Neem Plus Garlic Oils Biopesticide Formulation: Safe and Efficient in Watermelon Pest Management

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Summary

To investigate the impact of a novel biopesticide formulation (neem plus garlic oil emulsion - NGOE) in watermelon production, field trials were laid in the early- and late-cropping season of 2020. Watermelon seeds (var. Sweet Sangria F1) were sown in 36 plots which were grouped into 4 replicates of 9 treatments. Data collected include densities of leaf beetles, spiders and *Apis mellifera* L. (bees) using a modified Grizzly 2500/8 leaf blower-vac. All data were subjected to variance analysis after appropriate transformation. Results showed that the most efficient treatment with respect to leaf beetles was 0.125% Magicforce® - Lambda-cyhalothrin 15g/L + Dimethoate 300 g/L (MF) + 5% NGOE in the early-crop, and 0.25% MF in the late-crop. Assessment of change in population densities revealed that 0.25% MF, followed by 0.125% MF + 5% NGOE was most suppressive of *A. mellifera* in the early-crop. Corresponding ordering in the late-crop was: 0.25% MF followed by 0.125% MF + 3% NGOE. While treatment with NGOE suppressed ants by 27.99 – 56.21%, 0.25% MF did it by 45.49 – 63.51%. Similar trend was observed with respect to ant and spider densities. Though 0.125% MF + 5% NGOE treated plots consistently produced the highest number of fruits, followed by 0.25% MF, statistical analyses showed that except for 1% NGOE, other insecticide treatments were comparable in the late-, but not in the early-crop. While the effectiveness of Magicforce® in suppressing pest and improving yield of watermelon was highlighted, the potential of neem plus garlic oils formulation in managing pests of watermelon while relatively favouring the activities of beneficial arthropods, and improving yield was clearly noticeable.

Key words

Apis mellifera L., emulsifier, Grizzly 2500/8 leaf blower-vac, gum arabic solution, leaf beetle species

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Introduction

Across the world, rapid increase and indiscriminate use of insecticides aimed at optimizing crop performance have resulted in the development of resistance by some insect species, the disruption of the biocontrol systems, and the build-up of toxic residues in food as well as the environment (Campos et al., 2016; Okrikata et al., 2021). The growing awareness of the human health and environmental implication of this unchecked use of pesticide has heightened the need to explore viable and sustainable pest control options most especially for fruits and vegetables that are largely consumed raw. This is further escalated by the global shift in consumption patterns from conventionally produced foods to organically produced ones.

The consumption of Watermelon (*Citrullus lanatus* Thunb.) exceeds that of any other cucurbit in the world and it is attracting very high consumer interest in Nigeria as a result of rising understanding of its nutritional and health benefits (Okrikata et al., 2020). However, as reported in other parts of the world (Lima et al., 2014; Reetu and Tomar, 2017), watermelon can hardly be produced in Nigeria without pest control interventions (Okrikata et al., 2019). This is because a complex of arthropod pests, particularly leaf beetle species, infest the crop (from seedling to fruiting stage) with a resultant > 50% yield losses and sometimes even total crop failure (Okrikata et al., 2021).

In Nigeria, watermelon pest management has mainly been by synthetic pesticides whose misuse has led to the accumulation of residues in the fruits beyond the allowable limits as revealed by the investigations of Akan et al. (2015) and Mahmud et al. (2015) in Yobe State, and Omoyajowo et al. (2018) in Lagos State. This has serious human health and environmental consequences as previous studies revealed. For example, ailments and deaths due to consumption of foods with high levels of pesticides have been reported in Cross River, Gombe and Taraba States of Nigeria by Hopkins (2008). Also, a study conducted by Aikpokpodion et al. (2010) revealed that the application of Endosulfan (0.25% ai) in Ibadan, Nigeria significantly suppressed the availability of some important soil nutrients. Erhunmwunse et al. (2012) further reported the presence of DDT (Dichlorodiphenyltrichloroethane) and heptachlor pesticides above WHO (World Health Organization) limits in drinking water in Ibadan, Nigeria.

One of the viable and sustainable pest management alternatives which is currently gaining attention globally is the use of botanical pesticides. It has been shown that there are over 2,500 known plant species from 235 families with pesticidal properties (Stevenson et al., 2017). Of these, neem (*Azadirachta indica* A. Juss.) followed by chili pepper (*Capsicum frutescens* L.) and then garlic (*Allium sativum* L.) are the most recommended (Dougoud et al., 2019) as promising results have been obtained in their usage. Neem and garlic in particular, are among the botanical pesticides recommended by Natural Extension Partners in 20 countries within the Global Agricultural Plantwise Program with activity against different pest species (Dougoud et al., 2019). Botanicals have been reported to be used in combination with synthetic pesticides for better performance (due to synergistic effects), and also for economic and environmental reasons (Anis et al., 2010; Okrikata et al., 2019).

Despite the extensive evaluation of organic products for the control of pests of fruits and vegetable crops (Bushra et al., 2014; Abderrahmane and Lahcen, 2015) and the enormous quantity of these major pesticidal plants in Nigeria (neem and garlic in particular), available literature shows that their combination and/or compatibility with synthetic insecticides for the management of arthropod pests of crops, watermelon inclusive, has scantily been investigated. Additionally, the impact of botanicals on beneficial arthropods is hardly assessed on the assumption that since they are derived from natural sources, they will always be safer than synthetic pesticides - a view that has sometimes been found to be erroneous (Plata-Rueda et al., 2017). We therefore combined neem and garlic oils to formulate a novel biopesticide, and then evaluated its efficiency and compatibility with Magicforce® (Lambda-cyhalothrin 15g/L + Dimethoate 300 g/L) on arthropod pests of watermelon as well as their impact on beneficial organisms and the performance of watermelon.

Materials and Methods

Study Site

The research was conducted on the research field of Federal University, Wukari, Nigeria. Wukari lies within the Southern Guinea savanna agro-ecological zone (N7°50'37", E9°46'31") at an altitude of 187 m. The area is characterized by a warm tropical climate, and well defined rainy (commencing in April and ending in October with peaks in June and September), and dry seasons with average annual rainfall and temperature of 1205 mm and 26.8 °C, respectively (Okrikata et al., 2019).

