

Deltamethrin Resistance Tests: Alamogordo strain

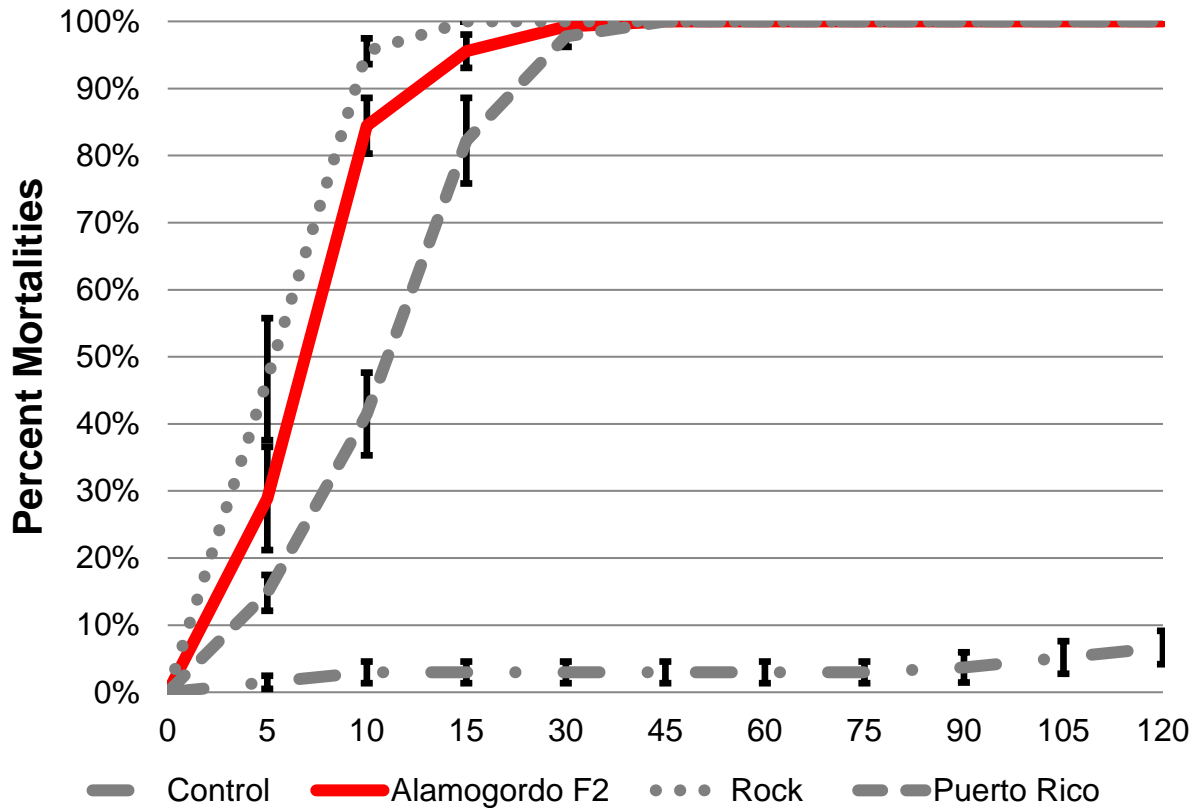


Figure A. Deltamethrin IR Bottle Test Results: 10.5 ug/bottle. Shown are means and standard error bars for different groups of mosquitoes. Rock is considered a pyrethroid-sensitive strain. Puerto Rico is considered a pyrethroid-resistant strain. The control was treated with acetone only.

Results- The control (Acetone) mortality curve was significantly different ($P < 0.0001$) from the curves for all other treatments with little to no mortalities. Kaplan Meier statistical analysis confirmed that there was a significant difference between the the curves plotted for the deltamethrin-exposed Rock Strain and the Alamogordo Strain ($P < .0001$). The curve plotted for the Puerto Rico Strain (a known resistant strain) is significantly different than that plotted for the Alamogordo Strain ($P < .0001$). The curves plotted for the Puerto Rico strain and the Rock strain were also significantly different from each other ($P < .0001$).

Conclusion- Although we found small but statistical significant differences in the survival curve of the ROCK control and the curve plotted for mosquitoes from Alamogordo, the comparison with the curve plotted for the resistant Puerto Rico strain suggests that the Alamogordo strain is sensitive to deltamethrin.

