STUDY ON THE COMBINED IMPACT OF CITOWETT AND COMMERCIAL NEEM EXTRACT AGAINST *PIERIS BRASSICAE* L. UNDER LABORATORY CONDITIONS (LEPIDOPTERA: PIERIDAE)

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ABSTRACT: To determine the impact of citowett oil and commercial neem extract mixture against the *Pieris brassica* these experiments were under taken. Citowett as a wetting agent was used in conjunction with neem extract against the larval stage of the pest in question. The LC_{50} values of neem extract and combination with citowett oil for second and third instars larvae were estimated, 4.40, 3.89 and 3.38, 2.16 ppm, respectively. The combination of neem extract with citowett increased the larval mortality rate up to 15%. This implies that the application of the mixture is a sound measure for enhancing lethality impacts. Based on collected data it could be concluded that combination of these two less hazardous chemicals is merit to be considered as a candidate control agent against *Pieris brassica*.

KEY WORDS: Compatibility, botanical origin compound, wetting agent, *Pieris brassica*.

Cabbage (*Brassica oleracea* L. var. *capitata*) is an important vegetable crop grown in many countries in the world (Žnidarčič et al., 2008). Several Lepidopterian pests attack cabbage vegetables and the most serious damage is caused mainly by the larvae of several species such as: small white butterfly (*Pieris rapae* L.), large white butterfly (*Pieris brassicae* L.), cabbage moth (*Mamestra brassicae* L.) and diamondback moth (*Plutella xylostella* L.). *Pieris brassicae* L., is a cosmopolitan insect, and is found wherever cruciferous plants are grown. Sometimes massive outbreaks of this pest may occur and injury on cabbage crops could be extensive (Metaspalu et al., 2009). The economic damage occurs when the percent destroyed leaf area reach within 3.97-10.41 in early cabbage, 10.75-13.76 in the late cabbage (Ashfag et al., 2006). The damage notably affects the value of this crop because its consumption and sale happen when it is still fresh (Cartea et al., 2009).

Azadirachtin, a chemical complex found in seeds of neem, *Azadirachta indica* A. Juss, is the main component responsible for the toxic, repellent, antifeedant, growth-inhibiting, oviposition-inhibiting and sterilizing effects in insects (Sukontason et al ., 2008). Neem extracts has also been shown to have direct detrimental and histopathological effects on many insect tissues, for example, muscles, fat body, and gut epithelial cells (Jogar et al., 2008). It is usually safe for beneficial organisms, such as bees, predators and parasitoids, mammals, and for the environment. This plant is an interesting option for integrated pest management programs, since it is selective, presents a less negative impact on the ecosystems and works in association with biological control organisms (Sukontason et al ., 2008).

Citowett as a wetting agent has been used in conjunction with neem extract, when it is added to a spray mixture, foliage wettability is greatly improved on the

surface of either "waxy" or "hairy" leaf surfaces or buds (Anonymus. 2008). The combination of two or more control options may minimize risk and costs of chemicals, reduce resistance development against the treatments and increase effectiveness of the treatments (Yuya et al., 2009).

With retrospect, the objectives of this study were to determine the combined effects of Citowett oil and Neem extracts against the Large White Butterfly, on cabbage, and to determine the minimum effective rate(s) of the combinations that can provide adequate protection to cabbage against *Pieris brassicae*.

MATERIALS AND METHODS

The eggs of cabbage butterfly were collected from the fields of Urmia. Newly hatched larvae were fed with fresh cabbage leaves. Larvae were reared under laboratory conditions 21 ± 2ºC and relative humidity 75±10 percent on fresh cabbage leaves. The combined effect of neem extract with Citowett oil on the second and third instars larvae of *P. brassicae* was investigated. To evaluate the synergistic effect of neem extract and citowett, we used a solution which contained 75 and 25% of nemm extract and citowit, respectively. To this end, concentrations of 1, 2, 4, 8, 16% of neem with 1% of citowett alongside untreated leaves were employed. Each experiment was replicated three times on different days. Freshly cut leaves of cabbage were immersed in a prepared insecticide solution for 10 s and hung vertically to air dry for 30 minutes. Control leaves were treated similarly with tap water. Filter paper was placed inside a plastic petri-dish and treated leaf was placed on top of the filter paper. Fifteen randomly selected second and third instars larvae were released in each petri dish, separately and held under laboratory conditions.