Field Layout and Experimental Design

Watermelon (var. Sweet Sangria F_1) was sown in 36 plots (each, 5 x 8 m) on a ploughed and harrowed land that had not been cultivated for three cropping years. This was done during the early and late cropping seasons of 2020 (sowing dates: 23rd April and 25th August, respectively). The plots were grouped into four replications of nine treatments (control inclusive) in a Randomized Complete Block Design (RCBD) (Gomez and Gomez, 1984) as shown in Fig. 1. The treatments evaluated were: the lowest field recommended dose (0.25%) of Magicforce® [manufactured by Anhui Zhongshen Chemical Industries Co. Ltd., China] – (0.25% MF); neem oil [manufactured by Mai Rana Islamic and Research, Nigeria] + garlic oil [manufactured by El Hawag Chemical Company, Badr City, Cairo, Egypt] emulsion (1:1, v/v) at 1% – (1% NGOE); 3% NGOE; 5% NGOE; 0.125% MF + 1% NGOE; 0.125% MF + 3% NGOE; 0.125% MF + 5% NGOE; 1% Emulsifier (2:1, v/v mixture of liquid soap manufactured by PZ cussons Nig Plc. and gum arabic solution); control (CT) – no spray.

The emulsion (NGOE) was prepared using the method described by Ukeh et al. (2007). Spraying at 400 Lha^{-1} output with the aid of 8 litre capacity Maxipro sprayer was done when the leaf beetles reached the economic injury level of 5 beetles/plant using visual observation (Foster, 2016; Okrikata et al., 2019), and this was maintained until early fruiting stage when the experiment was terminated.

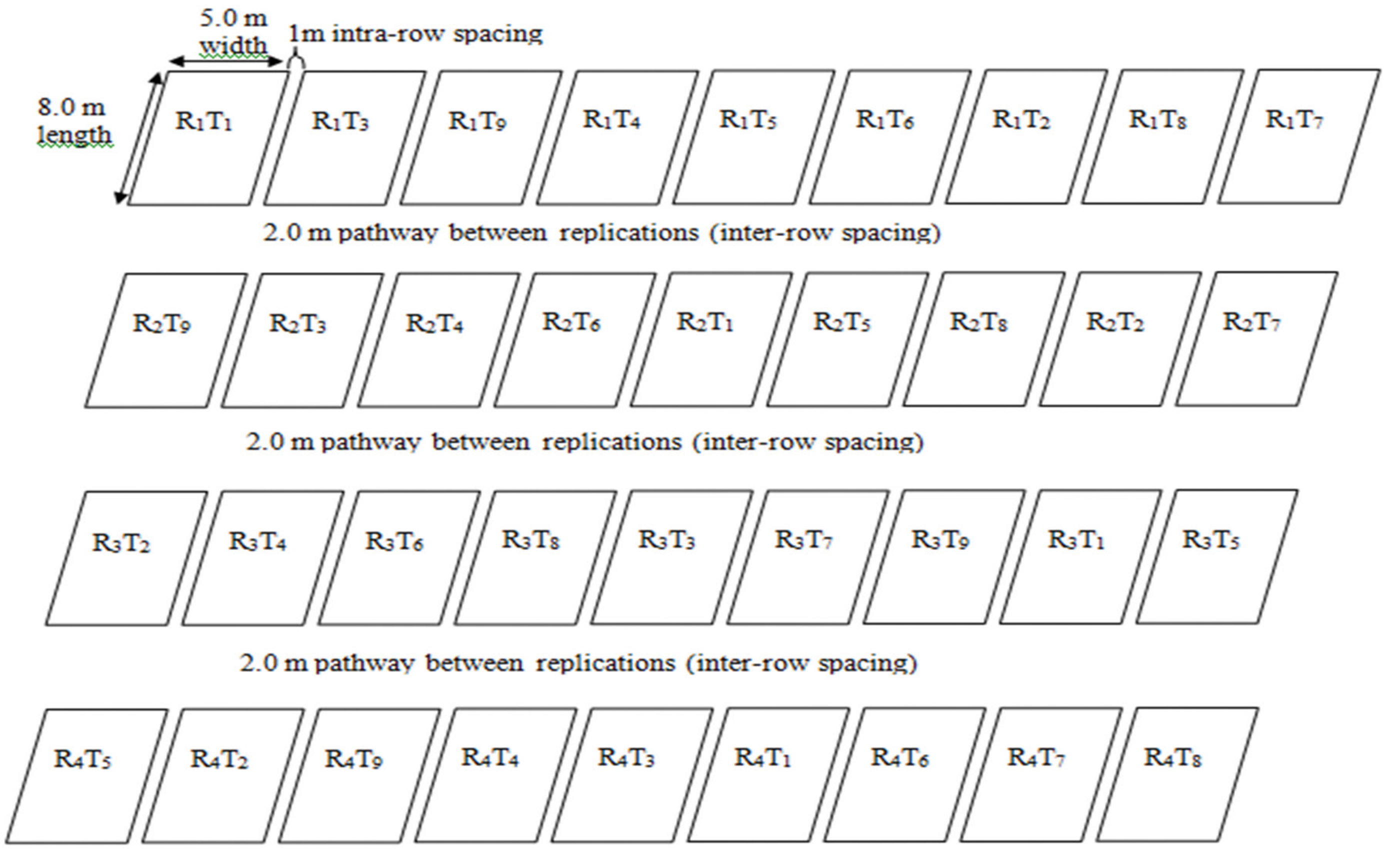

Figure 1. Experimental layout: **R** = Replicate; **T** = Treatment; $\mathbf{T_1}$ = 0.25% MF (Magicforce®); $\mathbf{T_2}$ = 1% NGOE (Neem + Garlic oil emulsion); $\mathbf{T_3}$ = 3% NGOE; $\mathbf{T_4}$ = 5% NGOE; $\mathbf{T_5}$ = 0.125% MF + 1% NGOE; $\mathbf{T_6}$ = 0.125% MF + 3% NGOE; $\mathbf{T_7}$ = 0.125% MF + 5% NGOE; $\mathbf{T_8}$ = 1% Emulsifier; $\mathbf{T_9}$ = Control

Data Collection

Sampling of Arthropod Species

Sampling of leaf beetles [predominated by *Epilachna chrysomelina* (Fabricius), *Monolepta nigeriae* (Bryant), *Asbecesta transversa* (Allard), *Asbecesta nigripennis* (Weise) and *Aulacophora africana* (Weise)], *Apis mellifera* (L.), predatory ants (predominated by *Pheidole* sp., *Crematogaster* sp. and *Camponotus* sp.) and spiders was done using a 5 cm internal diameter inlet coned modified Grizzly 2500/8 leaf blower-vac (Grizzly Gartengeräte GmbH & Co. KG, China) which was powered by a Tiger TG950DC portable generator (Agere et al., 2021). On each plot, the machine was swept through the 5 m length of the middle row at an approximate walking speed of 1m/second between the hours of 0600 – 0800. Arthropods of interest were collected following the method described by Agere et al. (2021), sorted and preserved in 70% alcohol for counting, and mean populations were expressed as number per 5 m length of row. Pre-treatment arthropod counts were taken one day before the treatment while post-treatment counts were taken at the 1st, 3rd and 7th day after treatments application (modified after Saleem et al., 2019).