Deltamethrin Resistance Tests: Las Cruces strain

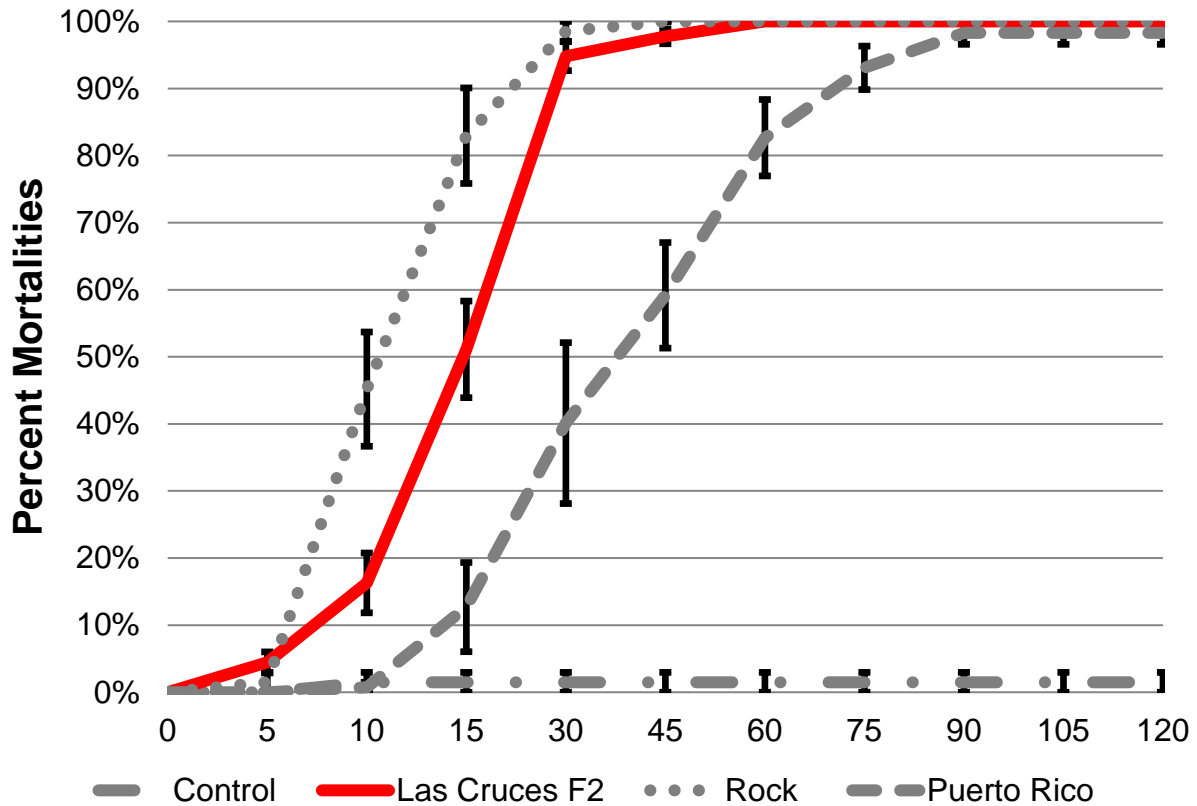


Figure B. Deltamethrin IR Bottle Test Results: 10.5 ug/bottle. Shown are means and standard error bars for different groups of mosquitoes. Rock is considered a pyrethroid-sensitive strain. Puerto Rico is considered a pyrethroid-resistant strain. The control was treated with acetone only.

Results- The control (Acetone) mortality curve was significantly different ($P < 0.0001$) from the curves for all other treatments with little to no mortalities. Kaplan Meier statistical analysis confirmed that there was a significant difference between the curves plotted for the deltamethrin-exposed Rock Strain and the Las Cruces Strain ($P < .0001$). The curve plotted for the Puerto Rico Strain (a known resistant strain) is significantly different than that plotted for the Las Cruces Strain ($P < .0001$). The curves plotted for the Puerto Rico strain and the Rock strain were also significantly different from each other ($P < .0001$).

Conclusion- the Las Cruces *Aedes aegypti* strain showed resistance toward deltamethrin.

Deltamethrin Resistance Tests: Carlsbad strain

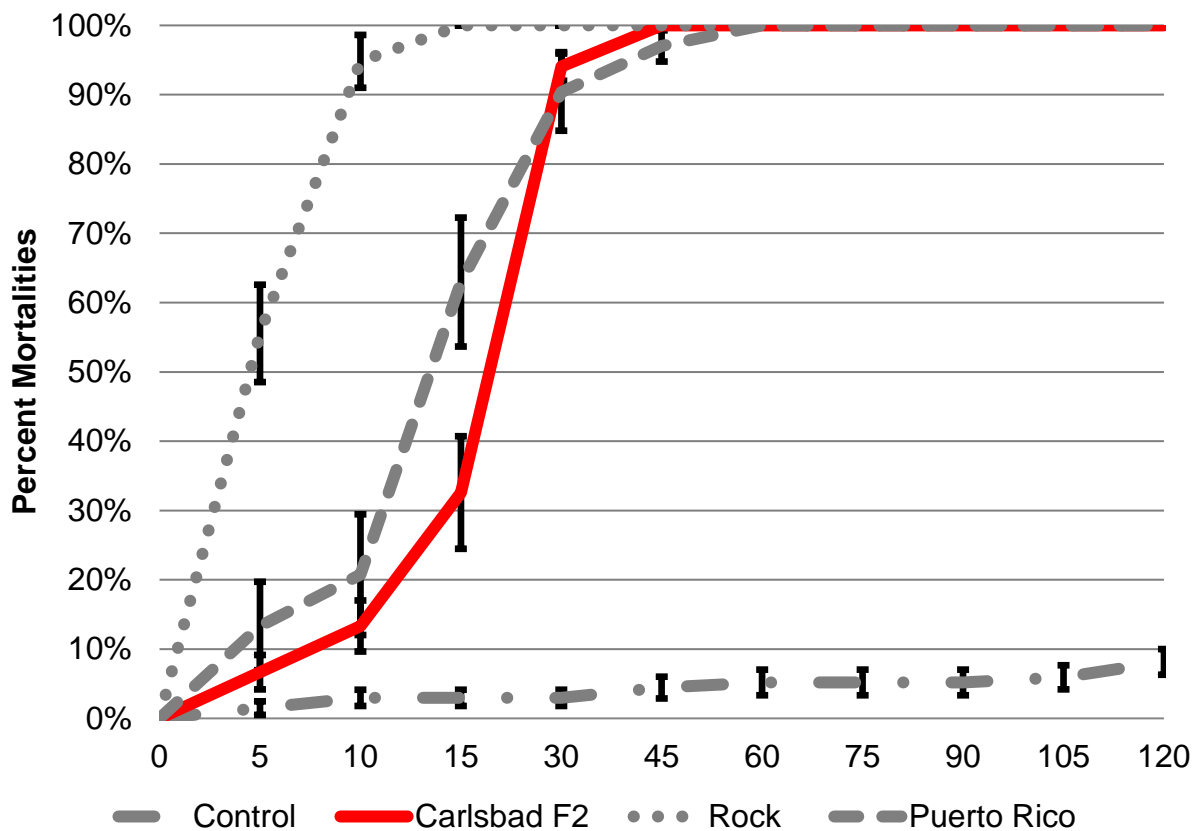


Figure C. Deltamethrin IR Bottle Test Results: 10.5 ug/bottle. Shown are means and standard error bars for different groups of mosquitoes. Rock is considered a pyrethroid-sensitive strain. Puerto Rico is considered a pyrethroid-resistant strain. The control was treated with acetone only.

Results- The control (Acetone) mortality curve was significantly different ($P < 0.0001$) from the curves for all other treatments with little to no mortalities. Kaplan Meier statistical analysis confirmed that there was a significant difference between the the curves plotted for the deltamethrin-exposed Rock Strain and the Carlsbad Strain ($P < .0001$). The curve plotted for the Puerto Rico Strain (a known resistant strain) is significantly different than that plotted for the Carlsbad Strain ($P < .003$). The curves plotted for the Puerto Rico strain and the Rock strain were also significantly different from each other ($P < .0001$).

Conclusion- The Carlsbad *Aedes aegypti* strain shows resistance to deltamethrin.