RESULTS

Antifeedant effects of neem extracts insecticides were indicated by patterns of larval development and foliage consumption. We observed that the color of the larvae became darker a few days after they fed on compounds-treated leaves, and turned almost complete black when they died. When second instars larvae were introduced on treated leaf disks, they ceased feeding, slowly dying, and did not cause noticeable foliage damage. When third instars larvae were introduced on treated leaf disks, they normally initiated feeding for a certain period of time, and eventually stopped feeding. Therefore, some degree of feeding damage occurred on the treated leaves, although significantly less foliage damage was caused by the younger and smaller larvae than that caused by the larger and older larvae (Table 1 and 2).

DISCUSSION

Botanical insecticides are an important group of naturally occurring, often slow-acting crop protectants that are usually safer to humans and the environment than conventional insecticides. Moreover, thanks to the fact that botanical insecticides contain mixtures of biologically active substances, no resistance is developed in pests and pathogens. Therefore the use of plant insecticides has been recommended ever more as a suitable alternative of plant protection with minimum negative risks (Pavela, R.2009). The results showed that neem was effective against *P. brassicae*, significantly reducing the survival of larvae feeding on cabbage leaves treated with the extract. In our tests, the extract

did not produce rapid mortality. Following ingestion of treated plant material, larvae became lethargic and exhibited slow locomotion and reactions. These compounds exhibited a significant lethal effect on second and third larval instars, although the effect was slow and varied among the larval instars. Therefore, the compound should be applied as early as possible when the larvae are in second instars of developmental stage. Neem might be enhanced the management measures of Lepidopterous pests in the vegetable agroecosystems because they do not persist in the environment, have unique modes of action, low mammalian toxicity, and may be potentially compatible with natural enemies.

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LITERATURE CITTED

Anonymus. 2008. Citowett Plus.http://pr-rp.pmra-arla. gc. ca/PR-SOL/pr-web.ve1?p-ukid=9862.

Ashfag, M., Gill, T. I. & Ali, A. 2006. Consumption and utilization of various plants by parasitized and unparasitized larvae of cabbage butterfly (*Pieris brassicae* L). Pak. Entomol., 28: 1-4.

Atawodi, S. E. & Atawodi J. C. 2009. Azadirachta indica (neem): a plant of multiple biological and pharmacological activities. Phytochem. Rev., 8: 601-620.

Calvo, D. & Molina, J. M. 2003. Effects of a commercial Neem (*Azadirachta indica*) extract on streblote panda larvae. Phytoparasitica, 31 (4): 365-370.

Cartea, M. E., Padilla, G., Vilar, M. & Velasco, P. 2009. Incidence of the major *Brassica* pests in Northwestern Spain. Plant Resistance. 767-773.

Greenberg, S. M., Showler, A. T. & Liu, T. 2005. Effects of neem-based insecticides on beet armyworm (Lepidoptera: Noctuidae). Insect Science, 12: 17-23.

Jogar, K., Kuusik, A., Metspalu, L., Hiiesaar, K., Grishakova, M. & Luik, A. 2008. Effects of Neem EC on gas exchange, tracheal ventilation, and water loss in diapausing pupae of *Pieris brassicae.*Entomologial Experimentalis et Applicata, 126: 165-173.

Jankwska, B. 2006. The occurrence of some Lepidoptera pests on different cabbage vegetables. Journal of Plant Protection Research, 46: 181-190.

Metaspalu, L., Hiiesaar, K., Jogar, K., Svilponis, E., Ploomi, A., Kivimagi, I., Luik, A. & Menshikova, N. 2009. Oviposition preference of Pieris *brassicae* (L) on different *Brassica oleracea* var. *Capitatal*. Cultivars. Agronomy Research, 7: 456-411.

