.5	27/10/2024)
	22		11 5	Week 5 (28-
	23		11.5	3/11/2024)

Figure 2: Chart Showing the Effect of Adding Harmal Leaves on Plant Height in the Study Sample

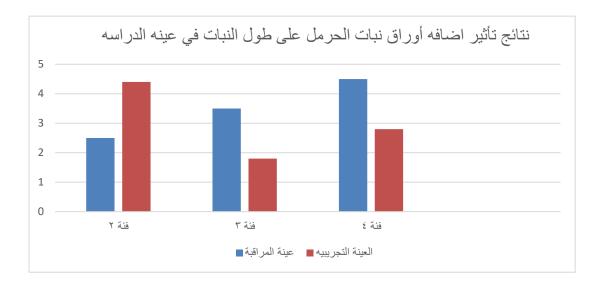


Table 7: Results of Analyzing Harmal Leaves Using HPLC

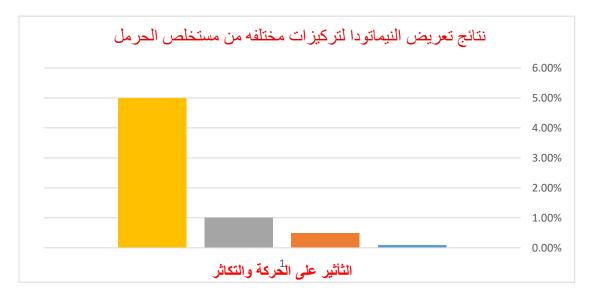
Concentration (mlg/g)	Element	Concentration (mlg/g)	Chemical Compound
1500	(Ca) Calcium	10.0	(Harmine)
600	(Mg) Magnesium	5.0	(Harmaline)
3000	(K) Potassium	2.5	(Harmalol)

200	(Fe) Iron	4.0	(Vasicine)
50	(Zn) Zinc	2.0	(Quercetin)
20	(Cu) Copper	1.5	(Kaempferol)
50	(Mn) Manganese	1.8	(Chlorogenic Acid)
500	(Na) Sodium	0.8	(Saponins)
0.1	(Pb)	1.2	(Tannins)
0.01	(As)		
0.01	(Hg)		

Table 8: Results of Exposing Nematodes to Different Concentrations of Harmal Extract

Additional Notes	Effect on Reproduction	Effect on Movement	Mortality Rate (%)	Volume of Solvent (ml)	Volume of Harmal Extract (ml)	Concentration (%)
Limited effect	Slight	Slight slowdown	10	99.9	0.1	0.1%
Moderate	Noticeable	Noticeable slowdown	30	99.5	0.5	0.5%
Strong effect	Reproduction stopped	Partial paralysis	60	99	1	1%
Severe effect	Complete stop	Complete paralysis	90	95	5	5%

Figure 3: Chart Showing the Results of Exposing Nematodes to Different Concentrations of Harmal Extract



Data were entered and submitted to the GLOBE program website (www.GLOBE.gov) via the DATA ENTRY application, where a new site was added, and water protocol data collected in the research were entered.

GLOBE Visualization System	Measurements Data Count	- 3 1	• wasa	A Sign In
s T 🖻 ? 💄 🔹 🔹	2019-02-14			
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Plot Date Range: 2018-10-14 10 2018-15-15	Almuelate by	vestigation Area Date Activated drology 2018-04-23 Unoisture 2018-04-23		
For optimum performance, the maximum recommended date range is 5 years	ALmueleos_UMther hy	Licharacteristics 2018-04-23 drology 2018-11-14	- 1	
Single Line Plot Stacked Plot Use Auto-Y Aris	Airabi_umthar2 hy	drology 2018-11-14 drology 2019-02-22 norphere 2018-10-15		

Discussion of Results:

After obtaining the results, they were presented to officials from Sultan Qaboos University, Dr. Jamal Al Subhi, and Engineer Mahmoud Al Nizwani from the Ministry of Agriculture in Al Buraimi to answer the first question: What is the effectiveness of using Harmal in fertilizing plants infected with nematodes?

