

# Dosage screening of *Heterorhabditis indica* for chafer grub control in peanut fields

LIU Qi-zhi\*<sup>1</sup>, WU Han-dong<sup>1</sup>, Arne Peters<sup>2</sup>, DU Xiao-kang<sup>1</sup>, ZHANG Li-juan<sup>1</sup>,  
LIANG Lin-lin<sup>1</sup>, XIE Na<sup>1</sup>, Ralf-Udo Ehlers\*<sup>3</sup>

1. Entomology and Nematology Laboratory, College of Agriculture and Biotechnology, China Agriculture University, Beijing 100193, China;

2. e-nema, Gesellschaft für Biotechnologie und biologischen Pflanzenschutz mbH Klausdorfer Str. 28-36 24223 Schwentinental, German;

3. Institute for Phytopathology, Christian-Albrechts-University, Department Biotechnology & Biology Control, Hermann-Rodewald-Str. 924118 Kiel, Germany

\* Corresponding Author: lqzzyx126@126.com ; ehlers@biotec.uni-kiel.de

**Abstract:** A trial of entomopathogenic nematode (*Heterorhabditis indica*) in the dosages of 2000, 3000, 4000, 5000 and 10000 IJs/pit (peanut plant) was conducted in this study for selecting an effective dosage to control chafer grubs in a peanut fields. The results showed that: 1) Damaged legumes (%) were significantly less in all treatments compared to the control treated with tap water only. The dosages of 4000, 5000 and 10000 IJs/pit resulted in 4.1%, 5.7% and 7.5% damage, respectively, which was significantly lower in the positive control treated with 40% isofenphos-methyl (10.2%); 2) A 95.7% decrease of grub numbers was achieved dosages of 4000 and 5000 IJs/pit, which was significantly higher than in the positive control (82.7%); 3) Peanut yield in the dosages of 4000 IJs/pit was 327.6 kg/667m<sup>2</sup>, significantly higher than in the positive control (292.6kg/667m<sup>2</sup>). The results demonstrated that *H.indica* could successfully control chafer grub and subsequently increase peanut yield and is superior to the chemical control strategy. The dosage of 4000 IJs/pit was the best one.

**Key words:** *Heterorhabditis indica*; isofenphos-methyl; chafer grub; *Holotrichia parallela*, *Holotrichia oblita*, *Holotrichia trichophora*, *Anomala corpulenta*, *Arachis hypogaea*

## Introduction

Peanut is a main oil crop in China. White grubs (Scarabaeidae) are a serious pest in peanut fields. They generally reduce peanut yield per year by 25 to 35% (Jiang, 2008). Chemical pesticides were used conventionally to control grubs. However, they were only moderately effective, induced grub resistance to chemical residues and gradually led to environment pollution.

Entomopathogenic nematodes (EPNs) of the genera *Steinernema* and *Heterorhabditis* are insect parasitic nematodes characterised by their symbiotic relationship to entomopathogenic bacteria of the genera *Xenorhabdus* and *Photorhabdus*, respectively. Because they have a broad host range and can be applied conveniently via irrigation systems or by spraying even combined with chemical and biological pesticides, EPNs have been used to control various soil living and concealed pests for many years (Glaser, *et al.* 1935; Gaugler, *et al.* 1992; Mart, 1995; Liu, *et al.* 1997; Georgis, *et al.* 1991; Liu, *et al.* 2002; Yadav, *et al.* 2004; Karunakar, *et al.* 2000; Sankaranarayanan, *et al.* 2006).

Only few papers have been published on the control of chafer grubs in peanut fields (Li, *et al.* 1993; Liu, *et al.* 2007). To evaluate the effect of EPNs, *Heterorhabditis indica* was

applied to control the chafer grub complex (*Holotrichia parallela* Motschulsky, *H. oblita* Faldermann, *H. trichophora* Fairmaire and *Anomala corpulenta* Motschulsky) in peanut fields at various dosages.

