IDENTIFICATION AND MANAGEMENT OF ROOT KNOT NEMATODES
(Meloidogyne spp.) ATTACKING CHICKPEA (Cicer arietinum L.) IN NAKURU COUNTY, KENYA USING NEMATICIDES AND CULTURAL PRACTICES

KIMANI ISHMAEL MAINA

A Thesis Submitted to the Graduate School in Partial Fulfillment for the Requirements of the Award of Master of Science Degree in Plant Pathology of Egerton University

EGERTON UNIVERSITY

AUGUST, 2016
DECLARATION AND RECOMMENDATION

DECLARATION

This is my original work and has not been submitted or presented for award of degree in any other University

Signature........................................ Date........................................

Kimani Ishmael Maina

SM15/3675/13

RECOMMENDATION

This Thesis has been submitted for examination with our approval as supervisors for examination as per the Egerton University regulations.

Signature........................................ Date........................................

Dr. Japhet M. Muthamia

Department of Biological Sciences,

Egerton University, Njoro

Signature........................................ Date........................................

Prof. D.O. Otaye

Department of Biological Sciences,

Egerton University, Njoro
COPYRIGHT
© 2016 Ishmael Maina Kimani

No part of this thesis may be reproduced in any retrieval system, transmitted in any form or means, mechanical, electronic, recording, photocopying or otherwise without prior permission from the author or Egerton University on that behalf.
DEDICATION

To my parents Mr. and Mrs. Kimani, my Aunt Ms. Esther Kahare.
ACKNOWLEDGEMENT

I would like to honour and thank the Almighty God for giving me a chance in life to do this work. Much gratitude goes to Egerton University and the Chairman, Department of Biological Sciences, Prof. A. Yasindi for allowing me to pursue my study, use the departmental glasshouse and laboratory services. I wish to sincerely thank my supervisors Dr. Japhet Muthamia and Prof. Daniel Otaye for their guidance, direction and support in all stages of this work. I would like to thank Dr. J. Muthamia again for encouraging me to take research work in Plant Nematology. I would also like to thank the Chief Technologist Botany section, Mr. Francis Ngumbu for his technical assistance, Mr. Bernard K. from Field 7, Egerton University for helping me acquire chickpea seeds. To my family and friends, much gratitude for prayers and perseverance during my study. Lastly, to my course mates Charei Munene, Fatuma Sharamo, Cynthia Wakhungu and Lucy Jepkemoi for their encouragement during both coursework and research work.
ABSTRACT

Root knot diseases are known to attack many crops especially legumes such as chickpea. Losses due to root knot nematodes (*Meloidogyne* spp.) in chickpea have been approximated at 13.7% of yield loss which translates to loss of billions of shillings annually. Four main *Meloidogyne* species; *M. hapla*, *M. javanica*, *M. arenaria* and *M. incognita* attack chickpeas. Experiments were conducted in the greenhouse to characterise, identify and control *Meloidogyne* spp. causing root knot disease in chickpea in Nakuru County. Characterization and identification were done using perineal patterns procedure on female root knot nematodes. Three treatments; poultry manure, two commercial nematicides and Sudan grass (*Sorghum sudanese*) were used in this study. Poultry manure was used at three levels; 250 g, 500 g, 750 g and control. Marshal 250 EC nematicide was used at 10 ml/ litre of water, 25 ml/ litre and 50 ml/ litre as low, recommended, high doses respectively. Nimbecidine nematicide was used at 3.5 ml/ litre of water, 7 ml/ litre and 14 ml/ litre as low, recommended, high doses respectively and control in each treatment. Completely randomized design was used in the study as a design for treatments. Thirty samples of *Meloidogyne* spp. female were used during characterisation and all had uniform perineal patterns similar to that of *M. javanica* distinguished from other species by a distinct lateral ridge separating dorsal and ventral arch. *M. javanica* was the main root knot species attacking chickpeas. There was significant difference (P=0.05) in root galling and *M. javanica* juveniles population reduction between positive control (zero grams) and other poultry manure treatments. The nematicides significantly reduced root galling and number of juveniles at recommended and high levels. Marshal 250 EC was an effective nematicide than Nimbecidine and Marshal 250 EC which recorded the lowest root galling and number of juveniles as compared to Nimbecidine. There was no significant difference (P=0.05) in results of 250 g, 500 g and 750 g poultry treatments. There was significant difference (P=0.05) between results of Sudan grass, positive and negative control. Poultry manure and Sudan grass treatments significantly reduced root galling and nematodes juvenile population. There was a relationship between root galling and juvenile number in soil, root galling and root weight per chickpea plant. The findings of this study will benefit farmers on choice of commercial nematicide to use against root knot nematodes to maximize yield of chickpea. The findings can also be used by farmers to manage root knot disease using cover crops such as Sudan grass.
# TABLE OF CONTENTS