The change in population density (CPD) of the arthropods and the efficacy percentage (EF) of the treatments were assessed using the methods described by Saleem et al. (2019) and Henderson and Tilton (1955), respectively:

$$CPD = (X_i - X_0)/\ X_0 \times 100$$

where: $\mathbf{X_0}$ – pre-treatment arthropod counts; $\mathbf{X_i}$ - post-treatment arthropod counts.

The CPD measures the extent of decrease/reduction (negative values), or increase (positive values) in arthropod density over time.

$$EF(\%) = 100 \times 1-(T_a \times C_b\ /\ T_b \times C_a)$$

where: $\mathbf{T_a}$ – post-treatment insect count; $\mathbf{T_b}$ – pre-treatment insect count; $\mathbf{C_a}$ – post-treatment insect count in control plots; $\mathbf{C_b}$ – pre-treatment insect count in control plots.

The EF assesses the level at which the treatment suppresses the insect pests in the individual treated plots vis-à-vis the untreated (control) plots - efficiency.

Assessment of Leaf Injury and Growth Indices

Ten leaves were randomly selected per plot and from these, the proportion and severity of leaves injured were computed at 50% vegetative, 50% flowering, and 50% fruiting stages as described by Okrikata and Anaso (2008) and Okrikata et al. (2020) in which the leaves were scaled 0 – 4 for severity of injury: **0** – 0% leaf injury; **1** – 1 – 25% leaf injury; **2** – 26 – 50% leaf injury; **3** – 51 – 75% leaf injury; **4** – 76 – 100% leaf injury.

Scores obtained per plot were transformed to Attack severity (%) using the equation described by Okrikata and Anaso (2008) and Okrikata et al. (2020):

$$\text{Attack severity}(\%) = \Sigma n \times 100 / N \times 4$$

where: **Σn** - summation of individual injury scores/plot; **N** - number of scores taken/plot (= 10); **4** - highest score on the scale.

Assessment of Survival Rate and Yield Parameter

The survival rate (SR) was estimated as:

$$SR(\%) = \frac{\text{Number of plants reaching maturity}}{\text{Number of plants per plot at 10 days after planting}}$$

At the early fruiting stage, the number of fruits/treatment were counted.

Data Analysis

Data collected were subjected to one-way analysis of variance (ANOVA) and significantly different treatment means were separated by Student Newman Keul's (SNK) test at 5% level of probability. Treatment effects were considered fixed, while replicate effects were considered random (Gomez and Gomez, 1984). However, the raw data collected were subjected to Shapiro-Wilk test at $P>0.05$ (Shapiro and Wilk, 1965; Okrikata et al., 2021), and in order to normalize the data and to meet the assumptions of parametric tests; count data were subjected to square root transformation while proportions were log transformed before they were subjected to analysis of variance. The transformed data were further subjected to Shapiro-Wilk test to confirm their normality at $P > 0.05$ (Shapiro and Wilk, 1965; Okrikata et al., 2021). The analyses were run using IBM® SPSS® version 23.0 (SPSS Inc., Chicago, Illinois) which was licensed in 2015.

Results

Impact of Magicforce® and Neem Plus Garlic Oils Derived Biopesticide on Leaf Beetles Infestation in Watermelon

Table 1 shows that the differences in population density of leaf beetles across the treatment plots before the treatments were imposed were due to random variations ($P>0.05$) in both the early- and late-sown crop. However, the reverse ($P<0.01$) was observed post-treatment. Across the post-spray sampling times of both seasons, density of leaf beetles was the least ($P<0.01$) in 0.25% MF treated plots and was statistically comparable with infestations in 0.25% MF + 5% NGOE treated plots. As at the 7th day post-treatment, the aforementioned treatments were statistically at par with only 5% NGOE in the early-crop, and also with 0.125% MF + 3% NGOE in the late-crop.

Table 1 also shows that the CPD and EF of the insecticide treatments were > 50% across the sampling days of both seasons. In the early crop, 0.125% MF + 5% NGOE had the highest population reduction (range; 84.60 – 85.90%) and efficiency (range; 85.77 – 86.52%). This was followed by 0.25% MF with corresponding range values of 83.89 – 84.40%, and 84.26 – 85.60%. In the late-sown crops however, 0.25% MF treated plots had the highest population decrease (range; 73.08 – 83.12%) and efficiency (range; 75.45 – 84.18%). This was followed by 0.125% MF + 5% NGOE on the 1st day of post-treatment with respective values of 74.03% and 74.68%, and by 5% NGOE on the 3rd- (values of 77.23% and 78.67%), and 7th day of post-treatment (values of 71.10% and 73.58%). Of all the treatments sprayed, 1% emulsifier had the least efficiency (range; 0.36 – 15.21%). Overall, MF + NGOE (pooled data of 0.125% MF + 1, 3 and 5% NGOE) was 1.15 and 1.06x more suppressive and 1.14 and 1.05x more efficient than sole NGOE (pooled data of 1, 3 and 5% NGOE) with respect to leaf beetles in the early and late crop, respectively. Corresponding values of sole MF (0.25% MF) over sole NGOE were 1.20 and 1.25x, and 1.19 and 1.23x.

Impact of Magicforce® and Neem plus Garlic Derived Biopesticide on Beneficial Arthropods in Watermelon Field

Table 2a shows that while there was overall increase in ant density in the control plots (except on the 7th day post-spray of the late crop which had 5.61% decrease) by an average of 46.47 and 4.02% in the early- and late-crop, respectively, 0.25% MF treated plots had a reduction in ant density by 45.49 and 63.51% in the respective seasons. Additionally, except on the 7th day post-spray of the early crop in which plots treated with 1% NGOE had a marginal increase in ant density (0.31%), the combination of MF and NGOE incurred an overall higher reduction in ants density by 1.03 and 1.06x than sole NGOE in the respective seasons as against sole MF that was 1.32 and 1.29x more suppressive than NGOE. Spraying with 1% emulsifier did not result in decrease of ants density in the early crop (mean ca. increase of 22.96% was recorded) but slightly did in the late crop by 1.61 to 4.46%.

