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BIOEFFICACY OF *METARHIZIUM ANISOPLIAE* WITH NEEM DERIVATIVES AGAINST *HELICOVERPA ARMIGERA* INFESTING CHICKPEA

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ABSTRACT
The field experiments were carried out during the year 2013-2014 at the experimental field of JNKVV, Jabalpur to evaluate the efficacy of *M. anisopliae* and neem derivatives alone and in combination against *Helicoverpa armigera* infesting chickpea. All the neem derivatives and microbials proved their superiority over the control in reducing the pod damage and increasing the grain yield. On the basis of effectiveness of different treatments pod damage by pod borer and grain yield, revealed that *Metarhizium anisopliae* + Neem soap @ 1 x 10^4 spores/ml +0.5% (w/v), was the most effective treatment. On the basis of cost benefit ratio the same combination gave maximum cost benefit ratio followed by *Metarhizium anisopliae* @ 1 x 10^8 spores/ml (1:8.39 and 1:5.40, respectively).

INTRODUCTION

Production of chickpea in our country is low and one of the major reasons is the losses caused by several pests and diseases, both in field and in storage. It is attacked by number of insect pests, among them, the gram pod borer *Helicoverpa armigera* Hubner (Lepidoptera: Noctuidae) is the most devastating insect pest inflicting serious damage to the crop (Sithanantham *et al*. 1984).

H. armigera is a polyphagous nature, the gram pod borer is also known as American cotton boll worm, corn ear worm, tomato fruit borer, tobacco bud worm, carnation worm, *etc*. It has been recorded feeding on 182 cultivated and uncultivated plant species belonging to 47 families and also seriously damages to 56 plant species (Pawar *et al*. 1986; Regupathy *et al*.1997). Hence, farmers are compelled to use chemical insecticides in order to cultivate lucratively, as traditional and cultural practices alone cannot not give satisfactory control over the pest menace (Vanlaldiki *et al*. 2013). This has necessitated the use of alternative eco-friendly insecticides to sustain the management of *Helicoverpa armigera*.

The growing awareness of pesticidal hazards on human being and environment is the real fact for the development of sustainable eco-friendly pest management program such as "Biological control". Biopesticides are inherently less harmful than conventional pesticides. Biopesticides are clearly and mostly target specific in contrast to broad spectrum conventional chemical pesticides that kill almost all living organism. Biopesticides are effective in very small quantities and are often quickly biodegradable. (Bhushan *et al*. 2012) Biological control includes use of bioagents and microbial derived from fungi, bacteria and virus to control disease and insect pests. The most considerable fungal species are *Metarhizium* spp., *Beauveria* spp., *Nomuraea rileyi, Verticillium lecanii* and *Hirsutella* spp (Alves and Lopes 2008). *Metarhizium* causes a disease known as 'green muscardine' in insect hosts because of the green colour of its conidial cells. With the current thrust on Sustainable agriculture and organic farming the use of *M. anisopliae* and neem derivatives have been reported efficacious against pod borer however, information are scanty and combined effect of *Metarhizium anisopliae* with neem derivatives not known(Murray *et al*. 2000; Kavitha *et al*. 2009). Keeping the fact in background the present investigation are undertaken under field condition on bioefficacy of *Metarhizium anisopliae* with neem derivatives against *Helicoverpa armigera* infesting chickpea.

MATERIALS AND METHODS

Field experiment was conducted at the experimental field of Department of Entomology, Adhartal, JNKVV, Jabalpur (M.P.) during *Rabi* 2013-14, under randomized block design. Chickpea variety JG-12 was sown on 12th December, 2013 in a plot size of 10 x 3.3 m^2 with a spacing of 30 x 10 cm. Other agronomic practices were followed as per local recommendation. There were eight treatments comprising of

Metarhizium anisopliae (1x10^8 spores/ml) and neem derivatives (Neem oil, NSKE, Neem soap) alone and combination of *M. anisopliae* with neem derivatives including control (Table 1). Neem seed kernel and crude neem oil were purchased from local market. Neem soap was purchased from Indian Institute of Horticulture Research, Hessarghatta Lake Post, Bengaluru. Preparation of NSKE was followed as per standard method (PURI *et al.,* 1998) i.e. 5 kg of dried neem seeds were taken and cleaned it thoroughly. A day before schedule spraying, it was powdered by grinding and soaked overnight in 10 litres of distilled water and stirred with wooden plank till solution became milky white. It was then filtered through double layer of muslin cloth and made its volume to 100 litre by adding distilled water. To it 200 ml of sticker was added and the resultant solution was sprayed to cover upper as well as lower foliar portions of the crop. Treatment wise application of biochemical's were made at pod formation stage (90 DAS) of crop at ETL (*i.e.* e 2 larvae/mrl) level of *H. armigera*. Observation were recorded on *H. armigera* larvae on randomly selected 10 sites (1 meter row length/site) on 3,7,10 days after application (DAA). Larval count was also taken at 24 hrs before initiation of treatments. Pods of ten plants per plot were counted and per cent pod damage were calculated based on healthy and bored pods. Grain yield/plot was also assessed after harvesting and C:B ratio was calculated based on cost of application , market price of biopesticides and chickpea.

