California
CELERY RESEARCH
ADVISORY BOARD

2018-19 Annual Report

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TABLE OF CONTENTS

The program was established in October 1976 under provisions of the California Marketing Act, following written assent of a majority of the industry's producers. The program is administered by the Celery Advisory Board, which is nominated at district meetings every three years by the industry and appointed by the Secretary of Food and Agriculture.

The function of the Board is to ascertain research needs that will benefit the widest spectrum of the industry in all producing districts, to establish priority of need, enter into agreements with approved agencies to initiate or continue such research, develop a budget for funding, give impetus, direction and evaluation to the conduct of the work, and report results to the industry periodically.

The Board represents the entire industry and all its actions and recommendations are subject to the approval of the California Secretary of Food and Agriculture. Board members serve without compensation. Funds budgeted by the Board for research projects amount to about one-fourth the total cost of the projects, with the University of California supplying the remaining three-fourths in supervisory research personnel, plant facilities and major equipment. Research initiated by the Board would not be conducted without the industry's contribution --- through equitable assessment --- for certain special equipment, supplies, staff research assistants, expenses, travel, etc., since neither funds nor nonpartisan direction are available from other sources.

RESEARCH PRIORITIES

Production	✓ Evaluate insect management tools, alternatives to organophosphates and carbamates (especially products which are subject to restriction or loss due to FQPA and other regulations - e.g., acephate, diazinon, carbamates)
	✓ Study the biology and management of soil pests
	✓ Study management of Sclerotinia (airborne and soil borne)
	✓ Evaluate new celery varieties for resistance to insects and diseases
	✓ Evaluate biofumigants as tools for pest control in celery
	✓ Develop resistance management strategies for all pest categories
	✓ Continue weed control research to find complimentary or replacement products for Lorox/linuron Caparol/prometry and Caparol/Prometryn
	✓ Evaluate the secondary effect of loss of methyl bromide as a tool used in rotational crops on celery production areas
Regulatory	✓ Registrants should insure that air and chemigation labels are a part of all new product registrations
	✓ Address REI issues/concerns for products used in late season (e.g. aphicides)
	✓ Multiple products should be allowed under 24(c) and Section 18's for resistance management
	✓ Expedite registration of Dual Magnum/S-metolachlor for nutsedge control
Educational	✓ Educate regulators, legislators, and policy makers on need for more than one product for a particular pest for effective resistance management
	✓ Educate regulators on what a suitable replacement product is and what constitutes commercially acceptable levels of control
	✓ Educate growers, PCAs, agencies and the urban community on the relationship of waterway management to weed and insect pests
	✓ Educate the urban population about agricultural practices necessary for celery production
	✓ Educate the public on the nutritional values of California grown celery and the high level of food quality standards established for this commodity

The Board's Pest Management Strategic Plan can be viewed on the Internet at:

http://www.ipmcenters.org/pmsp/pdf/CAcelery.pdf

CALIFORNIA CELERY RESEARCH BOARD

September 1, 2018 through August 31, 2019

DIST	MEMBER	ALTERNATE
2	**Adrian Zendejas** Desert Mist Farms	**Jeff Percy** Desert Mist Farms
DIST	**MEMBER**	**ALTERNATE**
3	**Hank Laubacher Jr** Laubacher Farms	**Alfredo Esquivias** Tanimura & Antle
3	**Geremy Olsen** Deardorff Family Farms	**VACANT**
3	**Danny Pereira – Chairman** Rio Farms - **RES COMM**	**Vicki Pierce** Duda Farm Fresh
3	**Steve Adams – Res/Vice Chairman** Boskovich Farms - **RES COMM**	**Mike Naumann** Naumann Farms
3	**Ryan Coultas** Coultas Vegetable	**VACANT**
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	Steve Donovan - **RES COMM**	

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CALIFORNIA CELERY RESEARCH ADVISORY BOARD

BUDGETARY COMPARISON SCHEDULE
FOR THE YEAR ENDED SEPTEMBER 30, 2019

REVENUE	BUDGET	ACTUAL	VARIANCE
Assessments	290,000	311,503	21,503
Interest	950	2,688	1,738
Total Revenue	290,950	314,191	23,241
EXPENDITURES			
Administrative:			
Annual Report	100	-	100
Audits	3,150	3,300	(150)
Insurance	600	535	65
Management Services	52,800	52,800	-
Meetings	500	327	173
Miscellaneous	100	-	100
Office Supplies	800	547	253
Postage	600	500	100
Telephone	700	509	191
Travel and Mileage	800	669	131
Website	250	239	11
	60,400	59,426	974
Research:			
Production Research	214,795	203,782	11,013
Departmental:			
Marketing Branch	13,750	14,463	(713)
Marketing Enforcement Branch	-	1,077	(1,077)
Total Expenditures	288,945	278,748	10,197
INCREASE (DECREASE) IN NET ASSETS	2,005	35,443	33,438
NET ASSETS, AT BEGINNING OF YEAR	142,118	142,118	-
NET ASSETS, AT END OF YEAR	144,123	177,561	33,438

The full financial statement for the period ending September 30, 2019 is available from the Board office upon request.

EPSTEIN

Fusarium Yellows in Celery: Breeding and Maintaining Resistance, and Integrated Control

Lynn Epstein, UC Davis, Department of Plant Pathology

We first observed a new *Fusarium oxysporum* f. sp. *apii* (*Foa*) in a single field in Camarillo in 2013, which we named race 4. As of Oct. 2019, we have documented *Foa* race 4 in all five celery production areas in Ventura County, and most recently in one field in Monterey Co. *Foa* race 4 isolates were highly virulent on all celery cultivars that we have tested, except for Rijk Zwaan's stick variety "Earthrace." We can diagnose *Foa* race 2 and race 4 *in planta* within two days with highly specific PCR primers, i.e., better than those that were previously published in Epstein et al. (2017). We also have developed semi-specific PCR primers and probes for *Foa* races 2 and 4 that are more sensitive and better suited for measuring the fungal growth of these strains in tissue and in some soils; this method will allow us to better evaluate the impact of cultural controls on inoculum levels. We have identified one celeriac accession with either resistance or tolerance to *Foa* race 4, and appear to have successfully produced celery cv. Challenger (*Foa* race 2-tolerant and *Foa* race 4-susceptible) x celeriac A0134 (*Foa* race 4-resistan or tolerant) F2 that have both *Foa* race 4-resistance/tolerance in greenhouse and field assays and progress towards development of a celery cultivar; we are preparing for both distribution of the F3s and backcrossing of the selected F2 with cv. Challenger. In order to facilitate future selection and breeding of resistant germplasm, we are participating in an effort led by UCD Professor Allen van Deynze to obtain a genetic map and full genome sequence of cv. Challenger. Because *Foa* race 4 is more severe at higher temperatures, we performed growth chamber experiments at soil temperatures of ca. 61, 64, 68, 72, 75, and 79 °F and monitored symptoms and fungal growth in crown tissue. We note that on celery extract agar, *Foa* race 2 has maximum growth at 76 to 78 °F, and *Foa* race 4 has maximal growth at 78 to 80 °F. In contrast, celery is considered to be a cool-weather crop with an optimum between 61 to 64 °F. In our trials, cv. Sonora and Challenger were grown in either uninfested soil, or soil infested with either *Foa* race 2 or *Foa* race 4. As expected, cv. Challenger tolerates (is relatively asymptomatic) to *Foa* race 2, regardless of temperature. However, *Foa* race 2 can infect and grow in Challenger crowns, and at the lower temperatures, to the same extent as it grows in Sonora. In contrast, symptom severity and growth of *Foa* race 4 in both cultivars increases with temperature, from few symptoms and growth at 64 °F to pronounced symptoms and growth at 72 °F and above. In keeping with the greater damage caused by *Foa* race 4 in the field than *Foa* race 2, in Sonora, which is susceptible to both races, there is more (2X to 11X) growth of *Foa* race 4 than *Foa* race 2 *in planta* in crown tissue at 72 °F and above.

