TEXAS A&M AGRILIFE

Update on Bt Insect Pest Resistance in Cotton and Current IPM Management Options

David Kerns Department of Entomology Texas A&M AgriLife Extension



United States Department of Agriculture National Institute of Food and Agriculture













Bt Resistance Monitoring



174 field populations with >267,264 insects from 2016-2023



Bt Resistance Monitoring Survey Bioassays

Bollworms/corn earworm collected from the field as larvae Overnight delivery to lab in College Station Reared to F1 or F2 generation and then bioassays Tested for response to Cry1Ac, Cry2Ab2, Cry1F and Vip3A Diet overlay bioassays

- Test 6-8 Bt concentrations and a control
- Used 16-32 neonate larvae, replicated 4 times for each concentration; allowed to feed for 7 days

Record number alive/dead, instar and weight of survivors Compare field populations to a standard laboratory strain (Benzon)

- Dead = Actual dead larvae + 1st instar larvae
- Dose response bioassay: Probit analysis for LC50 and their 95% CL.
- Resistance ratio = LC50 of a field population / LC50 of the susceptible strain.





Table 1. LC₅₀ and 95% confidence limits (CL) based on larval mortality of *Helicoverpa zea* to Cry1Ac protein in Texas in 2023, n=22

Insect strain	Ν	LC ₅₀ (95% CL) (µg/cm ²)	Slope ± SE	X ²	df	Resistance ratio
CBW-BZ-SS	512	0.10 (0.08, 0.13)	1.24 ± 0.10	36.9	26	1.0
CBW-Hutto TX-Intrasect	512	370.04 (143.48, 2570)	$\boldsymbol{0.89 \pm 0.17}$	24.5	26	3700.4
CBW-Hutto TX-NBT	512	304.20 (141.77, 1127)	0.87 ± 0.13	18.2	26	3042
CBW-Hutto TX-VT2P	512	268.14 (128.82, 921.05)	$\boldsymbol{0.87 \pm 0.13}$	24.9	26	2681.4
CBW-Malone TX-Intrasect	512	15.66 (8.93, 31.57)	$\textbf{1.14} \pm \textbf{0.17}$	72.9	26	156.6
CBW-Malone TX-NBT	512	250.73 (119.01, 850.19)	0.80 ± 0.12	19.8	26	2507.3
CBW-Malone TX-VT2P	512	85.04 (63.86, 127.0)	$\boldsymbol{1.84 \pm 0.26}$	12.5	26	850.4
CBW-Snook TX Early-Intrasect	512	46.02 (30.44, 78.35)	$\boldsymbol{0.96 \pm 0.10}$	22.4	26	460.2
CBW-Snook TX Early-NBT	512	36.84 (24.96, 59.98)	$\boldsymbol{0.98 \pm 0.09}$	31.9	26	368.4
CBW-Snook TX Early-VT2P	512	41.32 (29.13, 63.83)	$\textbf{1.14} \pm \textbf{0.12}$	11.2	26	413.2
CBW-Snook TX Late-Intrasect	512	594.24 (97.67, 126017)	$\boldsymbol{0.50 \pm 0.13}$	68.6	26	5942.4
CBW-Snook TX Late-NBT	512	1775121 (8990, 1.18E19)	$\boldsymbol{0.27 \pm 0.09}$	30.7	26	17751210
CBW-Snook TX Late-VT2P	512	6469 (642.06, 2416435)	$0.35{\pm 0.08}$	36.9	26	64690
CBW-Taylor TX-Intrasect	512	152.98 (83.95, 383.91)	$\boldsymbol{0.90 \pm 0.12}$	20.9	26	1529.8
CBW-Taylor TX-NBT	512	243.13 (67.76, 10598)	$\boldsymbol{0.80 \pm 0.22}$	57.4	26	2431.3
CBW-Taylor TX-VT2P	512	6265 (257.73, 7.35E10)	$\boldsymbol{0.27 \pm 0.09}$	52.9	26	62650
CBW-Thrall TX-Intrasect	512	189.69 (65.58,1332)	$\boldsymbol{0.56 \pm 0.09}$	41.8	26	1896.9
CBW-Thrall TX-NBT	512	996.74 (236.19, 16677)	$\textbf{0.45} \pm \textbf{0.08}$	35.8	26	9967.4
CBW-Thrall TX-VT2P	512	71.39 (43.06, 150.25)	$\boldsymbol{1.10\pm0.15}$	32.9	26	713.9
CBW-Comanche TX-NBT-F2	512	54.72 (32.04, 112.80)	0.74 ± 0.08	23.3	26	547.2
CBW-Comanche TX-Intrasect-F2	512	114.85 (61.00, 291.60)	0.74 ± 0.09	17	26	1148.5
CBW-Comanche TX-VT2P-F2	512	55.52 (36.53, 96.10)	0.98 ± 0.11	18.1	26	555.2
CBW-Wallis TX-WS3 Cotton	512	59.92 (38.69, 107.38)	0.96 ± 0.11	13.2	26	599.2

Resistance ratio = LC_{50} of a field population / LC_{50} of the susceptible strain.