Pavela, R. 2009. Effectiveness of some botanical insecticides against *Spodoptera littoralis* Boisduvala (Lepidoptera: Noctudiae), *Myzus persicae* Sulzer (Hemiptera: Aphididae) and *Tetranychus urticae* Koch (Acari: Tetranychidae). Plant Protection Science, 45: 161-167.

Sukontason, K. L., Olson, J. K., Chailapakul, O. & Sukontason, K. 2008. Efficacy of neem extract against the blowfly and housefly. Parasitol Res., 103: 535-544.

Younas, M., Naeem, M., Raqib, A. & Masud, S. 2004. Population dynamics of cabbage butterfly (*Pieris brassicae*) and cabbage aphids (*Brevicoryne brassicae*) on five cultivars of cauliflower at Peshawar. Asian Journal of Plant Sciences, 3 (3): 391-393.

Yuya, A. I., Tadesse, A. Azerefegne, F., & Tefera, T. 2009. Efficacy of combining Niger seed oil with malathion 5% dust formulation on maize against the maize weevil, *Sitophilus zeamais* (Coleoptera: Curculionidae). Journal of Stored Products Research, 45: 67-70.

Zabel, A., Manojlovic, B., Rajkovic, S., Stankovic, S. & Kostic, M. 2002. Effect of Neem extract on *Lymantria dispar* L. (Lepidoptera: Lymantriidae) and *Leptinotarba decemlineata* Say. (Coloptera: Chrysomelidae). J. Pest Science, 75: 19-25.

Žnidarčič, D., Valič, N. & Trdan, S. 2008. Epicuticular wax content in the leaves of cabbage (*Brassica oleracea* L. var. capitata) as a mechanical barrier against three insect pests. Acta Agriculturae Slovenica, 361-370.

Table 1- Mean mortality ± Standard Error (Mean ± SE) of neem extract against second and third instar larvae of *P. brassicae*.

Concentration	48 h		72 h		96 h		120 h
	L2	L3	L2	L3	L2	L3	L3
1	0.33±0.33	0.66±0.57	0.66±0.33	1.33±0.57	1.33±0.33	1.66±0.57	2±0
2	2.33±0.33	1.66±0.57	3.33±0.33	2.33±0.57	4±0.57	3±1	3.66±0.57
4	5±0.57	2.66±0.57	8±0.57	4.66±0.57	10.33±0.33	7.33±0.57	8.66±0.57
8	8.33±0.33	3.66±0.57	12.66±0.33	6.66±0.57	15.66±0.33	8.33±0.57	10.33±0.57
16	12±0.57	5.33±0.57	16.66±0.33	8.66±0.57	19.66±0.33	10.33±0.57	13.33±0.57

Table 2- Mean mortality ± Standard Error (Mean ± SE) of neem extract combination with citowett oilagainst second and third instar larvae of *P. brassicae*.

Concentration	48 h		72 h		96 h		120 h
	L2	L3	L2	L3	L2	L3	L3
1	1±0	0.66±0.57	1.33±0.57	2.33±0.57	1.66±0.57	2.66±0.57	4±1
2	2.33±0.57	2.33±0.57	4±1	3.66±0.57	5.33±0.57	5±1	5.66±0.57
4	4±1	2.66±0.57	8±1	6.33±0.57	12.33±0.57	8±0	10.66±0.57
8	8±1	3.66±0.57	13±1	8±1	16.33±0.57	10.66±0.57	14.66±0.57
16	11.33±0.57	6.33±0.57	16.66±1.15	11.33±0.57	19.66±0.57	12.66±0.57	15±0

Table 3- LC_{50} values Neem extract and Neem + citowett against second and third instar larvae of *P. brassicae*.

	LC50		CV	
	L2	L3	L2	L3
Neem extract	4.40	3.38	8.78%	11.28%
Neem + citowett	3.89	2.16	9.3%	10.91%