CALIFORNIA CELERY RESEARCH ADVISORY BOARD
RESEARCH PROJECT REPORT

for the period of:
October 1, 2018 - September 30, 2019

PROJECT TITLE: Fusarium yellows in celery: breeding and maintaining resistance, and integrated control.

PROJECT LEADER: Lynn Epstein, Department of Plant Pathology, University of California, Davis, California 95616, 530-754-7916, lepstein@ucdavis.edu

COOPERATING PERSONNEL: Sukhwinder Kaur, Peter Henry, Radwan Barakat, Alejandro Palma-Carias, Frank Martin, Oleg Daugovish, and Richard Hurstak,

II. OBJECTIVES**:**
The purpose of the research in this proposal is to provide long-term control of Fusarium yellows in California celery, caused by *Fusarium oxysporum* f. sp. *apii* (*Foa*).

Specific objectives:
1. To identify and breed germplasm for resistance/tolerance to the new *Foa* race 4, and specifically the following: a) to continue backcrosses with one selection (A0134 celeriac) with resistance/tolerance to the new *Foa* race 4 with cv. Challenger, b) to continue field testing promising selections, and c) to continue to identify and test additional *Apium graveolens* germplasm that has not been tested for resistance or tolerance to *Foa* race 4;
2. To a) continue to monitor any spread of the new *Foa* race 4 in celery fields in California, b) to develop a more rapid assay for whether the *Foa* race 4 isolate is the dominant *Foa* race 4 or a variant;
3. To identify a) the genes involved in cv. Challenger's resistance/tolerant responses to *Foa* races 2 and 3, and susceptible response to *Foa* race 4 and b) the genes involved in *Foa* races 2, 3 and 4 in both causing disease in the case of race 4, or in inducing resistance/tolerance in cv. Challenger to *Foa* race 2;
4. To assist with the UCD celery genome project so that resources will be available for our and other breeding efforts in the future;
5. To determine the effect of temperature on disease severity in cv. Sonora and Challenger and growth of *F. oxysporum* f. sp. *apii* race 2 and race 4 *in vitro* and *in planta* on Sonora and Challenger.
6. To continue to minimally maintain the UCD celery germplasm collection.

RESULTS. Please note that our last year's report actually had some results from the reporting period through Jan. 7, 2018. Data tables and figures are limited to those that are not in a previous report. We also note that these Objectives (except for #5) were written in June 2018, and, in some cases, results in the four month period before the start of the grant impacted our research.

Objective 1. *To identify and breed germplasm for resistance/tolerance to the new Foa race 4, and specifically the following: a) to continue backcrosses with one selection (A0134 celeriac) with resistance/tolerance to the new Foa race 4 with cv. Challenger, b) to continue field testing promising selections, and c) to continue to identify and test additional Apium graveolens germplasm that has not been tested for resistance or tolerance to Foa race 4.*

To date, we have screened 241 accessions for resistance/tolerance to *Foa* race 4: 125 celery (*Apium graveolens* var.*dulce*); 70 celeriac (*A. graveolens* ssp. *rapaceum)*; 26 smallage (*A. graveolens* var. *secalinum* (either a wild-type *A*. graveolens, or cultivated plants that are used as leaf/"cutting" celery or used as celery seed as a spice*)*; 5 celery x celeriac; and 15 miscellaneous *Apium spp*. Promising accessions were rescreened in an eight-week trial in the greenhouse in infested soil three times. Promising accessions were then tested in now three field trials. We have now focused on one celeriac accession A0134. We crossed A0134 x Challenger, with Challenger as both male and female, screened F1 in the greenhouse against *Foa* race 4, selected resistant/tolerant plants, induced flowering, and produced F2. We screened the F2 families, and selected resistant/tolerant individuals for induction of flowering and backcrossing to Challenger. We also screened some of the the F2 families in a field trial in *Foa* race 4-infested soil (Table 1).

Table 1. Selection of resistant/tolerant breeding lines to *Foa* race 4: response of F2 families from celeriac A0134 x Challenger[a] in the greenhouse and in an Aug. 2019 UCD-Dole field trial in Oxnard, CA.

F2 family	Greenhouse avg. vascular discoloration rating in the crown, 0=asymptomatic to 5 dead[c]	Field, 7 & 13 wk evaluations[b]	
		Acceptable disease resistance/ tolerance?	Developing desirable celery characteristics or more celeriac-type
76-12	1.0	yes	celeriac
77-12	1.0	NT[e], predicted insufficient celery-type	NT
76-21[d]	1.6	yes	celery
77-48	1.8	No? (yellowish)	celery
77-40	1.9	No? (yellowish)	celery
76-18	2.0	yes	celeriac
76-16	2.2	No? (yellowish)	celery
76-8[d]	2.3	yes	celery
77-29	2.3	No? (yellowish)	celery
77-51	2.3	NT	NT
76-15	2.3	NT	NT
77-53	2.3	NT	NT
77-33	2.4	NT	NT
77-4	2.7	NT	NT
77-67	2.8	NT	NT
76-41	4.2	NT	NT
76-11	4.6	NT	NT
(A0321 Challenger parent)	4.6	NO!	(celery)

[a]Both parents were used as both the male and female in separate crosses.
[b]Except for the susceptible parent control, the appearance of the F2 families in the field were quite homogeneous.
[c]Scores were based on 20 plants. Note that these scores are from families; only apparently highly-resistant individuals are used in subsequent crosses.
[d]Selected as a family of interest based on field performance.
[e]NT, not tested, i.e., we only screened the F2 families that had 1) the lower disease scores and promising celery characteristics.

Surprisingly, on a field level, the individuals within a family were fairly homogeneous.

Interestingly, although we select individuals for further screening, there is some correlation between the average family score in the greenhouse and the resistance phenotype of the family in the infested field; both families with an average rating of $\leq$ 1.6 in the greenhouse had acceptable levels of resistance in the field, but of the six tested families with a greenhouse rating of 1.8 to 2.3, only two families were deemed to have sufficient resistance in the field. Consequently, of the families in shown in Table 1, we are focusing on individuals from families 76-21 and 76-8.

Fig. 1. An example of an F2 from the 76-21 family seven-weeks post-transplant in the field trial in *Foa* race 4-infested soil. The family has good resistance/tolerance to *Foa* race 4 and promising celery characteristics, as indicated in Table 1.

In addition to our individual F2s that have already been screened for resistance/tolerance to *Foa* race 4, from the field trial at 13 weeks, we selected individuals from the 76-21 and 76-8 families that were the most celery-like, i.e., with solid petioles at approx. 18 inches above ground; (celeriac petioles are hollow in the center.) From these individuals, as part of next year's project, we are 1) producing clones that we can screen for *Foa* race 4 resistance, and 2) are inducing flowering. If these individuals have excellent resistance, we will produce F3's, test them for resistance and score them for celery traits, and make the best F3 seed available to breeders who want to introgress the resistance into their germplasm.

We hope to screen the indicated lines in Table 2 in a subsequent field trial in *Foa* race 4-infested soil starting in Aug. 2020.

