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Exploitation of Selected Plant Based Insecticides for Field Management and Control of Aphids in Organic Cabbage Production

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Abstract
Large colonies of cabbage aphids are capable of stunting and or killing small plants. They are also capable of contaminating harvested crop. The use of biodegradable botanicals for pest control is now considered one of the best alternatives to synthetic pesticides; and they are known to be safer and ecological friendly. This study was conducted to evaluate the effect of three plant based extracts and their mixtures for their efficiency in the control of aphids in a cabbage field. Extracts from *Azadirachta indica, Chrysanthemum cinerariifolium, Quassia amara*, their mixtures and a positive and negative control were used. Two field studies were carried out at different periods (spring and late summer studies). Decline in aphid infestation was determined by recording number of infested plants per plot and the average number of aphids per plant; before and after each treatment application. All extracts and their mixtures decreased aphid population significantly ($p<0.001$) in both seasons. A mean pest population decline of 86.78% and 85.48% for number of aphids/plant was recorded in the spring and late summer study respectively; and an 86.00% and 85.48% decline in number of plants infested for the spring and late summer study respectively. Decline in infestation produced by all extracts was comparable to that of the synthetic insecticide used for the study.
Keywords: Cabbage, Organic pest control, Bio-pesticide, Azadirachtin, Pyrethrin, Quassin, Plant extracts.

1. Introduction

Cabbage (*Brassica oleracea* L. var. capitata) is a member of the Brassicaceae family. Cabbage is produced globally including higher elevations in tropical and subtropical areas where temperatures are cooler. Cabbage is attacked by several insect pests including aphids. Per Ahmad and Akhtar (2013), under favourable conditions, feeding damage from large number of aphids can kill seedlings and young transplants, on larger plants, feeding results in curling and yellowing of leaves, stunting growth and damage to flowers. The success of this pest is most of the time attributed to evolution. The pace of evolution of these pests has hastened in order for them to compete with chemical control strategy intended against them. This has resulted in the expansion of insecticide resistance in aphid species (Sarwar et al., 2011; Sarwar, 2013). Thus, many pests including aphids have developed resistance to organophosphates, carbamates and pyrethroids insecticides (Ahmad and Akhtar, 2013); and, an increase in pest populations because of reduced populations of natural enemies (Dube et al., 1998).

Several other pest control progresses have also so far been achieved for this pest; these includes the exploitation of botanicals for its management. And in recent years, organic pest control agents have proved successful for pest management strategy (Sarwar, 2010; Ahmad et al., 2011; Sarwar et al., 2012). The use of natural pesticides is also progressively becoming recognized as one option to address yield losses due to pest attacks as well as reducing environmental degradation associated with the use of synthetic pesticides (Kopondo, 2004). Botanical components are also very suitable in comparison to synthetic insecticides because they have a reputation of being environmentally harmless and less dangerous to humans and other non-target organisms (McCloskey et al., 1993). They are also broad spectrum nature, unique in action, can easily be processed and are safe to apply. Locally available plant materials have been widely used in the past to protect plants from damage caused by insects (Golob and Webly, 1980). They include crude extracts and isolates or purified compounds from various plants species and commercial products (Liu et al., 2006).

To enhance the efficacies of botanical pesticides, and or to help reduce the over dependency of synthetic insecticides; and, to minimize the rate by which target species could develop resistance. It is believed that diversifying compounds used in pest control and the use of insecticides with different modes of action could delay resistance development. In a laboratory experiment conducted by Jababu et al (2016) against diamondback moth larvae, it was revealed that mixture of Neem and Pyrethrum; each with a different mode of action, proved effective against the diamondback moth. This can also be applicable to aphids, and can help minimize the rate at which resistance could be developed against botanical extracts.

The objective of this study was therefore to evaluate the efficacies of widely used botanical extracts from the Pyrethrum, Neem and Quassia amara and their mixtures for the control and management of aphids in a cabbage field.

