



The efficacy of two bio-rational pesticides on insect pests complex of two varieties of white cabbage (*Brassica oleracea* var. *capitata* L.) in the coastal savanna region of Ghana

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Abstract Insect population count and yield were used to determine the effectiveness of two bio-rational pesticides -a commercial formulation of *Pieris rapae* granulosis virus and *Bacillus thuringiensis*, Bypel 1[®] (PrGV + Bt) (1.5 g/l w/v) and aqueous neem kernel extract (ANKE) (50 g/l w/v), on the insect pests complex of two cabbage varieties, KK cross and Oxylus for two consecutive cropping seasons of 2016/2017. The experiment was laid in a split-plot design with three replications, with the cabbage varieties as main plots and the bio-rational pesticides as subplots. Cabbage plots treated with Bypel 1[®] and ANKE had the lowest population of *Plutella xylostella*, *Brevicoryne brassicae*, *Hellula undalis*, *Bemisia tabaci* and *Thrips tabaci* for both seasons. The control plot consistently had higher pest population in both seasons. There was no detrimental effect of bio-rational pesticides on natural enemies. The bio-rational pesticides-treated plots produced higher yield

than control plots. Mean numbers of *P. xylostella* was higher on KK (0.61, 1.46) compared to Oxylus (0.65, 1.70) in both seasons, respectively. The yield between the cabbage varieties was not significantly different in both seasons, although Bypel 1[®] and ANKE treated plots generally produced significantly higher yields than the control plot. Yield between both cropping seasons was significantly different ($p = 0.039$). These findings provide evidence that bio-rational pesticides may offer effective management of the pest complex of cabbage and may be successfully used as an integral part of IPM in Africa as a means to curb the abuse of synthetic insecticides.

Keywords Bio-rational pesticides · Bypel 1[®] · Aqueous neem kernel extract · Cabbage · *P. Xylostella*

Introduction

Brassica vegetables such as cabbage (*Brassica oleracea* var. *capitata* L.) which is often grown in Ghana and many other countries, are susceptible to damage by many insect pests which include: diamondback moth, *Plutella xylostella* (L) (Lepidoptera: Plutellidae), the cabbage webworm, *Hellula undalis* (F) (Lepidoptera: Crambidae), the cabbage aphid, *Brevicoryne brassicae* (L) (Homoptera: Aphididae), whiteflies, *Bemisia tabaci* (Gennadius) (Homoptera: Aleyrodidae), cabbage looper, *Trichoplusia ni* (Hubner) (Lepidoptera, Noctuidae), and thrips, *Thrips tabaci* (L) (Thysanoptera) (Chalfant et al. 1979; Shelton et al. 1988; Obeng-Ofori et al. 2007; Fail

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et al. 2013). In most areas where cabbages are cultivated, a complex of these pests occur leading to qualitative and quantitative yield losses. Due to the adverse effects of these pests on farmers' harvests, the implementation of effective management strategies is necessitated to meet market demand for healthy and pest-free cabbage heads, without blemish. Zalucki et al. (2012) and Wei et al. (2013) estimated that *P. xylostella* infestation can cause yield losses of up to 90%, with US\$ 4billion - US\$ 5 billion projected cost of control per annum. Considering that the leaves of the cabbage head are the marketable parts of the crop, the cumulative effect of insect pests feeding damage can, consequently, be excessive with a more adverse effect on the cosmetic value of the harvested cabbage head. This reduces the market value of the harvested head and consequently, the profit margins of farmers.

At the farmer level, management of insect pest infestation on cabbage has principally been using broad-spectrum synthetic insecticides. Preceding this study, earlier work done concluded that farmers in the Volta Region of Ghana applied synthetic insecticides indiscriminately to their crops, 12–15 times per growing season (N. E. Amengor per. Comm.). The net effect of these practices is the prevalence of unintended human and environmental health effects such as the development of insecticide resistance by commonly targeted insect pests and destruction of non-target organisms such as pollinators and natural enemies of pests, among others (Afreh-Nuamah 1996; Ntow et al. 2006; Fernandes et al. 2010; Fening et al. 2013, 2014). The blanket application of insecticides and disregard for pre-harvest intervals also compromises food safety as markets are flooded with cabbage heads with pesticide residues, sometimes above maximum residue limits (Asante and Ntow 2009; Armah 2011). The serious nature of the negative environmental and social impacts of the wanton use of broad-spectrum insecticides against *P. xylostella* and other cabbage insect pests calls for the re-evaluation of current and future pest management programmes to include ecologically sound and sustainable practices (Afreh-Nuamah 2000; Ntow et al. 2006; Coulibaly et al. 2007).