≥ 10 = resistant; 22:22



Table 3. LC₅₀ and 95% confidence limits (CL) based on larval mortality of *Helicoverpa zea* to Cry2Ab2 protein in Texas in 2023, n=22

CBW-BZ-SS5760.32 (0.24, 0.43)1.10 \pm 0.0814.5301.0CBW-Hutto TX-Intrasect5122.05 (0.95, 5.39)0.80 \pm 0.15111.3266.4CBW-Hutto TX-NBT51231.73 (14.76, 132.25)0.93 \pm 0.1721.92699.2CBW-Hutto TX-VT2P5127.07 (3.89, 16.25)1.05 \pm 0.1769.62622.1CBW-Malone TX-Intrasect5127.72 (4.89, 13.87)1.37 \pm 0.2140.22624.1CBW-Malone TX-NBT51220.14 (8.55, 116.67)0.99 \pm 0.22692662.9CBW-Malone TX-VT2P51248.43 (23.15, 154.90)0.71 \pm 0.0926.426151.3CBW-Malone TX-VT2P5126.54 (3.42, 16.49)1.00 \pm 0.1782.72620.4CBW-Snook TX Early-Intrasect5122.12 (1.13, 4.32)1.05 \pm 0.1775.7266.6CBW-Snook TX Early-VT2P51218.83 (10.82, 43.40)0.97 \pm 0.1328.72658.8CBW-Snook TX Late-Intrasect51225.00 (15.20, 51.97)0.96 \pm 0.1229.52678.1CBW-Snook TX Late-Intrasect5124.52 (2.49, 9.47)1.26 \pm 0.2255.12614.1	Insect strain	N	LC ₅₀ (95% CL) (µg/cm ²)	Slope ± SE	\mathbf{X}^2	df	Resistance rati
CBW-Hutto TX-Intrasect5122.05 (0.95, 5.39)0.80 ± 0.15111.3266.4CBW-Hutto TX-NBT51231.73 (14.76, 132.25)0.93 ± 0.1721.92699.2CBW-Hutto TX-VT2P5127.07 (3.89, 16.25)1.05 ± 0.1769.62622.1CBW-Malone TX-Intrasect5127.72 (4.89, 13.87)1.37 ± 0.2140.22624.1CBW-Malone TX-NBT51220.14 (8.55, 116.67)0.99 ± 0.22692662.9CBW-Malone TX-VT2P51248.43 (23.15, 154.90)0.71 ± 0.0926.426151.3CBW-Snook TX Early-Intrasect5122.12 (1.13, 4.32)1.05 ± 0.1782.72620.4CBW-Snook TX Early-NBT5122.12 (1.13, 4.32)1.05 ± 0.1775.7266.6CBW-Snook TX Late-Intrasect51225.00 (15.20, 51.97)0.96 ± 0.1229.52678.1CBW-Snook TX Late-NBT5124.52 (2.49, 9.47)1.26 ± 0.2255.12614.1	CBW-BZ-SS	576	0.32 (0.24, 0.43)	1.10 ± 0.08	14.5	30	1.0
CBW-Hutto TX-NBT512 $31.73 (14.76, 132.25)$ 0.93 ± 0.17 21.9 26 99.2 CBW-Hutto TX-VT2P 512 $7.07 (3.89, 16.25)$ 1.05 ± 0.17 69.6 26 22.1 CBW-Malone TX-Intrasect 512 $7.72 (4.89, 13.87)$ 1.37 ± 0.21 40.2 26 24.1 CBW-Malone TX-NBT 512 $20.14 (8.55, 116.67)$ 0.99 ± 0.22 69 26 62.9 CBW-Malone TX-VT2P 512 $48.43 (23.15, 154.90)$ 0.71 ± 0.09 26.4 26 151.3 CBW-Snook TX Early-Intrasect 512 $6.54 (3.42, 16.49)$ 1.00 ± 0.17 82.7 26 20.4 CBW-Snook TX Early-NBT 512 $2.12 (1.13, 4.32)$ 1.05 ± 0.17 75.7 26 6.6 CBW-Snook TX Early-VT2P 512 $18.83 (10.82, 43.40)$ 0.97 ± 0.13 28.7 26 58.8 CBW-Snook TX Late-Intrasect 512 $25.00 (15.20, 51.97)$ 0.96 ± 0.12 29.5 26 78.1 CBW-Snook TX Late-NBT 512 $4.52 (2.49, 9.47)$ 1.26 ± 0.22 55.1 26 14.1	CBW-Hutto TX-Intrasect	512	2.05 (0.95, 5.39)	$\boldsymbol{0.80 \pm 0.15}$	111.3	26	6.4