All the data were subjected to statistical analysis after appropriate transformation as suggested by Snedecor GW and Cochran WG, 1967.

RESULTS AND DISCUSSION

Effect of biopesticides on larval population of *H. armigera*

Pre- treatment

Differences in the *Helicoverpa armigera* mean larval population per meter row length (mrl) among different plots were not significant, indicating more or less uniform distribution of the pest but it was above the ETL (2.40 L/mrl) in the experimental field.

Three days after application

The mean *Helicoverpa armigera* larval population per mrl among different treatments ranged from 2.57 to 3.23 but without any significant difference indicating no immediate effect of treatments on larval population.

Seven days after application

As revealed from the Table 1. at seven days after treatment, all the treatments significantly reduced the larval population as compared to control (2.80 larvae / mrl). Among the treatments, *M. anisopliae* + Neem soap @ 1 × 10^4 spores/ml + 0.5% (w/v) was found to be the most effective as it recorded lowest larval population (0.80 larvae/mrl) this was followed by Neem seed kernel extract @ 5% (w/v) (0.87 larvae/mrl), but they did not differ significantly from each other. The next effective treatment was *M. anisopliae* + Neem oil @ 1 × 10^4 spores/ml + 2.5 % (v/v) (0.97 larvae/mrl), followed by *M. anisopliae* @ 1 × 10^8 spores/ml (1.07 larvae/mrl), *M. anisopliae* + NSKE @ 1 × 10^4 spores/ml + 2.5 % (w/v) (1.10 larvae/mrl), Neem soap @ 1 % (w/v)

Table 1: Bioefficacy of *Metarhizium anisopliae* and neem derivatives alone and their combinations on *Helicoverpa armigera* and chickpea grain yield

	Treatments	Dose	Mean *H. armigera* larval population / mrl*				Pod damage (%)**	Grain yield (kg/ha)
			Pre treatment	Days after spraying 3	7	10	Mean		
1	*Metarhizium anisopliae*	1l / ha (1 × 10^{8}spores/ml)min.	2.40(1.55)	2.50(1.58)	1.07(1.03)	0.94(0.97)	1.50(1.23)	12.00(20.23)	762.96
2	Neem seed kernel extract (NSKE)	5% (w/v)	2.27(1.50)	2.33(1.53)	0.87(0.93)	0.60(0.77)	1.27(1.13)	7.67(16.03)	842.59
3	Neem oil	5% (v/v)	2.53(1.59)	2.63(1.62)	1.17(1.08)	1.10(1.05)	1.63(1.28)	22.86(28.54)	556.48
4	Neem soap	1% (w/v)	2.50(1.58)	2.57(1.60)	1.13(1.06)	1.03(1.01)	1.58(1.26)	19.84(26.41)	568.52
5	*M. anisopliae* + NSKE	1× 10^4 spores /ml + 2.5% (w/v)	2.47(1.57)	2.53(1.59)	1.10(1.05)	0.97(0.98)	1.53(1.24)	16.83(24.20)	703.70
6	*M. anisopliae* + Neem oil	1× 10^4 spores /ml + 2.5% (v/v)	2.37(1.54)	2.43(1.56)	0.97(0.98)	0.67(0.81)	1.36(1.16)	9.32(17.72)	812.04
7	*M. anisopliae* + Neem soap	1×10^4 spores /ml + 0.5 % (w/v)	2.23 (1.49)	2.27 (1.51)	0.80 (0.89)	0.57 (0.74)	1.21 (1.10)	6.70 (14.96)	890.74
8	Control		2.57 (1.60)	2.73 (1.65)	2.80 (1.67)	2.87 (1.69)	2.80 (1.67)	25.95 (30.61)	523.15
		SEm ±	0.02	0.03	0.03	0.05	0.02	0.41	8.33
		CD at 5%	NS	NS	0.09	0.16	0.05	1.25	25.28

mrl = meter row length; * Figures in parentheses are ($\sqrt{x+0.5}$) square root transformed values; ** Figures in parentheses are arcsin transformed values; NS = Non significant

(1.13 larvae/mrl) and Neem oil @ 5% (v/v) (1.17 larvae/mrl) (T_3) but they did not differ significantly from each other.