A possible pathway to this among others is the use of selective bio-rational pesticides augmented with crop varieties that are more tolerant or less susceptible to pest attack. Bio-rational pesticides are toxic only to the target pest, have low toxicity to non-target organisms including beneficial insects and fewer environmental hazards

(Liu et al. 1999). One of such pesticide is Bypel 1[®] (PrGV+Bt), a fully registered insecticide in Ghana with registration number 13133/00648G (issue date, October 2015) and a hazard class of II (EPA 2015). Its active ingredient comprises a mixture of *Pieris rapae* Granulosis Virus and *Bacillus thuringiensis*. *Bacillus thuringiensis* produces a toxin that causes paralysis of the digestive tract (Guerena 2006) of a susceptible insect that ingests it. This toxin breaks down the gut wall allowing spores to invade the insect body and the insect ceases to feed, and consequently causes death by starvation, septicemia and/or osmotic shock within 24 to 48 h (Rowell and Bessin 2005). On the other hand, the neem tree, *Azadirachta indica* A. Juss (Meliaceae) contains Azadirachtin, a triterpenoid that has been shown to possess growth regulating and antifeeding materials against numerous pests (Saxena et al. 1988; Schmutterer 1990; Mordue (Luntz) and Blackwell 1993; Schmutterer and Singh 1995). Though, it has been used in other areas in Ghana (Afreh-Nuamah et al. 2006; Obeng-Ofori 2008; Baidoo and Adam 2012; Forchibe 2016), there is limited knowledge on the scope of its efficacy and effectiveness in the study area, under different ecological conditions. Smith (1989) stated that, despite the overwhelming success of resistant crop varieties, their adoption in the management of insect pests of vegetable crops, such as cabbage has been very limited. The current study explored the potential of two bio-rational pesticides as effective alternatives to synthetic pesticides in vegetable insect pest management. The study also evaluated the relative susceptibility of two commonly cultivated cabbage varieties to insect pests attack. It is expected that the use of these relatively safer insecticides for pest management could minimise insecticide misuse, while at the same time ensuring a healthy lifestyle for pesticide applicators and consumers within sub-Saharan Africa.

Materials and method

A field experiment was undertaken in the Ketu-South municipality of the Volta Region of Ghana (7° 0' 0" N, 0° 30' 0" E) (Fig. 1) during the first (July–October 2016) and second (November 2016–February 2017) cropping seasons. Two cabbage varieties, KK cross and Oxylus were treated with two bio-rational pesticides; a botanical (aqueous neem kernel extract) pesticide, a microbial pesticide (Bypel 1[®] (PrGV + Bt)) and untreated controls

(sprayed with water only). The varieties selected were farmers preferred cabbage varieties in the study area. Farmers' choice for Oxylus, (locally called 'Kpeve', meaning stone) relates to its compactness and ability to produce medium-sized heads which are heavy and tasty with a long shelf-life. The farmers indicated that KK cross is an early maturing variety and produces very big heads but may not be as heavy as that of Oxylus of comparable head size. Treatments and varieties were replicated three times in a randomized complete block design with a split-plot arrangement. Main plots were cabbage varieties and sub-plots were bio-rational pesticides and untreated controls. Each plot size was 5 m long and 2 m wide (10 m²), an alley of 1.5 m and 2 m were maintained between individual plots and varieties, respectively.

Transplanting of cabbage seedlings

Four weeks old healthy seedlings of the two cabbage varieties, raised in outdoor seedbeds, were planted 50 cm apart on raised beds spaced 50 cm apart on 2nd August and 8th December, corresponding to two cropping seasons, respectively. Standard agronomic practices including watering (morning and evening at planting, but later reduced to once every 2 days as the plants matured), hand weeding at two weeks interval and fertilizer application (180 ml/plant of NPK 15–15–15 and 3 g/plant of Sulphate of Ammonia at 7 and 42 days after transplanting, respectively) were undertaken.

Preparation and application of treatments

Freshly dropped neem fruits and fully ripped neem seeds were harvested/collected from neem trees. The fruits were de-pulped and dried in the shade for 14 days at room temperature (28 ± 2 °C). Dried seeds were later stored in baskets in a dry and well-ventilated room to prevent the development of mould. About 50 g of the seeds were crushed in a wooden mortar using a wooden pestle when needed, dissolved in one litre of water and stirred. Two drops of liquid soap (Morning Fresh®) were added to the mixture to break the surface tension and enhance its stickiness on to the leaf surfaces (Fening et al. 2014). The mixture was allowed to stand overnight and later filtered using a fine cloth so that the pure extract containing the active ingredient (azadirachtin) was used for spraying (Afreh-Nuamah 1996; Obeng-

