FINAL PROJECT REPORT

Project Title: Erythritol: An artificial sweetener with insecticidal properties

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Cooperators: None

Other funding sources: None

Total Project Funding: $16,310

Budget History:

Item	**Year 1: 2019**
WTFRC expenses	
Salaries	$14,690, USDA; $1560, WSU
Benefits	$1,175, USDA; $145, WSU
Wages	
Benefits	
Equipment	
Supplies	$445
Travel	$1917, WSU
Plot Fees	
Miscellaneous	
Total	$19,932

ORIGINAL OBJECTIVES

The project directly addressed several research priorities identified by the Pear Research Sub-committee, namely integrated control of pear psylla, mites, and codling moth. Specific objectives included:

1. Examine insecticidal and deterrent properties of erythritol against arthropod pests of pear under controlled laboratory conditions.

Subobjective 1a: Determine the lowest dose of erythritol needed to cause pear psylla mortality and determine whether erythritol has antibiotic (toxicity) or antixenotic (repellency) effects against psylla.

Subobjective 1b: Determine whether erythritol is lethal to pear rust mite and spider mite.

Subobjective 1c: Determine whether erythritol is lethal to codling moth larvae.

2. Determine whether foliar applications suppress arthropod pest populations in a pear orchard.

SIGNIFICANT FINDINGS

Objective 1:

Subobjective 1a: Erythritol was psyllicidal when applied to pear at mixtures of 20% w/v and killed nearly all psylla nymphs and adults within three days. The psyllicidal effects were both antibiotic and antienotic.

Subobjective 1b: Erythritol was lethal to both pear rust mite and spider mite.

Subobjective 1c: Erythritol was lethal to codling moth larvae when added to artificial diet.

Objective 2:

Foliar applications of erythritol significantly reduced pear psylla nymph populations under field conditions, but only when erythritol was completely dissolved into solution by aid of heat. Reductions in blister mite damage was also noted.

RESULTS & DISCUSSION

Subobjective 1a. There were significant differences among treatments in pear psylla feeding activity following a 24-h settling time (Fig. 1; F=9.1; d.f.=2, 39; P=0.006). Fewer pear psylla were observed feeding on erythritol diets compared with those consisting of sucrose or water only (Fig. 1). There were also significant differences among treatments in adult longevity (Fig. 2; $\chi 2$=49.6; d.f.=2; P<0.001). Survival time was significantly reduced for pear psylla that were fed erythritol compared with those that were fed pure water or 30% sucrose. After demonstrating that 30% erythritol was lethal to pear psylla adults when ingested from liquid diets, we examined the lethality of

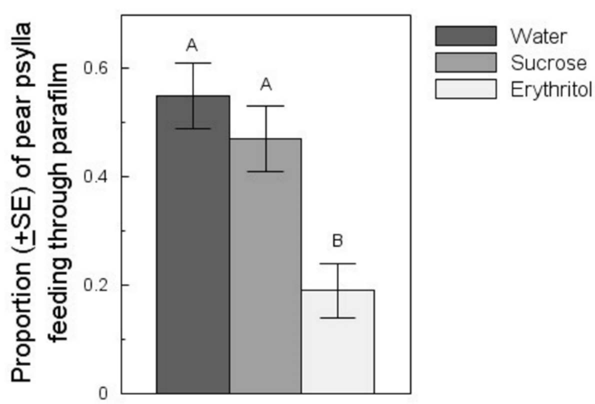

Figure 1. Proportion (±S.E.) of living psylla adults that were actively feeding after a 24-h settling time on liquid diets consisting of water only, 30% erythritol, or 30% sucrose. Different letters denote significant differences among treatment means.