Deltamethrin Resistance Tests: Roswell strain

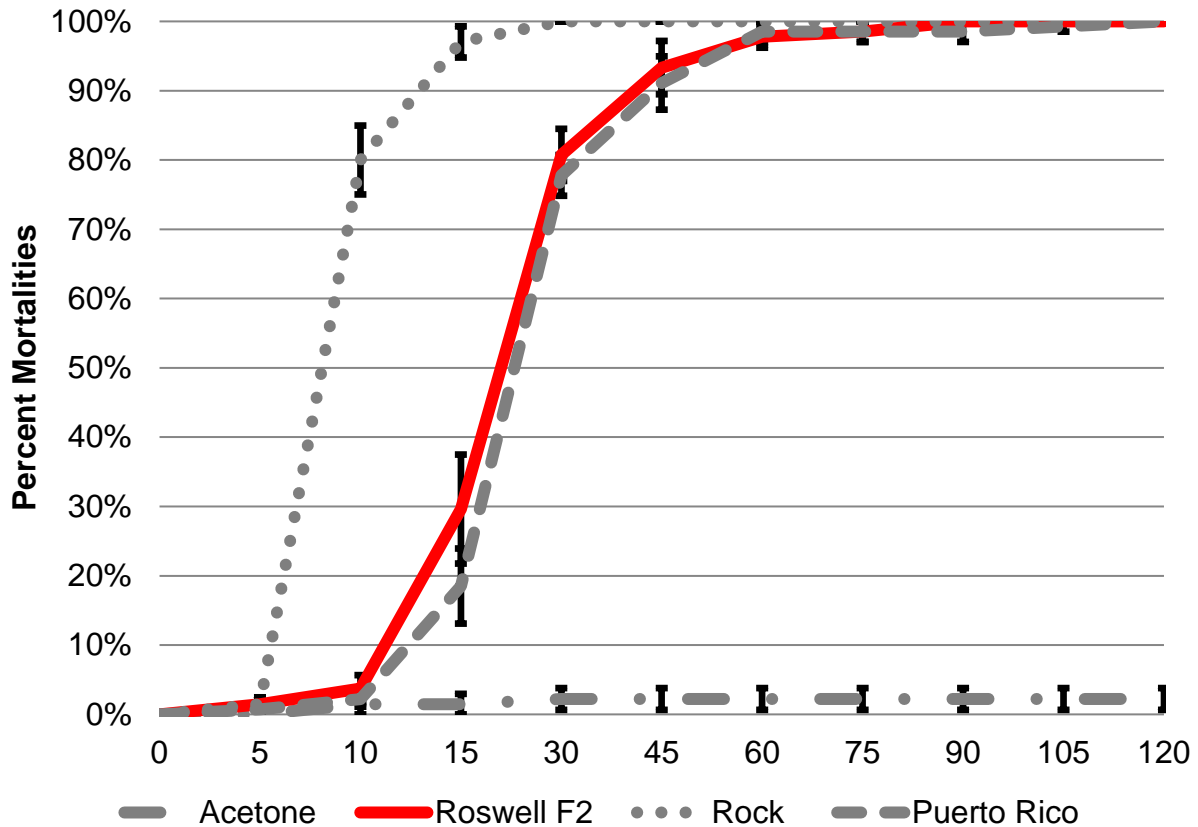


Figure D. Deltamethrin IR Bottle Test Results: 10.5 ug/bottle. Shown are means and standard error bars for different groups of mosquitoes. Rock is considered a pyrethroid-sensitive strain. Puerto Rico is considered a pyrethroid-resistant strain. The control was treated with acetone only.

Results- The control (Acetone) mortality curve was significantly different ($P < 0.0001$) from the curves for all other treatments with little to no mortalities. Kaplan Meier statistical analysis confirmed that there was a significant difference between the curves plotted for the deltamethrin-exposed Rock Strain and the Roswell Strain ($P < 0.0001$). The curve plotted for the Puerto Rico Strain (a known resistant strain) is not significantly different than that plotted for the Roswell Strain ($P = .091$). The curves plotted for the Puerto Rico strain and the Rock strain were also significantly different from each other ($P < 0.0001$).

Conclusion- The Roswell *Aedes aegypti* strain shows resistance towards deltamethrin.

Etofenprox Resistance Tests: Roswell strain

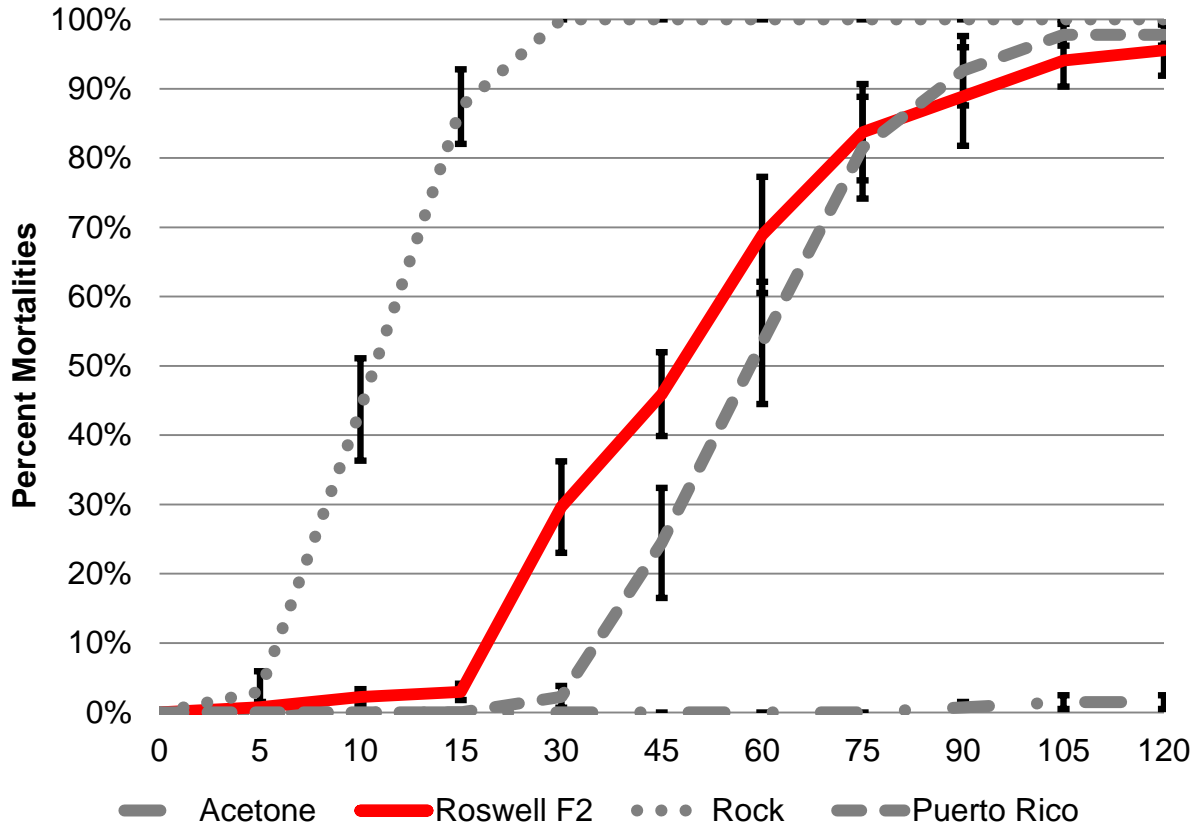


Figure E. Etofenprox IR Bottle Test Results: 37.5 ug/bottle. Shown are means and standard error bars for different groups of mosquitoes. Rock is considered a pyrethroid-sensitive strain. Puerto Rico is considered a pyrethroid-resistant strain. The control was treated with acetone only.

