



## Research Article

# Management of white grub, *Holotrichia consanguinea* (Blanchard) using biocontrol agents in sugarcane in coastal Andhra Pradesh

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**ABSTRACT:** White grubs are one of the major pests of sugarcane crop in coastal Andhra Pradesh, India. The present study was taken up to assess the efficacy of biocontrol agents viz., entomopathogenic fungi and entomopathogenic nematodes for management of white grub, *Holotrichia consanguinea* in sugarcane ecosystem during 2015-16 and 2016-17. Two entomopathogenic fungi, *Metarhizium anisopliae* (NBAIR Ma4 strain) and *Beauveria bassiana* (NBAIR Bb5a), two entomopathogenic nematodes (*Heterorhabditis indica* NBAIR-H38 and *Steinernema carpocapsae* NBII Sc 05), Neem cake, Phorate 10G were evaluated through soil application as different treatments in white grub infested sugarcane field in coastal Andhra Pradesh for two seasons during 2015 and 2016-17. All biocontrol agent treatments were better than the insecticidal/neem cake application in the management of sugarcane white grubs. However, based on cost benefit ratio of the treatments, soil application of *M. anisopliae* @ 2.5 kg along with 250 kg farm yard manure per hectare (twice application in July and August) proved superior to other biocontrol agent treatments. Soil application of biocontrol agents, Phorate and Neem cake did not show any deleterious effect on non-target organisms like, earthworms, earwigs, spiders, coccinellids and chrysopids.

**KEY WORDS:** Biocontrol agents, *Holotrichia consanguinea*, sugarcane, white grubs

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## INTRODUCTION

Sugarcane is an important cash crop grown in India and India is second largest producer of sugarcane in the world. In Andhra Pradesh, sugarcane is grown in 2.40 lakh hectares and 136 lakh tons of sugarcane is produced in the state. It is largely grown in Visakhapatnam, West Godavari, East Godavari, Chittoor, Krishna, Vizayanagaram, Srikakulam and Nellore Districts with 90 per cent of the area under this crop (Kumar and Suneetha, 2016). White grub, *Holotrichia consanguinea*, Blanchard (Coleoptera: Scarabaeidae) is an important soil pest in sugarcane crop in India. It causes severe damage to commercial crops in India. In recent years, white grubs extending its spatial range due to monoculture of sugarcane and minimal varietal diversity. It has affected the sugarcane crop in certain parts of Andhra Pradesh. The adult beetles feed on foliage of host trees like neem, acacia etc., late larval stages feed on roots of sugarcane and underground portions of stalks. White grub infestation on roots results in yellowing of leaves, wilting and drying of clumps. Infested canes lose weight, dry up, collapse and become unfit for crushing. White grub damage generally occurs in patches leaving gaps and severe infestation results in devastation

of entire field. Severe symptoms of damage by older grubs generally observed late in the season resulting in complete loss of crop as curative chemical control is ineffective against late stage grubs. The yield loss due to white grubs was reported to be as high as 100 per cent in Tamil Nadu (Selvi *et al.*, 2010). Several practices were adopted for the management of white grubs including cultural, mechanical, biological, chemical and integrated methods (Sahayaraj and Borgio, 2009; Srikanth and Singaravelu, 2011). Pest management strategy depends primarily on the use of highly poisonous poor graded chemical pesticides. Chemical control is practically uneconomical, difficult and associated with high cost, environmental pollution and pesticide residues. Hence, there is need for effective ecofriendly and economically feasible strategy for the control of white grubs in sugarcane. Entomofungal Pathogens (EPF) like *Beauveria bassiana*, *Metarhizium anisopliae*, Entomopathogenic Nematodes (EPN) like, *Heterorhabditis* sp. and *Steinernema* sp. were reported to be cost effective and eco-friendly management of white grubs pests in various crops. These biocontrol agents are also self-perpetuating in nature. Hence, studies on field evaluation of EPF and EPN were carried out for the

management of white grubs in sugarcane in an endemic area of Andhra Pradesh.

## MATERIALS AND METHODS

### Field evaluation

The field trials were conducted during 2015-16 & 2016-17 against white grubs in sugarcane field (variety, 2003V46) at RARS, Anakapalli, Andhra Pradesh, India (Latitude-17.6896° N and Longitude-83.0024° E). The field trials were laid out with nine treatments as given below in Randomized Block Design (RDB) in 5 ha area. Each treatment had three replications distributed randomly in 0.555 hectare area. First year sugarcane was ratoon crop which was started in January 2015 and was harvested in December 2015. Second year crop was fresh planting sown in March 2016 and was harvested in January 2017. All the agronomic practices with recommended dose of fertilizers were followed till the crop harvest to maintain good plant health as per the package of practices of sugarcane crop (Anon 2018).