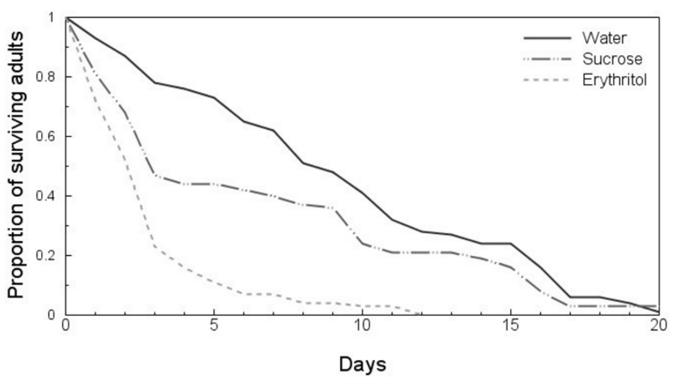

Figure 2. Survival rates of pear psylla adults on liquid diets consisting of water, 30% sucrose, or 30% erythritol. Although the x-axis is limited to 20 days to allow visual comparisons, survival on water and sucrose exceeded 30 days.

various erythritol concentrations ranging from 0 to 30%. Analysis of the 3-d mortality rates of pear psylla indicated significant differences among treatments (F=16.0; d.f.=4, 38; P<0.001) where morality rates generally increased with increasing concentrations of erythritol (Fig. 3).

Results of these assays demonstrate that erythritol has dose-dependent insecticidal properties when ingested from liquid solutions by pear psylla adults. We did not specifically compare the effects of erythritol on winterform and summerform psylla, but our assays included either morphotype depending upon availability and demonstrated that erythritol is lethal to both seasonal morphotypes. Results of these assays are consistent with reports that erythritol is insecticidal (Baudier et al. 2014, O'Donnell et al. 2016, Choi et al. 2017, Burgess and King 2018, Sampson et al. 2018, Gilkey et al. 2018, Caponera et al. 2019), but our study is the first to demonstrate that erythritol is lethal to a phloem-feeding insect.

Observations after the 24-h settling time revealed altered behavior or impaired motor skills by pear psylla that ingested erythritol. Pear psylla that were fed erythritol appeared to spend more time grooming their stylets compared with psylla that were fed sucrose or water. In addition, many of the erythritol-fed psylla appeared to have an impaired ability to climb on the lid of the petri dish. While these observations should be interpreted cautiously because our experiment was not specifically designed to examine the effects of erythritol on the behavior of pear psylla, these observations are indeed consistent with a report of impaired motor functions by fruit flies after ingesting erythritol. In that experiment, *Drosophila melanogaster* adults that were fed erythritol exhibited a decreased ability to climb compared with adults that were fed water or other artificial sweeteners (Baudier et al. 2014).

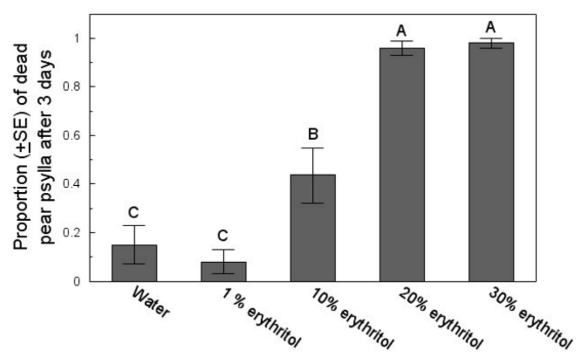

Figure 3. Three-day mortality rates (±S.E) of pear psylla fed increasing doses of erythritol in liquid diets. Different letters denote significant differences among treatment means.

Laboratory assays with excised leaves were used to examine the effects of foliar treatment of erythritol on mortality of pear psylla adults and nymphs. Mortality rates of both nymphs (F=6.7; d.f.=2, 11; P=0.013) and adults (F=6.0; d.f.=2, 23; P=0.008) were significantly different among treatments (water, 30% sucrose, and 30% erythritol). Mortality rates of nymphs confined to leaves treated with water or sucrose were similar, but mortality was significantly higher for nymphs that were confined to leaves treated with erythritol (Fig. 4A). All pear psylla adults that were confined to leaves treated with water survived, so this

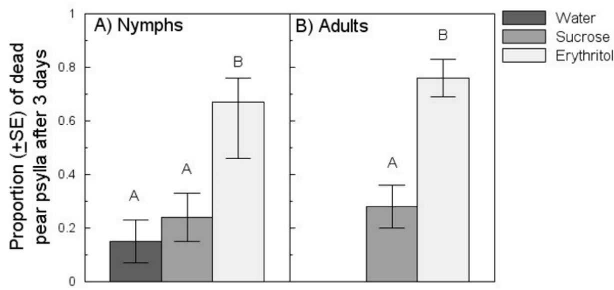

Figure 5. Three-day mortality (±S.E.) of pear psylla adults (A) and nymphs (B) confined to pear leaves treated with water, 30% sucrose, or 30% erythritol. Different letters denote significant differences among treatment means.