## Material and methods

### *Trial materials*

*H. indica* was produced in Germany, by *e-nema* GmbH; 40% isofenphos-methyl EC (emulsifiable concentrate) was produced in China, by Luxi-yinong Chemical Co., Ltd..

### *Trial methods*

The field trial was conducted in a peanut field seriously infected with grubs in Henan province of China. Peanuts (strain Yuhua 15) were planted on May 11, 2008 in the density of 10000 plants/667m<sup>2</sup>. The plants are planted singly in pits. *Heterorhabditis indica* were applied at dosages of 2000, 3000, 4000, 5000 and 10000IJs/pit and 40% isofenphos-methyl EC at 400ml/667m<sup>2</sup> was used as a chemical pesticide control (CK1). Tap water was applied as a negative control (CK2). They were applied on July 19, 2008, after peanut bloom period, with flooding irrigation.

Each treatment or control was conducted on three plots. One plot (60 m<sup>2</sup>) had 900pits (peanut plants). The data were collected two days before harvest (22-23th, September, 2008). In each plot, 15x5 plants from 5 points (1 m<sup>2</sup>/point) were collected on a **Zigzag** line in each plot. The peanut legumes and the rhizosphere soil were dug out. The number of living grubs, injured legumes and fresh peanut legume weight were recorded per point and then calculated per 5 points. Further 15 points on the decrease rate of grub, the rate of injured legume, the weight of dry legume (0.7 times of fresh legume weight), and the increasing weight rate of dry legume of each plot. The calculation followed the formula of Liu *et al* (2007).

This paragraph is hard to understand. Let me see, if I understood: You sampled 15 x 5 plants per plot and recorded the number of living grubs, the (number or percentage of ?) injured legumes and the fresh weight of the legumes per point (5 points per plot). Then you calculated further parameters from these values which is decrease rate (better reduction) of grubs the dry weight and the gain in legume dry weight for each plot. If I am right the text should be:

In each plot, 5 x 15 plants were collected from 5 patches of 1 m<sup>2</sup> along a Zigzag line transecting the plot. The peanut legumes and the soil around the roots (how much approximately ? surface area and depth of sample) were dug out and the number of living grubs, the number of damaged legumes and the fresh-weight of the peanuts per plant was recorded. From these values, the reduction in grubs numbers, the percentage of legumes damaged and the increase in legume dry-weight was calculated.

## Results and discussion

### *Effect of different dosages of H. indica on legume damage*

The porportion of damaged legumes (IL) was significantly lower in all treatments compared to the untreated control (Fig.1). The dosages of 4000, 5000 and 10000IJs/pit resulted in 4.1%, 5.7% and 7.5% IL, respectively. Dosages of 4000 and 5000 IJs/pit were most effective resulting in significantly lower damage than in plots treated with 40% isofenphos-methyl control (10.2%). Among the five dosages of *H. indica*, 4000IJs/pit was the best one. Considering grub control effect and economic aspects, the doses of 2000 and 10000IJs/pit are least promising.