Table 2b shows that differences in predatory spider density among the treatments before spray in both seasons were insignificant ($P>0.05$). However, observations on the post-spray sampling days showed significant differences ($P<0.01$) among the treatments. Higher and statistically comparable spider densities were recorded in control and 1% emulsifier treated plots across sampling time and in both seasons which were only statistically at par with those in 1% NGOE treated plots on the 3rd day post-spray in both seasons. The highest reduction in spider density was observed in plots treated with 0.25% MF (range; 75.47 – 89.65%), followed by 0.125% MF + 5% NGOE with a range of 65.59 – 84.48%. Overall, while sole MF resulted in 1.64 and 1.82x decrease in spider densities compared to NGOE in the early and late crop, combination of MF and NGOE was 1.33 and 1.37x more suppressive than sole NGOE. The exception to this was on the 7th day post-spray of the early crop in which control and 1% emulsifier treated plots respectively had 2.67 and 11.88% increase in spider density, and they had an overall lesser spider reduction (1.50 – 22.96%) than the other treatments.

Table 2c indicates significant differences ($P<0.01$) among the treatments with respect to *A. mellifera* density; in both seasons there were no significant differences in *A. mellifera* density between control plots and those treated with 1% emulsifier. More so, while the control plots and those sprayed with 1% emulsifier had increases in *A. mellifera* density across sampling days after spray and in both seasons (range; 7.20 – 37.59%), the insecticide treated plots largely had reductions. The exception was for plots sprayed with 1% NGOE which had marginal increases of 2.32 and 1.16% on the 3rd and 7th day post-spray of the early crop.

Table 1a. Individual and composite effect of Magicforce® and neem plus garlic oils derived biopesticide on pest infestation in Watermelon at Wukari in 2020: Leaf beetles[a] density (no. per 5 m row)

Treatment	PD before spray	After 1 day of spray			After 3 days of spray			After 7 days of spray		
		PD	CPD (%)	EF (%)	PD	CPD (%)	EF (%)	PD	CPD (%)	EF (%)
Early-sown										
0.25% MF	9.87	1.54[c]	-84.40	84.26	1.56[d]	-84.19	85.23	1.59[e]	-83.89	85.60
1% NGOE	10.20	3.52[b]	-64.49	65.18	3.55[b]	65.20	67.48	3.90[c]	-61.76	65.82
3% NGOE	9.87	3.49[b]	-64.64	64.33	2.30[cd]	-76.70	78.23	2.83[d]	-71.33	74.37
5% NGOE	10.13	3.43[b]	-66.14	65.84	1.90[cd]	-81.24	82.47	2.10[de]	-79.27	81.47
0.125% MF + 1% NGOE	10.57	1.91[c]	-81.93	81.78	2.68[c]	-74.65	76.31	2.80[d]	-73.51	76.32
0.125% MF + 3% NGOE	11.46	1.84[c]	-83.94	83.80	2.20[cd]	-80.80	82.06	2.63[d]	-77.05	79.48
0.125% MF + 5% NGOE	12.34	1.74[c]	-85.90	85.77	1.78[cd]	-85.58	86.52	1.90[e]	-84.60	86.24
1% Emulsifier	10.28	9.34[a]	-9.14	8.33	10.08[a]	-1.95	8.36	9.75[b]	-5.15	15.21
Control (no spray)	10.28	10.19[a]	-0.88		11.00[a]	7.00		11.50[a]	11.87	
F-value (*df* = 8, 27)	0.55	68.42			60.09			90.40		
P-value	0.87	<0.01			<0.01			<0.01		
Late-sown										
0.25% MF	14.75	2.93[e]	-80.14	80.63	2.49[e]	-83.12	84.18	3.97[c]	-73.08	75.45
1% NGOE	15.10	6.96[b]	-53.91	55.06	6.89[b]	-54.37	57.25	7.60[b]	-49.67	53.98
3% NGOE	13.98	5.66[bc]	-59.51	60.52	4.43[cd]	-68.31	70.31	5.17[c]	-63.02	66.19
5% NGOE	15.33	4.74[cd]	-69.02	69.85	3.49[de]	-77.23	78.67	4.43[c]	-71.10	73.58
0.125% MF + 1% NGOE	13.73	5.55[bc]	-59.58	60.59	5.48[bc]	-59.01	62.61	6.64[b]	-51.64	55.78
0.125% MF + 3% NGOE	15.15	4.13[d]	-72.74	73.42	4.37[cd]	-71.16	72.98	5.30[c]	-65.02	68.01
0.125% MF + 5% NGOE	14.75	3.83[de]	-74.03	74.68	3.60[de]	-75.59	77.13	4.44[c]	-69.90	70.62
1% Emulsifier	14.57	14.89[a]	2.20	0.36	14.62[a]	0.34	6.00	15.36[a]	5.42	3.61
Control (no spray)	14.83	15.21[a]	2.56		15.83[a]	6.74		16.22[a]	9.37	
F-value (*df* = 8, 27)	0.59	68.13			57.11			78.67		
P-value	0.76	<0.01			<0.01			<0.01		

PD = Population density; **CPD** = Change in population density; **EF** = Efficacy percentage; Means are values of 4 replications; [a] = Leaf beetles (mean of E. *chrysomelina, M. nigeriae, A. transversa, A. nigripennis* and *A. africana*); Means followed by the same superscript letter(s) within a column are not significantly different using Student-Newman Keul's (SNK) test ($P \leq 0.05$)

Table 2a. Individual and composite effect of Magicforce® and neem plus garlic oils derived biopesticide on density of beneficial arthropods in Watermelon at Wukari in 2020: Predatory ants[b] (no. per 5 m row)