We did not have time to test new accessions, partly because Objective 5 on temperature effects became a larger objective than in the original proposal.

Table 2. Selection of additional resistant/tolerant breeding lines to *Foa* race 4: response of F2 families from celeriac A0134 x Challenger[a] in the greenhouse that have not yet been tested (and selected) in the field.

F2 Family	Greenhouse avg. vascular discoloration rating in the crown, 0=asymptomatic to 5 dead	Interest in field testing F2 family?
76-2	0.4	Yes!
76-47	0.6	No, insufficient celery type
76-6	0.6	Yes!
76-41	0.9	No, insufficient celery type
A0134 (celeriac parent)	1.1	(as a control)
76-17	1.2	No, insufficient celery type
76-55	1.3	Yes
77-50	1.4	Yes
76-64	1.6	No, insufficient celery type
76-22	1.7	No, insufficient celery type
76-66	1.7	No, insufficient celery type
76-27	1.8	Yes
76-57	1.9	No, insufficient celery type
76-32	2.0	No, insufficient celery type
76-34	2.0	Yes
76-44	2.3	No
76-56	2.6	No
76-52	3.6	No
A0321 (Challenger parent)	4.2	(as a control)

[a]Both parents were used as the male and female in separate crosses.

Objective 2. *To a) continue to monitor any spread of the new Foa race 4 in celery fields in California, b) to develop a more rapid assay for whether the Foa race 4 isolate is the dominant Foa race 4 or a variant.*

As of 28 Oct. 2019, we have detected *Foa* race 4 in 33 fields in Ventura Co.: Camarillo (20 fields), Oxnard (6 fields), Santa Paula (1 field), Saticoy (2 fields), Ventura (2 fields), and Fillmore (2 fields). Unfortunately, in 2019, we detected *Foa* race 4 in one field in King City in Monterey Co. Thus, *Foa* race 4 spread from a single field in Camarillo in 2013 to all celery-growing regions within Ventura Co. by 2017, and then to Monterey Co. (200 miles to the north) in 2019.

How is the pathogen being introduced into new fields? I believe that harvest equipment is the most common source of inoculum to previously uninfested fields, partly because if these vehicles are not washed after being in an infested field, they carry comparatively large volumes of celery and soil debris. In the first year an infection is observed, there is frequently a relatively small "hot spot" close to a field road. Certainly, once inoculum is in a field, it will be moved by horticultural operations. And there may be some movement of infested soil from one field to adjacent ones during high winds. But sanitation and exclusion of *Foa* race 4 are the most important controls! Every farm should have a cleaning protocol for equipment that will be moved from an infested to an uninfested field.

We note that we have better PCR primers that can be used to specifically identify either *Foa* race 4 or *Foa* race 2 from either infected celery or from cultures within a day than those that were reported in Epstein et al. (2017).

Based on the 628 bp from an intron-rich EF-1α and 640 bp from rDNA IGS (a very small sample of the total DNA), all but one field with *Foa* race 4 appear to be infested with the same

strain; indeed, when we returned to the field in which we isolated the variant, we identified the "regular" *Foa* race 4. The *Foa* race 4 DNA variant appears to be as virulent as the other isolate. In order to determine if the variant is just a minor variant or a different strain, we performed next generation sequencing (=2nd generation, Illumina Hi-Seq) sequencing of the variant's DNA. As far as we can tell, the race 4 DNA variant is a *bona fide* variant (we confirmed the single nucleotide polymorphism), but we have no compelling evidence that it is another strain. Except for special circumstances, we will no longer test for the DNA variant.

Objective 3. *To identify a) the genes involved in cv. Challenger's resistance/tolerant responses to Foa races 2 and 3, and susceptible response to Foa race 4 and b) the genes involved in Foa races 2, 3 and 4 in both causing disease in the case of race 4, or in inducing resistance/tolerance in cv. Challenger to Foa race 2.*

This is the first year of a multi-year objective that will require the completion of the celery genome project to finish part a of the objective. Using a method called 3' TagSeq (Lohman et al. 2016), we first quantified gene expression (mRNA) of *Foa* races 2, 3, and 4 in celery extract as a baseline control; typically, genes that are specifically involved in host-pathogen interactions are either not expressed or are expressed at comparatively low levels when the pathogen is growing saprophytically (in dead tissue). Using 3' TagSeq, we detected expression of 10,430 genes in *Foa* race 2, 10,250 genes in *Foa* race 3 and 10,556 genes in *Foa* race 4. The computer algorithm predicts that there are ca. 20,000 genes per strain, although this is likely an over-estimate due to inactive transposases ("selfish or junk DNA"). Regardless, for the next step, we initially tried to quantify *Foa* gene expression in Tall Utah celery plants early in the infection process, because all races are virulent on this cultivar. However, this is much more difficult than in celery extract because almost all of the mRNA from celery crowns is from celery, and not from the pathogen. Consequently, we focused on *Foa* race 4 in Tall Utah and were able to do sufficient sequencing so that we detected 35 genes that are expressed at significantly higher levels *in planta* than in celery extract, and that are likely to be specifically involved in causing disease (Table 4, which is shown on the last page of this report). It is unknown whether targeting any of these 35 genes will help with devising disease control measures. Of the 35 genes, 22 (63%) are putative "effectors," which are small secreted proteins that affect the host by either making it more susceptible or by interfering with the normal resistance response. Characteristically, effectors in *F. oxysporum* are on "accessory" chromosomes, and, in *Foa* race 4, 15 of the putative effectors are on accessory chromosomes. Two of the effectors are very similar to "Secreted in xylem 1" (SIX1), which has been reported in several other *F. oxysporum* interactions, and has been shown to be required for full virulence in *F. oxysporum* f. sp. *conglutinans* in cabbage (Li et al. 2016), f. sp. *cubense* tropical race 4 in banana (Widinugraheni et al. 2018) and in f. sp. *lycopersici* in tomato (van der Does 2008). We also detected a very similar protein to Pep1, which was previously speculated to be an effector in *F. oxysporum* f. sp. *vasinfectum* in cotton (Chakrabarti et al. 2010). Of the 13 other putative pathogenicity factors that we quantified that had increased and comparatively high levels of expression *in planta*, interestingly, two are putative transposons (NS.12180 and NS.11338), i.e., repetitive elements that can cause genetic changes.

In addition to identifying these 35 genes of interest in *Foa* race 4, we now know specifically how to conduct the 3' TagSeq experiment in cv. Challenger for part a of this objective in order to identify the gene(s) for resistance/tolerance to *Foa* race 2; this experiment is now in progress.

Objective 4. *To assist with the UCD celery genome project so that resources will be available for our and other breeding efforts in the future.*

a. We provided the celery genome project with axenic Challenger seedlings for second (short read) and third (long read) generation DNA sequencing.
b. We provided purified genomic DNA for short read sequencing of three accessions: the most likely celeriac parent of Challenger (PI 169001); and two celery cultivars that are presumed celery parents of Challenger -- Tall Utah 5270R Improved and Tendercrisp. Both Tall Utah and Tendercrisp are susceptible to *Foa* race 2.
c. We provided Challenger in fifteen stages of development for determination of mRNA sequence (to assist with both selection of algorithms for prediction of gene models and annotation of the genome):

1. 15-day old seedling roots (axenic)
2. 15 day-old seedling stems & 1st leaves (axenic)
3. Leaves-range of ages, from just unfurled to fully expanded but not senescent
4. Petioles - range of ages, from very young to 1 cm diameter
5. Crown –younger & older (not grown in infested soil, as are treatments 13-15)
6. Vegetative buds
7. Floral buds (not opened)
8. Flowers open but not fertilized
9. Flowers ca. 1 week after fertilization
10. Young green seeds, ca. 3-4 weeks after fertilization
11. Mature seeds
12. Seeds, just pre-germ tube emergence (5 days post-hydration)
13. Crowns from Challenger grown in soil w/ *Foa* race 2, 7 days post-transplant (new infections)
14. Crowns from Challenger grown in soil w/ *Foa* race 2, 14 days post-transplant (fungus at max of growth; plant starting to produce a limited quantity of phenolics)
15. Crowns from Challenger grown in soil w/ *Foa* race 2, 21 days post-transplant (fungus contained; apparently post-phenolic production)

d. We started preparation of 12 accessions for genotyping by sequencing (GBS). These accessions will be reported in next year's report.