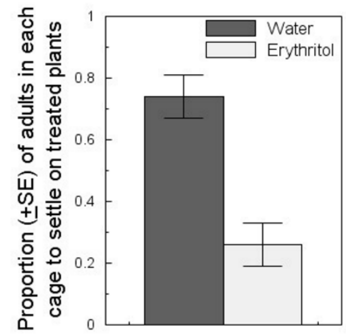

Figure 4. Mean proportion (±S.E.) of the total number of pear psylla adults in each cage to settle on pear leaves treated with water (control) or 30% erythritol in preference assays.

treatment was removed from the analysis. As observed for nymphs, significantly more adults died after three days when confined to erythritol-treated leaves compared with those confined to sucrose-treated leaves (Fig. 4B). Nearly all nymphs, regardless of treatment were still living three days after being briefly submersed in erythritol, sucrose, or water, suggesting that the psyllicidal properties are due to ingestion of erythritol, not residual activity. Results of our choice-preference assays showed a clear preference by adults for untreated versus erythritol-treated leaves (Fig. 5; F=16.9; d.f.=1, 12; P=0.002).

Subobjective 1b. Erythritol was moderately toxic to twospotted spider mite on contact, but only at the highest (30%) rate; the lower rates were not different from the check (Fig. 6). Runoff was low throughout the test, indicating the materials tested were not repellent. The low mortality in the check (8%) and the high mortality in the FujiMite standard (98%) indicate a valid bioassay response.

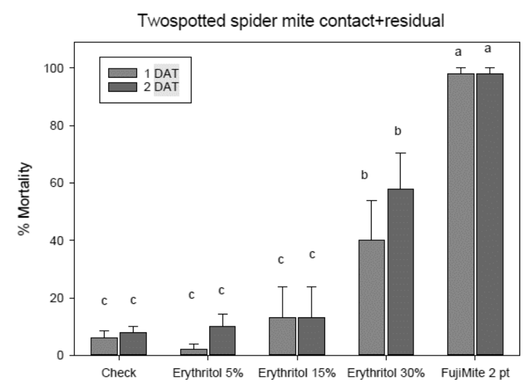

Figure 6. Mortality of twospotted spider mite following contact and residual exxposure to erythritol and FujiMite.

Mortality following exposure to residues was low to negligible in all treatments, although significantly higher in the FujiMite treatment; the erythritol treatments were not significantly different from the checks. Egg numbers were significantly lower in the FujiMite and 30% erythritol treatments in comparison to the check (Fig. 7).

In the contact+residual bioassay at 2 DAT, mortality of female *G. occidentalis* was 100% in the FujiMite standard, but moderate to high in the erythritol treatments (Fig. 8). Inexplicably, the highest rate of the erythritol produced mortality that was lower (20%) than the two lower rates, which averaged about 58% mortality. Egg numbers at 2 DAT were lower in the FujiMite and erythritol 15 and 30% treatments.

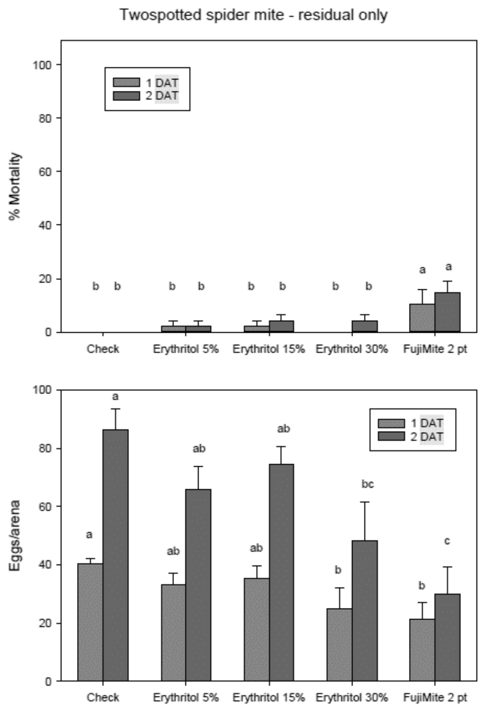

Figure 7. Mortality (upper) and fecundity (lower) of twospotted spider mites exposed to residues of erythritol and FujiMite.