Treatment	PD before spray	After 1 day of spray		After 3 days of spray		After 7 days of spray	
		PD	CPD (%)	PD	CPD (%)	PD	CPD (%)
Early-sown							
0.25% MF	6.36±0.34	2.67±0.20[d]	-58.02	3.68±0.08[d]	-42.14	4.05±0.10[f]	-36.32
1% NGOE	6.38±0.37	4.13±0.24[b]	-35.27	5.07±0.17[b]	-20.53	6.40±0.33[b]	0.31
3% NGOE	7.03±0.37	3.43±0.09[c]	-51.21	4.18±0.08[cd]	-40.54	5.65±0.13[c]	-19.35
5% NGOE	6.42±0.37	3.15±0.06[cd]	-50.93	3.85±0.06[d]	-40.03	4.78±0.09[de]	-25.55
0.125% MF + 1% NGOE	5.44±0.23	3.35±0.13[c]	-38.42	4.48±0.03[c]	-17.65	5.20±0.22[cd]	-4.41
0.125% MF + 3% NGOE	6.47±0.33	2.95±0.17[cd]	-54.40	3.78±0.09[d]	-41.58	4.75±0.09[de]	-26.58
0.125% MF + 5% NGOE	6.76±0.24	2.83±0.15[cd]	-58.14	3.70±0.09[d]	-45.27	4.40±0.71[ef]	-34.91
1% Emulsifier	6.43±0.56	7.52±0.15[a]	16.95	8.37±0.32[a]	30.17	7.83±0.24[a]	21.77
Control (no spray)	5.43±0.29	7.62±0.09[a]	40.33	8.35±0.27[a]	53.78	7.89±0.20[a]	45.30
F-value (*df* = 8, 27)	2.25	163.12		138.56		59.33	
P-value	0.05	<0.01		<0.01		<0.01	
Late-sown							
0.25% MF	7.81±0.40	2.48±0.52[e]	-68.25	2.94±0.32[d]	-62.36	3.13±0.33[e]	-59.92
1% NGOE	7.59±0.49	4.09±0.17[bc]	-46.11	4.64±0.12[b]	-38.87	5.80±0.09[b]	-23.58
3% NGOE	8.49±0.28	3.26±0.11[d]	-61.60	3.90±0.04[c]	-54.06	4.15±0.06[d]	-51.12
5% NGOE	8.00±0.26	2.85±0.07[de]	-64.38	3.76±0.02[c]	-53.00	3.90±0.04[d]	-51.25
0.125% MF + 1% NGOE	7.71±0.50	3.50±0.12[b]	-54.60	4.48±0.03[b]	-41.89	4.68±0.12[c]	-39.30
0.125% MF + 3% NGOE	7.51±0.59	2.70±0.04[bc]	-64.05	3.73±0.14[c]	-50.33	4.00±0.21[d]	-46.74
0.125% MF + 5% NGOE	7.89±0.78	2.63±0.08[cd]	-66.67	3.85±0.13[c]	-51.20	3.60±0.10[d]	-54.37
1% Emulsifier	8.07±0.40	7.92±0.25[a]	-1.86	7.94±0.22[a]	-1.61	7.71±0.20[a]	-4.46
Control (no spray)	7.84±0.52	8.21±0.32[a]	4.72	8.10±0.31[a]	3.32	7.40±0.11[a]	-5.61
F-value (*df* = 8, 27)	0.35	79.85		92.15		85.88	
P-value	0.94	<0.01		<0.01		<0.01	

PD = Population density; **CPD** = Change in population density; Means (± SE) are values of 4 replications; [b] = Predatory ants (mean of *Camponotus* sp., *Crematogaster* sp. and *Pheidole* sp.); Means (± SE) followed by the same superscript letter(s) within a column are not significantly different using Student-Newman Keul's (SNK) test ($P \leq 0.05$)

Table 2b. Individual and composite effect of Magicforce® and neem plus garlic oils derived biopesticide on density of beneficial arthropods in Watermelon at Wukari in 2020: Spiders[c] (no. per 5 m row)

Treatment	PD before spray	After 1 day of spray		After 3 days of spray		After 7 days of spray	
		PD	CPD (%)	PD	CPD (%)	PD	CPD (%)
Early-sown							
0.25% MF	6.85±0.53	0.78±0.15[f]	-88.61	1.68±0.33[d]	-75.47	1.59±0.30[f]	-76.79
1% NGOE	6.39±0.30	3.71±0.03[b]	-41.94	4.48±0.13[a]	-29.89	4.77±0.12[b]	-25.35
3% NGOE	5.78±0.52	2.58±0.10[c]	-55.36	3.34±0.16[b]	-42.21	3.40±0.17[c]	-41.18
5% NGOE	6.44±0.49	1.24±0.05[e]	-80.75	2.40±0.16[bc]	-62.73	2.48±0.15[de]	-61.49
0.125% MF + 1% NGOE	6.53±0.12	2.25±0.06[cd]	-65.54	2.78±0.23[bc]	-57.43	3.28±0.33[cd]	-49.77
0.125% MF + 3% NGOE	6.75±0.35	1.83±0.24[d]	-72.89	2.63±0.17[bc]	-61.04	2.45±0.18[de]	-63.70
0.125% MF + 5% NGOE	6.28±0.34	1.13±0.28[ef]	-82.01	1.99±0.27[cd]	-68.31	2.10±0.29[ef]	-66.56
1% Emulsifier	5.89±0.24	5.62±0.10[a]	-4.58	4.88±0.36[a]	-17.15	6.59±0.10[a]	11.88
Control (no spray)	6.36±0.32	5.40±0.26[a]	-15.05	4.90±0.29[a]	-22.96	6.53±0.13[a]	2.67
F-value (df = 8, 27)	0.86	72.04		23.25		47.98	
P-value	0.57	<0.01		<0.01		<0.01	
Late-sown							
0.25% MF	8.31±0.25	0.86±0.18[e]	-89.65	1.95±0.38[d]	-76.53	1.74±0.33[f]	-79.06
1% NGOE	7.42±0.29	4.85±0.05[bc]	-34.64	6.28±0.21[a]	-15.36	6.21±0.17[b]	-16.31
3% NGOE	7.40±0.31	3.12±0.12[d]	-57.84	4.35±0.20[b]	-41.22	4.17±0.23[c]	-43.65
5% NGOE	6.67±0.33	1.37±0.06[de]	-79.46	2.93±0.21[c]	-56.07	2.75±0.17[de]	-58.77
0.125% MF + 1% NGOE	7.28±0.60	2.85±0.07[b]	-60.85	4.12±0.12[b]	-43.41	3.95±0.25[c]	-45.74
0.125% MF + 3% NGOE	6.91±0.33	2.01±0.15[bc]	-70.91	3.14±0.24[c]	-54.56	2.96±0.26[d]	-57.16
0.125% MF + 5% NGOE	7.15±0.23	1.11±0.10[cd]	-84.48	2.46±0.29[cd]	-65.59	2.25±0.13[e]	-68.53
1% Emulsifier	7.80±0.40	6.42±0.15[a]	-17.69	6.86±0.50[a]	-12.05	7.47±0.16[a]	-4.23
Control (no spray)	7.34±0.28	5.92±0.16[a]	-19.35	6.87±0.40[a]	-6.40	7.23±0.16[a]	-1.50
F-value (df = 8, 27)	1.84	79.85		39.04		71.84	
P-value	0.11	<0.01		<0.01		<0.01	