Results- The control (Acetone) mortality curve was significantly different ($P < 0.0001$) from the curves for all other treatments with little to no mortalities. Kaplan Meier statistical analysis confirmed that there was a significant difference between the curves plotted for the etofenprox-exposed Rock Strain and the Roswell Strain ($P < .0001$). The curve plotted for the Puerto Rico Strain (a known resistant strain) is significantly different than that plotted for the Roswell Strain ($P = .034$). The curves plotted for the Puerto Rico strain and the Rock strain were also significantly different from each other ($P < .0001$).

Conclusion- The Roswell *Aedes aegypti* strain shows resistance towards etofenprox.

Etofenprox Resistance Tests: Las Cruces strain

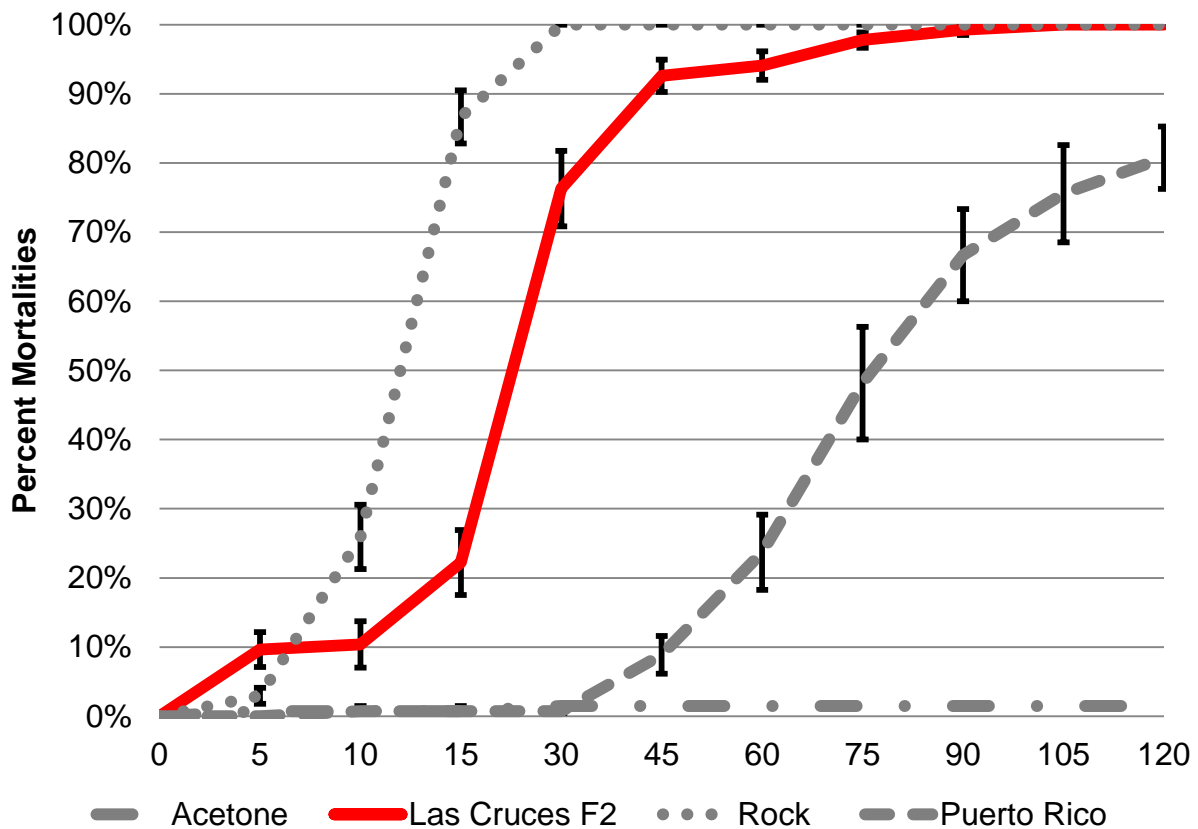


Figure F. Etofenprox IR Bottle Test Results: 37.5 ug/bottle. Shown are means and standard error bars for different groups of mosquitoes. Rock is considered a pyrethroid-sensitive strain. Puerto Rico is considered a pyrethroid-resistant strain. The control was treated with acetone only.

Results- The control (Acetone) mortality curve was significantly different ($P < 0.0001$) from the curves for all other treatments with little to no mortalities. Kaplan Meier statistical analysis confirmed that there was a significant difference between the curves plotted for the etofenprox-exposed Rock Strain and the Las Cruces Strain ($P < 0.0001$). The curve plotted for the Puerto Rico Strain (a known resistant strain) is significantly different than that plotted for the Las Cruces Strain ($P < 0.0001$). The curves plotted for the Puerto Rico strain and the Rock strain were also significantly different from each other ($P < 0.0001$).

Conclusion- The Las Cruces *Aedes aegypti* strain shows resistance towards etofenprox.