2. Materials and Methods

2.1. Study Site

The experiment was conducted in the experimental field of the Faculty of Horticulture of the Mendel University in Brno in two separate fields and seasons (for the summer crop and late summer crop) in the 2016 cropping year. Cabbage seeds (Hornet F1, obtained from Moravo Seed Ltd in Czech Republic) were sown and maintained under Greenhouse conditions until transplanting.

2.2. Botanical Extracts/Treatments Used for the Study

The experiments consisted of 9 treatments including a positive (+) and a negative (-) control, and each replicated five times. Each treatment consisted of two concentrations (C1 and C2; C2 being 50% dilution of C1 concentration of the various formulations). The (+) and (-) controls were Karate (Zeon 5 CS) and plain water with few drops a detergent respectively. Other treatments were extracts of Neem; *Azadirachta indica*, Pyrethrum; *Chrysanthemum cinerariifolium* and Quassia wood chips from *Quassia amara* and a 1:1 mixture of the individual extracts and 1:1:1 mixture of all the three species. The neem and pyrethrum were commercial formulations with azadirachtin and pyrethrin being the active ingredients respectively, and azadirachtin and pyrethrin contents equivalent to 10.6 g/l, and 4.59 g/l respectively. Treatments with quassin being the active component were prepared by soaking quassia wood chips overnight in water and heated for 30 minutes, following procedures similar to those described by Dodia et al. (2008) and Zijp and Blommers (2002) until wood chips settled at the bottom. The supplier did however not specify the amount of quassins in the wood chips, but it is known that the source of quassin and neoquassin is the wood of *Quassia amara* L. (Sapindales: Simaroubaceae); and contains 0.14–0.28% of quassinoids (quassin and neoquassin), depending on the age (Villalobos et al., 1999).

Treatment concentrations (C1) were as follows: Azadirachtin (A): [2% v/v], Pyrethrin (P): [2% v/v], Quassin (Q): [3% w/v], A + P: [1:1of 2% A + 2 % P], A + Q: [1:1 of 2% A + 3% Q], P + Q: [1:1 of 2% P + 3% Q], A + P + Q: [1:1:1 of 2% A + 2% P + 3% Q], plain water with drops of a detergent and 0.01% Karate (Zeon 5 CS).

2.3. Experimental design and Procedure

The experiment consisted of two separate Randomized Complete Block Designs (RCBD) opposite to each other by 1.5m. Each represented treatment for the C1 and C2 treatment concentrations used for the study. The study was undertaken on two different field locations and planting seasons. Six weeks old cabbage seedlings were transplanted on 5th May 2016 and 10th July 2016 unto the field for the Summer crop and Late Summer planting season respectively. After transplanting, seedlings were protected from field insects by covering them with pest-exclusion nets for two weeks; to allow better stand establishment. For the summer crop, the study span from May-July, and July - September; for the late summer crop.

Four spraying regimes (treatment applications) were made with a 2L knapsack sprayer for both the summer and late summer crop. Treatment applications were carried out for each of the concentrations (C1 and C2) on the same day at a ten-day interval after each application (spraying). For the summer crop, treatment applications were made on 06/06/16, 16/06/16, 26/06/16 and 06/07/16 for the 1st to 4th treatment applications respectively. For seedlings transplanted for the late summer crop, treatments were applied on 08/08/16, 18/08/16, 28/08/16 and 07/09/16 for the 1st to 4th treatment applications respectively.

2.4. Data Collection and Analysis

Number of plants infested with aphids on each plot, and the average number of wingless aphids on the leaves and terminals on 3 selected plants per replication were counted and recorded one day before and after each treatment application.

This was followed by determining the rate of decline/increase in number of plants infested by aphids and the average number of aphids on the selected plants at the end of all the spraying regimes. Decline/Increase in aphid infestation was calculated with the formula: % Decline/Increase = [(Population before Treatment) – (Population after treatment)] ÷ [Population before treatment] × 100. Data obtained was analyzed with the statistical package STATISTICA 12 (Stat Soft Inc.). Factorial ANOVA ($P < 0.05$) was performed to evaluate the effect of the extracts at the end of the study. This was followed by a Tukey's HSD test ($P < 0.05$) to identify significant differences among means.