CBW-Hutto TX-VT2P5127.07 (3.89, 16.25)1.05 ± 0.1769.62622.1CBW-Malone TX-Intrasect5127.72 (4.89, 13.87)1.37 ± 0.2140.22624.1CBW-Malone TX-NBT51220.14 (8.55, 116.67)0.99 ± 0.22692662.9CBW-Malone TX-VT2P51248.43 (23.15, 154.90)0.71 ± 0.0926.426151.3CBW-Snook TX Early-Intrasect5126.54 (3.42, 16.49)1.00 ± 0.1782.72620.4CBW-Snook TX Early-NBT5122.12 (1.13, 4.32)1.05 ± 0.1775.7266.6CBW-Snook TX Early-VT2P51218.83 (10.82, 43.40)0.97 ± 0.1328.72658.8CBW-Snook TX Late-Intrasect51225.00 (15.20, 51.97)0.96 ± 0.1229.52678.1CBW-Snook TX Late-NBT5124.52 (2.49, 9.47)1.26 ± 0.2255.12614.1	CBW-Hutto TX-NBT	512	31.73 (14.76, 132.25)	0.93 ± 0.17	21.9	26	99.2
CBW-Malone TX-Intrasect512 $7.72 (4.89, 13.87)$ 1.37 ± 0.21 40.2 26 24.1 CBW-Malone TX-NBT 512 $20.14 (8.55, 116.67)$ 0.99 ± 0.22 69 26 62.9 CBW-Malone TX-VT2P 512 $48.43 (23.15, 154.90)$ 0.71 ± 0.09 26.4 26 151.3 CBW-Snook TX Early-Intrasect 512 $6.54 (3.42, 16.49)$ 1.00 ± 0.17 82.7 26 20.4 CBW-Snook TX Early-NBT 512 $2.12 (1.13, 4.32)$ 1.05 ± 0.17 75.7 26 66.6 CBW-Snook TX Early-VT2P 512 $18.83 (10.82, 43.40)$ 0.97 ± 0.13 28.7 26 58.8 CBW-Snook TX Late-Intrasect 512 $25.00 (15.20, 51.97)$ 0.96 ± 0.12 29.5 26 78.1 CBW-Snook TX Late-NBT 512 $4.52 (2.49, 9.47)$ 1.26 ± 0.22 55.1 26 14.1	CBW-Hutto TX-VT2P	512	7.07 (3.89, 16.25)	$\boldsymbol{1.05\pm0.17}$	69.6	26	22.1
CBW-Malone TX-NBT51220.14 (8.55, 116.67)0.99 ± 0.22692662.9CBW-Malone TX-VT2P51248.43 (23.15, 154.90)0.71 ± 0.0926.426151.3CBW-Snook TX Early-Intrasect5126.54 (3.42, 16.49)1.00 ± 0.1782.72620.4CBW-Snook TX Early-NBT5122.12 (1.13, 4.32)1.05 ± 0.1775.7266.6CBW-Snook TX Early-VT2P51218.83 (10.82, 43.40)0.97 ± 0.1328.72658.8CBW-Snook TX Late-Intrasect51225.00 (15.20, 51.97)0.96 ± 0.1229.52678.1CBW-Snook TX Late-NBT5124.52 (2.49, 9.47)1.26 ± 0.2255.12614.1	CBW-Malone TX-Intrasect	512	7.72 (4.89, 13.87)	1.37 ± 0.21	40.2	26	24.1
CBW-Malone TX-VT2P51248.43 (23.15, 154.90) 0.71 ± 0.09 26.426151.3CBW-Snook TX Early-Intrasect512 6.54 (3.42, 16.49) 1.00 ± 0.17 82.7 2620.4CBW-Snook TX Early-NBT512 2.12 (1.13, 4.32) 1.05 ± 0.17 75.7266.6CBW-Snook TX Early-VT2P512 18.83 (10.82, 43.40) 0.97 ± 0.13 28.72658.8CBW-Snook TX Late-Intrasect512 25.00 (15.20, 51.97) 0.96 ± 0.12 29.52678.1CBW-Snook TX Late-NBT512 4.52 (2.49, 9.47) 1.26 ± 0.22 55.12614.1	CBW-Malone TX-NBT	512	20.14 (8.55, 116.67)	0.99 ± 0.22	69	26	62.9
CBW-Snook TX Early-Intrasect5126.54 (3.42, 16.49)1.00 ± 0.1782.72620.4CBW-Snook TX Early-NBT5122.12 (1.13, 4.32)1.05 ± 0.1775.7266.6CBW-Snook TX Early-VT2P51218.83 (10.82, 43.40)0.97 ± 0.1328.72658.8CBW-Snook TX Late-Intrasect51225.00 (15.20, 51.97)0.96 ± 0.1229.52678.1CBW-Snook TX Late-NBT5124.52 (2.49, 9.47)1.26 ± 0.2255.12614.1	CBW-Malone TX-VT2P	512	48.43 (23.15, 154.90)	$\boldsymbol{0.71 \pm 0.09}$	26.4	26	151.3