### Treatments : 9

T1: *Metarhizium anisopliae* (NBAIR Ma4) 2.0% W.P. talc formulation @ 2.5 kg ha<sup>-1</sup> 1 x10<sup>8</sup> Cfu/g (minimum) in 250 kg FYM (2 times soil application, July and August)

T2: *Beauveria bassiana* (NBAIR Bb5a) 2.0% W.P. talc formulation @ 2.5 kg ha<sup>-1</sup> 1 x10<sup>8</sup> Cfu/g in 250 kg FYM (2 times soil application, July and August)

T3: *Heterorhabditis indica* (NBAIR H38) WP @ 20 kg ha<sup>-1</sup> in 150 kg moist sand ha<sup>-1</sup> (Single soil application in July)

T4: *Heterorhabditis indica* (NBAIR H38) WP @ 20 kg ha<sup>-1</sup> in 150 kg moist sand ha<sup>-1</sup> soil application two times at two month interval (July and September)

T5: *Steinernema carpocapsae* (NBAII Sc05) WP @ 20 kg ha<sup>-1</sup> in 150 kg moist sand ha<sup>-1</sup> (Single soil application in July)

T6: *Steinernema carpocapsae* (NBAII Sc05) WP @ 20 kg ha<sup>-1</sup> in 150 kg moist sand ha<sup>-1</sup> soil application two times at two month interval (July and September)

T7: Neem cake @ 500 kg ha<sup>-1</sup> (Single soil application in July)

T8: Phorate 10G @ 15 kg ha<sup>-1</sup> (Single soil application in July)

T9: Untreated control 250 kg of FYM without enrichment of fungus (2 times application, July and August)

### Preparation of talc formulation (2% WP) of *M. anisopliae* and *B. bassiana*

The fungi, *M. anisopliae* NBAIR-Ma4 and *B. bassiana* NBAIR Bb-5a were grown in 1 litre conical flask (containing 500 ml medium) of Sabouraud's Dextrose Yeast extract Broth (SDYB) (Dextrose 20 g, Mycological peptone 10 g, yeast extract 5 g in 1L of distilled water) in orbital shaker at 25 ± 2°C temperature, 150rpm for 8 days. The 8 days old culture broth was centrifuged at 5000rpm for 10mins, supernatant was discarded and the pellet was mixed in sterilized talcum powder at 2% (20grams of pellet in 1 kg talc) in sterilized tray under laminar air flow and dried to 8% moisture. The talc formulations of *M. anisopliae* NBAIR-Ma4 and *B. bassiana* NBAIR Bb-5a contained 1.8 x10<sup>8</sup> and 1.0 x10<sup>8</sup> cfu/g respectively. The formulations were stored in milky white polypropylene pouches for further use in field evaluation trials.

### Preparation of *M. anisopliae*/*B. bassiana* enriched farm-yard manure

2.5 kg of talc formulation (2%WP) of each fungus was mixed thoroughly with 250 kg FYM in shaded area in the field and incubated for 15 days with intermittent sprinkling of water. After 15 days, the fungus enriched Farmyard manure was used for soil application.

### Application of treatments

In T1 and T2 treatments, soil application of *M. anisopliae*/*B. bassiana* enriched Farmyard manure @ 250 kg/ha was done two times in the furrows adjoining the root zone of sugar cane clumps. The first application was in July and the second application was done in August after one month of first application in each year. In T3 and T5 treatments, *H. indica*/*S. carpocapsae* were applied to the soil @ 20 kg ha<sup>-1</sup> in 150 kg moist sand ha<sup>-1</sup> in July in each year. In T4 and T6 treatments, *H. indica*/*S. carpocapsae* were applied to the soil two times at two months intervals @ 20 kg ha<sup>-1</sup> in 150 kg moist sand ha<sup>-1</sup> in July and September in each year. In case of T7 treatment, soil application of Neem cake @ 500kg/ha and in T8 treatment, Phorate 10 G @ 15 kg/ha was done in July each year. In T9 treatment, soil application of Farmyard manure (without fungus enrichment) @ 250 kg/ha was done similarly two times in July and August in each year.

### Observations

#### Effect on plant damage, white grub population and crop yield

Observations on number of plants damaged by white grubs (%), number of white grubs per 10 meter row were recorded at monthly interval till the crop was harvested (July-

December 2015 during 2015 crop season & July 2016-January 2017 during 2016-17 crop season). Yield was recorded in December 2015 in the first year of trial and in January 2017 in second year trial and expressed as tons per hectare. Monthly data on plant damage, white grub population and cane yield were analyzed statistically for drawing inferences. The cost benefit ratio of the treatments has been calculated based on the formula given below.