Data from Table 2, Table 3, and Table 4 were used. It was found that Harmal is rich in antimicrobial elements at high levels, such as alkaloids: harmine and harmaline, and flavonoids such as quercetin and kaempferol, and saponins. Additionally, it contains many antioxidant elements that can also be beneficial for human use, such as chlorogenic acid and kaempferol. Elements such as calcium, magnesium, and potassium are also present in high proportions in Harmal leaves and play an important role in plant biochemical interactions (Buhner, 2012). Additionally, these elements improve plant growth and enhance root, stem, and leaf growth. This is shown in the results of Table 3 and Table 4. Additionally, these elements can increase disease resistance by strengthening cell walls and improving the plant's natural immunity. They also enhance the photosynthesis process (Al Rashid, 2017).

To answer the second question: What are the appropriate concentrations of Harmal needed to achieve an anti-nematode effect? Harmal extract was prepared using a solvent (ethanol). Data from Table 5 and Table 6 were observed, and it was found that at a concentration of 0.1%, there is a slight effect on nematodes, with a low mortality rate and slight slowdown in movement. At a concentration of 0.5%, the effect is moderate, with an increase in mortality rate and noticeable slowdown in movement. At a concentration of 1%, there is a strong effect, with a high mortality rate and partial paralysis of nematodes. At a concentration of 5%, there is a severe effect, with a very high mortality rate and complete paralysis of nematodes. The results also showed that very high concentrations of Harmal may negatively affect plant cell growth due to the presence of arsenic, mercury, and lead. This is shown in the results of Table 5. These heavy and toxic elements, at high levels, lead to

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coagulation and curling of plant leaves, thus affecting the photosynthesis process, so the plant does not get enough food, weakens, and then dies (Abdul Rahman, 2015).

To answer the third question: Can commercial products be developed from Harmal to replace chemical fertilizers, and what challenges might this development face? The research results and findings were discussed with relevant authorities, such as the Ministry of Agriculture and Dr. Jamal from Sultan Qaboos University, to find solutions that serve the environment. The solutions were to chemically and technically treat Harmal extract, and these compounds can be extracted and developed into liquid or powder formulations that are easy to apply and benefit from, to generate income for the country. However, this requires careful planning and overcoming technical, economic, and regulatory challenges. If these challenges are overcome, Harmal could become a promising option in the field of biological control of nematodes, especially in organic and sustainable agriculture, as indicated by Dr. Jamal Al Subhi, head of the Central Laboratory at Sultan Qaboos University. Harmal can also be a suitable option as a natural pesticide for nematodes, as the cost is relatively low. Harmal is a plant available in many regions, especially in arid and semi-arid areas, and is widespread in the wadis of Oman. This may reduce production costs and reliance on expensive chemical pesticides. Given the increasing demand for natural alternatives, with increasing environmental and health awareness, there is a growing demand for organic and natural products in agriculture. The research results are consistent with the findings of (Abdul Rahman 2015).

Challenges that may face development:

1. Determining Effective and Safe Concentrations: The optimal concentrations effective against nematodes must be determined without harming plants or beneficial organisms in the soil, as high concentrations may be toxic to plants or the environment.

- 2. Environmental Impacts: The environmental impacts of using Harmal on nontarget organisms, such as beneficial soil microorganisms and beneficial insects, must be studied.
- 3. **Registration and Regulation**: Developing any commercial product requires obtaining regulatory approvals from relevant authorities. The registration process may be costly and time-consuming, especially if detailed toxicity studies are required.
- Economic Cost: Although Harmal is available in nature, the process of extracting active compounds and manufacturing them in large quantities may be costly.
- 5. **Competition with Chemical Pesticides**: Traditional chemical pesticides are often readily available and faster–acting, which may make it difficult for natural products to compete in the market.
- 6. **Farmer Awareness and Acceptance**: Farmers may need training on how to effectively use Harmal-based products. There may also be hesitation in adopting new products that are not widely known.

Steps to Develop Commercial Products Based on Harmal:

- 1. **Conduct Scientific Studies**: Identify active compounds and their effectiveness against nematodes. Study the effects on plants, soil, and non-target organisms.
- 2. **Improve Extraction Methods**: Develop efficient and low-cost methods for extracting active compounds.
- 3. **Manufacture and Test the Product**: Develop different formulations (liquid, powder, granular) and test them in field conditions.
- 4. **Marketing and Awareness**: Educate farmers about the benefits of the product and how to use it, and market the product as a natural and safe alternative.