3. Results

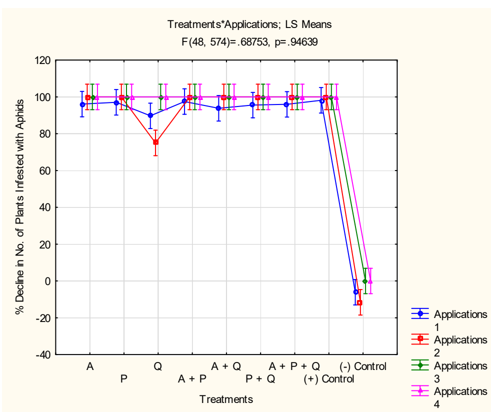
All botanical extracts used in this experiment exhibited varying levels of efficacies against the cabbage aphid; with most them resulting in a reduction of infestation comparable to that of Karate (Zeon 5 CS) by the end of the fourth treatment application in both the summer and late summer crop. In table 1; a summary of mean reduction in pest infestation for number of plants infested with aphids, and average number of aphids per plant for both the summer crop and late summer crop is recorded.

Table 1. Decline in Number of plants infested and Number of Aphids per Plant

Active Components of Treatments	% Decline in Number of Plants Infested with Aphids				% Decline in Average Number of Aphids per Plant			
	Spring Transplants		Late Summer Transplants		Spring Transplants		Late Summer Transplants	
	Mean	SD	Mean	SD	Mean	SD	Mean	SD
Azadirachtin (A)	99.03	3.53	97.38	6.75	98.37	5.02	86.00	32.13
Pyrethrin (P)	99.27	3.25	97.91	8.33	99.21	3.54	96.00	16.80
Quassin (Q)	91.18	32.52	94.49	12.08	92.83	15.13	80.14	42.78
A + P	99.38	2.92	99.05	3.39	98.92	3.91	93.14	23.11
A + Q	98.46	4.50	95.88	8.42	97.51	6.04	87.88	32.09
P + Q	98.89	4.02	98.35	4.72	98.72	4.06	92.53	24.03
A + P + Q	98.99	3.71	98.54	4.54	98.80	4.76	93.40	20.84
(+) Control	99.54	2.03	99.50	3.16	100.00	0.00	95.00	22.07
(-) Control	-4.43	9.47	-11.73	17.58	-3.33	16.24	-10.74	19.02
Total	**86.70**	**34.37**	**85.49**	**35.54**	**86.78**	**33.00**	**79.26**	**41.85**

Average Decline in Number of plants infested and Average Decline in Number of Aphids per Plant at the end of the Fourth Treatment Application (Means expressed in %).

Effect of the different treatment concentrations used for the study were significant in both planting seasons; treatment with highest concentrations resulted in the highest infestation decline. Decline in aphid infestation and trends of the decline is shown in figures 1-8.



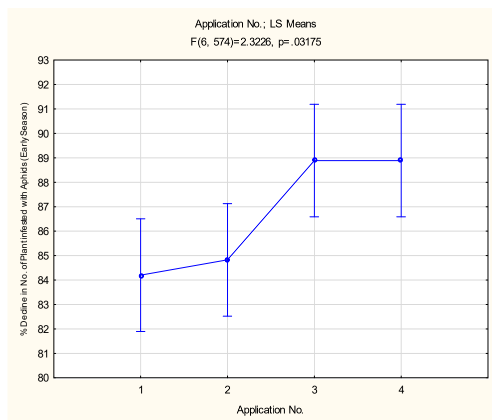
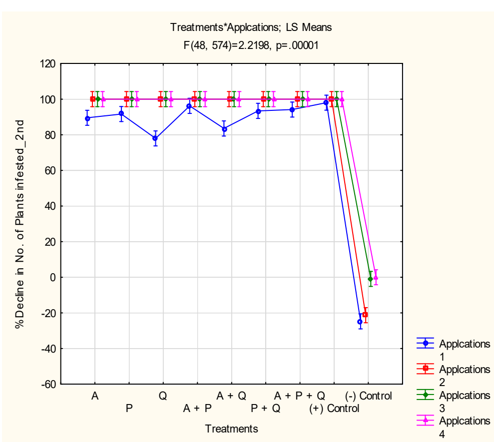