BC Ratio = NR/CC; NR- Net Returns; CC- Cost of Cultivation

### Effect on non-target organisms

Non target organisms which were inhabiting soil *viz.*, earthworms and earwigs were recorded in a cubic meter area by removing the soil a day before treatment imposition and after 30 days of treatment. The predators like spiders, coccinellids and chrysopids per plant were recorded in each treatment a day before treatment imposition and after 30 days of treatment which were averaged and subjected to statistical analysis.

## RESULTS AND DISCUSSION

The experimental results on the efficacy of different treatments are presented in Table-1 (2015), Table-2 (2016-17) and Table-3 (Pooled analysed data for 2 years). The results on the effect of different treatments on Non-Target organisms are presented in Table-4 (2015) and Table-5 (2016-17)

### Plant damage due to white grubs (%)

During the first year field trial (2015), the per cent plants damaged by white grubs was lowest (1.47%) in *M. anisopliae* (Strain no. NBAIR Ma4) 2.0% W.P. Formulation @ 2.5kg ha<sup>-1</sup> (twice application) treated plots which was statistically at par with other treatments like, T4: *H. indica* (NBAIR H38) WP @ 20kg ha<sup>-1</sup> (twice application) with plant damage of 2.17%, T6 and T5: *S. carpocapsae* (NBAIL Sc05) WP @ 20kg ha<sup>-1</sup> (twice and single application) with damage of 2.79 and 3.16% and T2: *B. bassiana* (NBAIR-Bb5a) 2.0% W.P. Formulation @ 2.5kg ha<sup>-1</sup> (twice application) with 3.0% plant damage (Table 1). Other treatments like, Phorate 10G/Neem cake application/untreated control recorded significantly very high plant damage of 12.65, 25.0 and 48.0% (Table 1).

During the second year field trial (2016-17), the lowest per cent of plants damaged by white grubs was observed in T4 treatment (1.6%), *H. indica* (NBAIR H38) WP @ 20 kg ha<sup>-1</sup> (twice application) which was statistically at par with other treatments like, T3, *H. indica* (NBAIR H38) WP @ 20 kg ha<sup>-1</sup> (single application) with plant damage of 2.81%, T1, *M. anisopliae* (NBAIR Ma4) @ 2.5kg ha<sup>-1</sup> (twice application)

with plant damage of 4.04%, T6 and T5: *S. carpocapsae* (NBAIL Sc05) WP @ 20 kg ha<sup>-1</sup> (twice and single application) with plant damage of 4.58 and 4.12% and T2: *B. bassiana* (NBAIR Bb5a) @ 2.5kg ha<sup>-1</sup> (twice application) with 5.12% plant damage (Table 1). Other treatments like, Phorate 10G/Neem cake application/untreated control recorded significantly high plant damage of 9.17, 11.29 and 16.41% (Table 2).

The pooled data analysis of two years indicates that all biocontrol agent treatments (T1, T2, T3, T4, T5 and T6 treatments were at par with each other with regard to percent plant damage (2.75-4.06%), although T1 (*M. anisopliae*) and T4 (*H. indica* NBAIR H38) treatments showed lowest percent plant damage (2.275 and 2.99%) (Table 3).

### Incidence of white grub population

During 2015 trial, the white grub population was significantly low (0.67 grubs/ 10 m row) in *M. anisopliae* (Strain no. NBAIR Ma4) 2.0% W.P. Formulation @ 2.5kg ha<sup>-1</sup> (twice application) treated plots and superior to all other treatments (T2, T3, T4, T5, T6, T7 and T8) which showed white grub population in the range of 1.33 to 9.00 grubs/10 m row. Untreated control recorded significantly very high grub population 13.67 grubs/10 m row (Table 1).

In 2016-17 trial, the lowest white grub population was recorded in T4 treatment (1.46 grubs/10 m row), *H. indica* (NBAIR H38) (twice application) which was statistically at par with other treatments like, T3, *H. indica* (NBAIR H38) (single application) with 2.67 grubs/10m, T2, *M. anisopliae* treatment with 2.67 grubs/10m, T5: *S. carpocapsae* (NBAIL Sc05) (single application) with 3.62 grubs/10m and T2, *B. bassiana* with 4.08 grubs/10 m and T6 *S. carpocapsae* (NBAIL Sc05) (twice application) with 4.31 grubs/10m row (Table 1). Other treatments like, Phorate 10G/Neem cake application/untreated control recorded significantly high grub population of 9.12, 7.01 and 13.82 grubs/10m row (Table 2).