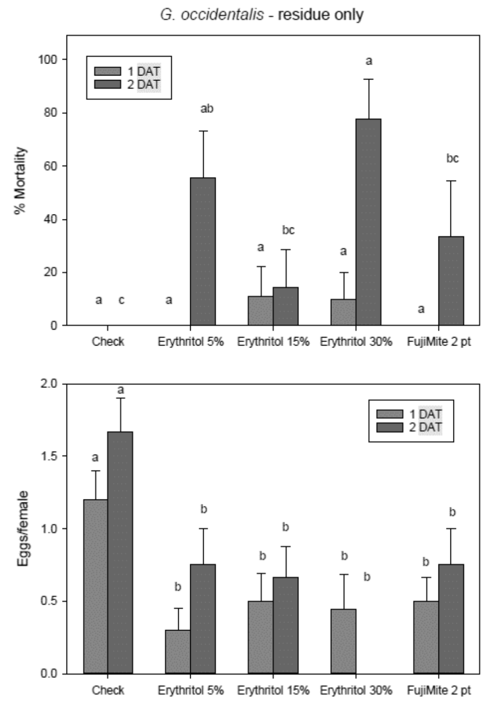

Figure 8. Mortality (upper) and fecundity (lower of western predatory mites exposed to residues of erythritol and FijiMite.

In the residual-only bioassay, there was very little mortality in any of the treatments, including the FujiMite standard, after 24 h. Mortality increased considerably at 48 h, with the highest levels in two of the erythritol treatments (5 and 30%). There was no rate effect due to the low levels of mortality in the 15% treatment; however, there was a trend for the erythritol treatments to cause more mortality than FujiMite. Erythritol appears to have only slightly lower toxicity to *G. occidentalis* by exposure to residues as it is by direct contact. Egg numbers were reduced by all treatments relative to the check.

Pear rust mite mortality was low in the check and high (100%) in the standard (Nexter) treatment at 2 DAT, bracketing the erythritol treatments (Fig. 9). The three rates tested ranged from 33 to 74%, indicating a moderate degree of activity. While several applications at the high rate may be necessary, this material shows promise of an organic rust mite material.

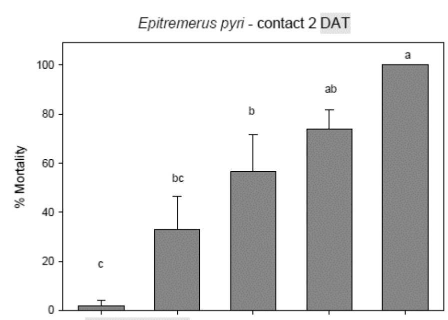

Figure 9. Mortality of pear rust mites exposed by contact of erythritol and Nexter.

Subobjective 1c. Because of uncertainties about when codling moth larvae enter fruit (Garczynski, personal communication), the effects of erythritol on codling moth was tested by mixing 30% erythritol into standard codling moth rearing diet and examining for mortality. Within a week, all codling moth larvae confined to diet mixed with erythritol had died and were still located on the surface of the diet. In contrast, all larvae confined to control diets were still living and had burrowed into the diet.

