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Section L: Laboratory Bioassays

Laboratory Bioassays of Biological/Organic Insecticides to Control Corn Earworm on Hemp in Virginia, 2019

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Hemp Fiber | *Cannabis sativa* ssp. Indica

Corn earworm (CEW) | *Helicoverpa zea* (Boddie)

Hemp acreage in the United States is increasing and outdoor crops are susceptible to corn earworm (CEW) feeding injury. Two separate bioassays were conducted in fall 2019 to evaluate the effects of biological/organic insecticide products on CEW in hemp.

Bioassay 1 was initiated on 16 Sep 2019 and included the following treatments: Gemstar (*Helicoverpa zea* nuclear polyhedrosis virus [HzNPV]), Javelin (*Bacillus thuringiensis* var. *kurstaki*), DiPel (*Bacillus thuringiensis* var. *kurstaki*), XenTari (*Bacillus thuringiensis* var. *aizawai* + *kurstaki*), Venerate (94.5% Heat-killed *Burkholderia* spp. strain A396 cells and spent fermentation media), Grandevo (30% *Chromobacterium subtsugae* strain PRAA4-11 and spent fermentation media), Entrust (Spinosad), and an untreated check (Table 1). Third and fourth instar CEW larvae were collected from ears from an untreated field of sweet corn (*Zea mays*) established at Virginia Tech's Kentland Farm in Whitethorne, VA (Kentland). Only vigorous larvae with fresh color were used for the experiment. On 16 Sep 2019, hemp seed heads ('Felina-32') were collected from field plots at Kentland, brought to the laboratory, and cut into ~9 cm^3 sections. Forty hemp seed head sections were dipped into spray-tank concentrations of each treatment (Table 1) and placed individually into 1 oz plastic diet cups with a single CEW larva. A tray of 10 cups represented a replicate and four replicates were established for each treatment and placed in a different stack on the laboratory bench for the duration of the experiment. Diet cups were placed on the laboratory benchtop and held at laboratory ambient light and temperature (20–25°C) for 96 h and checked daily for mortality. Percent mortality data were analyzed with ANOVA procedures and means separated with Tukey's HSD.

Bioassay 2 was initiated on 2 Oct 2019 and included the following treatments: Agree (*Bacillus thuringiensis* var. *aizawai*), Javelin (*Bacillus thuringiensis* var. *kurstaki*), Deliver (*Bacillus thuringiensis* var. *kurstaki*), XenTari (*Bacillus thuringiensis* var. *aizawai* + *kurstaki*), Pyganic (pyrethrins), Entrust (spinosad), and an untreated check (Table 2). The experiment was conducted using the same aforementioned procedures except that rather than using field-collected CEW, which were depleted from the field, we used third instars raised on artificial diet that were purchased from Benzon Research Inc., Carlisle, PA.

In bioassay 1, CEW mortality varied greatly among treatments. As control mortality remained low (<15%) for the duration of the experiment, the 4 DAT data are probably the most useful because many of the treatments tested take a few days to actually kill larvae (Table 1). Entrust resulted in a significantly higher mortality than any other product with 95% mortality after 4 d. XenTari (67.5%) had a significantly higher mortality than any of the other *Bacillus thuringiensis* products and its efficacy was similar to Venerate (55%). Javelin and DiPel resulted in a significantly higher mortality (32.5% and 35%, respectively) than Gemstar (10%) and the untreated check (15%). It should be noted that Gemstar frequently takes more than 96 h to have a lethal effect on larger CEW larvae.

In bioassay 2, Pyganic and Entrust performed significantly better than all other treatments, resulting in 100% and 97.5% mortality, respectively. XenTari, again, had the highest mortality among the tested *Bacillus thuringiensis* products (75%). However, it only had significantly higher mortality than Agree (47.5%) and the untreated check (2.5%). Javelin and Deliver obtained 67.5% and 60% mortality, respectively, which was almost double what these treatments achieved with field-collected larvae in Bioassay 1. Resistance to Cry1AB Bt proteins is widespread in Virginia CEW populations, and likely explains this difference. It should be noted that resistance to pyrethroid/pyrethrin insecticides is also observed widely in Virginia CEW populations, and thus, although not tested in Bioassay 1, Pyganic would not be expected to result in 100% mortality of field-collected CEW larvae.[1]

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Table 1.

Treatment	Rate/acre	Rate/1,500 ml water	Average % mortality			
			1 DAT	2 DAT	3 DAT	4 DAT
Untreated check			0.0c	12.5cd	12.5de	15.0d
Gemstar	5 fl. oz.	1.7 ml	2.5c	2.5d	5.0e	10.0d
Javelin	16 oz.	5.21 g	0.0c	12.5cd	25.0cd	32.5c
DiPel	16 oz.	5.21 g	5.0c	17.5cd	32.5c	35.0c
XenTari	16 oz.	5.21 g	12.5abc	52.5ab	57.5b	67.5b
Venerate	128 fl. oz.	43.5 ml	15.0abc	30.0bc	37.5c	55.0b
Grandevo	48 oz.	15.6 g	20.0ab	22.5cd	22.5cde	22.5cd
Entrust	5 fl. oz.	1.7 ml	27.5a	65.0a	87.5a	95.0a
P-value			0.0142	0.0004	0.0001	0.0001

Means within columns followed by the same letter are not significantly different.

Table 2.

Treatment	Rate/acre	Rate/1,500 ml water	Average % mortality			
			1 DAT	2 DAT	3 DAT	4 DAT
Untreated control			0.0c	2.5c	2.5d	2.5d
Agree	16 oz.	5.21 g	2.5c	7.5c	37.5c	47.5c
Javelin	16 oz.	5.21 g	0.0c	10.0c	55.0bc	67.5bc
Deliver	16 oz.	5.21 g	5.0c	10.0c	47.5bc	60.0bc
XenTari	16 oz.	5.21 g	5.0c	12.5c	62.5bc	75.0b
Pyganic	59 fl. oz.	20.0 ml	97.5a	97.5a	100.0a	100.0a
Entrust	5 fl. oz.	1.7 ml	37.5b	82.5b	92.5a	97.5a
P-value			0.0001	0.0001	0.0001	0.0001

Means within columns followed by the same letter are not significantly different.