The pooled data analysis of two years indicates that all biocontrol agent treatments (T1, T2, T3, T4, T5 and T6 treatments were at par with each other with regard white grub population (1.40-2.99 grubs/10m row), although T4 (*H. indica* NBAIR H38) treatment showed lowest grub population of 1.4 grubs/10m row (Table-3).

### Cane yield

In the first year trial (2015), the cane yield was significantly high (87.41t/ha) in *M. anisopliae* (NBAIR-Ma4) 2.0% W.P. Formulation @ 2.5kg ha<sup>-1</sup> (twice application) treated plots and superior to all other treatments (T2, T3, T4, T5, T6, T7 and T8) which recorded cane yield in the range of

**Table 1. Efficacy of bioagents against white grubs in sugarcane crop during 2015-16**

Treatment	Per cent plants damaged by white grubs	Per cent reduction in plant damage by white grubs over control	White grubs/ 10m row	Per cent reduction in white grub population over control	Cane yield t/ha	Yield increase over control (%)	C:B ratio
T1: <i>Metarhizium anisopliae</i> (NBAIR Ma4) 2.0% W.P. @ 2.5 kg ha <sup>-1</sup> 1 x10 <sup>8</sup> Cfu/g (min.) in 250 kg FYM (2 times application)	1.47 <sup>a</sup>	96.94	0.67 <sup>a</sup>	95.10	87.41 <sup>a</sup>	71.48	2.0
T2: <i>Beauveria bassiana</i> (NBAIR Bb5a) 2.0% W.P. @ 2.5 kg ha <sup>-1</sup> 1 x10 <sup>8</sup> Cfu/g in 250 kg FYM (2 times application)	3.0 <sup>a</sup>	93.76	1.67 <sup>b</sup>	87.78	74.99 <sup>b</sup>	66.76	1.71
T3: <i>Heterorhabditis indica</i> (NBAIR H38) WP @ 20 kg ha <sup>-1</sup> in 150 kg moist sand ha <sup>-1</sup>	4.61 <sup>ab</sup>	90.40	2.67 <sup>c</sup>	80.47	70.77 <sup>c</sup>	64.77	1.49
T4: <i>Heterorhabditis indica</i> (NBAIR H38) WP @ 20 kg ha <sup>-1</sup> in 150 kg moist sand ha <sup>-1</sup> two times at two month interval	2.17 <sup>a</sup>	95.48	1.33 <sup>b</sup>	90.27	77.08 <sup>b</sup>	67.66	1.50
T5: <i>Steinernema carpocapsae</i> (NBAIL Sc05) WP @ 20 kg ha <sup>-1</sup> in 150 kg moist sand ha <sup>-1</sup>	3.16 <sup>a</sup>	93.42	1.67 <sup>b</sup>	87.78	70.09 <sup>c</sup>	64.43	1.48
T6: <i>Steinernema carpocapsae</i> (NBAIL Sc05) WP @ 20 kg ha <sup>-1</sup> in 150 kg moist sand ha <sup>-1</sup> two times at two months interval	2.79 <sup>a</sup>	94.19	1.67 <sup>b</sup>	87.78	73.42 <sup>bc</sup>	66.04	1.43
T7: Neem cake @ 500 kg ha <sup>-1</sup>	25.00 <sup>c</sup>	47.96	9.0 <sup>c</sup>	34.16	61.90 <sup>d</sup>	59.72	1.36
T8: Phorate 10G @ 15kg ha <sup>-1</sup>	12.65 <sup>b</sup>	73.67	6.33 <sup>d</sup>	53.69	61.59 <sup>d</sup>	59.52	1.30
T9: Untreated control only 250kg FYM	48.04 <sup>d</sup>		13.67 <sup>f</sup>	-	24.93 <sup>c</sup>		0.57
CD (P=0.05)	8.35		0.51		3.70		
CV%	21.23		14.13		2.42		

Values in column followed by different letter (a,b,c) are significantly different from each other