Ofori 2008). The commercial biological insecticide, Bypel 1® (PrGV + Bt) consisting of a mixture of *Pieris rapae* Granulosis Virus (PrGV) (10,000 PIB/mg) and *Bacillus thuringiensis* (Bt) (16,000 IU/mg) was used as a standard check. The spray liquid was obtained by mixing 1.5 g/l of water as per the manufacturer's recommendation. Bio-rational insecticide treatments were applied after scouting at 21 days after transplanting of cabbage seedlings when insect pests were detected during the first season and 14 days after transplanting in the minor season due to the early occurrence of insect pests on the field. The treatments were applied using a 15 l lever operated Jacto knapsack sprayer with a cone nozzle. Spraying was carried out in the evening (5:00–6:00 pm) to prevent photo-breakdown of chemicals and maximum coverage of leaves including the lower leaf surfaces where pests normally hide. This was repeated weekly until the cabbage heads were fully matured (firm to hand pressure when touched), about 14 days to harvesting (Fening et al. 2013, 2014).

Data collection

For two consecutive cropping seasons, treatment plots were sampled weekly for pests for six and seven weeks (starting 3 weeks and 2 weeks after planting), respectively. The leaves of 10 randomly selected plants from the two inner rows of each plot were closely inspected and all the lepidopteran insect larvae (*P. xylostella*, *H. undalis*) observed were counted, the aphid, *B. brassicae* (nymphs and adults) was scored using a scale of 0–5 (Fening et al. 2014), ranging from the least to the highest infestation, thrips and whiteflies (nymphs and adults) observed on leaves of selected plants were equally counted. Observed natural enemies were also counted on the 10 randomly selected plants from the inner rows of each plot. Data were taken weekly after treatment application in the morning between 6:00–8:30 am when pests were less active. At harvest, 15 plants per treatment plot were selected at random from the two innermost rows for yield assessment. The yield per unit area was extrapolated into tonnes per hectare (t/ha).

Identification of insect pests and natural enemies

The insect pests and their natural enemies collected in this study were taken to the Entomology Laboratory of the African Regional Post Graduate Programme in

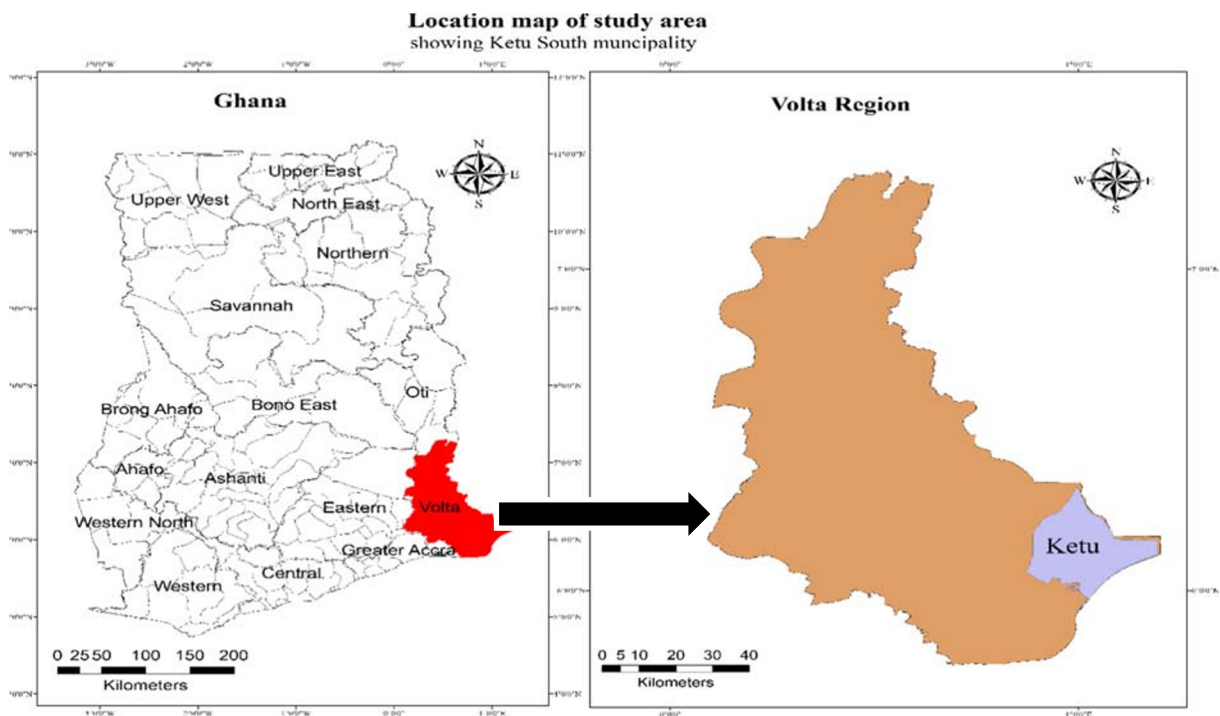


Fig. 1 Map of the study area

Insect Science for identification using morphological features and comparing with reference specimens at the Entomology laboratories of the Departments of Crop Science and Animal Biology and Conservation Science, University of Ghana.