Chlorpyrifos Resistance Tests: Las Cruces strain

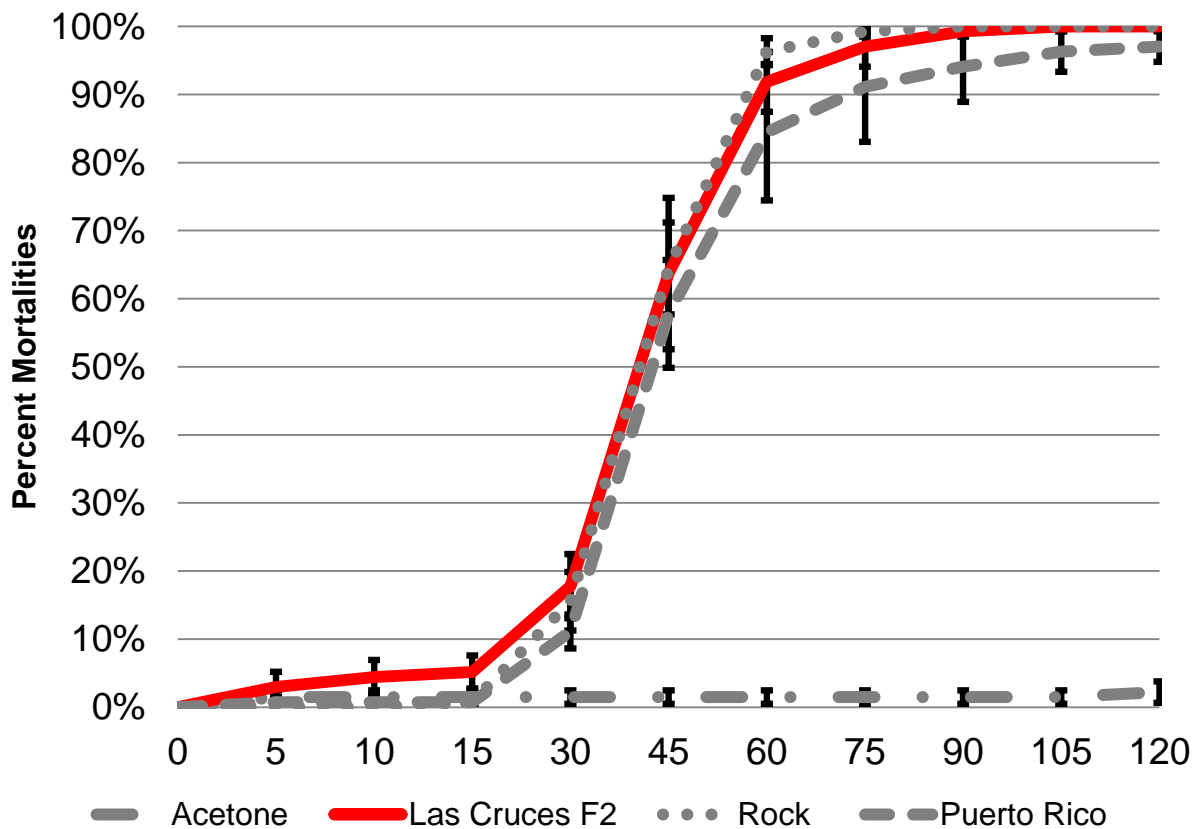


Figure G. Chlorpyrifos IR Bottle Test Results: 40 ug/bottle. Shown are means and standard error bars for different groups of mosquitoes. Rock is considered a pyrethroid-sensitive strain. Puerto Rico is considered a pyrethroid-resistant strain. The control was treated with acetone only.

Results- The control (Acetone) mortality curve was significantly different ($P < 0.0001$) from the curves for all other treatments with little to no mortalities. Kaplan Meier statistical analysis confirmed that there was no significant difference between the curves plotted for the chlorpyrifos-exposed Rock Strain and the Las Cruces Strain ($P = .589$). The curve plotted for the Puerto Rico Strain (a known resistant strain) is significantly different than that plotted for the Las Cruces Strain ($P = .014$). The curves plotted for the Puerto Rico strain and the Rock strain were also significantly different from each other ($P = .005$).

Conclusion- The Las Cruces *Aedes aegypti* strain is sensitive towards chlorpyrifos.

Chlorpyrifos Resistance Tests: Alamogordo strain

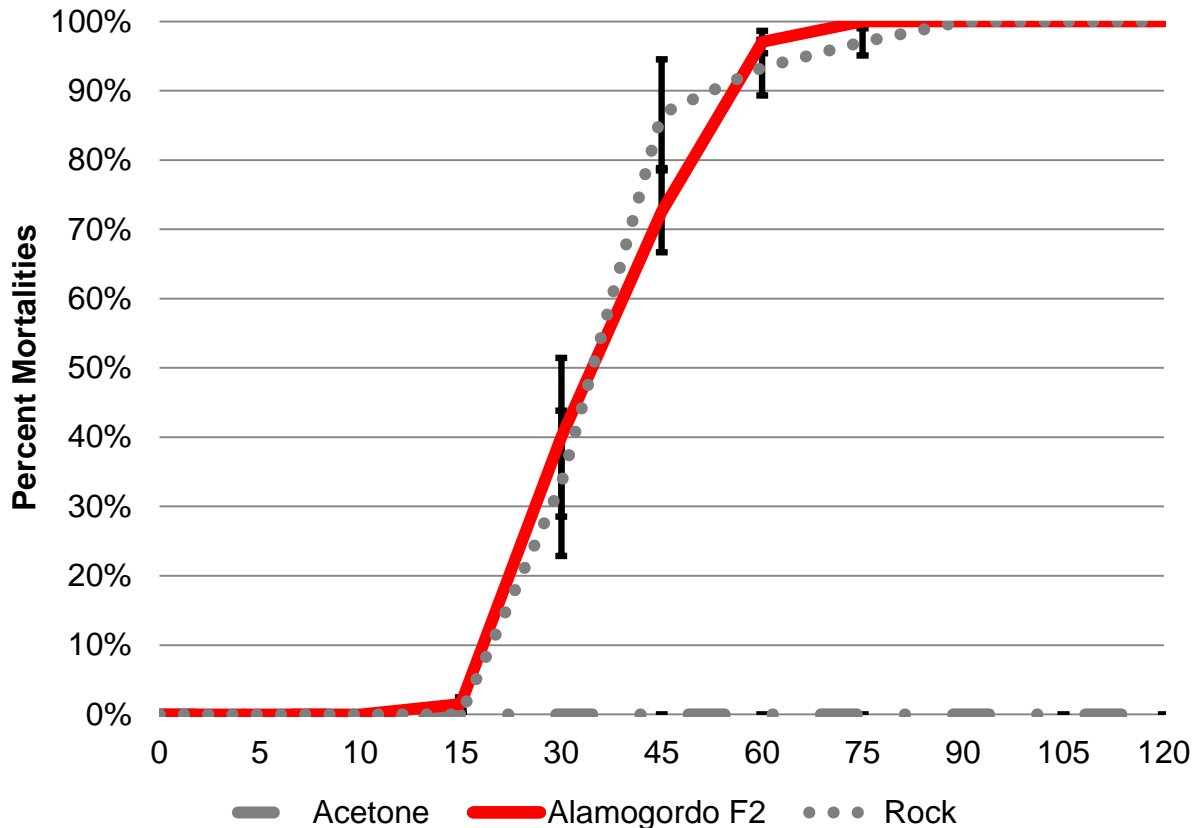


Figure H. Chlorpyrifos IR Bottle Test Results: 40 ug/bottle. Shown are means and standard error bars for different groups of mosquitoes. Rock is considered a pyrethroid-sensitive strain. The control was treated with acetone only.

Results- The control (Acetone) mortality curve was significantly different ($P < 0.0001$) from the curves for all other treatments with little to no mortalities. Kaplan Meier statistical analysis confirmed that there was not a significant difference between the curves plotted for the chlorpyrifos-exposed Rock Strain and the Alamogordo Strain ($P = .984$).