**Table 2. Efficacy of bioagents against white grubs in sugarcane crop during 2016-17**

Treatment	Per cent plants damaged by white grubs	Per cent reduction over control	White grubs/ 10m row	Per cent reduction in white grub population over control	Cane yield t/ha	Yield increase over control (%)	C:B ratio
T1: <i>Metarhizium anisopliae</i> (NBAIR Ma4) 2.0% W.P.@ 2.5 kg ha <sup>-1</sup> 1 x10 <sup>8</sup> Cfu/g (min.) in 250 kg FYM (2 times application)	4.04 <sup>ab</sup>	75.38	2.96 <sup>ab</sup>	78.58	97.73 <sup>a</sup>	51.79	2.03
T2: <i>Beauveria bassiana</i> (NBAIR Bb5a) 2.0% W.P. @ 2.5 kg ha <sup>-1</sup> 1 x10 <sup>8</sup> Cfu/g in 250 kg FYM (2 times application)	5.12 <sup>ab</sup>	68.8	4.08 <sup>ab</sup>	70.48	92.82 <sup>ab</sup>	49.23	1.93
T3: <i>Heterorhabditis indica</i> (NBAII H38) WP @ 20 kg ha <sup>-1</sup> in 150 kg moist sand ha <sup>-1</sup>	2.81 <sup>a</sup>	82.88	2.67 <sup>a</sup>	80.68	99.8 <sup>a</sup>	52.78	1.93
T4: <i>Heterorhabditis indica</i> (NBAII H38) WP @ 20 kg ha <sup>-1</sup> in 150 kg moist sand ha <sup>-1</sup> two times at two months interval	1.60 <sup>a</sup>	90.25	1.46 <sup>a</sup>	89.44	101.1 <sup>a</sup>	53.39	1.82
T5: <i>Steinernema carpocapsae</i> (NBAII Sc05) WP @ 20 kg ha <sup>-1</sup> in 150 kg moist sand ha <sup>-1</sup>	4.12 <sup>ab</sup>	74.89	3.62 <sup>ab</sup>	73.81	98.49 <sup>a</sup>	52.16	1.90
T6: <i>Steinernema carpocapsae</i> (NBAII Sc05) WP @ 20 kg ha <sup>-1</sup> in 150 kg moist sand ha <sup>-1</sup> two times attwo months interval	4.58 <sup>ab</sup>	72.09	4.31 <sup>ab</sup>	68.81	98.70 <sup>a</sup>	52.26	1.77
T7: Neem cake @ 500 kg ha <sup>-1</sup>	11.29 <sup>bc</sup>	31.20	9.12 <sup>b</sup>	34.01	86.02 <sup>b</sup>	45.22	1.72
T8: Phorate 10G @ 15kg ha <sup>-1</sup>	9.17 <sup>b</sup>	44.12	7.01 <sup>b</sup>	49.28	69.74 <sup>c</sup>	32.43	1.45
T9: Untreated control only 250kg FYM	16.41 <sup>c</sup>		13.82 <sup>c</sup>	-	47.12 <sup>d</sup>		0.98
CD (P=0.05)	5.37		4.33		8.88		
CV%	22.15		19.51		5.79		

Values in column followed by different letter (a,b,c) are significantly different from each other

**Table 3. Efficacy of bioagents against white grubs in sugarcane crop (Pooled data of 2015 and 2016-17)**

Treatment	Per cent plants damaged by white grubs	Per cent reduction in plant damage over control	White grubs/ 10m row	Per cent reduction in white grub population over control	Cane yield t/ha	Yield increase over control(%)	C:B ratio
T1: <i>Metarhizium anisopliae</i> (NBAIR Ma 4) 2.0% W.P.@ 2.5 kg ha <sup>-1</sup> 1 x 10 <sup>8</sup> Cfu/g (min.) in 250 kg FYM (2 times application)	2.75 <sup>a</sup>	91.47	1.99 <sup>a</sup>	85.53	92.58 <sup>a</sup>	61.09	2.02
T2: <i>Beauveria bassiana</i> (NBAIR Bb5a) 2.0% W.P. @ 2.5 kg ha <sup>-1</sup> 1 x 10 <sup>8</sup> Cfu/g in 250 kg FYM (2 times application)	4.06 <sup>ab</sup>	87.4	2.88 <sup>a</sup>	79.05	83.90 <sup>b</sup>	57.07	1.82
T3: <i>Heterorhabditis indica</i> (NBAIR H38) WP @ 20 kg ha <sup>-1</sup> in 150 kg moist sand ha <sup>-1</sup>	3.71 <sup>ab</sup>	88.49	2.67 <sup>a</sup>	80.58	85.29 <sup>ab</sup>	57.77	1.71
T4: <i>Heterorhabditis indica</i> (NBAIR H38) WP @ 20 kg ha <sup>-1</sup> in 150 kg moist sand ha <sup>-1</sup> two times at two months interval	2.99 <sup>a</sup>	90.72	1.40 <sup>a</sup>	89.82	89.09 <sup>ab</sup>	59.57	1.69
T5: <i>Steinernema carpocapsae</i> (NBAII Sc05) WP @ 20 kg ha <sup>-1</sup> in 150 kg moist sand ha <sup>-1</sup>	4.00 <sup>ab</sup>	87.58	2.65 <sup>a</sup>	80.73	84.29 <sup>b</sup>	57.27	1.66
T6: <i>Steinernema carpocapsae</i> (NBAII Sc05) WP @ 20 kg ha <sup>-1</sup> in 150 kg moist sand ha <sup>-1</sup> two times at two months interval	3.69 <sup>ab</sup>	88.55	2.99 <sup>a</sup>	78.25	86.06 <sup>ab</sup>	58.15	1.60
T7: Neem cake @ 500 kg ha <sup>-1</sup>	14.09 <sup>b</sup>	56.28	9.07 <sup>c</sup>	34.04	73.97 <sup>c</sup>	51.30	1.54
T8: Phorate 10G @ 15kg ha <sup>-1</sup>	10.91 <sup>b</sup>	66.15	6.67 <sup>b</sup>	51.49	65.67 <sup>d</sup>	45.14	1.38
T9: Untreated control only 250kg FYM	32.23 <sup>c</sup>	-	13.75 <sup>d</sup>	-	36.02 <sup>e</sup>	-	0.78
CD (P=0.05)	7.6		1.78		4.53		
CV%	20.56		20.88		3.35		