Ten days after application

At ten days after treatment, All the treatments significantly reduced the larval population as compared to control (2.87 larvae / mrl). Among the treatments, *M. anisopliae* + Neem soap @ 1×10^4 spores/ml + 0.5% (w/v) was found to be most effective as it recorded lowest larval population (0.57 larvae/mrl), this was followed by Neem seed kernel extract @ 5% (w/v) (0.60 larvae/mrl) and *M. anisopliae* + Neem oil @ 1×10^4 spores/ml + 2.5 % (v/v) (0.67 larvae/mrl), but all the three treatments were at par with each other. The next effective treatment was *M. anisopliae* @ 1×10^8 spores/ml (0.94 larvae/mrl), followed by *M. anisopliae* + NSKE @ 1×10^4 spores/ml + 2.5 % (w/v) (0.97 larvae/mrl), Neem soap @ 1 % (v/v) (1.03 larvae/mrl), Neem oil @ 5% (v/v) (1.10 larvae/mrl), but they did not differ significantly from each other .

Overall mean

On the basis of overall mean, the differences in the mean larval population among different treatments were significant. All the treatments significantly reduced the larval population as compared to control (2.80 larvae / mrl). Among the treatments, *M. anisopliae* + Neem soap @ 1×10^4 spores/ml + 0.5% (w/v) was found to be most effective as it recorded lowest larval population (1.21 larvae/mrl). The next effective treatment was Neem seed kernel extract @ 5% (w/v) (1.27 larvae/mrl), followed by *M. anisopliae* + Neem oil @ 1×10^4 spores/ml + 2.5 % (v/v) (1.36 larvae/mrl) but they differ significantly from each other. The next effective treatment was *M. anisopliae* @ 1×10^8 spores/ml (1.50 larvae/mrl), followed by *M. anisopliae* + NSKE @ 1×10^4 spores/ml + 2.5 % (w/v) (1.53 larvae/mrl), but both of them were at par with each other. The least effective treatments were Neem soap @ 1 % (v/v) (1.58 larvae/mrl) and Neem oil @ 5% (v/v) (1.63 larvae/mrl) and were at par with each other.

Effect of biopesticides on pod damage and grain yield of chickpea

Chickpea Pod damage

Based on per cent infestation at harvest, chickpea pod damage caused by *H. armigera* was in the range of 6.70% to 22.86% in the biopesticides treatments as against 25.95% in untreated control. The lowest pod damage (6.70%) was due to combination treatment of *M. anisopliae* + Neem soap @ 1×10^4 spores/ml + 0.5% (w/v) this was followed by Neem seed kernel extract @ 5% (w/v) recording (7.67%) pod damage but both were at par with each other. The treatment of *M. anisopliae* combined with Neem oil with their half doses also recorded lower (9.32%) pod damage. The higher pod damage in the range of 12 to 19.84% were recorded in *M. anisopliae* @ 1×10^8 spores/ml, *M. anisopliae* + NSKE @ 1×10^4 spores/ml + 2.5 % (w/v), Neem soap @ 1 % (w/v). Application of Neem oil @ 5% (v/v) was least effective with high pod damage of 22.86%.

Chickpea grain yield

The yield of net plot area of each treatment was recorded and converted into kg/ha. All the treatments registered significantly higher grain yields as compared to the control (523.15 kg/ha). The highest grain yield was recorded in *M. anisopliae* + Neem soap @ 1×10^4 spores/ml + 0.5% (w/v) treated plots (890.74 kg/ha) which was significantly superior than the other treatments. Subsequent higher grain yield was recorded with Neem seed kernel extract @ 5% (w/v) (842.59 kg/ha) followed by *M. anisopliae* + Neem oil @ 1×10^4 spores/ml + 2.5% (v/v) (812.04 kg/ha), but they differed significantly from each other. The next effective treatment were *M. anisopliae* @ 1×10^8 spores/ml (762.96 kg/ha), followed by *M. anisopliae* + NSKE @ 1×10^4 spores/ml + 2.5 % (w/v) (703.70 kg/ha), but they differed significantly from each other. The least effective treatments were Neem soap @ 1% (w/v) (568.52 kg/ha) and Neem oil @ 5% (v/v) (556.48 kg/ha) and they were at par with each other.