Objective 5*. To determine the effect of temperature on disease severity in cv. Sonora and Challenger and growth of F. oxysporum f. sp. apii race 2 and race 4 in vitro and in planta on Sonora and Challenger.*

This objective is not explicitly stated in our original grant proposal because it was developed with the CCRAB board at their September 2018 meeting in Santa Maria.

We first note that *Foa* race 2 and *Foa* race 4 are from different genetic lineages. *Foa* race 4 has a slightly higher optimum temperature than *Foa* race 2; estimates of the 95% confidence intervals for peak hyphal extension on celery extract agar were 76 to 78 °F for *Foa* race 2 and at 78 to 80 °F for *Foa* race 4 (Fig. 2). In contrast, celery is considered to be a cool-weather crop with an optimum between 61 to 64 °F. Thus, temperatures that are optimum for *Foa* race 2 and even more so with *Foa* race 4 are temperatures of some heat-stress in celery.

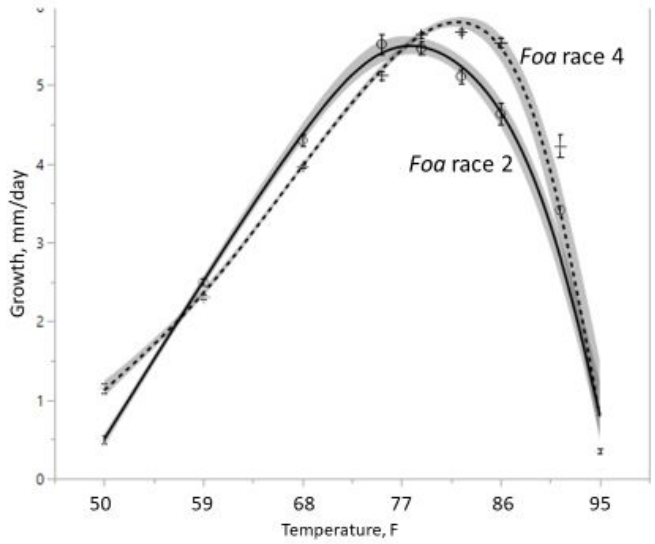

Fig. 2. Effect of temperature on the growth of *Foa* race 2 and race 4 in celery extract.

For this experiment, we concurrently operated growth chambers at either 60.8, 68, and 75.2 °F for two trials, or at 64.4, 71.6, and 78.8°F for two trials. In each growth chamber, there was a two-factor experiment, with two cultivars (Sonora and Challenger) and three levels of pathogen (uninfested, infested with *Foa* race 2 and infested with *Foa* race 4). There were five replicate plants of each treatment for each of the two trials. Two month-old seedlings were transplanted and after five weeks, plants were harvested and scored for disease severity. As expected cv. Challenger tolerates *Foa* race 2, regardless of temperature (Fig. 3). In contrast, in Challenger, the severity of *Foa* race 4 increases with temperature, from virtually no disease symptoms at 64 °F and increasing severity throughout the range tested to 79 °F. In cv. Sonora, which is susceptible to both *Foa* race 2 and *Foa* race 4, in general, severity increased with temperature for both *Foa* race 2 and *Foa* race 4.

For this objective, in order to detect the *Foa* at lower concentrations *in planta* at the lower tempearatures, we developed new multi-copy qPCR primer pairs and probe that amplify just 1) *Foa* race 2, and not either *Foa* race 4 or celery grown in the greenhouse and 2) *Foa* race 4 and not *Foa* race 2 or celery grown in the greenhouse. We note that these primers/probe work well in greenhouse/growth chamber assays, but may not be suitable for at least all fields because the *Foa* race 2 primers/probe amplify *F. oxysporum* f. sp. *congultinans* (the cabbage pathogen), and the *Foa* race 4 primers/probe amplify *Foa* race 3 and *F. oxysporum* f. sp. *coriandrii*, the cilantro pathogen. Nevertheless, in a preliminary trial with freshly-dried CCRAB Fusarium trial plot soil from Santa Maria in 2019 (which probably has an unusually high Foa race 2 inoculum load), we detected 619 *Foa* race 2 cell equivalents per g soil with the *Foa* multi-copy race 2 primers and 805 *Foa* race 2 cell equivalents per g soil with what we believe is a more specific but less sensitive single copy race 2 primer. Thus, it appears that the new primers are not detecting other strains in the CCRAB trial soil. Regardless, while these primers and probe may not be useful for at least some field analyses, with the proper controls, they will be acceptable and useful for quantification of *Foa* biomass in a range of greenhouse experiments. We note that the primers and probes are from transposable elements; there are 155 copies/cell of the *Foa* race 2 amplicon

and 60 copies/cell of the *Foa* race 4 amplicon. This allows us to accurately quantify as little as 0.9 fg and 2.2 fg of *Foa* race 2 and race 4 DNA, respectively (Fig. 4).

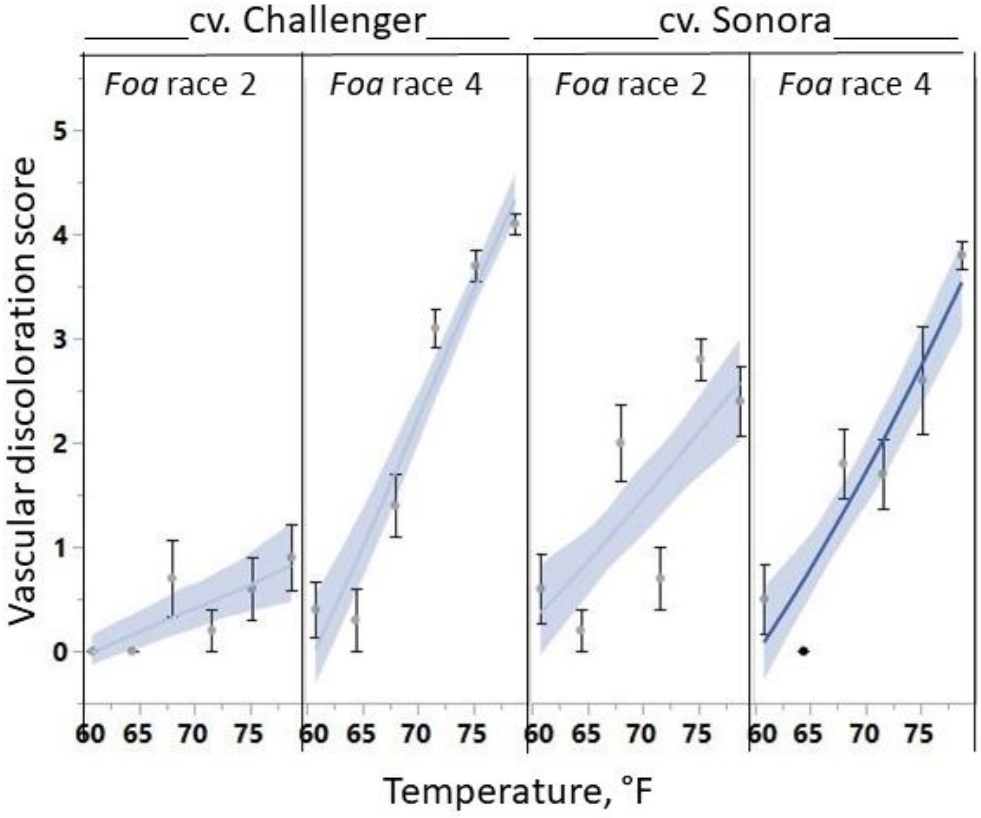

Fig. 3. Effect of growth chamber (and soil) temperature on disease severity based primarily on the extent of vascular discoloration, rated from 0 (asymptomatic) to 5 (dead). Error bars are 1 SE.