Figure 1 and 2 (Right). Effects of Extracts on Number of Plant infested with Aphids after Spraying of Extracts, and Trend of infestation Decline after sprayings in the Early Season



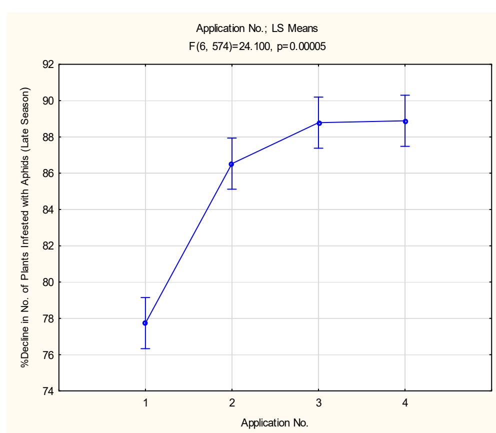


Figure 3 and 4 (Right). Effects of Extracts on Number of Plant infested with Aphids after Spraying of Extracts, and Trend of infestation Decline after sprayings in the Late Season

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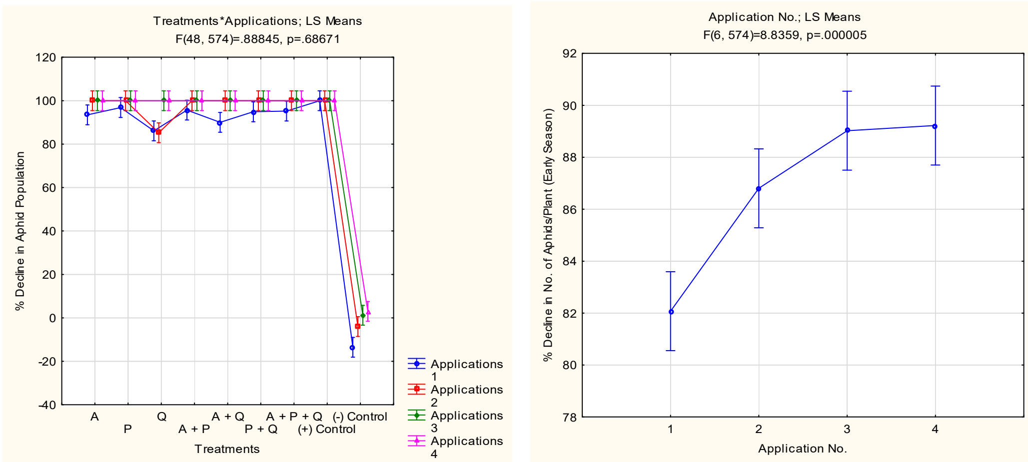

Figure 5 and 6 (Right). Effects of Extracts on Number of Aphids per Plant after Spraying of Extracts, and Trend of infestation Decline after sprayings in the Early Season