All the neem derivatives and *M. anisopliae* alone and their combination proved their superiority over control in reducing the pest population and pod damage and in increasing the grain yield. Several workers have also reported similar findings, that application of neem derivatives and *M. anisopliae* effectively reduced the damage due to pod borer with increased grain yield than control (Katole *et al.,* 2000; Kumar and Chowdhary 2004; Nahar *et al.* 2004; Kulkarni *et al.,* 2005; Singh and Yadav 2005; Gundannavar *et al.* 2007; Haque and Ghosh 2007; Ali *et al.,* 2008; Kale and Men 2008; Rijal *et al.,* 2008; 2008a; Wakil *et al.* 2008; Bhushan *et al.,* 2011; Moorthy *et al.,* 2011; Rao *et al.* 2011; Wakil and Ghaznafar 2011; Singh *et al.,* 2012; Ahmad *et al.,* 2014 and Chandel *et al.,* 2014).

REFERENCES

Ahmad, Salman., Ansari, Mohmmad Shafiq. and Hussain, Mazhar. 2014. Effect of insecticides on management of *Helicoverpa armigera* in chickpea agro ecosystem. *Annals of Plant Protection Science.* **22(1):** 107-111.

Ali, Ghulam Abbas., Saleem, Akhtar., Zafar, Abbas. and Muhammad, Aslam. 2008. Efficacy of Neem (*Azadirachta indica A.* Juss) products against *Helicoverpa armigera* (Hubner) on chickpea. *Science International (Lahore).* **20(4):** 281-283.

Alves, S. B. and Lopes, R. B. 2008. Controle microbiano de pragas na América Latina. *avanços e desafi os. Piracicaba.* FEALQ. p. 414.

Bhushan, S., Singh, R. P. and Shanker, R. 2011. Bioefficacy of neem and Bt against pod borer, *Helicoverpa armigera* in chickpea. *J. Biopesticides.* **4(1):** 87-89.

Bhushan, S., Singh, R. P. and Shanker, R. 2012. Biopesticidal management of yellow stem borer(*Scirpophaga incertulas* Walker) in rice. *The Bioscan.* **7(2):** 317-319.

Chandel, Rajeev., Lyall, Hemant. and Bhamba, D. R. 2014. Efficacy of insecticides and neem products against *Helicoverpa armigera* on chickpea. *Ann. Pl. Protection Science.* **22(1):** 205-206.

Gundannavar, K. P., Lingappa, S. and Giraddi, R. S. 2007. Study of dose mortality response between instars of *Helicoverpa armigera* (Hubner) and *Metarhizium anisopliae* (Metschinkoff) Sorokin. Karnataka *J. Agricultural Sciences.* **20(1):** 140-141.

Haque, J. and Ghosh, A. B. 2007. Effect of *Beauveria bassiana* (Bals.) Vuill, *Metarhizium anisopliae* (Metsch.) Sorokin and Nimbecidine on some insect pests. *Environment and Ecology.* **25(1):** 209-211.

Kale, S. N. and Men, U. B. 2008. Efficacy of microbial insecticides and their combinations against *Helicoverpa armigera* (Hubner) on chickpea. *J. Biological Control.* **22(1):** 205-208.

Katole, S. R., Nimbalkar, S. A., Kolhe, A. V., Ghuguskar, H. T. and Yadgirwar, P. V. 2000. Performance of some IPM modules against *Helicoverpa armigera* on chickpea. *PKV Research J.* **24(1):** 51-53.

Kavitha, E., Kingsley, S., Revathi, N. and Sathivel, M. 2009. Insecticidal activity of neem derivatives against okra fruit borer *Helicoverpa armigera* Hubner. *International J. Agricultural Sciences.* **5(2):** 528-530.

Kulkarni, K. A., Kambrekar, D. N. and Gundannavar, K. P. 2005. Management of *Helicoverpa armigera* Hubner on chickpea through biopesticides. *Karnataka J. Agricultural Sciences.* **18(4):**1114-1116.

Kumar, V. and Chowdhry, P. N. 2004. Virulence of entomopathogenic fungi *Beauveria bassiana* and *Metarhizium anisopliae* against tomato fruit borer, *Helicoverpa armigera*. *Indian Phytopathology.* **57(2):** 208-212.