To examine fungal growth in tissue, we extracted and purified total DNA from 50 mg samples of freeze-dried celery crowns, and then assayed duplicate samples of 10 ng total DNA. To demonstrate that we had no PCR inhibitors that affected our quantification, we also assayed each sample at 3 ng total DNA. The results in Fig. 5 and Table 3 show the following. 1) Temperatures at 71.6 °F and above stimulate growth of *Foa* race 4 *in planta*; temperatures of 68 °F and below do not appear to stimulate growth of the pathogen. 2) In keeping with field observations, *Foa* race 4 grows *in planta* substantially more than *Foa* race 2. 3) Although the vascular discoloration score is actually more of a measure of the plant's response to infection rather than a measure of fungal growth, there is a very good correlation between the vascular discoloration score and *Foa* race 4 growth in the crown of Challenger ($r=0.83$, $P<0.0001$) and Sonora ($r=0.72$, $P<0.0001$). Overall, the correlation between the vascular discoloration score and *Foa* race 2 growth is significant, but not as strong as for *Foa* race 4. 4) Growth of *Foa* race 2 is stimulated in cv. Sonora at 75.2 °F and above, but not to the extent that *Foa* race 4 is stimulated. Growth of *Foa* race 2 in the tolerant cv. Challenger is relatively unaffected by temperature.

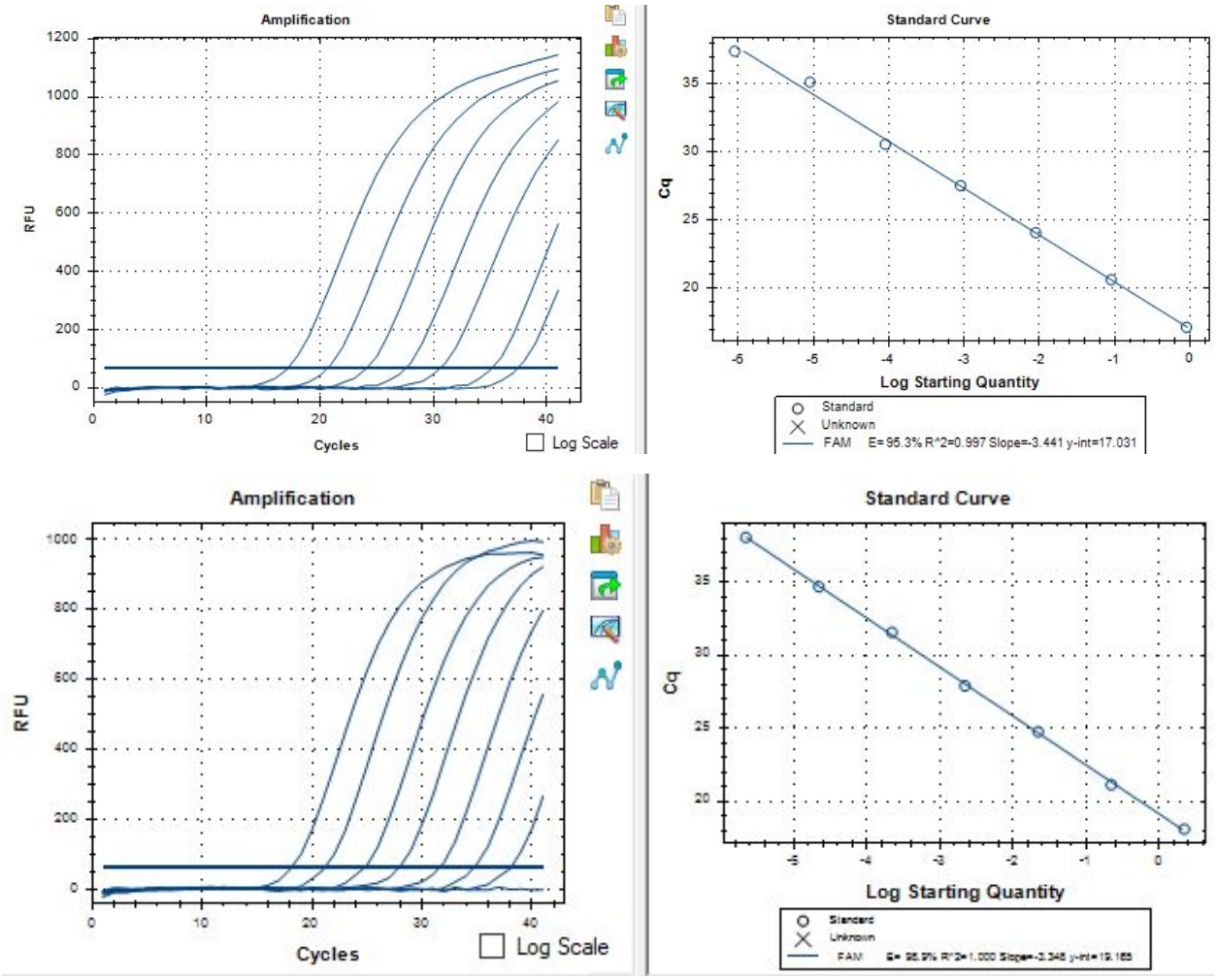

Fig. 4. Examples of quantitative PCR amplification of (upper) *Foa* race 2 and (lower) *Foa* race 4 standard DNA using fluorescently-labeled DNA probes for specific multi-copy sequences. The race 2 primers and probe do not amplify either *Foa* race 4 or celery, and the race 4 primers and probe do not amplify either *Foa* race 2 or celery (data not shown). The most dilute standards for *Foa* race 2 and race 4 have only 0.9 fg and 2.2 fg total DNA, respectively. i.e. our assays are quantitative and extremely sensitive!