PD = Population density; **CPD** = Change in population density; Means (± SE) are values of 4 replications; [c] = Spider species were treated as a single population/taxon; Means (± SE) followed by the same superscript letter(s) within a column are not significantly different using Student-Newman Keul's (SNK) test ($P \leq 0.05$)

Table 2c. Individual and composite effect of Magicforce® and neem plus garlic oils derived biopesticide on density of beneficial arthropods in Watermelon at Wukari in 2020: *Apis mellifera* L. (no. per 5 m row)

Treatment	PD before spray	After 1 day of spray		After 3 days of spray		After 7 days of spray	
		PD	CPD (%)	PD	CPD (%)	PD	CPD (%)
Early-sown							
0.25% MF	6.23±0.12^{a}	3.16±0.05^{f}	-49.28	3.69±0.07^{f}	-40.77	3.68±0.05^{h}	-40.93
1% NGOE	6.03±0.25^{a}	5.95±0.17^{a}	-1.32	6.17±0.20^{a}	2.32	6.10±0.02^{a}	1.16
3% NGOE	6.00±0.24^{a}	5.00±0.21^{b}	-16.67	5.81±0.10^{ab}	-3.17	5.78±0.04^{b}	-3.67
5% NGOE	5.95±0.23^{a}	4.11±0.09^{d}	-30.92	5.37±0.19^{bc}	-9.75	5.39±0.06^{c}	-9.41
0.125% MF + 1% NGOE	6.31±0.15^{a}	4.57±0.10^{c}	-27.62	4.95±0.12^{cd}	-21.55	4.91±0.01^{d}	-22.19
0.125% MF + 3% NGOE	6.25±0.17^{a}	3.85±0.16^{de}	-38.40	4.75±0.08^{cde}	-24.00	4.75±0.05^{e}	-24.00
0.125% MF + 5% NGOE	6.28±0.13^{a}	3.63±0.05^{e}	-42.20	4.55±0.12^{de}	-27.55	4.54±0.05^{f}	-27.71
1% Emulsifier	3.61±0.14^{b}	3.87±0.05^{de}	7.20	4.15±0.33^{ef}	14.96	3.88±0.03^{g}	7.48
Control (no spray)	3.40±0.09^{b}	3.88±0.07^{de}	14.12	4.17±0.20^{ef}	22.65	3.89±0.02^{g}	14.41
F-value (*df* = 8, 27)	43.46	49.31		21.51		451.65	
P-value	<0.01	<0.01		<0.01		<0.01	
Late-sown							
0.25% MF	6.23±0.12^{a}	3.13±0.05^{f}	-49.76	3.63±0.06^{e}	-41.73	3.63±0.05^{g}	-41.73
1% NGOE	6.78±0.43^{a}	5.79±0.17^{a}	-14.60	5.90±0.18^{a}	-12.98	5.90±0.03^{a}	-12.98
3% NGOE	6.25±0.10^{a}	4.89±0.20^{b}	-21.76	5.61±0.09^{ab}	-10.24	5.65±0.03^{b}	-9.60
5% NGOE	6.20±0.25^{a}	4.06±0.08^{d}	-34.52	5.25±0.18^{b}	-15.32	5.31±0.06^{c}	-14.35
0.125% MF + 1% NGOE	6.31±0.15^{a}	4.43±0.11^{c}	-29.79	4.73±0.11^{c}	-25.04	4.78±0.03^{d}	-24.25
0.125% MF + 3% NGOE	6.75±0.40^{a}	3.40±0.13^{de}	-49.63	4.60±0.07^{c}	-31.85	4.64±0.04^{e}	-31.26
0.125% MF + 5% NGOE	6.28±0.13^{a}	3.60±0.07^{e}	-42.68	4.45±0.11^{cd}	-29.14	4.50±0.04^{f}	-28.34
1% Emulsifier	3.10±0.29^{b}	3.83±0.10^{de}	23.55	3.98±0.31^{de}	28.39	3.72±0.03^{g}	20.00
Control (no spray)	2.90±0.20^{b}	3.83±0.13^{de}	32.07	3.99±0.20^{de}	37.59	3.73±0.03^{g}	28.62
F-value (*df* = 8, 27)	37.86	47.12		21.65		499.67	
P-value	<0.01	<0.01		<0.01		<0.01	

PD = Population density; **CPD** = Change in population density; Means (± SE) are values of 4 replications; Means (± SE) followed by the same superscript letter(s) within a column are not significantly different using Student-Newman Keul's (SNK) test ($P \leq 0.05$)

Impact of Magicforce® and Neem Plus Garlic Oils Derived Biopesticide on Leaf Damage and Yield Parameters in Watermelon

Table 3 shows significant differences among the treatments with respect to proportion and severity of leaf injury in both seasons. Higher and statistically comparable leaf damage (proportion and severity) was observed in leaves in control plots and those sprayed with 1% emulsifier. The least proportion of leaf damage was observed in crops sprayed with 0.25% MF which was statistically at par with 5% NGOE and 0.125% MF + 5% NGOE in both seasons. The severity of leaf injury followed a similar trend. Overall, while sole MF suppressed proportion of leaves injured by 82.13%, sole NGOE did it by 68.73% and MF + NGOE by 71.44% in the early crop. Comparable values for late crop were 82.25, 66.67 and 71.51%, respectively. With respect to severity of leaves injured, overall suppression over control was 84.14% (for MF), 69.42% (for sole NGOE) and 76.58% (for MF + NGOE) in the early crop. Late crop values were 89.01, 72.11 and 80.64%, respectively (Table 3).

Table 4 shows that ≈ 30% of the crops reached maturity without insecticide intervention. Proportion of plants that reached maturity stage in 1% emulsifier sprayed plots did not differ from those in control plots. Table 4 also indicates that overall, 62 – 96% of the insecticide treated crops reached maturity. Survival rate (%) was topmost in 0.125% + 5% NGOE treated crops in the early crop (96.98±1.66) and in 0.25% MF treated plots in the late crop (90.05±3.69). The aforementioned treatments were statistically comparable with each other in both seasons and also with all other insecticide treatments except with 1% NGOE in the late crop, and also with 3% NGOE in the early crop. Crops treated with 1% NGOE had the least survival rate (62.00±3.29 and 59.56±3.89% in the early and late crop respectively) among the insecticide treatments. This was followed by those treated with 3% NGOE (75.17±2.81 and 71.03±2.45% in the respective seasons).