CBW-Snook TX Early-NBT5122.12 (1.13, 4.32)1.05 ± 0.1775.7266.6CBW-Snook TX Early-VT2P51218.83 (10.82, 43.40)0.97 ± 0.1328.72658.8CBW-Snook TX Late-Intrasect51225.00 (15.20, 51.97)0.96 ± 0.1229.52678.1CBW-Snook TX Late-NBT5124.52 (2.49, 9.47)1.26 ± 0.2255.12614.1	CBW-Snook TX Early-Intrasect	512	6.54 (3.42, 16.49)	1.00 ± 0.17	82.7	26	20.4
CBW-Snook TX Early-VT2P51218.83 (10.82, 43.40)0.97 ± 0.1328.72658.8CBW-Snook TX Late-Intrasect51225.00 (15.20, 51.97)0.96 ± 0.1229.52678.1CBW-Snook TX Late-NBT5124.52 (2.49, 9.47)1.26 ± 0.2255.12614.1	CBW-Snook TX Early-NBT	512	2.12 (1.13, 4.32)	$\boldsymbol{1.05 \pm 0.17}$	75.7	26	6.6
CBW-Snook TX Late-Intrasect51225.00 (15.20, 51.97)0.96 ± 0.1229.52678.1CBW-Snook TX Late-NBT5124.52 (2.49, 9.47)1.26 ± 0.2255.12614.1	CBW-Snook TX Early-VT2P	512	18.83 (10.82, 43.40)	0.97 ± 0.13	28.7	26	58.8
CBW-Snook TX Late-NBT512 $4.52 (2.49, 9.47)$ 1.26 ± 0.22 55.12614.1	CBW-Snook TX Late-Intrasect	512	25.00 (15.20, 51.97)	0.96 ± 0.12	29.5	26	78.1
	CBW-Snook TX Late-NBT	512	4.52 (2.49, 9.47)	1.26 ± 0.22	55.1	26	14.1
CBW-Snook TX Late-VT2P51225.15 (14.97, 53.38) 0.91 ± 0.11 21.022678.6	CBW-Snook TX Late-VT2P	512	25.15 (14.97, 53.38)	0.91 ± 0.11	21.02	26	78.6
CBW-Taylor TX-Intrasect5128.51(5.01, 17.66)1.00 ± 0.14 412626.6	CBW-Taylor TX-Intrasect	512	8.51(5.01, 17.66)	1.00 ± 0.14	41	26	26.6
CBW-Taylor TX-NBT5128.83 (5.46, 16.80) 0.97 ± 0.12 30.32627.6	CBW-Taylor TX-NBT	512	8.83 (5.46, 16.80)	0.97 ± 0.12	30.3	26	27.6
CBW-Taylor TX-VT2P5127.39 (4.14, 16.60) 1.02 ± 0.16 54.72623.6	CBW-Taylor TX-VT2P	512	7.39 (4.14, 16.60)	1.02 ± 0.16	54.7	26	23.6
CBW-Thrall TX-Intrasect512 $326.56 (74.22, 6611)$ 0.48 ± 0.09 36.8261020.5	CBW-Thrall TX-Intrasect	512	326.56 (74.22, 6611)	0.48 ± 0.09	36.8	26	1020.5
CBW-Thrall TX-NBT5128.86 (5.12, 19.32)1.15 \pm 0.1955.62627.7	CBW-Thrall TX-NBT	512	8.86 (5.12, 19.32)	1.15 ± 0.19	55.6	26	27.7
CBW-Thrall TX-VT2P512133.32 (36.30, 6920) 0.85 ± 0.23 25.526416.6	CBW-Thrall TX-VT2P	512	133.32 (36.30, 6920)	0.85 ± 0.23	25.5	26	416.6
CBW-Comanche TX-NBT-F251226.68 (14.03, 75.99) 0.92 ± 0.14 24.62683.4	CBW-Comanche TX-NBT-F2	512	26.68 (14.03, 75.99)	0.92 ± 0.14	24.6	26	83.4
CBW-Comanche TX-Intrasect-F25129.23 (6.62, 13.89)1.46 \pm 0.1814.42628.8	CBW-Comanche TX-Intrasect-F2	512	9.23 (6.62, 13.89)	1.46 ± 0.18	14.4	26	28.8
CBW-Comanche TX-VT2P-F2512 $34.70 (20.02, 81.42)$ 0.97 ± 0.13 12.726108.4	CBW-Comanche TX-VT2P-F2	512	34.70 (20.02, 81.42)	0.97 ± 0.13	12.7	26	108.4
CBW-Wallis TX-WS3 Cotton51219.35 (10.54, 49.64) 0.89 ± 0.13 21.22660.8	CBW-Wallis TX-WS3 Cotton	512	19.35 (10.54, 49.64)	0.89 ± 0.13	21.2	26	60.8