Data analysis

The number of insect pests and natural enemies per 10 plants was divided by sampling weeks to give mean insect number per plot. Data were analysed using a split-plot design option of the analysis of variance (ANOVA) in GenStat statistical software (version 12.1) to determine the effect of treatment, varieties and treatment*variety interaction. Shapiro-Wilk test was used to check for normality of the data ($p > 0.05$) after which square root transformation was performed on all count data before analysis. The analysis was performed separately for each season. Untransformed means are reported in the figures presented. Where significant differences were observed, the mean separation was performed using Fisher's least significant difference (LSD) test at 5%. A student's T test was used to compare the pest population and yield data between the two cropping seasons.

Results

The lowest mean number of diamondback moth, *P. xylostella* was recorded on Bypel 1[®] (PrGV + Bt) plots, followed by aqueous neem kernel extract (ANKE) plots, the control plots recorded the highest population for both seasons (Fig. 2a and b). Bypel 1[®] and ANKE treatments recorded significantly lower numbers than the control ($F_{2, 8} = 5.05$, $P = 0.038$ and $F_{2, 8} = 101.70$, $P < 0.001$) for the two seasons. The population of *P. xylostella* was higher on KK cross than on Oxylus for both seasons. There was no significant statistical interaction between cabbage varieties and treatments in the first ($F_{1, 2} = 0.04$, $P = 0.960$) and second cropping seasons ($F_{1, 2} = 0.86$, $P = 0.458$). Results of the Student's t test revealed a significantly higher DBM infestation in the second season ($t = -2.87$, $p = 0.011$).

The population of the cabbage aphid, *B. brassicae* was less on Bypel 1[®] (PrGV + Bt) and ANKE - treated plots, with the control plots having the highest numbers in both seasons (Fig. 3a and b). It was observed that differences were not significant between varieties in both seasons ($F_{1, 2} = 0.01$, $P = 0.927$ and $F_{1, 2} = 0.05$, $P = 0.727$), but KK cross had a higher *B. brassicae*

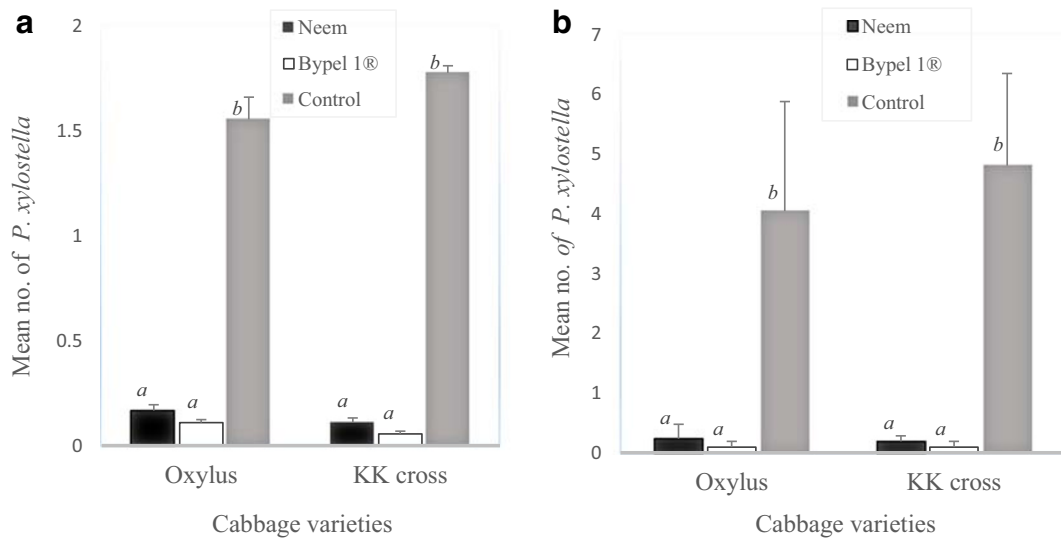


Fig. 2 Mean number (\pm S.E) of *Plutella xylostella* per plot in the first (a) and second (b) cropping seasons of 2016/2017

population than Oxylus. *Brevicoryne brassicae* infestation was higher in the second than the first season.