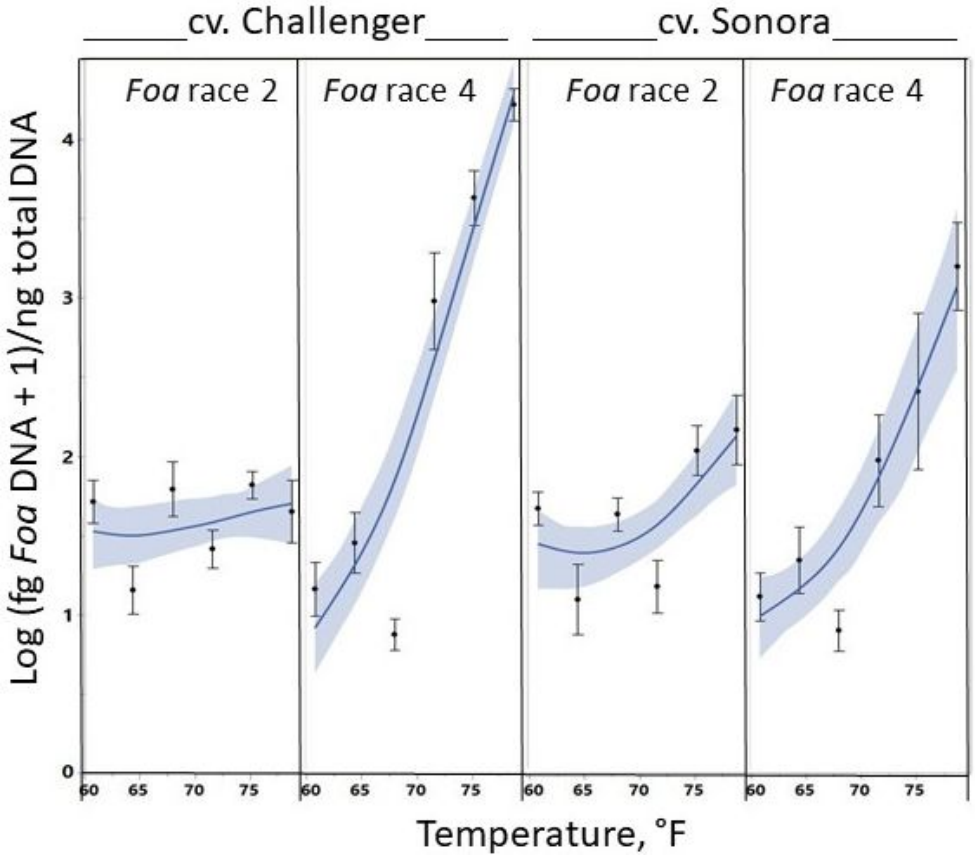

Fig. 5. The quantity of *Foa* race 2 and race 4 in Sonora and Challenger celery crowns after growth for 2 months in infested soil in growth chambers at varying temperatures. Fungal growth is estimated as the log of femtograms of DNA of the specific race per nanogram of total DNA (which is almost all celery DNA). At log 1, the fungal DNA is 0.001% of total DNA; at log 2, the fungal DNA is 0.01% of total DNA; at log 3, the fungal DNA is 0.1% of total DNA; and at log 4, the fungal DNA is 1% of total DNA. (Note: each plant cell has ca. 100X the amount of DNA that's in a single *Foa* cell). The single dead cv. Challenger in *Foa* race 4-infested soil at 79 F was excluded. Error bars are 1 SE.

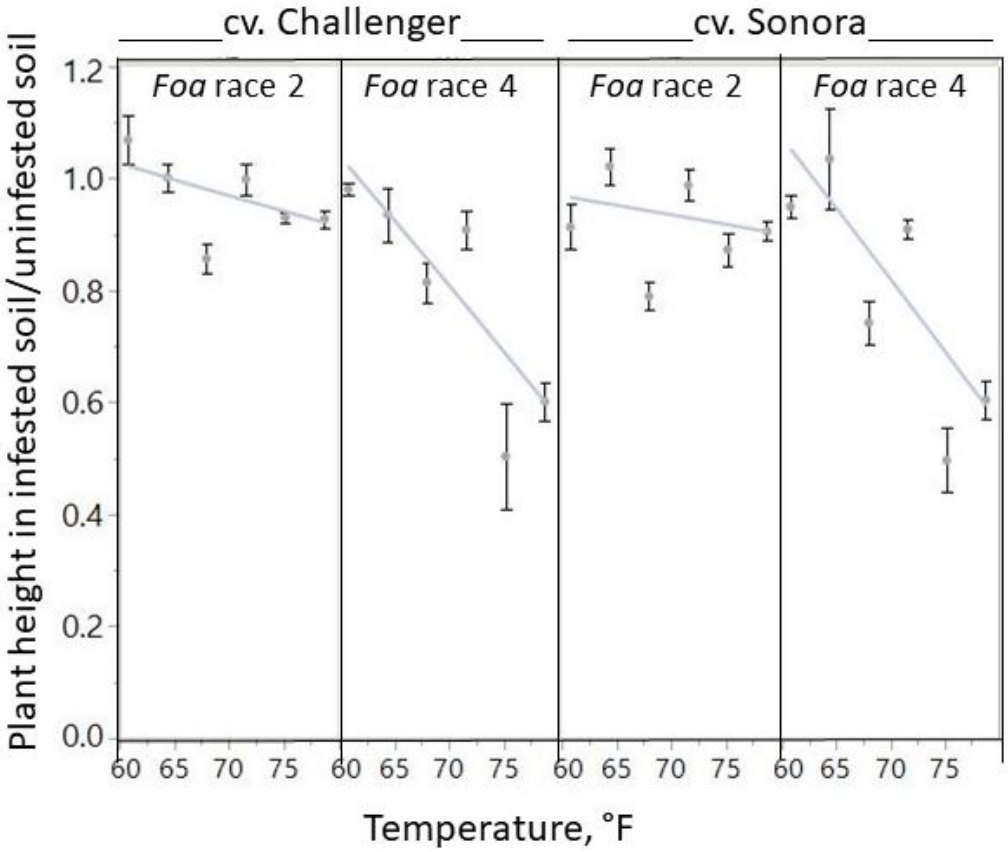

Fig. 6. Effect of growth chamber (and soil) temperature on plant height, shown as the ratio of the height of a plant grown in infested soil/the average height of that cultivar grown in uninfested soil in the same growth chamber. The single dead cv. Challenger in *Foa* race 4-infested soil at 79 F was excluded. Error bars are 1 SE.

Table 3. For each plant in the study that was grown in infested soil, the pairwise correlation of the vascular discoloration/disease severity score, the *Foa* race 2 or race 4 biomass in the crown by qPCR, and the ratio of plant height/average height of the uninoculated controls[a].

Group	Variable 1	Variable 2	Correlation, r[b]	*P*-value[c]
cv. Challenger in *Foa* race 4-infested soil	*Foa* biomass	Vascular discoloration score	0.83	<0.0001
cv. Challenger in *Foa* race 4-infested soil	*Foa* biomass	Plant height as a fraction of the average height of uninoculated controls	-0.66	<0.0001
cv. Challenger in *Foa* race 4-infested soil	Vascular discoloration score	Plant height as a fraction of the average height of uninoculated controls	-0.64	<0.0001
cv. Sonora in *Foa* race 4-infested soil	*Foa* biomass	Vascular discoloration score	0.72	<0.001
cv. Sonora in *Foa* race 4-infested soil	*Foa* biomass	Plant height as a fraction of the average height of uninoculated controls	-0.68	<0.0001
cv. Sonora in *Foa* race 4-infested soil	Vascular discoloration score	Plant height as a fraction of the average height of uninoculated controls	-0.72	<0.0001

cv. Challenger in *Foa* race 2-infested soil	*Foa* biomass	Vascular discoloration score	0.35	0.007
cv. Challenger in *Foa* race 2-infested soil	*Foa* biomass	Plant height as a fraction of the average height of uninoculated controls	(not significant)	0.12
cv. Challenger in *Foa* race 2-infested soil	Vascular discoloration score	Plant height as a fraction of the average height of uninoculated controls	-0.47	0.001
cv. Sonora in *Foa* race 2-infested soil	*Foa* biomass	Vascular discoloration score	0.58	<0.0001
cv. Sonora in *Foa* race 2-infested soil	*Foa* biomass	Plant height as a fraction of the average height of uninoculated controls	(not significant)	0.09
cv. Sonora in *Foa* race 2-infested soil	Vascular discoloration score	Plant height as a fraction of the average height of uninoculated controls	-0.52	0.0003

[a]Data from the uninfested controls were not included in the correlation analysis; they would have inappropriately improved the correlations. All uninfested controls had a vascular discoloration score of 0 and no detected *Foa* biomass by qPCR.
[b]The r correlation value can range from -1.00 (a perfect negative correlation between two scales, that are in opposite directions) to 0.00 with absolutely no correlation to 1.00 (a perfect positive correlation between two scales that are in the same direction).
[c]The smaller the *P*-value, the more likely that the variables are truly associated.