Objective 2, Field trials. A previous report indicated that higher concentrations of erythritol is harmful to both corn and tomato seedlings (Scanga et al. 2018), but erythritol did not appear to damage the foliage of pear. Repeated measures analysis of the numbers of pear psylla nymphs collected in an orchard near Moxee, WA did not indicate a significant treatment by week interaction indicating that the effects of treatment were similar among weeks (F=2.3; d.f.=4, 12; P=0.118). Numbers of nymphs on all treatments declined from week 1 to week 3 (Fig. 10A; F=16.6; d.f.=2, 6; P=0.004), and were lower on trees treated with erythritol or erythritol plus Regulaid compared with controls (Fig. 10B; F=6.2; d.f.=2, 6; P=0.035). Numbers of adults did not differ among treatments (F=0.22; d.f.=2, 36; P=0.805) and there was no treatment by week interaction (F=0.9; d.f.=10,36; P=0.547), but numbers of adults generally declined each week regardless of treatment (F=22.0; d.f.=5, 18; P<0.001).

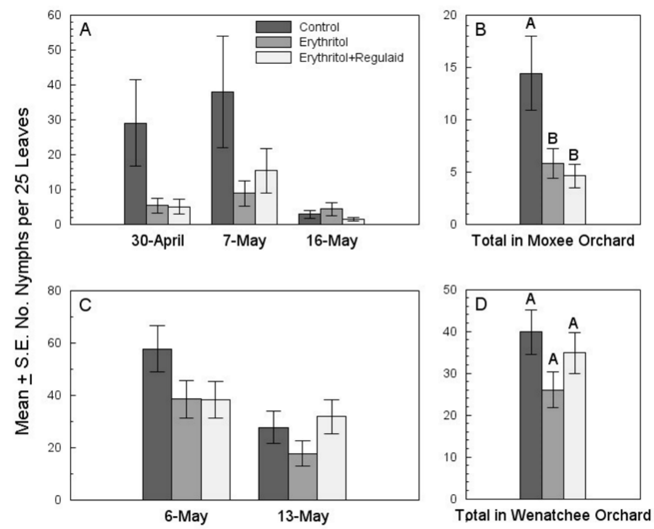

Figure 10. Mean number (±S.E.) of pear psylla nymphs per 25 leaves on trees treated with control (water mixed with surfactant), erythritol, or erythritol mixed with a surfactant in orchards near Moxee, WA (A and B) or Wenatchee, WA (C and D). Different letters within dates denote significant differences among treatment means.

Repeated measures analysis of the numbers of pear psylla collected in an orchard near Wenatchee, WA did not indicate a significant treatment by week interaction for nymphs (F=1.4; d.f.=2, 8; P=0.312) or adults (F=0.7; d.f.=12, 48; P=0.760). As observed in Moxee, populations of nymphs (Fig. 10C; F=11.1; d.f.=1, 4; P=0.029) and adults (F=50.2; d.f.=6, 24; P<0.001) declined from the initial sampling date. Although there were numerically fewer nymphs on erythritol-treated trees than on control trees on week 1 (Fig. 10D), the analysis did not indicate significant differences among treatments for nymphs (F=2.0; d.f.=2, 8; P=0.195) or adults (F=0.5 d.f.=2, 8; P=0.608).

The experiment conducted at the Moxee location was consistent with the results obtained from laboratory assays but results of field experiments from the two locations conflicted. The major difference in methods used at the two locations is the method in which the erythritol treatments were prepared. Solutions were heated to dissolve the erythritol before treatment of trees at the Moxee location, but not before treatment at the Wenatchee location. We conducted a follow-up laboratory experiment using excised leaves to compare pear psylla survival on leaves treated with erythritol that was dissolved into solution by heating versus erythritol that was suspended in water without heat. That experiment indicated that heat-dissolving is required for erythritol to be effective as an insecticide against pear psylla when applied to pear leaves (Fig. 11; F=9.4; d.f.=2, 11; P=0.004). Overall, results or our field experiments are consistent with other reports that demonstrated insecticidal properties of erythritol under field conditions (Sampson et al. 2017, 2018).