Conclusion- The Alamogordo *Aedes aegypti* strain is sensitive to chlorpyrifos.

Chlorpyrifos Resistance Tests: Carlsbad strain

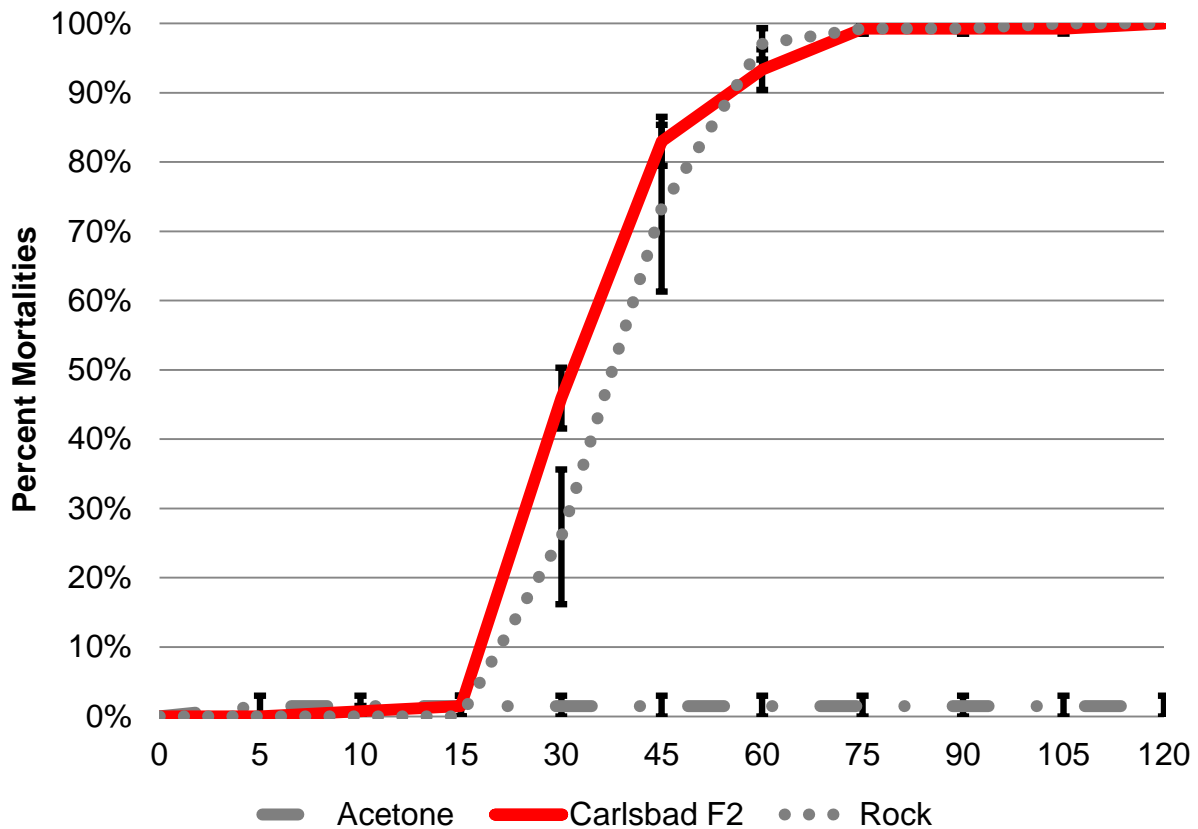


Figure I. Chlorpyrifos IR Bottle Test Results: 40 ug/bottle. Shown are means and standard error bars for different groups of mosquitoes. Rock is considered a pyrethroid-sensitive strain. The control was treated with acetone only.

Results- The control (Acetone) mortality curve was significantly different ($P < 0.0001$) from the curves for all other treatments with little to no mortalities. Kaplan Meier statistical analysis confirmed that there was a significant difference between the curves plotted for the chlorpyrifos-exposed Rock Strain and the Carlsbad Strain ($P = .028$).

Conclusion- The Carlsbad *Aedes aegypti* strain shows sensitivity towards chlorpyrifos.