<table>
<thead>
<tr>
<th>Section</th>
<th>Page</th>
</tr>
</thead>
<tbody>
<tr>
<td>DECLARATION AND RECOMMENDATION</td>
<td>2</td>
</tr>
<tr>
<td>COPYRIGHT</td>
<td>3</td>
</tr>
<tr>
<td>DEDICATION</td>
<td>4</td>
</tr>
<tr>
<td>ACKNOWLEDGEMENT</td>
<td>5</td>
</tr>
<tr>
<td>ABSTRACT</td>
<td>6</td>
</tr>
<tr>
<td>TABLE OF CONTENTS</td>
<td>7</td>
</tr>
<tr>
<td>LIST OF TABLES</td>
<td>11</td>
</tr>
<tr>
<td>LIST OF FIGURES</td>
<td>12</td>
</tr>
<tr>
<td>LIST OF PLATES</td>
<td>13</td>
</tr>
<tr>
<td>LIST OF ABBREVIATIONS AND ACRONYMS</td>
<td>14</td>
</tr>
<tr>
<td>CHAPTER ONE</td>
<td>1</td>
</tr>
<tr>
<td>INTRODUCTION</td>
<td>1</td>
</tr>
<tr>
<td>1.1 Background Information</td>
<td>1</td>
</tr>
<tr>
<td>1.2 Statement of the problem</td>
<td>2</td>
</tr>
<tr>
<td>1.3 Objectives</td>
<td>2</td>
</tr>
<tr>
<td>1.3.1 General objective</td>
<td>2</td>
</tr>
<tr>
<td>1.3.2 Specific objectives</td>
<td>2</td>
</tr>
<tr>
<td>1.4 Hypotheses</td>
<td>3</td>
</tr>
<tr>
<td>1.5 Justification of the study</td>
<td>3</td>
</tr>
</tbody>
</table>
CHAPTER TWO

LITERATURE REVIEW

2.1 Chickpea origin and center of diversity .................................................................5

2.1.1 Plant description and ecology .............................................................................5

2.2 Varieties of chickpea ...............................................................................................5

2.3 Economic importance of chickpea .........................................................................6

2.3.1 Carbohydrates ....................................................................................................6

2.3.2 Proteins .................................................................................................................6

2.3.3 Fat and fatty acids ...............................................................................................6

2.3.4 Vitamins ................................................................................................................7

2.3.5 Health benefits .....................................................................................................7

2.4 Chickpea production in Kenya ...............................................................................7

2.5 Diseases of chickpea ..............................................................................................7

2.5.1 Ascochyta blight ................................................................................................8

2.5.2 Bacterial blight ....................................................................................................8

2.5.3 Chickpea stunt virus disease ...............................................................................9

2.6 Nematode diseases .............................................................................................9

2.6.1 Root knot nematode ..........................................................................................9

2.6.2 Pathogenesis ......................................................................................................10

2.6.3 Life and disease cycles of Meloidogyne spp. ......................................................11

2.6.4 Symptoms .........................................................................................................11
2.7 Control of root knot nematodes ........................................................................................................ 11
  2.7.1 Crop rotation .................................................................................................................................. 11
  2.7.2 Use of Chemical formulations ......................................................................................................... 12
  2.7.3 Use of Sudan grass (S. sudanese) to manage nematodes ................................................................. 12
  2.7.4 Biological management .................................................................................................................... 13
  2.7.5 Host resistance .................................................................................................................................. 13
  2.7.6 Use of soil amendments ................................................................................................................... 13

CHAPTER THREE ........................................................................................................................................ 15

MATERIALS AND METHODS .................................................................................................................... 15
  3.1 Description of study area ..................................................................................................................... 15
  3.2 Sample collection of Meloidogyne spp. ................................................................................................. 15
    3.2.1 Extraction of root knot nematodes from root galls for characterisation and identification ................................................................................................................................................................................................. 16
  3.3 Inoculum preparation .......................................................................................................................... 16
    3.3.1 Procedure for inoculation .............................................................................................................. 16
  3.4 Determination of the effectiveness of common commercial nematicides against Meloidogyne spp. .................................................................................................................................................................................................. 16
  3.5 Determination of the effect of poultry manure on the control of root knot nematodes ..................... 17
  3.6 Determination of the effect of Sudan grass (S. sudanese) in Meloidogyne spp. population 18

CHAPTER FOUR ........................................................................................................................................... 19

RESULTS ..................................................................................................................................................... 19
4.1 Characterisation and identification of *Meloidogyne* spp. attacking chickpea in Nakuru County

4.2 Effectiveness of two common commercial nematicides against root knot nematode disease in chickpea

4.3 Effect of poultry manure in the control of root knot nematode diseases in chickpea

4.4 Effect of Sudan grass (*S. sudanese*) in the *Meloidogyne* spp. population as a management strategy against root knot disease

CHAPTER FIVE

DISCUSSIONS

5.1 Characterisation and identification of *Meloidogyne* spp. attacking chickpea in Nakuru County

5.2 Effectiveness of two common commercial nematicides against root knot nematode diseases in chickpea

5.3 Determination of the effect of poultry manure on the control of root knot nematodes

5.4 Determination the effect of Sudan grass (*S. sudanese*) on the *Meloidogyne* spp. population as a management strategy against root knot disease