Conclusion:

In this research, we attempted to prove the effectiveness of fertilization with Harmal plant in killing or inhibiting the movement of nematodes in plants. We applied GLOBE environmental protocols and conducted field visits. We reached the following conclusions: Harmal plant contains effective compounds with anti-nematode properties. Harmal extracts can be used as a natural and safe alternative to chemical pesticides in controlling nematodes. Further studies are needed to determine the optimal concentrations and the most effective application method. Through this research, a set of recommendations were made with the assistance of the environmental team, the most important of which are:

- The necessity of utilizing Harmal as a suitable option as a natural pesticide for nematodes.
- Developing efficient and low-cost methods for extracting active compounds from Harmal extract for utilization.
- Obtaining healthy seedlings from a reliable source and spacing them to prevent the spread of infection, and keeping trees away from any environmental or agricultural stressors.
- Educating farmers about the benefits of Harmal and how to use it as a natural and safe alternative.

We recommend disseminating the research idea to the international community for benefit, and conveying the problem and recommendations to the relevant authorities to play an active role in reducing agricultural pests. The strengths of the research, in my opinion, are contributing to solving an environmental problem using locally available plants, and the research contributed to developing cognitive, skill, and research and experimentation strategies to find environmental solutions. We believe that this research is very useful and can contribute to solving a Gulf problem in general if attention is given by officials and recommendations are adopted.

Acknowledgments:

We would like to extend our sincere thanks and appreciation to Mr. Ahmed Al Balushi, the National Coordinator of the GLOBE environmental program in the Sultanate of Oman, for all the information he provided, and to the members of the program's central team and the program team in the governorate for their continuous follow–up and encouragement in preparing and presenting the research in the appropriate manner, especially Ms. Amna Al Saadi and the STEEM team.

We also extend our thanks to Ms. Nawal Al Shamsi, the school principal, for her cooperation and role in providing advice and guidance on everything related to the research. To Ms. Rahma Al Badi, the laboratory specialist at Um Dhar Al Ghafari School, for her cooperation with us. To Engineer Mahmoud Al Nizwani from the Ministry of Agriculture for his cooperation with us in collecting and analyzing data, and to Dr. Jamal Al Subhi and Engineer Mohammed Al Shamsi for their support in contributing to finding solutions to the problem.

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2- Duke, J. A. (2002). Handbook of medicinal herbs (2nd ed.). CRC Press.

3- Buhner, S. H. (2012). Herbal antibiotics: Natural alternatives for treating drug-resistant bacteria (2nd ed.). Storey Publishing.

Appendix (1):

Photos of interviews with specialists at Sultan Qaboos University and Department of Agriculture Research Center



Appendix (2):

Results of Harmal analysis from Sultan Qaboos University.



Appendix (3):

Results of soil and root sample analysis

مركز بحوث وقاية النبات قسم بحوث أمراض النبات	تقرير في	المديرية العامة للبحوث الزراعية والحيوانية
التاريخ: 25-12-2024م		مصدر العينة: محافظة البريمي
نوع العينة: تربة من حقل زراعي		فاحص العينة: م. محمد العبدلي

<u>1</u>. نتائج التشخيص:

بعد فحص عينات التربة والجذور بقسم بحوث أمراض النبات أظهرت النتائج ما يلي:

الإصابة النيماتودية	نوع العينة	العينة	م
Root Knot Nematode (Meloidogyne sp.)	تربة وجذور	البريمي 1	1
لا توجد إصابة	تربـــة	البريمي 2	2

3. التوصيات:

- يُفضّل التخلص من المحصول بالحرق إذا كانت الإصابة عالية، لمنع انتشار النيماتودا.
- معاملة التربة بالتعقيم الحراري مع ضرورة تغطيتها بغطاء أسود لمنع الرطوبة ونمو جديد وإصابات أخرى.
 - معاملة التربة والمحصول المصاب بمبيد (Velum Prime) بمعدل 202مل لكل فدان على أن تكون المعاملة بعد الري مباشرة وتعاد مرة واحده بعد 15 يوم لمكافحة النيماتودا.
 - يرجى عدم استخدام مبيد Velum Prime في فترة الحصاد وذلك لسميته العالية ويستخدم قبل زراعة المحصول القادم.

.....قسم بحوث أمراض النبات.....قسم