Values in column followed by different letter (a,b,c) are significantly different from each other

**Table 4. Effect of bioagents on non-target organisms in sugarcane ecosystem during 2015-16**

Treatments	*Mean number/3 plants or places														
	Earthworm (No./m3)			Earwig Population (No./ m3)			Spiders			Coccinellids			Chrysopids		
	1 DBT	30 DAT	60 DAT	1 DBT	30 DAT	60 DAT	1 DBT	30 DAT	60 DAT	1 DBT	30 DAT	60 DAT	1 DBT	30 DAT	60 DAT
T1 : <i>Metarhizium anisopliae</i> (NBAIR Ma4) 2.0% W.P @ 2.5 kg ha <sup>-1</sup> 1x10 <sup>8</sup> Cfu/gm (min.) in 250 kg FYM (2 times)	1.0	2.00	2.33	0.00	0.33	0.67	0.67	0.67	1.0	2.0	1.67	1.33	2.33	2.67	1.33
T2: <i>Beauveria bassiana</i> (NBAIR Bb5a) 2.0% W.P @ 2.5 kg ha <sup>-1</sup> 1x10 <sup>8</sup> Cfu/g in 250 kg FYM(2 times)	0.67	1.33	2.0	0.33	0.33	0.00	1.33	1.67	1.00	1.67	1.33	1.33	3.00	2.67	2.33
T3: <i>Heterorhabditis indica</i> (NBAIR H38) WP @ 20 kg ha <sup>-1</sup> in 150 kg moist sand ha <sup>-1</sup>	0.67	1.33	1.67	0.33	0.67	0.33	0.67	0.33	0.33	1.67	1.33	1.00	2.67	2.67	2.00
T4: <i>Heterorhabditis indica</i> (NBAIR H38) WP @ 20 kg ha <sup>-1</sup> in 150 kg moist sand ha <sup>-1</sup> two times at two month interval	1.00	1.00	1.33	0.00	0.33	0.00	0.00	0.33	1.00	1.33	1.00	0.67	2.00	1.33	0.67
T5: <i>Steinernema carpocapsae</i> (NBAIL Sc05) WP @ 20 kg ha <sup>-1</sup> in 150 kg moist sand ha <sup>-1</sup>	1.00	0.67	1.67	0.00	0.33	0.00	0.67	0.33	1.00	2.33	1.33	1.33	2.67	2.33	1.00
T6: <i>Steinernema carpocapsae</i> (NBAIL Sc05) WP @ 20 kg ha <sup>-1</sup> in 150 kg moist sand ha <sup>-1</sup> two times at two month interval	1.00	1.33	1.67	0.67	0.00	0.00	1.00	0.67	0.67	0.67	1.00	1.33	2.00	2.00	1.33
T7: Neem cake @ 500 kg ha <sup>-1</sup>	0.67	1.00	1.33	0.00	0.00	0.00	1.67	0.33	0.67	1.00	1.00	1.0	2.33	2.33	1.00
T8: Phorate 10G @ 15kg ha <sup>-1</sup>	1.00	0.67	0.0	0.00	0.00	0.00	0.67	1.00	0.67	1.00	1.00	0.67	2.33	1.33	1.33
T9: Untreated control only 250kg FYM	0.67	0.00	0.00	0.00	0.00	0.00	0.67	0.67	0.67	1.33	1.33	1.00	1.67	1.67	1.67
CD (0.05)	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS

**DBT - Days before treatment DAT - Days after treatment**  
**NS- Non Significant**

**Table 5. Effect of bioagents on non target organisms in sugarcane ecosystem during 2016-17**

Treatments	*Mean number/3 plants or places														
	Earthworm (No./m3 )			Earwig Population (No./ m3)			Spiders			Coccinellids			Chrysopids		
	1 DBT	30 DAT	60 DAT	1 DBT	30 DAT	60 DAT	1 DBT	30 DAT	60 DAT	1 DBT	30 DAT	60 DAT	1 DBT	30 DAT	60 DAT
T1: <i>Metarhizium anisopliae</i> (NBAIR Ma4) 2.0% W.P @ 2.5 kg ha <sup>-1</sup> 1x10 <sup>8</sup> Cfu/ gm (min.) in 250 kg FYM (2 times)	0.00	1.67	2.00	0.33	2.00	0.67	1.00	1.00	2.00	1.00	1.00	1.67	2.33	1.67	2.67
T2: <i>Beauveria bassiana</i> (NBAIR Bb5a) 2.0% W.P@ 2.5 kg ha <sup>-1</sup> 1x10 <sup>8</sup> Cfu/g in 250 kg FYM(2 times)	0.00	1.00	1.00	0.00	1.67	1.00	1.00	1.67	1.67	1.33	0.67	1.33	1.67	2.33	2.33
T3: <i>Heterorhabditis indica</i> (NBAIR H38) WP @ 20 kg ha <sup>-1</sup> in 150 kg moist sand ha <sup>-1</sup>	0.33	1.00	1.33	0.00	0.67	0.00	0.33	0.33	0.33	1.00	0.67	1.33	1.67	2.00	2.00
T4: <i>Heterorhabditis indica</i> (NBAIR H38) WP @ 20 kg ha <sup>-1</sup> in 150 kg moist sand ha <sup>-1</sup> two times at two month interval	0.00	1.00	0.67	0.67	0.00	0.00	0.33	0.33	1.00	0.67	0.33	1.00	1.33	1.67	1.33
T5: <i>Steinernema carpocapsae</i> (NBAlI Sc05) WP @ 20 kg ha <sup>-1</sup> in 150 kg moist sand ha <sup>-1</sup>	0.67	0.67	2.33	0.00	0.00	0.33	1.00	1.00	1.33	1.33	1.00	1.33	1.33	2.00	1.67
T6: <i>Steinernema carpocapsae</i> (NBAlI Sc05) WP @ 20 kg ha <sup>-1</sup> in 150 kg moist sand ha <sup>-1</sup> two times at two month interval	0.00	1.33	0.00	0.00	0.00	0.00	0.67	1.00	0.67	1.33	0.67	1.00	1.67	1.33	2.00
T7: Neem cake @ 500 kg ha <sup>-1</sup>	0.67	1.00	0.67	0.00	0.33	0.67	0.67	1.00	0.33	1.00	0.33	1.00	1.67	1.00	2.00
T8: Phorate 10G @ 15kg ha <sup>-1</sup>	1.00	0.67	0.00	0.00	0.00	0.00	0.67	1.00	0.33	0.67	0.33	1.00	1.33	1.67	1.67
T9: Untreated control only 250kg FYM	0.67	0.00	0.00	0.00	0.00	0.00	0.67	0.67	0.67	1.00	0.67	1.33	1.33	1.33	0.67
CD (0.05)	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS

DBT - Days before treatment DAT - Days after treatment NS- Non-Significant

61.59 to 77.08t/ha. Untreated control recorded significantly very low cane yield of 24.93t/ha (Table 1).

In the second year trial (2016-17), the cane yield was significantly high in T4 *H. indica* (NBAIR H38) (twice application) treatment (101t/ha), which was statistically at par with other treatments like, T3, *H. indica* (NBAIR H38) (single application) (99.8t/ha), T6 and T5: *S. carpocapsae* (NBAII Sc05) (twice and single application) with an yield of 98.7 and 98.49 t/ha T1, *M. anisopliae* treatment (97.73t/ha) and T2, *B. bassiana* (92.82t/ha) (Table 2). Other treatments like, Phorate 10G/Neem cake application/untreated control recorded significantly low cane yields of 69.74, 86.02 and 47.12 t/ha respectively (Table 2).

The pooled data analysis of two years indicates that all biocontrol agent treatments (T1, T2, T3, T4, T5 and T6) treatments were at par with each other with regard cane yield (83.9-92.58t/ha), although T1 (*M. anisopliae* NBAIR Ma4) treatment showed highest yield of 92.58t/ha (Table 3).