Resistance ratio = LC_{50} of a field population / LC_{50} of the susceptible strain.

≥ 10 = resistant; 20:22



Table 5. LC₅₀ and 95% confidence limits (CL) based on larval mortality of *Helicoverpa zea* to Vip3Aa39 protein in Texas in 2023, n=21

Insect strain	N	LC ₅₀ (95% CL) (µg/cm ²)	Slope ± SE	X ²	df	Resistance rati
CBW-BZ-SS	512	0.30 (0.18, 0.51)	$\boldsymbol{1.26 \pm 0.17}$	61.8	26	1.0
CBW-Hutto TX-Intrasect	448	0.050 (0.041, 0.059)	3.03 ± 0.41	6.1	22	0.17 (4.25)
CBW-Hutto TX-NBT	448	$0.049 \ (0.042, 0.058)$	$\boldsymbol{3.78 \pm 0.50}$	3.8	22	0.16 (4.00)
CBW-Hutto TX-VT2P	448	$0.070 \ (0.057, 0.084)$	$\boldsymbol{2.62 \pm 0.30}$	5.6	22	0.23 (5.75)
CBW-Malone TX-Intrasect	448	0.028 (0.019, 0.035)	2.80 ± 0.53	7	22	0.09 (2.25)
CBW-Malone TX-NBT	448	$0.071 \ (0.059, 0.085)$	3.02 ± 0.36	12.6	22	0.24 (6.00)
CBW-Malone TX-VT2P	448	0.047 (0.039, 0.056)	3.37 ± 0.46	5.6	22	0.16 (4.00)
CBW-Snook TX Early-Intrasect	448	0.099 (0.084, 0.118)	3.19 ± 0.35	16.4	22	0.33 (8.25)
CBW-Snook TX Early-NBT	448	0.031 (0.024, 0.036)	4.18 ± 0.84	25.9	22	0.10 (2.50)
CBW-Snook TX Early-VT2P	448	0.039 (0.034, 0.046)	4.61 ± 0.70	3.6	22	0.13 (3.25)
CBW-Snook TX Late-Intrasect	448	0.049 (0.038, 0.060)	2.44 ± 0.33	8.5	22	0.16 (4.00)
CBW-Snook TX Late-NBT	448	0.063 (0.051, 0.076)	2.66 ± 0.33	16.5	22	0.21 (5.25)
CBW-Taylor TX-Intrasect	448	0.099 (0.084, 0.118)	3.09 ± 0.34	11.1	22	0.33 (8.25)
CBW-Taylor TX-NBT	448	0.085 (0.069, 0.104)	2.49 ± 0.27	22.7	22	0.28 (7.00)
CBW-Taylor TX-VT2P	448	0.087 (0.073, 0.103)	3.14 ± 0.35	12.9	22	0.29 (7.25)
CBW-Thrall TX-Intrasect	448	$0.030 \ (0.021, 0.038)$	2.52 ± 0.44	3.8	22	0.10 (2.5)
CBW-Thrall TX-NBT	448	0.028 (0.020, 0.034)	3.37 ± 0.67	3.6	22	0.09 (2.25)
CBW-Thrall TX-VT2P	448	0.013 (0.002, 0.021)	2.12 ± 0.61	10.7	22	0.04 (1.00)
CBW-Comanche TX-NBT-F2	448	0.035 (0.029, 0.040)	$\textbf{4.65} \pm \textbf{0.85}$	3.4	22	0.12 (3.00)
CBW-Comanche TX-Intrasect-F2	448	0.035 (0.030, 0.040)	$\textbf{4.70} \pm \textbf{0.85}$	3	22	0.12 (3.00)
CBW-Comanche TX-VT2P-F2	448	0.036 (0.031, 0.042)	$\textbf{4.90} \pm \textbf{0.85}$	3.1	22	0.12 (3.00)
CBW-Wallis TX-WS3 Cotton	448	0.11 (0.093, 0.139)	2.41 ± 0.25	10	22	0.37 (9.25)

Resistance ratio = LC50 of a field population / LC50 of the susceptible strain.