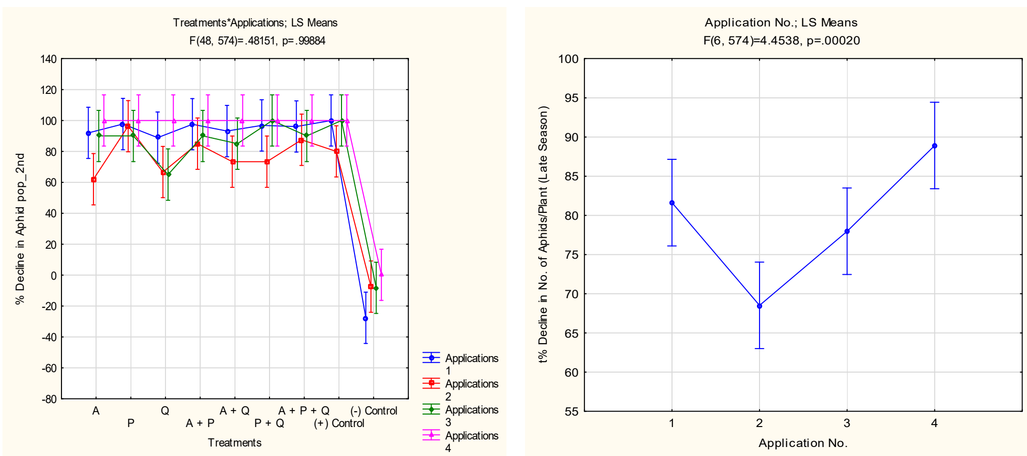

Figure 7 and 8 (Right). Effects of Extracts on Number of Aphids per Plant after Spraying of Extracts, and Trend of infestation Decline after sprayings in the Late Season

3.1 Extracts effect on Number of Plants infested with Aphids and Population of Aphids per Plant

Results from both the early and late season indicated that all treatments compared to the negative control produced a highly significant effect ($p=0.00001$). Decline in pest population produced by extracts from the pesticidal plants and their mixtures were also comparable to population decline produced by the Karate (Zeon 5 CS).

Results also revealed a similar rate of decline on 'number of plants' infested with the pest in both the summer and late summer study. All treatments also produced a significant effect in comparison to the negative control ($p=0.00001$). Decline in number of plants infested with the pest after extracts application was also again similar to that of the synthetic pesticide (Zeon 5 CS). In all cases, decline by C1 treatment concentration were higher than in the C2 treatment concentrations ($P < 0.05$).

In both results for 'number of plants infested with aphids' and the 'average number of aphids per plant', formulation with only Quassin as the active component produced the least effect. Even though infestation decline of 91.18% and 80.14% for number of plants infested with aphids and the average number of aphids per plant respectively produced by the formulation.

Comparing the effects of application regimes; also revealed some differences in efficacies between the 1st to the 4th treatment applications even though not statistically significant in most cases for both the summer and late summer crops for both the number of plants infested with aphids and the average population of aphids per plant. The only exception is seen in fig 7; where the treatment application regime resulted in a significant effect ($p< 0.05$). Also, in most instances, decline in aphid infestation was gradual; thus, decline in infestation was highest after the 4th treatment application.

4. Discussion

It is obvious everybody would prefer a clean and good quality farm produce devoid of foreign particles including infestation by pests at the kitchen. It is however unfortunate that nature alone does not guarantee that. But putting in place the right strategies and mechanisms can limit the impact of organisms and conditions that are a threat to achieving the desired quality. In recent years, attention has been paid towards exploitation of higher plant products as novel chemotherapeutics for plant protection because they are mostly non-phytotoxic and easily biodegradable; however, quantitatively the most important botanical is pyrethrum, followed by neem, rotenone and essential oils (Isman, 2006). Isman also believes pyrethrum, Quassia extract, and neem; are among the most used botanical insecticides. As a result, it is not surprising that all extracts used in this study produced a satisfactory pest infestation decline in all stages of the experiment. The decline in infestation was comparable to the decline recorded by the synthetic insecticide [Karate (Zeon 5 CS)] used in the study; this in a way could be a reason for their wide usage in the fight against most pests.

Among all the extracts used for the study, treatments with Pyrethrin as the only active component produced the best effects and the least effects was recorded from treatments with only Quassin as the active component. Results from the mixtures also revealed that treatments with Pyrethrin as one of the active component performed better than the rest. However, Pyrethrin and azadirachtin mixture is preferable the best, even though differences in treatments effect was insignificant in most cases. In spite of the minor differences in effect recorded among extracts from the three species, our findings are still in line with observations made by Isman (2006), with regards to the efficacies of the selected plant species. This also suggests that the selected botanical extracts used for this study and their mixtures could be valuable in control and management of aphids in the field.