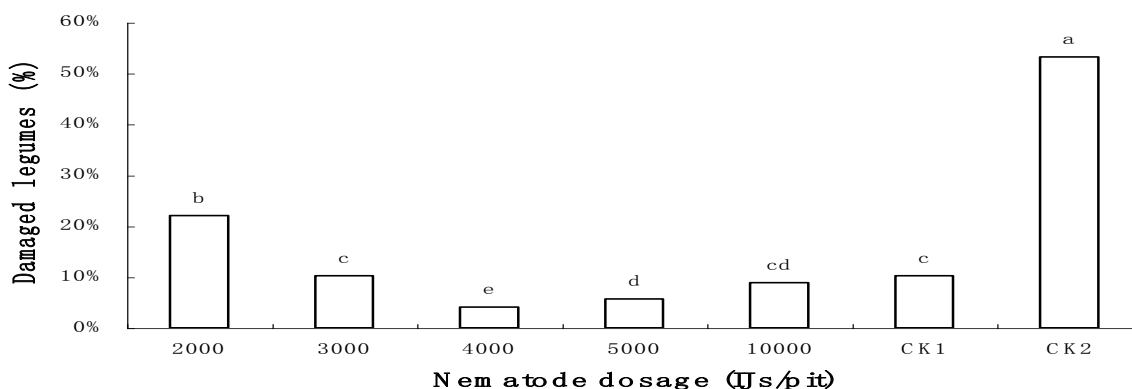


Figure 1. Proportion of damaged legumes in peanuts treated with different dosages of *Heterorhabditis indica* and 40% Isufenphos-methyl (CK1)

**Effect of different dosages of *H. indica* on grub numbers**

The average amount and reduction of grubs in all treatments were significantly different to the untreated tap water control. On average the number of grubs were reduced by 95.7% (Fig. 3) to 0.33 individuals/m<sup>2</sup> (Fig.2) the reduction the best performing dosages of 4000 and 5000 IJs/pit. In the plots treated with 40% isofenphos-methyl EC grub reduction was 82.7%.

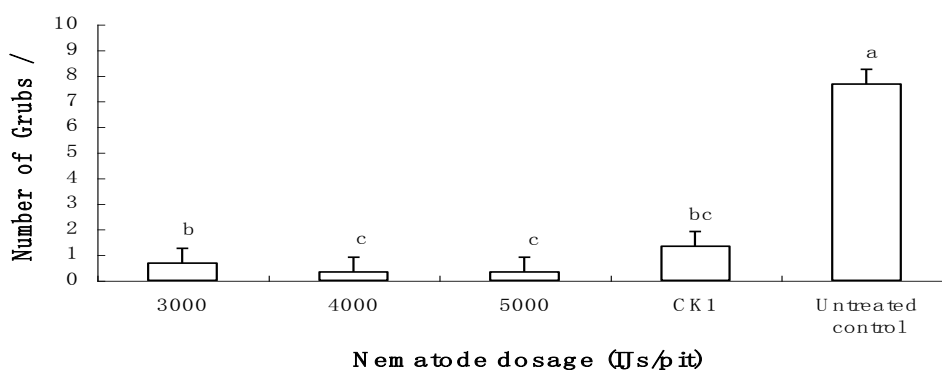


Figure 2. Number of grubs after treatment with different dosages of *Heterorhabditis indica* and 40% isofenphos-methyl EC (CK1).

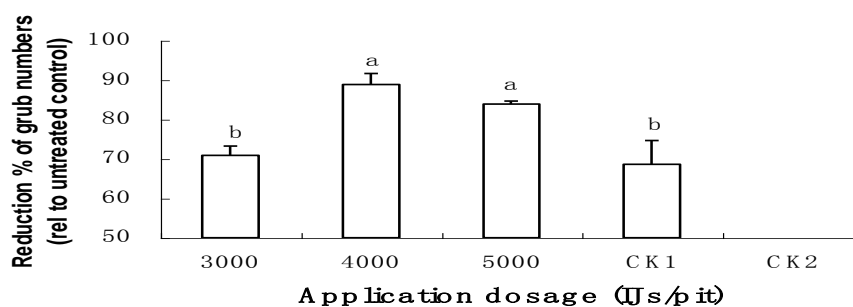


Figure 3. The reduction in grub numbers after treatment with different dosages of *Heterorhabditis indica* and 40% isofenphos-methyl EC (CK1).

### **Effect of different dosages of *H. indica* on peanut yield**

The peanut yields (Fig. 4) was 327.6 and 319.0 kg/667m<sup>2</sup>, respectively at the dosages of 4000 and 5000 IJs/pit. Both were significantly higher than the yield in the plots treated with 40% isofenphos-methyl control (292.6kg/667m<sup>2</sup>) and in the untreated tap water control (173.5kg/667m<sup>2</sup>) (p<0.05).