Table 4 also shows that 0.125% MF + 5% NGOE effected the production of the highest number of fruits in both seasons. In the late crop it was statistically comparable with all other insecticide treatments except 1% NGOE. The least number of fruits was recorded in the control plots and was statistically at par with those in 1% emulsifier treated plots in both seasons. The insecticide treatments affected 6.55 – 12.03x more fruits than control in the early-crop. Values for the late crop were 5.45 – 11.78x.

Discussion

In this study, a novel plant-based oil emulsion derived from neem and garlic oils mixture was formulated and field-tested vis-à-vis key pest and beneficial arthropods associated with watermelon. This is instructive as available literature indicates that only a few studies on arthropod pests of watermelon pay attention to the impact of the control strategies on beneficial arthropods.

Watermelon is one of the crops that heavily rely on bees (16 – 24 flower visits required for maximum pollination) to optimally produce fruits. However, it is prone to serious attacks by a variety of arthropod pests (Okrikata et al., 2019; Ternest et al., 2020). Hence, pest control strategies must be amenable to pollinators and pollination for optimum productivity of the crop. In the study area, as in other areas around the world, leaf beetles are highly pestiferous on watermelon (Foster and Brust, 1995; Okrikata et al., 2019). Aside vectoring bacterial pathogens, the adults are critical defoliators – they are capable of causing total crop failure. Hence, early detection and application of pest suppressing strategies is critical in watermelon production.

Neem oil contains a minimum of 100 bioactive compounds reported to be effective against over 550 pest species (Anuradha and Annadurai, 2008). The major ones are the triterpenes also known as limonoids of which azadirachtin is the most important (Campos et al., 2016). Neem has contact and systemic properties (Okrikata and Anaso, 2008). It inhibits feeding, disrupts juvenile hormone functions and suppresses growth and fertility of arthropod pests (Saleem et al., 2019). However, it also has pathogenicidal properties but findings show it is less, if at all toxic to pollinators and natural enemies (Mkindi et al., 2017; Dougoud et al., 2019). Garlic oil, on the other hand, possesses an alliaceous compound; allyl-epropyle-disulphide which is very pungent and has fumigant, repellent, anti-feedant, and insecticidal properties on a number of pest species (Aziza and Asma, 2015). It is also bactericidal in action (Prowse et al., 2006). However, the active ingredient of garlic is known to degrade very quickly (Koch and Lawson, 1996). Garlic is non-selective and as such, it is broad-spectrumed in effect – investigations revealed lethal and sub-lethal effects of garlic on bees (Xavier et al., 2015).

Observations in this study revealed that none of the rates of sole NGOE and its combination with Magicforce® showed any symptom of phytotoxicity on the crop. However, it was evident that all the rates of the insecticidal treatments used suppressed pest infestation. This highlights the efficacy and compatibility of neem with garlic oil, and also with Magicforce®. Pest suppressing effects of the combination of MF and NGOE, and also 5% NGOE largely matched that of sole MF (0.25%). This buttresses the findings of Štefanidesová et al. (2017) which showed that botanical pesticides had suppressive effects on agricultural pests to extents comparable and in certain instances, more efficacious than synthetic chemicals owing to their varied mechanisms of action.

An important observation in this study was that aside from reducing both the dosage required and toxicity towards the beneficial arthropods (predatory ants, spider species, and bees), the combination of the synthetic pesticide (Magicforce®) with neem plus garlic oil emulsion, improved the efficacy of the biopesticide formulation. The constituents of the biopesticide formulation (neem and garlic oils) have each been shown to be efficient in field pest management (Plata-Rueda et al., 2017). Neem-based pesticides have been successful in the control of blattodean, hemipteran, lepidopteran and thysanopteran pests (Aziz et al., 2013; Okrikata et al., 2019).

Addition of adjuvants is a common activity in pesticide formulation. This is aimed at improving the pesticide efficiency via longer persistence and better coverage (Witt, 2012). Soap (bar flakes or liquid) is the most widely used emulsifier or adjuvant. While laboratory studies revealed that small concentrations (≈ 1ml/l) of domestic soap prepared with distilled water might be highly insecticidal (Lee et al., 2006), field trials revealed otherwise (Amoabeng et al., 2014). The current study however revealed that the emulsifier used (liquid soap + gum arabic solution) was ineffective in suppressing leaf beetles at 1%.

Table 3. Individual and composite effect of Magicforce® and neem plus garlic derived biopesticide on leaf damage in Watermelon at Wukari in 2020

Treatment	Proportion of leaves injured (%)	Severity of leaves injured (%)
Early-sown		
0.25% MF	8.34±0.96 (2.10±0.11^{f})	4.55±0.08 (1.52±0.02^{f})
1% NGOE	20.84±2.10 (3.02±0.10^{b})	15.10±0.18 (2.71±0.01^{b})
3% NGOE	14.17±0.84 (2.65±0.06^{bcd})	7.50±0.24 (2.01±0.03^{d})
5% NGOE	9.17±0.83 (2.20±0.10^{ef})	5.56±0.11 (1.71±0.02^{ef})
0.125% MF + 1% NGOE	16.67±1.36 (2.80±0.08^{bc})	10.56±0.13 (2.36±0.01^{c})
0.125% MF + 3% NGOE	13.33±1.36 (2.57±0.10^{cde})	6.40±0.08 (1.86±0.01^{de})
0.125% MF + 5% NGOE	10.00±1.36 (2.27±0.14^{def})	4.54±0.11 (1.51±0.02^{f})
1% Emulsifier	46.67±1.93 (3.84±0.04^{a})	30.30±4.08 (3.39±0.12^{a})
Control (no spray)	46.67±7.07 (3.80±0.18^{a})	30.61±4.01 (3.40±0.12^{a})
F-value (*df* = 8, 27)	31.76	30.49
P-value	<0.01	<0.01
Late-sown		
0.25% MF	9.17±0.83 (2.20±0.10^{e})	4.69±0.09 (1.55±0.02^{g})
1% NGOE	24.17±2.85 (3.16±0.13^{b})	19.99±0.42 (2.99±0.02^{b})
3% NGOE	16.67±1.36 (2.80±0.08^{c})	9.30±0.45 (2.23±0.05^{d})
5% NGOE	10.83±1.59 (2.35±0.16^{de})	6.41±0.17 (1.86±0.03^{ef})
0.125% MF + 1% NGOE	19.17±2.10 (2.93±0.12^{bc})	12.40±0.20 (2.52±0.02^{c})
0.125% MF + 3% NGOE	14.17±0.84 (2.65±0.06^{cd})	7.00±0.22 (1.94±0.03^{e})
0.125% MF + 5% NGOE	10.83±0.84 (2.37±0.07^{de})	5.39±0.21 (1.68±0.04^{fg})
1% Emulsifier	50.83±2.85 (3.92±0.06^{a})	41.44±6.11 (3.70±0.13^{a})
Control (no spray)	51.67±3.11 (3.94±0.05^{a})	42.66±5.77 (3.73±0.12^{a})
F-value (*df* = 8, 27)	42.23	29.16
P-value	<0.01	<0.01