CHAPTER 6

CONCLUSIONS AND RECOMMENDATIONS

REFERENCES

APPENDICES
LIST OF TABLES

Table 1: Diseases attacking chickpea ........................................................................................................2

Table 2: Taxonomic characters of perineal patterns of the four common root knot nematodes ...10

Table 3: Fresh root, dry root weight, galls, gall indices and number of *M. javanica* juveniles/100 g soil in Marshal 250 EC treatment eight weeks after inoculation .........................................................21

Table 4: Fresh root, dry root weight, galls, gall indices and number of *M. javanica* juveniles/100 g soil in Nimbecidine treatment eight weeks after inoculation ....................................................22

Table 5: Root knot disease parameters in Marshal 250 EC and Nimbecidine treatments eight weeks after inoculation .................................................................................................................22

Table 6: Comparison of Marshal 250 EC and Nimbecidine treatments in root knot disease parameters evaluated eight weeks after inoculation into chickpea .................................................23

Table 7: Relationship between gall indices, fresh and dry root weights eight weeks after poultry manure treatment ..........................................................................................................................25

Table 8: Number of *M. javanica* juveniles per 100 g of soil, eight weeks after poultry manure treatment ........................................................................................................................................26

Table 9: Root gall indices eight weeks after inoculation and poultry manure treatment ......................27

Table 10: Root gall indices, fresh and dry roots weight eight weeks after inoculation with *M. javanica* juveniles in Sudan grass treatment ..........................................................................................30

Table 11: Relationship between root gall index and juveniles in 100 g soil eight weeks of Sudan grass treatment ........................................................................................................................................31
LIST OF FIGURES

Figure 1: Map of study area showing study sites .................................................................15

Figure 2: Mean root gall index for each level of nematicide treatment .............................24

Figure 3: Average number of *M. javanica* juveniles/100 g of soil for each level of nematicide treatment .................................................................................................................................24

Figure 4: Mean number of *M. javanica* juveniles/100 g of soil in each level of poultry manure treatment ..........................................................................................................................27

Figure 5: Average fresh root and dry root weight per plant for each level of poultry manure treatment ........................................................................................................................................28

Figure 6: Mean root gall indices per plant in each level of poultry manure treatment ........28

Figure 7: Relationship between root galling indices and the number of *M. javanica* juveniles in 100 g of soil on poultry manure treatment ..................................................................................................................29

Figure 8: Mean root gall index per plant in Sudan grass treatments ...............................31

Figure 9: Relationship between root galling indices per plant and average number of *M. javanica* in 100 g soil in Sudan grass treatment .................................................................................................................32
LIST OF PLATES

Plate 1: Perineal patterns of *M. javanica* .................................................................19
Plate 2: Features in the perineal patterns .................................................................20
Plate 3: Uninoculated (A) and inoculated (B) chickpea plant .............................33
Plate 4: Heavily galled (C) and non-galled chickpea roots (D) .........................33
Plate 5: Chickpea plant cropped with Sudan grass ................................................34
# LIST OF ABBREVIATIONS AND ACRONYMS

<table>
<thead>
<tr>
<th>Abbreviation</th>
<th>Full Form</th>
</tr>
</thead>
<tbody>
<tr>
<td>ACA</td>
<td>Agriculture crops alternatives</td>
</tr>
<tr>
<td>BeYDV</td>
<td>Bean yellow dwarf virus</td>
</tr>
<tr>
<td>BMI</td>
<td>Basal metabolic index</td>
</tr>
<tr>
<td>CRD</td>
<td>Completely randomized design</td>
</tr>
<tr>
<td>DF</td>
<td>Dietary fiber</td>
</tr>
<tr>
<td>DM</td>
<td>Dry matter</td>
</tr>
<tr>
<td>DNA</td>
<td>Deoxyribonucleic acid</td>
</tr>
<tr>
<td>FAO</td>
<td>Food and Agriculture Organization</td>
</tr>
<tr>
<td>FAOSTAT</td>
<td>Food and Agriculture Organization statistics</td>
</tr>
<tr>
<td>ICRISAT</td>
<td>International Crops Research Institute for Semi-arid Tropics</td>
</tr>
<tr>
<td>LA</td>
<td>Linoleic acid</td>
</tr>
<tr>
<td>LSD</td>
<td>Least significant difference test</td>
</tr>
<tr>
<td>RKN</td>
<td>Root knot nematodes</td>
</tr>
<tr>
<td>SCFA</td>
<td>Short chain fatty acid</td>
</tr>
<tr>
<td>USDA</td>
<td>United State Department of Agriculture</td>
</tr>
<tr>
<td>CpCDV</td>
<td>Chickpea chlorotic dwarf virus</td>
</tr>
</tbody>
</table>