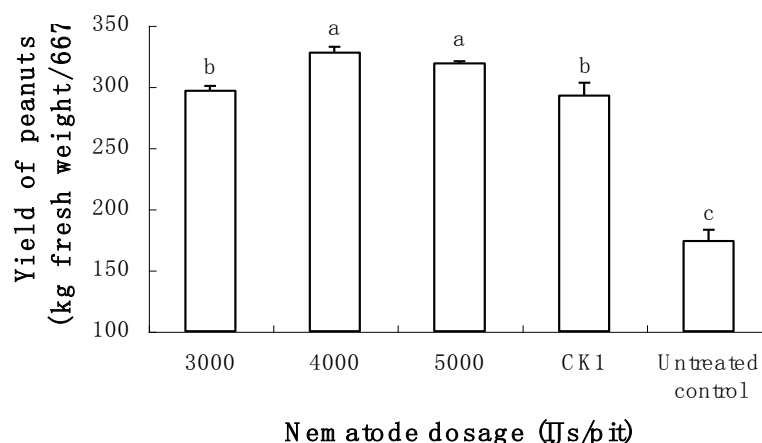


Figure 4. Peanut yield after treatment with different dosages of *Heterorhabditis indica* and 40% isofenphos-methyl EC (CK1).

### **Acknowledgements**

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### **References**

- Gaugler, R., Bednarek, A., Campbell, J. F. 1992: Ultraviolet inactivation of Heterorhabditid and Steinernematid nematodes. J. Invertebr. Pathol. 59: 155-160.
- Georgis, R. & Gaugler, R. 1991: Predictability in biological control using entomopathogenic nematodes. J. Econ. Entomol. 84: 713-720.
- Glaser, R.W. & Farrell, C.C. 1935: Field experiments with the Japanese beetle and its nematode parasite. J. New. York. Entomol. S. 43: 345-371.
- Jiang, X.L. 2008: Occurrence and integrated control of grub species in peanut fields. Bulletin of Agricultural Science and Technology. 5: 124-125.
- Karunakar, G.; Easwaramoorthy, S.; David, H. 2000: Pathogenicity of Steinernematid and Heterorhabditid nematodes to whitegrubs infesting sugarcane in India. Int. J. Nematol. 10(1): 19-26.
- Li, S.C., Ding, W.D., Han, F.S. 1993: The application of entomophagous nematode *Taishan* No.1 against peanut grubs. J. Plant. Protect. 20 (1): 55-60.
- Liu, Q.Z., Wang, Y.Z., Zhou, H.Y., et al. 1997: A study on the application of entomopathogenic nematodes for controlling larvae of red longicorn beetle. Acta. Agric. Boreali-Sinica. 12 (1): 97-101.
- Liu, Q.Z., Zhao, Y.X., Yan, Y.H., et al. 2002: Progress of applied research on entomopathogenic nematodes in biological control in China. J. China Agri. Univ. 7 (5): 65-69.

- Liu, Q.Z., Li, J.X., Xu, X.J., *et al.* 2007: The preliminary study on grub control with *Rhabditis (Oscheius) spp.* in peanut fields. *Acta. Agric. Boreali-Sinica.* 22 (suppl.): 250-253.
- Mart, G.C. 1995: Viewpoint entomopathogenic nematodes for the biological control of insects. *J. Nematol.* 27 (4): 529-534.
- Sankaranarayanan, C., Somasehar, N., Singaravelu, B. 2006: Biocontrol potential of entomopathogenic nematodes *Heterorhabditis* and *Steinernema* against pupae and adults of white grub *Holotrichia serrata*. *Sugar. Tech.* 8(4):268-271
- Yadav, B. R.; Veer, Singh; Yadava, C. P. S. 2004: Application of entomogenous nematode, *Heterorhabditis bacteriophora* and fungi, *Metarhizium anisopliae* and *Beauveria bassiana* for the control of *Holotrichia consanguinea* by soil inoculation method. *Ann. Agri. Bio. Res.* 9(1): 67-69.