≥ 10 = resistant; 0:21



Diet-overlay Bioassays (2016-2023)

Percentage of populations with RR > 10X

Bt protein	2016 (5)	2017 (14)	2018 (34)	2019 (30)	2020 (5)	2021 (12)	2022 (37)	2023 (3
Cry1Ac	/	100%	94%	96%	100%	92%	100%	100%
Cry2Ab2	80%	77%	73%	73%	100%	92%	74%	97%
Vip3Aa	0%	0%	0%*	0%*	0%	0%	0%	0%







Frequency of Bt resistant alleles



F₂ screen principle for isolating Bt resistant alleles in isofamily lines



Andow, D. A., and D. N. Alstad. 1998. F2 Screen for Rare Resistance Alleles. Journal of Economic Entomology 91: 572-578.

(RR) 6.25%

Assuming resistance is controlled by a single gene and recessive alleles



H. zeα-F₂ families surviving a discriminating concentration of 10μg Cry1Ac/cm² in Texas

Year of collection	Methods to establish the F_2 families	Number of F ₂ families Screened ^{1, 2}	Number of surviving F ₂ families ³	Percentage (%) of surviving families	Estimated number of resistance alleles ⁴	Estimated resistance allele frequency	Confidence Interva (95%)
2018	Cross with SS♂	12	10	83.33	13	0.5417	(0.3507 -0.7211)
2019	Light trap	94	89	94.68	Min: 153 Max: 186	0.4069 0.4947	(0.3584 – 0.4573 (0.4445 - 0.5450
Overall	Cross with SS Light trap	106	99	93.40	Min: 166 Max: 199	0.4150 0.4975	(0.3677 – 0.4639 (0.4488 – 0.5463

- Total insects assayed in 2018 and 2019 = 13,568 larvae 1.
- Based on 128 larvae per bioassay/F₂ family 2.
- 5 survivors \geq 2nd instar with at least 1 larva \geq 3rd instar 3.
- Number of resistant alleles based on results from simple monogenic inheritance models ($\chi^2 < 3.841$ with 1 df, p > 0.05) 4.

An allele frequency of <0.001 is considered rare





H. zeα-F₂ families surviving a discriminating concentration of 10μg Cry2Ab2/cm² in Texas

Year of collection	Methods to establish the F_2 families	Number of F ₂ families Screened ^{1, 2}	Number of surviving F ₂ families ³	Percentage (%) of surviving families	Estimated number of resistance alleles ⁴	Estimated resistance allele frequency	Confidence Interva (95%)
2018	Cross with SS♂	12	7	58.33	11	0.4583	(0.2789 -0.6493
2019	Light trap	108	35	32.41	Min: 39 Max: 45	0.0903 0.1042	(0.0667 – 0.1210 (0.0788 – 0.1365
Overall	Cross with SS Light trap	120	42	35.00	Min: 50 Max: 56	0.1097 0.1228	(0.0842 – 0.1417 (0.0958 – 0.1561

- 1. Total insects assayed in 2018 and 2019 = 15,360 larvae
- Based on 128 larvae per bioassay/F₂ family 2.
- 5 survivors \geq 2nd instar with at least 1 larva \geq 3rd instar 3.
- Number of resistant alleles based on results from simple monogenic inheritance models ($\chi^2 < 3.841$ with 1 df, p > 0.05) 4.

An allele frequency of <0.001 is considered rare



H. zea- F_2 families surviving a discriminating concentration of $3\mu g$ Vip3Aa39/cm² in Texas

Collection site	Year of collection	Methods to establish the F_2 families	Number of F ₂ families screened2	Number of surviving F ₂ families ³	Percentage (%) of surviving families	Estimated resistance allele frequency ⁴	Confidence Interval (95%) ⁵
Texas	2019 ¹	Light trap	114	2	1.59	0.0065	(0.0014 – 0.0157)

- 1. Total insects assayed in 2019 = 14,592 larvae
- 2. Based on 128 larvae per bioassay/ F_2 family
- 3. 5 survivors \geq 2nd instar with at least 1 larva \geq 3rd instar
- 4. (Andow and Alstad, 1998)
- 5. (Andow and Alstad, 1999)

An allele frequency of <0.001 is considered rare



What is driving Bt resistance?





VT2P = Cry1A.105 + Cry2Ab2

Two Types of Vip3Aa Resistance?

CEW from Leptra corn



Low level of resistance (probably minor genes?)





Complete resistance (major gene controlled)

Early Warning of Resistance to Vip3Aa



Increase from 2016 to 2020 in the Vip3Aa resistance ratio relative to the BZ lab strain for 71 field-derived strains of CEW. Linear regression: $\log(y) = 0.14X - 282$, $R^2 = 0.12$, df = 69, P = 0.003.