Objective 6. *To continue to minimally maintain the UCD celery germplasm collection.*
This past year, we rejuvenated and increased the available seed of three accessions in the collection: A0026, A0034 and A0224.

Details about methods are available by request.

Literature cited

Chakrabarti A, Rep M, Wang B, Ashton A, Dodds P, Ellis J. 2011. Variation in potential effector genes distinguishing Australian and non-Australian isolates of the cotton wilt pathogen *Fusarium oxysporum* f. sp. *vasinfectum.* Plant Pathol 60:232–243. https://doi.org/10.1111/j.1365-3059.2010 .02363.x.

Epstein L. Kaur S, Chang P, Carrasquilla-Garcia N, Lyu G, Douglas Cook D, Subbarao K, Kerry O'Donnell K. 2017. Races of the celery pathogen *Fusarium oxysporum* f. sp. *apii* are polyphyletic. *Phytopathology* 107:463-473.

Li E, Wang G, Xiao J, Ling J, Yang Y, Xie B. A *SIX1* homolog in *Fusarium oxysporum* f. sp. *conglutinans* is required for full virulence on cabbage. PLoS One. 2016; 11: 1–15. https://doi.org/10.1371/journal.pone.0152273 PMID: 27010418

Lohman BK, Weber JN, Bolnick DI. 2016. Evaluation of TagSeq, a reliable low-cost alternative for RNA seq. Molecular Ecology Resources16:1315-21.

van der Does HC, Duyvesteijn RGE, Goltstein PM, van Schie CCN, Manders EMM, Cornelissen BJC, et al. Expression of effector gene *SIX1* of *Fusarium oxysporum* requires living plant cells. Fungal Genet Biol. 2008; 45: 1257–1264.

Widinugraheni S, Niño-Sánchez J, van der Does HC, van Dam P, García-Bastidas FA, Subandiyah S, et al. (2018) A *SIX1* homolog in *Fusarium oxysporum* f.sp. *cubense* tropical race 4 contributes to virulence towards Cavendish banana. PLoS ONE 13(10): e0205896. https://doi.org/10.1371/journal.pone.0205896

Table 4. Thirty-five up-expressed potential effectors or pathogenicity/virulence factors in *Foa* race 4 *in planta*[a].

Gene	EdgeR analysis of 3'TagSeq				Predicted protein			Genome location		Postulated function[f]
	Avg. cpm[b] *in vivo*	Avg. cpm[d]b*i n vitro*	LogFC	Adjusted *P*-value	Cellular local-ization[c]	Protein mass, kDa	No. cyste-ine resi-dues	Accessory, fast core, or core[d]	Contig: start bp[e]	
NS.09678	25300	2284	2.46	2.5E-07	secr	12.8	3	Fast Core, OUT	SS4:3084010	Effector
NS.05815	18471	1.2	13.2	1.3E-08	secr	30.0	8	Acc	SS14:228927	SIX1 effector
PGN.06282	18379	0.1	15.4	4.5E-09	secr	5.6	0	Acc	SS17:11290	Effector
NS.06742	14139	5.3	10.0	2.2E-09	secr	29.0	8	Acc	SS19:499478	Effector
NS.14450	12854	598	3.20	1.7E-04	intra	3.6	5	Core chr10, W/IN	uni19:1108440	Unknown but not an effector
NS.06525[k]	11596	3.1	10.7	1.4E-09	secr	12.8	3	Acc	SS17:1662847	Effector
PGN.05952	9116	2.7	10.5	4.5E-09	secr	12.5	6	Acc	SS14:222108	Effector
NS.05812	7873	5.3	9.5	2.1E-10	intra	15.2	0	Acc	SS14:223943	Unknown, but not secreted
PGN.20363	7809	0.4	12.95	4.8E-08	secr	10.8	8	Core, W/IN	uni7:4951240	Effector
PGN.05922	7745	36	6.5	1.4E-09	secr	11.9	6	Acc	SS14:122611	Effector
PGN.15680	6730	57	5.65	1.5E-09	secr	12.4	8	Core chr10, W/IN	uni19:3727811	Effector
PGN.06635	6133	0.2	13.5	5.5E-10	secr	13.8	1	Acc	SS17:1461842	Effector
NS.06362	5038	0.1	13.2	2.0E-08	secr	9.9	5	Acc	SS17:1000217	Effector
PGN.09917	4632	480	2.23	1.3E-08	secr	12.8	7	Fast core, OUT	SS4:3084950	Effector
NS.05829	4407	0.3	12.1	6.4E-08	secr	30.5	6	Acc	SS14:278932	SIX1 effector
PGN.07042	4099	1.1	10.7	1.2E-09	secr	13.7	6	Acc	SS19:912650	Effector
NS.06821[i]	3935	0.1	13.1	7.1E-09	intra	31.3	0	Acc	SS19:758783	Abhydrolase_1
NS.10798	3622	158	3.33	5.5E-06	secr	45.2	12	Fast core, OUT	SS5:2865460	Metalloproteinase
NS.10798	3622	158	3.33	5.5E-06	secr	45.2	12	Fast core, OUT	SS5:2865460	Metalloproteinase
NS.17257	2979	2.3	9.17	2.5E-10	secr	51.2	3	Fast core, W/IN	uni26:184597	Amine oxidase or dehydrogenase
PGN.06691[j]	2938	0.0	13.0	4.6E-09	secr	12.5	7	Acc	SS17:1660905	Effector
NS.12180	2830	14.0	6.5	6.0E-08	TM?	28.0	3	Acc	uni111:57177	Transposable element
NS.01422	2533	121	3.01	8.6E-05	secr? TM?	25.2	7	Core, W/IN	SS10:4596446	Unknown
NS.09643	2463	0.3	11.07	5.4E-08	secr	29.3	8	Fast core, OUT	SS4:2943454	Effector; *F. oxysporum* f. sp. *vasinfectum* Pep1 homolog

NS.16793	2461	0.2	11.61	1.5E-07	secr	18.2	14	Fast core, W/IN	uni26:670796	Effector
NS.06528	2038	55.7	3.8	1.6E-07	secr?	15.1	9	Acc	SS17:1671928	Effector
NS.06820[i]	1874	0.1	12.3	4.0E-08	intra	33.9	3	Acc	SS19:758056	Alcohol dehydrogenase GroES-like domain & NAD(P) binding (Panther)
PGN.06376	1768	0.2	11.6	1.9E-08	secr	12.3	8	Acc	SS17:415228	Effector
NS.14958	1743	1.6	8.82	1.3E-07	TM	69.0	11	Core chr10, W/IN	uni19:2687638	Polyol transporter
NS.07235	1665	45.3	4.1	2.3E-07	intra	81.2	3	Acc	SS2:1462977	Catalase/peroxidase
NS.06359	1636	1.0	9.4	2.6E-09	secr	73.2	12	Acc	SS17:990399	Extracellular glucosidase
NS.17886	1589	4.5	6.1	5.7E-04	Intra/nuclear	106.7	9	Acc	uni56:301050	Unknown, but not an effector
NS.15045	1545	95	2.93	4.0E-08	secr	28.8	0	Core chr10, W/IN	uni19:2886040	Effector; related to early nodulin 75 precursor
NS.11338	1262	2.7	6.7	8.5E-04	intra	119.6	11	Acc	SS6:142150	Retrotransposon
PGN.05959	1060	0.8	9.4	8.2E-08	secr	9.8	4	Acc	SS14:239989	Effector
PGN.06650	1040	0.0	11.6	4.6E-09	secr	12.3	8	Acc	SS17:1511071	Effector

[a]Genes 1) had significantly (adjusted $P<0.05$) higher expression *in planta* in celery crowns than *in vitro* celery extract; 2) accounted for more than 0.1% of the total fungal reads *in planta,* i.e., were relatively highly expressed *in planta*; and 3) were not "house-keeping" genes.