Cabbage plants in Bypel 1® plots did not record any *H. undalis* infestation in the first season but had the least population in the second season, whilst ANKE had consistently fewer numbers of this pest with the highest on control plots in both seasons (Fig. 4a and b). The susceptibility of cabbage varieties was not significantly different in both seasons ($F_{1, 2} = 0.14$, $P = 0.742$ and $F_{1, 2} = 0.05$, $P = 0.843$). The statistical interaction between

treatments and cabbage varieties did not significantly improve the management of this pest in both seasons ($F_{1, 2} = 0.52$, $P = 0.612$ and $F_{1, 2} = 0.31$, $P = 0.744$, respectively).

Bypel 1® and ANKE treated plots had a significantly lower whitefly, *B. tabaci* population than that observed in the control plots ($F_{2, 8} = 29.08$, $P < 0.001$ and $F_{2, 8} = 26.36$, $P < 0.001$). The least infestation occurred on Bypel 1® treated plots (1.67) in the first season but in the second season, it was on ANKE plots (55.7) (Fig. 5a and

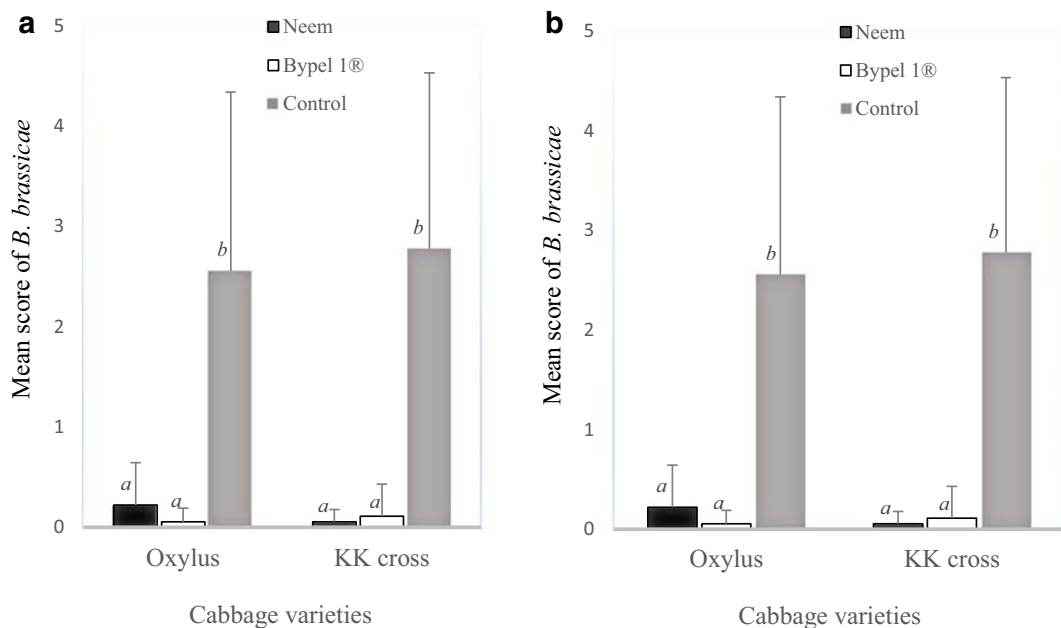


Fig. 3 Mean score (\pm S.E) of *Brevicoryne brassicae* per plot in the first (a) and second (b) cropping seasons of 2016/2017

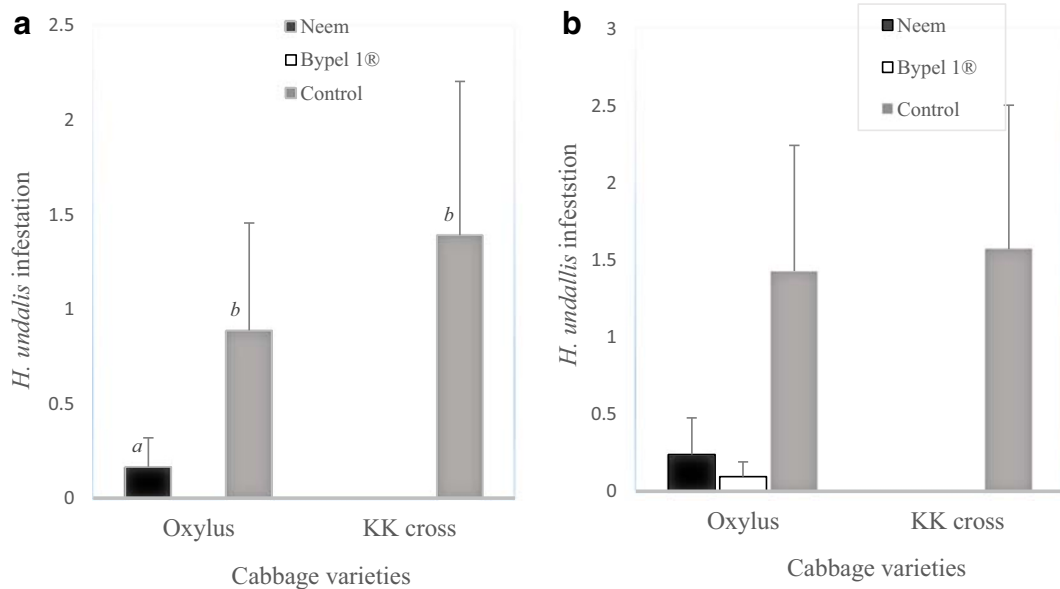


Fig. 4 Mean number (\pm S.E) of *Hellula undalis* per plot in the first (a) and second (b) cropping seasons of 2016/2017