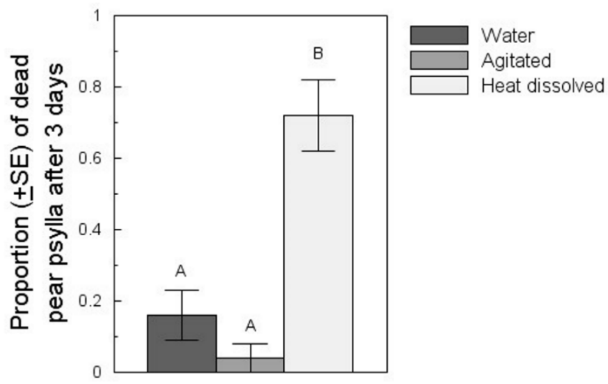

Figure 11. Three-day mortality (±S.E.) of pear psylla adults confined to pear leaves treated with water, 30% erythritol that was agitated into solution, or 30% erythritol that was dissolved into solution by heat treatment. Different letters denote significant differences among treatment means.

LITERATURE CITED

Braudier, K. M., S. D. Kaschock-Marenda, N. Patel, K. L. Diangelus, S. O'Donnell, and D. R. Marenda. 2014. Erythritol, a non-nutritive sugar alcohol sweetener and the main component of Truvia, is a palatable ingested insecticide. PLoS One 9: e9849.

Burgess, E. R., and B. H. King. 2018. Insecticidal potential of two sugar alcohols to *Musca domestica* (Diptera: Muscidae). J. Econ. Entomol. 110: 2252-2258.

Caponera, V., M. Barrett, D. R. Marenda, and S. O'Donnell. 2019. Erythritol ingestion causes concentration-dependent mortality in eastern subterranean termites (Blattodea: Rhinotermitidae). J. Econ. Entomol. in press.

Choi, M. Y., S. B. Tang, S. J. Ahn, K. G. Amarasekare, P. Shearer, and J. C. Lee. 2017. Effect of non-nutritive sugars to decrease the survivorship of spotted wing drosophila, *Drosophila suzukii*. J. Insect Physiol. 99: 86-94.

Gilkey, P. L., D. T. Bolshakov, J. G. Kowala, L. A. Taylor, S. O'Donnell, D. R. Marenda, and L. K. Sirot. 2018. Lethal effects of erythritol on the mosquito *Aedes aegypti* Linnaeus (Diptera: Culicidae). J. Appl. Entomol. 142: 873-881.

Marenda, D. R. and S. O'Donnell. 2015. Use of erythritol or compositions comprising same as mammal-safe insecticides. Patent # W2015/153957.

O'Donnell, S., K. Baudier, and D. R. Marenda. 2016. Non-nutritive polyol sweeteners differ in insecticidal activity when ingested by adult *Drosophila melanogaster* (Diptera: Drosophilidae). J. Insect Sci. 16: 47.

O'Donnell, S., K. Baudier, K. Fiocca, and D. R. Marenda. 2017. Erythritol ingestion impairs adult reproduction and causes larval mortality in *Drosophila melanogaster* fruit flies (Diptera: Drosophilidae). J. Appl. Entomol. 142: 37-42.

Sampson, B. J., C. T. Werle, S. J. Stringer, and J. J. Adamczyk. 2017. Ingestible insecticides for spotted wind drosophila control: A polyl, erythritol, and an insect growth regulator, Lufenuron. J. Appl. Entomol. 141: 8-18.

Sampson, B. J., M. W. Easson, S. J. Stringer, C. T. Werle, D. Magee, and J. J. Adamczyk. 2019. Laboratory and field assessments of erythritol derivatives on the survival, reproductive rate, and control of *Drosophila suzukii* (Diptera: Drosophilidae). J. Econ. Entomol. 112: 173-180.

Scanga, S. E., B. Hasanspahic, E. Zvornicanin, J. S. Kozenjic, A. K. Rahme, and J. H. Shinn-Thomas. 2018. Erythritol, at insecticidal doses, has harmful effects on two common agricultural crop plants. PLoS One 13: e0192749.

EXECUTIVE SUMMARY

Project Title: Erythritol: An artificial sweetener with insecticidal properties

Key words: pear psylla, twospotted spider mite, rust mite, codling moth

Abstract:

Although safe for human consumption, the artificial sweetener, erythritol, has been shown to be insecticidal against fruit flies. We found that 20-30% erythritol is also insecticidal to pear psylla, twospotted spider mites, rust mite, and codling moth. Erythritol could be developed into an organic insecticide to manage key pear pests.