Chlorpyrifos Resistance Tests: Roswell strain

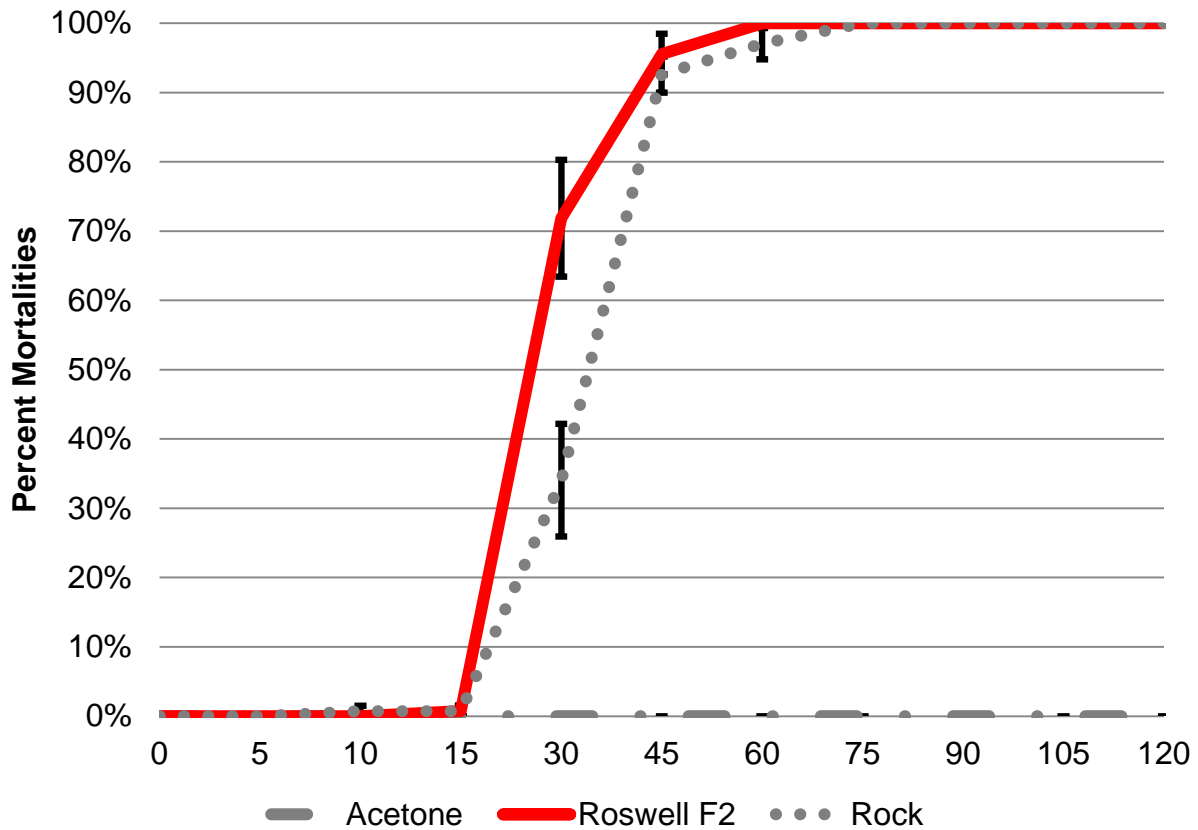


Figure J. Chlorpyrifos IR Bottle Test Results: 40 ug/bottle. Shown are means and standard error bars for different groups of mosquitoes. Rock is considered a pyrethroid-sensitive strain. The control was treated with acetone only.

Results- The control (Acetone) mortality curve was significantly different ($P < 0.0001$) from the curves for all other treatments with little to no mortalities. Kaplan Meier statistical analysis confirmed that there was a significant difference between the the curves plotted for the chlorpyrifos-exposed Rock Strain and the Roswell Strain ($P < .0001$).

Conclusion- The Roswell *Aedes aegypti* strain is sensitive to chlorpyrifos.

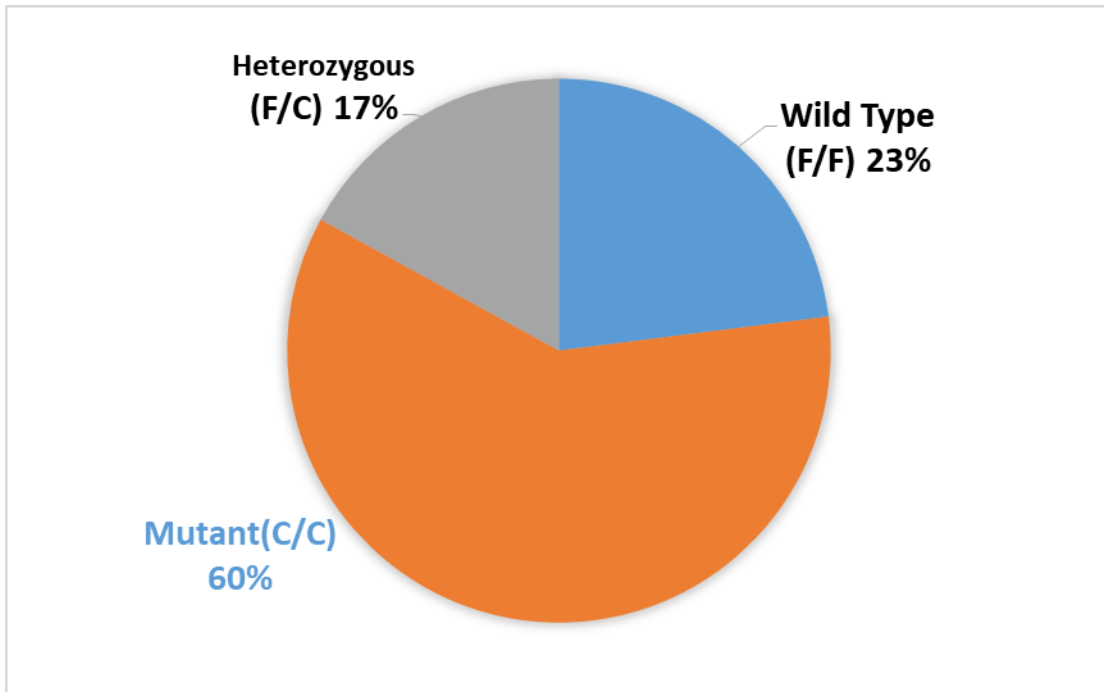


Figure K: kdr position 1534 Genotypes in 50 mosquitoes from New Mexico. The figure shows the percentage of heterozygous and homozygous mosquitoes. The resistant mutants are shown in orange, wild type in blue, and heterozygous in grey.