Per Mrak (1973); Pyrethrum has quick insecticidal action against a broad range of insects, including cockroaches, mosquitoes, fleas, flies and aphids. Brown (2006) also believes pyrethrum immediately paralyses the insects; known as "knockdown", which is followed later by death. These properties exhibited by pyrethrins, makes it a better alternative pest control product. It can also be used to boost the insecticidal activity of other extracts in the form of mixtures. In our experiment, the insecticidal action of the pyrethrins was widely noticed. Formulations with pyrethrin as the only active ingredient and mixtures containing pyrethrin as one of the active components, were very effective against the cabbage aphid; they produced the highest population decline in comparison to all other extracts.

Azadirachtin, on the other hand is a chemical complex found in seeds of neem and is the main component responsible for the toxic, repellent, antifeedant, growth-inhibiting, oviposition-inhibiting and sterilizing effects in insects (Ahmed and Grainge, 1986; Schmutterer, 1990; Martinez, 2002). It is also known to be effective against more than 400 insect species belonging to the orders Diptera, Hymenoptera, Coleoptera, Lepidoptera, Orthoptera and Hemiptera (Martinez, 2002). Extract of the neem according to Ulrichs et al., (2001), reduces the population of several aphid species in many crops, causing high mortality and decreasing fecundity, as well as inhibiting population growth.

Our observations, proved the efficacies of pyrethrin and azadirachtin for cabbage aphid control; both extracts, individually and their mixtures produced a significant decline in the pest population in both seasons. Decline in infestation was however lower for azadirachtin compared to the pyrethrins. Nonetheless, the decrease in infestation produced by the formulations of azadirachtin was highly very significant; thus, even though its effect was second to the pyrethrin, the decline in aphid population produced was substantial.

This study also confirmed the insecticidal action of Quassin for the management of the cabbage aphid. But contrary to conclusions made by Weihrauch et al (2008) that the systemic application of quassin has to be regarded currently as the method of choice for aphid control in organic hop growing, our findings proved otherwise; formulations with Quassin as a component or a sole agent, produced the least effect in all cases for aphid control in cabbage.

Throughout the experiment; for the summer crop and late summer studies, a consistent decline in the average number of aphids per plant and number of plants infested with aphids was observed, except in the negative controls where a rise in population of aphids/plant and number of plants infested was recorded. The decrease in infestation observed in both studies also followed a similar pattern. The declines recorded was also in line with the number of applications made; a progressive decline in infestation was realized from the first to the fourth extract application regimes, even though not significant in all cases. Results also revealed that treatments with only *Azadirachta indica* (azadirachtin) and *Chrysanthemum cinerariifolium* (Pyrethrin) as the only active component, were the most effective. In the case of the mixtures, pyrethrin + azadirachtin combination was the best. Also, all the mixtures (Azadirachtin + Pyrethrin, Azadirachtin + Quassin, Pyrethrin + Quassin and Azadirachtin + Pyrethrin + Quassin) recorded very similar decline in infestations comparative to the positive control. The mixtures also revealed an efficient interaction among their individual components, and thus suggests the use of formulations with different active components could play a great role in controlling the pest and to minimise the rate by which the pest can develop resistance against the individual components.

5. Conclusion

Formulations with azadirachtin, pyrethrins, quassin, and their mixtures are all effective for the control and management of cabbage aphids; all extracts and their mixtures resulted in a significant reduction in aphid infestation by the end of the fourth treatment application regime; an indication that these formulations could serve as an efficient alternative for the control and management of the pest. However, azadirachtin, and pyrethrin in their individual form and the following binary mixtures: azadirachtin + pyrethrin, and pyrethrin + quassin, were found more favourable; and could provide a very effective field control for aphids when incorporated with an effective integrated pest management strategy.

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