Figures in parentheses are logarithmic transformed values; Means (± SE) are values of 4 replications; Means (± SE) followed by the same superscript letter(s) within a column are not significantly different using Student-Newman Keul's (SNK) test ($P \leq 0.05$)

Table 4. Individual and composite effect of Magicforce® and neem plus garlic derived biopesticide on plant survival and fruit production of Watermelon at Wukari in 2020

Treatment	Survival rate (%)	Number of fruits/ha
Early-sown		
0.25% MF	90.69±0.47 (4.51±0.01^{ab})	17165.16±488.17 (130.98±1.73^{b})
1% NGOE	62.00±3.29 (4.12±0.05^{c})	10137.79±275.29 (100.66±1.38^{e})
3% NGOE	75.17±2.81 (4.32±0.04^{b})	13858.68±158.22 (117.72±0.67^{d})
5% NGOE	82.91±0.68 (4.42±0.01^{ab})	15576.75±738.80 (124.70±2.98^{c})
0.125% MF + 1% NGOE	79.38±1.91 (4.37±0.02^{ab})	15607.75±455.11 (124.89±1.82^{c})
0.125% MF + 3% NGOE	86.07±0.61 (4.46±0.01^{ab})	16336.70±235.57 (127.81±0.92^{bc})
0.125% MF + 5% NGOE	96.98±1.66 (4.57±0.02^{a})	18615.06±457.00 (136.41±1.67^{a})
1% Emulsifier	28.63±3.45 (3.33±0.1^{d})	1585.80±52.06 (39.81±0.66^{f})
Control (no spray)	31.07±2.89 (3.42±0.10^{d})	1547.26±48.26 (39.32±0.61^{f})
F-value (df = 8, 27)	70.57	288.07
P-value	<0.01	<0.01
Late-sown		
0.25% MF	90.05±3.69 (4.50±0.04^{a})	15708.50±1411.93 (124.92±5.91^{a})
1% NGOE	59.56±3.89 (4.08±0.07^{b})	7596.94±275.51 (87.12±1.59^{b})
3% NGOE	71.03±2.45 (4.26±0.03^{ab})	12896.14±382.32 (113.52±1.71^{a})
5% NGOE	79.45±0.43 (4.38±0.01^{a})	14413.84±1162.40 (119.74±5.01^{a})
0.125% MF + 1% NGOE	75.75±2.25 (4.33±0.03^{a})	13946.24±723.80 (117.97±3.11^{a})
0.125% MF + 3% NGOE	81.51±1.02 (4.40±0.01^{a})	14525.08±1073.87 (120.25±4.62^{a})
0.125% MF + 5% NGOE	87.73±0.85 (4.47±0.01^{a})	16404.02±1526.25 (127.62±6.23^{a})
1% Emulsifier	27.09±3.37 (3.28±0.12^{c})	1417.86±85.69 (37.60±1.15^{c})
Control (no spray)	28.84±3.12 (3.34±0.12^{c})	1392.90±82.02 (37.27±1.10^{c})
F-value (df = 8, 27)	53.07	37.27
P-value	<0.01	<0.01

Figures in parentheses are logarithmic transformed values for survival rate, square root transformed for number of fruits/ha; Means (± SE) are values of 4 replications; Means (± SE) followed by the same superscript letter(s) within a column are not significantly different using Student-Newman Keul's (SNK) test ($P \leq 0.05$)

However, it positively impacted the surface tension of the oil mixture thereby promoting miscibility with water which apparently improved coverage and efficacy of the active ingredient/formulation. Aside from effecting mortality of insects, the efficacy of the formulated biopesticide as observed by way of lower leaf damage, higher plant survival rate and number of fruits produced vis-à-vis the control, could be attributed to the feeding deterring activities of the biopesticide constituents, particularly neem.

Natural enemies are important in suppressing pest populations. Cloyd (2012) opined that the application of either synthetic or natural pesticides could be harmful to the natural enemies. The use of broad spectrum synthetic insecticides, in particular, adversely affects natural enemies and other non-target pests due to their rapid killing action. The current study shows that the population density of predatory ants and spiders (which are polyphagous natural enemies) was suppressed by each of the synthetic and natural pesticides and their combination. However, sole synthetic pesticide was the most suppressive and this agrees with the report of Mkindi et al. (2017) that shows that botanicals may be less toxic to natural enemies.

That pure azadirachtin can be classified as moderately toxic to bees, as reported (Cluzeau, 2002). A similar finding was also reported with respect to a commercial garlic product (Xavier et al., 2015). The current study however showed that application of NGOE attracted comparable and even higher visits by bees to the botanically treated plots than control. While this may be attributed to higher number of flowers to visit due to more plant population in NGOE treated plots, it also showed that NGOE had little if any suppressive effects on bees.

One key limitation of botanicals however, is the need for frequent applications due to their slow action and rapid degradation. This can, however, be addressed via combination with synthetic insecticides if they are compatible. The improvement of the efficacy of NGOE when combined with Magicforce® indicates compatibility. In the long run however, the deployment of nanotechnology holds the potential to deal with this drawback since formulating biopesticides as nanoparticles could provide an effective strategy of preventing the rapid breakdown of their active ingredients.

Conclusion

Producing watermelons without suppressing pest infestation leads to very low productivity. Application of Magicforce® at 0.25%, which is the lowest field recommended dose, was efficient in watermelon pest management. However, 0.125% MF + 5% NGOE was the most efficient treatment. Neem plus garlic oil biopesticide emulsion was observed to be compatible with Magicforce®, and to be relatively more favourable to the activities of beneficial arthropods (ants, spiders and bees - *A. mellifera*) as comparably higher densities were recorded in plots so treated.

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