b). Mean numbers of *B. tabaci* was highest on Oxylus (3.09) compared to KK cross (2.69) in the first season. A significantly high whitefly population was recorded in the second than the first cropping season ($t = -5.92$, $p < 0.001$).

Bypel 1® and ANKE treated plots recorded lower populations of *T. tabaci* for the two seasons compared to the control plot which had the highest population in both seasons (Fig. 6a and b). Differences in the susceptibility of Oxylus and KK cross to thrips infestation was not

significant for both cropping seasons ($F_{1, 2} = 2.19$, $P = 0.175$ and $F_{1, 2} = 0.47$, $P = 0.565$). The population of thrips did not differ significantly between the first and second cropping seasons ($t = 0.33$, $p = 0.745$).

Bypel 1® and ANKE plots had a lower mean population of other pests, mainly *Trichoplusia ni* and *Zonocerus variegatus*, in both seasons as opposed to the highest population in the control plot. Differences between treatments were not significant ($F_{1, 2} = 0.36$, $P = 0.710$ and $F_{1, 2} = 0.37$, $P = 0.701$). Oxylus had

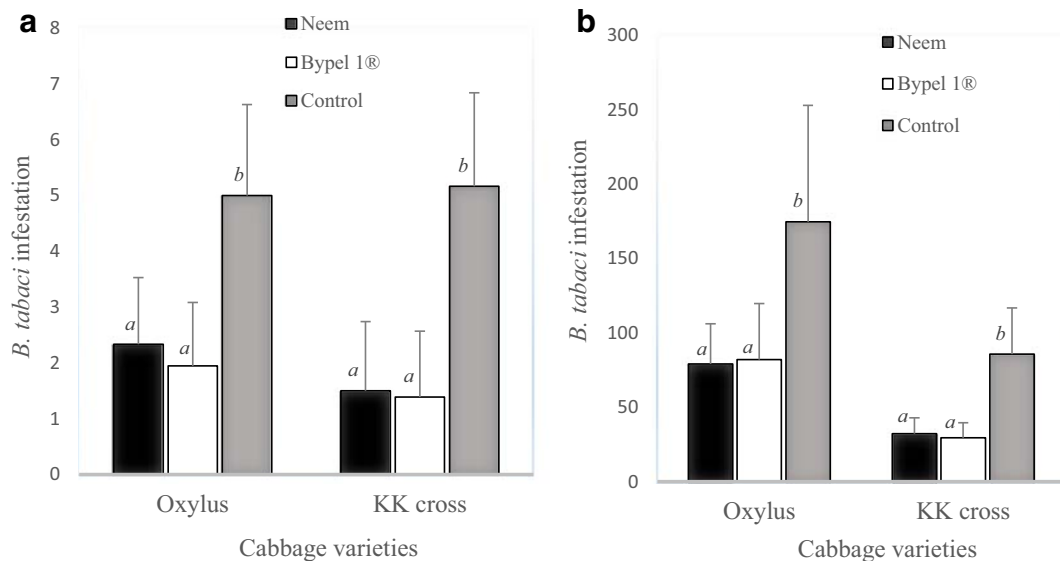


Fig. 5 Mean number (\pm S.E) of *Bemisia tabaci* per plot in the first (a) and second (b) cropping seasons of 2016/2017