Moorthy, Dakhsana., Anandhi, P., Elamathi, S. and Simon, Sobita. 2011. Evaluation of bio- rational insecticides for management of *Helicoverpa armigera* in chickpea. Annals of Plant Protection Science. **19(1):** 207-209.

Murray, D. A. H., Lioyd, R. and Buddington, J. 2000. Potential in Australia for a *Helicoverpa baculovirus*. Abstract. International Congress of Entomology, 21- 25 Aug 2000, Igassu Falls, Brazil.

Nahar, P., Yadav, P., Kulye, M., Hadapad A., Hassani, M., Tuor, U., Keller, S., Chandele, A. G., Thomas, B. and Deshpande, M. V. 2004. Evaluation of indigenous fungal isolates, *Metarhizium anisopliae* M34412, *Beauveria bassiana* B3301 and *Nomuraea rileyi* N812 for the control of *Helicoverpa armigera* (Hubner) in pigeonpea field. *J. Biological Control.* **18(1):** 1-7.

Pawar, C. S., Bhatnagar, V. S. and Jadhav, D. R. 1986. Heliothis species and their natural enemies with their potential for biological control. In Proceed. *Indian Acad. Sci. (Animal Sciences).* **95:** 697-703.

Puri, S. N., Sharma, O. P., Murthy, K. S. and Rajshe, O. 1998. Preapration of neem seed kernel extract. " Handbook on diagnosis and integrated management of cotton pests." *NCIPM* New Delhi, p. 106.

Rao, G. V., Rao, V. R., Gottumukkala, S. J., Vidya, M. S., Srinivas, V. and Linga, O. R. 2011. Efficacy of botanical extracts and entomopathogens on control of *Helicoverpa armigera* and *Spodoptera litura*. *African J. Biotechnology.* **10(73):** 16667-16673.

Regupathy, A., Armes, N. J., Asokan, G., Jadhav, D. R., Soundarajan, R. P. and Russell, D. A. 1997. Best method for insecticide resistance management of *Helicoverpa armigera*. In:International Conf. Integrated Approach to Combating Resistance. *A.L. Devonshire (ed.), IACR, Rothamsted, Harpenden, UK.* p. 116.

Rijal, J. P., Yubak Dhoj, G. C., Thapa, R. B. and Kafle, L. 2008. Efficacy of *Metarhizium anisopliae* and *Beauveria bassiana* against *Helicoverpa armigera* in chickpea under field condition in Nepal. *Formosan Entomology.* **28:** 249-258.

Rijal, J. P., Yubak Dhoj, G. C., Thapa, R. B. and Kafle, L. 2008 a. Virulence of native isolates of *Metarhizium anisopliae* and *Beauveria bassiana* against *Helicoverpa armigera* in in chickpea under field condition in Nepal. *Formosan Entomology.* **28:** 21-29.

Singh, B. and Yadav, R. P. 2005. Field efficacy of some microbial agents against *Helicoverpa armigera* Hub. on chickpea. *J. Applied Zoological Researches.* **16(1):** 5-6.

Singh Rajendra, Singh Sarika, Apalwal Meena and Anandhi, P. 2012. Management *Helicoverpa armigera* on chickpea through biopesticides. *Annals of Plant Protection Sciences.* **20(1):** 215-216.

Sithanantham, S., Rao, V. and Ghalar, M. A. 1984. International review of crop losses caused by insects on chickpea. In: Proceed. National Semi. *Crop losses due to Insect Pests.* pp. 269-284.

Snedecor, G. W. and Cochran, W. G.1967. Statistical Methods, Oxford and IBH Publishing Company, New Delhi. pp. 1-292.

Vanladiki, H., Singh, Premjit, M. and Sarkar, P. K. 2013. Efficacy of ecofriendly insecticides on the management of diamondback moth (*Plutella xylostella*) Linn. On cabbage. *The Bioscan.* **8(4):**1225-1230.

Wakil, Waqas., Muhammad, Ashfaq., Ghazanfar, M. U., Saleem, Akhtar. and Malhi, Z. A. 2008. Management of *Helicoverpa armigera* on chickpea. *Pakistan Entomologist.* **30(1):** 51-54.

Wakil, W. and Ghazanfar, M. U. 2011. Effects of *Metarhizium anisopliae*, *Bacillus thuringiensis* and new chemistry insecticide on the mortality and pupation of field populations of *Helicoverpa armigera* Hubner (Lepidoptera:Noctuidae). *IOBC/WPRS Bulletin.* **66:** 105-108.