Open Access Article

Early Warning of Resistance to Bt Toxin Vip3Aa in Helicoverpa zea

by 🙁 Fei Yang ^{1,*} 🖾 💿, 🙁 David L. Kerns ¹ 🗠, 🙁 Nathan S. Little ² 🗠, 🙁 José C. Santiago González ¹ 🗠 and Ruce E. Tabashnik ^{3,*} 🖂 💿

- ¹ Department of Entomology, Texas A&M University, College Station, TX 77843, USA
- ² USDA Agricultural Research Service, Stoneville, MS 38776, USA
- ³ Department of Entomology, University of Arizona, Tucson, AZ 85721, USA
- Authors to whom correspondence should be addressed.

Toxins 2021, 13(9), 618; https://doi.org/10.3390/toxins13090618



Unexpected Injury in Vip Corn







Technology	Traits
NBT-1 Dekalb	None
VT2P	Cry1A.105 + Cry2Ab2
NNT-2 Pioneer	None
Intrasect	Cry1Ab + Cry1F
Leptra	Cry1Ab + Cry1F + Vip3Aa

Vip3Aa Resistant Populations

Insect population	Collected location (Year)	LC50 (95% CL) (µg/cm²)	Resistance ratio	Inheritance
CBW-BZ-SS	/	0.11 (0.09, 0.13)	1	/
CBW-TX-VIP-RR	Snook, TX (2019)	> 100	> 909.1	Recessive, Autosomal, single-gene
CBW-LA-M1-VIP-RR	Alexandria, LA (2019)	> 100	> 909.1	Recessive, Autosomal, single-gene
CBW-MS-R2-VIP-RR	Stoneville, MS (2020)	> 100	> 909.1	Recessive, Autosomal, single-gene
CBW-MS-R15-VIP-RR	Stoneville, MS (2020)	> 100	> 909.1	Recessive, Autosomal, single-gene
CBW-LA-AC4-VIP-RR	Winnsboro, LA (2020)	> 100	> 909.1	Recessive, Autosomal, single-gene

Vip-RR Interstrain Complementation Tests



Among these 5 strains there appears to be 3 different major gene loci conveying resistance

The MS strains are similar to each other CBW-MS-R2-RR CBW-MS-R15-RR One LA strain is unique CBW-LA-AC4-RR

ect strain cross	No. tested	Survival at Vip3Aa 10.0 ug/cm2	Genetic Basis
/-MS-R2-RR X X-LT#70-RR	256	0	Different
/-MS-R15-RR X -LT#70-RR	256	0	Different
/-LA-AC4-RR X X-LT#70-RR	256	0	Different
/-LA-AC4-RR X S-R15-RR	256	0	Different
/-MS-R2-RR X S-R15-RR	256	255	Similar
/-LA-M1-RR X -LT#70-RR	256	256	Similar

The TX strain is similar to one of the LA strains CBW-TX-LT#70-RR CBW-LA-MI-RR



2023 Vip3Aa Cotton Unexpected Injury Events

			F	Resistance Ra	tio
Location	Technology	% damaged fruit	Cry1Ac	Cry2Ab2	Vip3Aa39
Starkville, MS	TwinLink Plus	17%	10298	1215	0.30 (7.50)
Wallis,TX	WideStrike 3	25%	599.2	60.8	0.37 (9.25

- Vip3Aa failures in 2023 occurred in cotton that was cut out
- Damage was almost exclusively to the bolls

• Vip3Aa resistance was slightly elevated but not high enough to warrant concern





Bt Protein Concentrations



Changes in Cry2A Concentrations in BG2 Terminal Leaf Tissue Over Time





Days After Planting

Differences in Cry2A Concentrations Among BG2 Cotton Tissues







Survivorship of Cry Resistant **Bollworms on BG2**







Changes in Vip3Aa Concentrations in BG3 Terminal Leaf Tissue Over Time









Differences in Vip3Aa Concentrations Among BG3 Cotton Tissues 0.015 A 0.010 Vip3A 0.005 0.000-







Survivorship of Cry Resistant **Bollworms on BG3**





0.05



Managing Bollworms in Cotton



Bollworm Injury to Bt Cotton - High Bollworm Pressure

College Station, TX - July 17, 2017





Benefit from Spraying



College Station, TX (2) - 2018

·····BC			
		BCD	
	D		D
, TL	TLP	BG2	BG3

Bt Cotton Trait Performance Texas





Percent Reduction in Fruit Damage Relative to Non-Bt							
Technology	June 28	July 3	July 12	July 17	July 25	Aug 1	Mean
BG2	97.79	94.47	90.91	75.47	71.52	63.09	82.21
WS3	97.04	96.67	94.00	98.21	100.00	94.64	96.76
BG3	96.34	98.87	96.91	91.26	100.00	100.00	97.23





Insecticide Efficacy





Managing Bollworms in Cotton





- 1. Plant cotton that produces the Vip3Aa Bt protein.
- 2. Vip cotton will usually provide >95% control.
- 3. Dual-gene cotton will provide ~80% control.