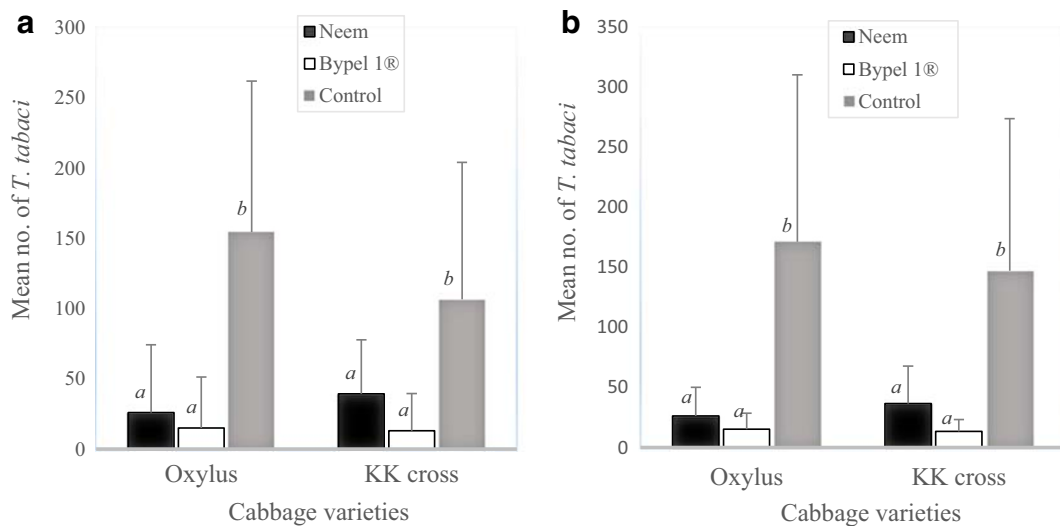


Fig. 6 Mean number (\pm S.E) of *Thrips tabaci* per plot in the first (a) and second (b) cropping seasons of 2016/2017

higher pest numbers than KK cross. Their populations were generally very low in the first cropping season.

Differences in the abundance of predators (Table 1), notably hoverflies, ladybird beetles, *Cheilomenes* spp. (Coccinellidae) and spiders (Araneae) during the first season ($F_{2, 8} = 0.51$, $P = 0.619$, $F_{2, 8} = 1.50$, $P = 0.280$ and $F_{2, 8} = 0.39$, $P = 0.686$), were respectively not significant. In the second season, lady bird beetles were absent and hoverflies and spiders population was not significant among treatments ($F_{2, 8} = 3.36$, $P = 0.086$, $F_{2, 8} = 0.47$, $P = 0.641$). The larval endoparasitoid of DBM, *Cotesia plutellae* (Hymenoptera: Braconidae) was observed only in the second season and was highest on Bypel 1® - treated plots. Oxylus had a significantly higher spider population in the second season than KK cross ($F_{1, 2} = 48.00$, $P = 0.020$).

Differences in yield obtained between the cabbage varieties in both seasons were not significant ($F_{1, 2} = 0.25$, $P = 0.667$ and $F_{1, 2} = 2.25$, $P = 0.272$, respectively), however cabbage plots sprayed with Bypel 1® and ANKE generally obtained significantly higher yields than the untreated control plots.

Means with the same letter(s) are not significantly different ($P < 0.05$, LSD).

Discussion

With a complex of pests damaging cabbage heads, a reduction in yield or complete crop failure might have resulted if effective management strategies were not

instituted. Cabbage insect pest management strategies that increases yields and enhance food and environmental safety are currently more desirable. The two bio-rational pesticides evaluated in this study were both effective in reducing insect pest damage and yield losses and were also less detrimental to non-target arthropods. The results are in consonance with work by Sow et al. (2013) who recorded an increase in head size due to effective control of *P. xylostella* when neem, biobit® (*B. thuringiensis*) and neem/biobit® rotated were used on cabbage, with no significant differences between them. Li and Sengonca (2003) observed high efficacy of GCSC-BtA (Germany-China Scientific Cooperation - *Bacillus thuringiensis* - Abamectin) biocide on key cabbage pests whilst Ivey and Seth (1997) in a related study, reported that products containing effective strains of *B. thuringiensis* can be successfully used to manage *P. xylostella* with a resultant increase in yields. According to Guereña (2006), early instar larvae of *P. xylostella* are more susceptible to *B. thuringiensis* whilst older larvae are harder to kill. In this study, the dead *P. xylostella* larvae recovered from Bypel 1® plots consisted of individuals of different larval stages suggesting that the action of Bypel 1®, which consists of a mixture of *P. rapae* granulosus virus and *Bacillus thuriengensis* was synergistic. Aqueous neem kernel extract did not significantly differ from Bypel 1® in terms of effectiveness and significantly reduced *P. xylostella* and *H. undalis* population. The effectiveness of crude seed extracts of neem have been reported in other studies (Obeng-Ofori 2008; Lidet et al. 2009;

Table 1 Mean number (\pm S.E) of natural enemies and yield (t/ha) per plot during the first and second cropping seasons