#### Cost Benefit Ratio

The cost benefit ratio (2.0) was found highest in *M. anisopliae* (NBAIR Ma4) 2.0% W.P. formulation treatment in both years (2.0 and 2.03) which was followed by *B. bassiana* (NBAIR Bb5a) treatment (1.71 and 1.93) and rest of the treatments showed C:B ratio in the range of 0.57-1.50 (2015) and 0.98-1.82 (2016-17) (Table 1 and 2).

#### Effect of non target organisms

Statistically significant differences were not observed in the population levels of non-target organisms like soil beneficial organisms like, Earthworms and Earwigs and predators like, Spiders, Coccinellids and Chrysopids in all treated plots and in the untreated control plots in both years (2015 and 2016-17) (Table 3 and 4).

The pooled data analysis of two years indicates that all biocontrol agent treatments were at par with each other with regard to percent plants damaged by white grubs and grub population. Regarding yield, treatments with *M. anisopliae*, *H. indica* (single and twice application) and *S. carpocapsae* (twice application) showed significantly higher yields compared to the yields in *B. Bassiana* and *S. carpocapsae* (single application). However, when cost benefit ratio is taken in to consideration, treatment with *M. anisopliae* proved superior to the other biocontrol agent treatments. Other treatments like, Phorate 10G/Neem cake application/untreated control recorded significantly high percent of plant damage, grub population and low yield compared the treatments with biocontrol agents. Significant differences were not observed in the population levels of non-target

organisms like earthworms, earwigs, spiders, coccinellids and chrysopids in all treatments and in untreated control plots indicating their non-deleterious effect on the beneficial soil organisms and natural enemies of insect pests.

Kulye and Pokharkar (2009) reported the efficacy of soil application of *M. anisopliae* @  $2 \times 10^{12}$  conidia ha<sup>-1</sup> 46.74% reduction of grub population of *H. consanguinea* and increased yield of 27.6t ha<sup>-1</sup> in potato. Application of *M. anisopliae* at the dose of  $4 \times 10^9$  conidia ha<sup>-1</sup> against sugarcane white grub *Holotrichia serrata* (Blanch) was found effective with 92% reduction of grub population and increased yield of 100.6t ha<sup>-1</sup> and was found next to chlorpyrifos treatment (Manisegaran *et al.*, 2011). Samson *et al.* (1999) showed that, soil application of *M. anisopliae* @  $3.3 \times 10^{13}$  conidia ha<sup>-1</sup> against gray back cane grub (*Dermolepida albobirtum*) in Australia showed 50-60 per cent reduction in grub population in sugarcane. *M. anisopliae* and *B. bassiana* @  $5 \times 10^8$  conidia ha<sup>-1</sup> along with chlorpyrifos @ 2lit. ha<sup>-1</sup> was found effective in reducing grub population (Bhagat *et al.*, 2003). Kumbhar *et al.* (2019) reported superior field efficacy of *M. anisopliae*-talc based at the dose of 5 gm per litre against the white grub, *Leucopholis lepidophora* in sugarcane in Maharashtra with minimal clump mortality (9.36%) followed by the application of entomopathogenic Nematode-Powder with the dose of 5 gm per litre (11.68% clump mortality). In Germany, natural epizootics of *Heterorhabditis sp* was reported in grub populations achieving 71% control of the pest in sugarcane field (Akhurst *et al.*, 1992). In our study, soil application of biocontrol agents during two-year field trials showed 68-95% reduction of pest population and 45-62% increased yield of over control in sugarcane crop in coastal Andhra Pradesh. The present findings also indicated that all biocontrol agents tested showed no adverse effect on non-target organisms like earthworms, earwigs, spiders, Coccinellids and Chrysopids. Thungrabeab and Tongma (2007), reported safety of *B. bassiana* Bb.5335 and *M. anisopliae* Ma.7965 to non-target insects such as natural enemies viz., *Coccinella septempunctata* L., *Chrysoperla carnea* (Stephens) and *Dicyphus tamaninii* (Wagner) and beneficial soil insect *Heteromurus nitidus* (Templeton).

#### CONCLUSION

The results showed that soil application of biocontrol agents effectively reduced the grub population of *H. consanguinea* and increased yield in white grub infested sugarcane crop in Coastal Andhra Pradesh, India. Based on cost-benefit ratio, the treatment with *M. anisopliae*, (ICAR-NBAIR Ma-4) was found superior to all other treatments in the management of sugarcane white grubs in coastal Andhra Pradesh with no deleterious effect on non-target organisms. This treatment was better than the insecticidal application in the management of sugarcane white grubs.

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