- 1. Base treatment decisions on risk of economic damage.
- 2. 6% fruit damage is a good threshold.
- 3. Be aware of nickel & diming.
- 4. In dual-gene cotton spraying on a 20% egg lay is usually justified.



- **1. Avoid pyrethroids.**
- 2. Use products that contain chlorantraniliprole.
- 3. Get them before they get you!
- 4. Good coverage is vital.
- 5. Use higher rate when longer residual control is needed.





What does the ThryvOn Bt technology bring to the table and is it worth the cost?

ThryvOn Availability 2024

Zone D

- Seed treatment required
- \$558.50 per bag with Acceleron Standard
- \$604.50 per bag with Acceleron Elite
- Cost for ThryvOn = \$16.49

Zone E (includes all of Oklahoma)

- Seed treatment optional
- \$517.50 per bag with Acceleron Standard
- \$556.50 per bag with Acceleron Elite
- Cost of ThryvOn = \$10.78

*Prices from Bayer Crop Science for Deltapine varieties, prices and availability may vary by seed supplier



TEXAS A&M AGRILIFE

ThryvOn Activity on Thrips and Lygus







Untreated Non Bt





Thrips Injury





Thrips Damage Ratings- Lubbock



Suhas Vyavhare, AgriLife Extension





Thrips Damage Ratings- Snook



Season Average Large Nymphs Drop Cloth – Numbers per 10 Row Ft (Interaction)



No Trait

Averaged over 2 years in 2 locations

Scott Stewart – University of Tennessee



Yield Interaction Pounds of Seed Cotton per Acre (Interaction)



No Trait

Trait

Averaged over 2 years in 2 locations

Scott Stewart – University of Tennessee



TEXAS A&M AGRILIFE

ThryvOn Activity on Cotton Fleahopper







- High Infestation
- Spray treatment significantly lowered populations
- No individual trait effect on populations

2019 Fruit Retention



High Infestation

- No individual trait effect on populations
- ThryvOn Nonsprayed was statistically the same as the **Non-traited** Sprayed
- ThryvOn plots were higher than the Nontraited counterpart



2019-2021 Combined Population Structure







100



- Significantly different population structures (*P=0.0001*)
- Ratio of small to large nymphs in ThryvOn plots was 2.6:1
 - Only 2 of every 5 nymphs were able molt to the later instars
 - Survivorship of nymphs was lower than non-traited
 - The ThryvOn trait appears to delay nymph development
- Ratio of small to large nymphs in Nontraited was 1.1:1
 - Almost all nymphs that hatched were able to survive to later instars

Cumulative Insect Days vs Fruit Retention



- Cumulative insect days against fruit retention across years by variety
 - Significantly different regressions (*P=0.023*)
- Faster rate of fruit abscission in nontraited variety

Electropenetrography (EPG)





Cell Rupturing Events vs Ingestion Events



· Non-traited best fit model was linear regression $(R^2=0.7641)$

• ThryvOn best fit model was logistic growth regression ($R^2=0.5262$)

· Similar trajectory until ingestion events plateaued at around 6 on ThryvOn squares

40

Conclusions

- I'd rate efficacy as ...
 - Thrips if if if (a potential game changer)
 - Plant bugs 🧰 🧀 (a tool in the toolbox)
 - Cotton fleahoppers i (will make insecticides look better)

There is clear evidence of behavioral avoidance by thrips (strong), and tarnished plant bug and cotton fleahopper (relatively weak)

- Negatively effects oviposition and feeding behavior
- We won't know everything until the technology is has been used on a large scale
 - Development of other traits is ongoing







NEW MEXICO Clovis . • Lubbock Roswell Midland . Odessa El Paso

Amarillo





Antelope Wells (& Animas ?) New Mexico. - and across AZ border

- Small valleys isolated by desert
- Many years of Cry 1F, then Cry1F + 1Ab
- Part of acres planted to SSTX in 2024
 - Cry1F + Cry 1A.102 + Cry2Ab2
- Pyramids: 5% seed blend refuge
- Obvious resistance in 2023
 - Collections made by TAMU
- 70-90% of SSTX plants infested in 2024
 - Official UXI investigation
 - 9 Bayer employees, 3 TAMU
 - Bayer & TAMU made collections 7/9/24
 - Vip3a is still holding up
 - Said to be low dose







David Kerns Texas A&M University, College Station, TX Email: <u>David.Kerns@ag.tamu.edu</u> Phone: 318-439-4844



United States Department of Agriculture National Institute of Food and Agriculture