Treatment	Variety	Hoverfly		Spiders		Ladybird		<i>C. pluteellae</i>		Yield(t/ha)	
		First	Second	First	Second	First	Second	Second	First	First	Second
Bypel 1 [®]	Oxylus	0.167 \pm 0.096	0.476 \pm 0.191	2.722 \pm 0.747	1.810 \pm 0.252	–	–	0.111 \pm 0.110	–	77.16 \pm 13.83a	61.33 \pm 4.37a
ANKE	Oxylus	0.278 \pm 0.200	0.095 \pm 0.095	2.556 \pm 0.433	1.333 \pm 0.530	0.056 \pm 0.054	–	0.056 \pm 0.054	–	64.44 \pm 4.15a	57.87 \pm 6.94a
Control	Oxylus	0.167 \pm 0.096	1.048 \pm 0.191	4.889 \pm 1.123	0.762 \pm 0.343	–	–	0.111 \pm 0.110	–	30.87 \pm 5.44b	35.87 \pm 1.94b
Bypel 1 [®]	KK cross	0.833 \pm 0.674	0.381 \pm 0.095	4.333 \pm 1.776	1.143 \pm 0.594	0.222 \pm 0.111	–	0.222 \pm 0.111	–	73.48 \pm 7.67a	54.53 \pm 3.43a
ANKE	KK cross	0.167 \pm 0.096	0.286 \pm 0.165	3.556 \pm 1.106	0.667 \pm 0.191	0.056 \pm 0.005	–	0.054 \pm 0.053	–	63.32 \pm 5.21a	46.53 \pm 0.93ab
Control	KK cross	0.444 \pm 0.200	0.095 \pm 0.095	2.778 \pm 1.169	0.952 \pm 0.415	–	–	–	–	30.67 \pm 0.67b	31.87 \pm 2.69b
F		0.51	3.36	0.39	0.47	1.50	–	0.94	–	17.4	25.62
P		0.619	0.0861	0.6862	0.641	0.280	–	0.429	–	0.001	< 0.001

Kibrom et al. 2012; Sow et al. 2013; Prasannakumar et al. 2014; Forchibe 2016; Ezena et al. 2016) against insect pests of tomato, cabbage, cucumber, okra, pepper and garden eggs. The use of neem in pest management relates to its repellent, oviposition deterrent and natural insecticidal properties as explained by Saxena et al. (1988).

The results of this study suggest that the treatment of cabbage plants with Bypel 1[®] and ANKE had no or minimal detrimental effects on natural enemies of pests, similar to previous studies by Owusu-Ansah et al. (2001); Obeng-Ofori and Ankrah (2002) and Rowell and Bessin (2005). Other studies also reported that whilst neem oil formulation was effective against *Lipaphis erysimi*, it did not have any detrimental effect on its predator, the hoverfly *Ischiodon scutellaris* (Boopathi and Pathak 2011). These low-risk pesticides appear to conserve natural biocontrol agents which play a significant role in reducing insect pest population on infested cabbage farms.

Although Oxylus produced medium-sized cabbage heads relative to the larger heads of KK cross, the observed weights of heads of the two varieties were similar. This could be attributed to the compact nature of Oxylus heads, with well-arranged wrapper leaves. Close observations of infested cabbage plants in treatment plots appear to suggest the low infestation of Oxylus variety by *P. xylostella* is based on leaf texture. Physical examination of the leaves of both varieties indicates that the leaf epidermis of Oxylus is relatively thicker than that of KK cross, a feature which may hinder the mining activity of early instar *P. xylostella* larvae. Although KK cross recorded higher numbers of aphids in this study, Lal. (1989) reported that the variety was less susceptible to aphid (*B. brassicae*) infestation. The lack of significant interaction in the populations of major pests sampled and yield is indicative that the efficacy of the different insect pest management strategies evaluated in this study was similar on both varieties.

The heavier cabbage head weights recorded in the plots treated with the two bio-rational insecticides used in this study could be attributed to the effective reduction of insect pest population and damage on treated cabbage plants. This, coupled with the low observed adverse effects on non-target arthropods may provide a clue to the potential utility of the two bio-rational insecticides as part of a broad integrated pest management (IPM) strategy for cabbage pests in the study area. Of

recent, since most consumers are conscious of the safety of what they eat, the widespread adoption of such an IPM programme by farmers will minimize the risk of vegetable contamination and ensure consumer safety, since most vegetables such as cabbages are eaten raw (Fening et al. 2013, 2014).

Conclusion

The two bio-rational pesticides (Bypel 1[®] and ANKE) evaluated effectively managed the insect pest complex on cabbage while conserving their natural enemies leading to increased yields. Results of the study also indicates that the use of the two bio-rational insecticides is effective regardless of the cabbage variety. Bypel 1[®] and ANKE are environmentally friendly pest management options. Substituting the use of broad-spectrum synthetic insecticides in the cultivation of cabbage with these could increase vegetable yield and ensure food and environmental safety and enhanced nutrition.

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Compliance with ethical standards

Conflict of interest The authors declare no conflicting interest.

Code availability Not applicable.

Consent to participate Not applicable.

Ethical approval Not applicable.

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