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NC_035109_egypt	MLVGDVSCIPFFLATVVIGNLVGLNLFLLALLSNFGSSSL SAPTADNETNKIAEAFNRIS	1060		
NW_004765908_Musca	MYVGDVSCIPFFLATVVIGNLVGLNLFLLALLSNFGSSSL SAPTADNDTNKIAEAFNRIS	1053		
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1011	1016			
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NW_004765908_Musca	RFKNWVKRNIADCFKLIRNKLTNQISDQ-----PSEHGDSNE	1089		
	:			
NC_035109_egypt	LELTPDDILADGLLKKGVKEHNQLEVAIGDGMEFTIHGDLKNGKKNKQLMNSKIVIGNS	1180		
NW_004765908_Musca	LELGHDEIMGDGLIKKGMKGETQLEVAIGDGMEFTIHGDMKNKPKKSKFINNNTMIIGNS	1149		
	*** * : * : . * * : * * * * : * . . * * * * * * * * * * * * * * * * : * * : * : * * * * : * * * * * * *			
NC_035109_egypt	ISNHQDNKLEHELNHRGMSLQDDDTASIKSYGSHKRNPFKDESHKGSAAETMEGEKRDVS	1240		
NW_004765908_Musca	I-NHQDNRELEHELNHRGLSIQDDDTASINSYGSHKRNPFKDESHKGSAAETIEGEKRDVS	1208		
	* * * * * : *			
NC_035109_egypt	KEDLGIDEELDDECDGEEGPLDGLIIHADE-DEVIEDSPADCCPDNCYKKFPVLGDDD	1299		
NW_004765908_Musca	KEDLGLDEELDEEAGDEQLDGLDIIHAQNDDIIDDYPADCFPDSYKFKPILAGDED	1268		
	* * * * * : * * * * * * * : * : * * * * * : * * : *			
NC_035109_egypt	APFQGWANLRLKFTQLIENKVFETAVITMILLSLALALEDVHLPHRPILQDVLVYMDR	1359		
NW_004765908_Musca	SPFQGWGNLRLKFTQLIENKVFETAVITMILMSSLALALEDVHLPRPVMQDILVYMDR	1328		
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NC_035109_egypt	IFTVIFLEMLIKWLALGFRVYFTNAWCWLD FIVMVSLINVASLCGAGGIQAFKTMRT	1419		
NW_004765908_Musca	IFTVIFLEMLIKWLALGFKVYFTNAWCWLD FIVMVSLINLVAVWVGLNDIAVFRSMRT	1388		
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NC_035109_egypt	LRLRPLRAMSRMQMRVVNALVQAIPISFNVLVCLIFWLI FAIMGVQLFAGKYFKCV	1479		
NW_004765908_Musca	LRLRPLRAVSRWEGMKVVNALVQAIPISFNVLVCLIFWLI FAIMGVQLFAGKYFKCK	1448		
	* * * * * * * : * * : *			
NC_035109_egypt	DNKNTTLSHEIIPDVNACVAENYTWNSPMNFDHVKGAYLCLFQVATFKGWIQIMND AID	1539		
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NC_035109_egypt	SREVGKQPIRETNIYMVLYFVFFIFIGSFRTLNLFI GV I IDNFNQK KAGGSLEMFMTE	1599		
NW_004765908_Musca	SREVDKQPIRETNIYMVLYFVFFIFIGSFRTLNLFI GV I IDNFNQK KAGGSLEMFMTE	1568		
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NW_004765908_Musca	DQKKYNNAMKMGSKKPLKAI PRWRPQAIVFEIVTDKKFDI IIMLFI GLNMF TMTLDR	1628		
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NC_035109_egypt	YKQTDTFSAVLDYLNMFICIFSSCELMKIFALRYHYFIEPWNLFDFVVLVLSILGLVLS	1719		
NW_004765908_Musca	YDASEAYNVDLKNLGI FVVFISGECLLIFALRYHYFKEPWNLFDFVVLVLSILGLVLS	1688		
	* . : : : . * * * * * * * : *			
NC_035109_egypt	DLIEKYFVSPPTLLRVRVAVGRVLRVLKGA GIRTLLFALAMSLPALFNICLLLV FVMF	1779		
NW_004765908_Musca	DLIEKYFVSPPTLLRVRVAVGRVLRVLKGA GIRTLLFALAMSLPALFNICLLLV FVMF	1748		
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NC_035109_egypt	IFAIFGMSFFMHVYKXSGLDVYVFKTFGQSMILLFQ MSTSAGWDGVL DGI INEDECLPP	1839		
NW_004765908_Musca	IFAIFGMSFFMHVKKXSGINAVYVFKTFGQSMILLFQ MSTSAGWDGVL DAI INEEDCPP	1808		
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NW_004765908_Musca	DNDKGYPGNCGSATVGTIFLLSYLVISFLIVINMYIAVILENYSQATEDVQEGLTDDD YD	1868		
	* *			
NC_035109_egypt	MYYEIWQQFDPDGTQYIRYDQLSDFLDVLEPPLQIHKPNKYKI ISMDIPICRGDMMFCVD	1959		
NW_004765908_Musca	MYYEIWQQFDPEGTQYIRYDQLSEFLDVLEPPLQIHKPNKYKI ISMDIPICRGDMMYCV	1928		
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NC_035109_egypt	ILDALTKDFFARKGNPIEETAELGEVQARPDEVGYEPV SSTLWRQREEYCARVIQHAWRK	2019		
NW_004765908_Musca	ILDALTKDFFARKGNPIEETGEIGEARPDETYD PVSSTLWRQREEYCAKLIQNAWRR	1988		
	* *			
NC_035109_egypt	HKERQAGGGGDDTDADACNDDGDDGGGGAGDGGG SAGGGV T-SPGVGSGSIVGGGTTP	2078		
NW_004765908_Musca	YKNGPPQEGDEG---EAAGGEDG AEGGEGEGSGGGGGGDDGG SATGATAAAAGATSP	2044		
	: * : * * : * * . * * * * . * * * * . . . * : * * : * * * * : *			

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NC_035109_aegypt      GSGGGGSQANLGI VVEHNLSPKESP DGNNDPQGRQTAVLVESDGFVTKNGHRVVIHSRSP 2138
NW_004765908_Musca   SDPDAGEA--DGASVGGPLSPGCV---SGGSNGRQTAVLVESDGFVTKNGHKVVIHSRSP 2099
.. ..*      * * ***      ... :*****:*****

NC_035109_aegypt      SITRSADV      2147
NW_004765908_Musca   S-----      2100
*
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Figure L. Sequence alignment of the Para protein of *Aedes aegypti* and *Musca domestica*. Kdr mutation sites are red and the respective location is given in boxes above and below the alignment.

City	Pesticides - Product Name	Pesticides - Active Ingredient
Las Cruces	not provided	Etofenprox Deltamethrin Chlorpyrifos Bifenthrin Lambda Cyhalothrin
T or C	Prentox Aqua Perm-X UL 30-30 Altosid XR Briquets Aqualuer 20-20	Permethrin and Piperonyl Butoxide (S)-Methoprene Permethrin and Piperonyl Butoxide Technical
Roswell	no info	no info
Lordsburg	none	n/a
Alamogordo	Vectobac	<i>Bacillus thuringiensis</i> subsp. <i>israelensis</i>
Clovis	Delta Guard Atrapa	Deltamethrin Malathion
Sunland Park	Altosid products (2015, 2016, 2017) Fyfanon (2016, 2017) Kontrol 4x4 (2015) Vectolex CG products (2013, 2014) Evoluer 30-30 (2013, 2014) Sustain MBG Pellets (2013)	(S)-Methoprene Malathion Permethrin and Piperonyl Butoxide <i>Bacillus sphaericus</i> Permethrin and Piperonyl Butoxide <i>Bacillus thuringiensis</i> subsp. <i>israelensis</i>
Portales	no info	no info
Carlsbad	Mosquito Master Delta Guard Altosid Fourstar BTI Natar™ products Permex Pursuit PyroFos 1.5 ULV (discontinued by the manufacturer)	Chlorpyrifos and Permethrin Deltamethrin (S)-Methoprene <i>Bacillus thuringiensis</i> subsp. <i>Israelensis</i> Spinosad (soil bacterium) Permethrin Permethrin and Piperonyl Butoxide Chlorpyrifos
Lovington	Mosquito Master BTI Briquets	Chlorpyrifos and Permethrin <i>Bacillus thuringiensis</i> subsp. <i>Israelensis</i>
Deming	not provided	Etofenprox Deltamethrin Chlorpyrifos Bifenthrin Lambda Cyhalothrin
Socorro	not provided	Permethrin

Table A. Pesticide-use in selected cities across New Mexico.

City	# sites contributing to adults	# adults added to cage
Alamogordo	9	28
Carlsbad	8	144
Deming	6	28
Las Cruces	9	60
Lovington	5	20
Roswell	8	22
Sunland Park	12	72

Table B. Number of founding individuals for New Mexico *Aedes aegypti* strains used in this study. Traps were set for 24 hours at the respective sites.