[b]cpm, number of reads per million fungal reads.

[c]intra, intracellular, based on WoLF PSORT and the absence of a secretion signal and a transmembrane domain; secr, secreted based on SignalP-5.0; secr? based on the absence of a secretion signal by SignalP but an extracellular localization by WoLF PSORT; TM, transmembrane based on Geneious.

[d]Based on comparisons with progressiveMauve of *Foa* race 4 with the *Fol* 4287 reference. Contigs were assigned as follows: Acc, accessory contigs (no homology to a core chromosome); fast core [Likkens et al. 2018] on contigs that are homologs of *Fol* chromosomes 11, 12 and 13; on Core chr 10, on core chromosome 10; or on other core chromosomes. Genes on core chromosomes were further designated as W/IN, within the core region, i.e., within a progressiveMauve colinear block, or OUT, outside of the core region, i.e., between colinear blocks.

[e]SS, superscaffold; uni, unitig

[f] DNA and predicted amino acid sequences were BLASTed on GenBank and primarily analyzed with Panther, respectively. Genes that were identified as putative effectors were 1) secreted based SignalP-5.0, 2) relatively small proteins with a molecular mass <35 kDa, and 3) have no known biochemical function.

[g]NS.06820 and NS.06821 are adjacent to each other. [h]PGN.06691 and NS.06525 are adjacent to each other.

HURSTAK

Celery Cultivar and Germplasm Evaluations and Field Demonstrations for Observations of Fusarium Yellows Resistance and Crop Performance

Richard Hurstak, Crop Science Services, LLC, Private Consultant

Introduction: Fusarium Yellows, thought to be caused by *Fusarium oxysporum f. sp. apii* can induce stunting and yield reductions based on the level of tolerance or resistance within the individual celery varieties. The soil born disease enters the plant through the root system, damaging the vascular tissue of the host plant. Once damaged, the plant becomes more vulnerable to environmental stresses that often causes the commodity to fail in meeting market quality parameters. Currently, the only viable non-chemical means of controlling losses is by the use of resistant varieties.

We continued to evaluate several advanced lines, new entries, and current commercially available varieties in the 2019 testing program, with entries from Bejo Seeds Inc., Syngenta Seeds, Rijk Zwaan, Cal Grow, Tozer Seeds Ltd, and 3 Star Lettuce. Newer hybrid varieties along with open pollinated varieties were planted into a historically Fusarium infested site in the Santa Maria growing district for evaluation of disease resistance and general crop characteristics observations. A field day was scheduled at the trial location and direct observations were made by growers, celery plant breeders, and other allied celery production personnel.

Celery Research Board Annual Report 2019

Title: Celery Cultivar and Germplasm Evaluations and Field Demonstration for Observations of Fusarium Yellows Resistance and Crop Performance.

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Objective Evaluate current celery cultivars and newly developed germplasm for resistance to Fusarium Yellows thought to be caused by *Fusarium oxysporum f. sp. apii* "race 2" and evaluate crop characteristics important to California celery growers in the Santa Maria production region of California. Establish one research trial in a historically Fusarium infested field, schedule a field day at the location for direct observations by growers, celery plant breeders, and other allied celery production personnel.

Establishment Procedures: Seed of varieties in or near market release were requested from cooperating celery plant breeders and private seed companies. Varieties were seeded into transplant trays by Plantel Nursery and grown under commercial production practices until ready for field transplanting. The experimental design at the field location consisted of a randomized complete block with three replications of each variety. Test plots were planted by a commercial crew using a tractor mounted transplanter, in single rows with two plant rows per bed at 7-7.5-inch plant spacing, measuring 40" x 30'. The trial was placed within the grower's field and managed with the same inputs and growing systems as typical of commercial celery for the region.

Table 1. Study details:

2019 Study Schedule

Location: Betteravia Farms Ranch 12 Plot 04	
Activity	***Santa Maria***
Seeded	16-Apr
Transplant	18-Jun
Harvest Evaluation	22 thru24-Sep
Fusarium Pressure	High
Irrigation Type	Sprinkler/Drip
Transplant Operator	Plantel Nursery Santa Maria
Transplant Grower	Mike Leedom
Field Grower	Tom Minetti
Cooperator	Betteravia Farms

Table 2. List of celery varieties and breeders planted at the trial location.

Variety	Breeder	Variety	Breeder
Hudson	Tozer	Conquistador	Bejo
Rivary	Tozer	Challenger	Syngenta
Hadrian	Tozer	Command	Syngenta
TZ3537	Tozer	Sonora	Syngenta
TZ6010	Tozer	Mission	Syngenta
TZ4068	Tozer	Ag121767	Cal Grow
TZ4053	Tozer	Sequoia	Cal Grow
TZ5107	Tozer	Enterprise	Cal Grow
Bachata	Bejo	1001	Rijk Zwaan
Frevo	Bejo	1002	Rijk Zwaan
Balada	Bejo	1003	Rijk Zwaan
Samba	Bejo	1004	Rijk Zwaan
Merengo	Bejo	1005	Rijk Zwaan
A10331	Bejo	1006	Rijk Zwaan
A10322	Bejo	Tax112	3 Star Lettuce
A10324	Bejo	Tax116B	3 Star Lettuce
A10345	Bejo	Tax327	3 Star Letuce
Celex147	Syngenta		

Evaluation Procedures

Celery harvest was conducted as close to commercial harvest timing as scheduling demands allowed. At that time, 10 adjoining plants from the center of each plot were harvested for evaluation. Plants were dug from the soil with roots intact, soil covering the roots was removed and the roots were trans-sectioned multiple times to determine where Fusarium infection began to affect root color. Plants were then trimmed, and plant height, weight, color, ribbiness, and suckering were evaluated and recorded. Several varieties died due to fusarium infection prior to the harvest evaluation, these varieties were recorded as "dead" and received a 5 score for fusarium but no height, weight, ribbiness, suckering, or color scores.

Fusarium: The level of Fusarium infection was quantified using the common diagnostic discoloration seen in the vascular tissues of the roots and crown of infected plants. The following rating scale was used based on root discoloration; 0: none, 1: visible in fine roots, 2: visible in several roots, 3: visible in most roots slightly affecting crown, 4: clearly visible in crown, 5: crown rotting /dead.

Stalk Weight: Average weight for 10 plants in pounds, commercially trimmed to 14 inches.

Ribbiness: This is a fairly arbitrary rating estimation of the celery petiole texture or smoothness rated from 1 to 3 at harvest. Plants given a rating of 1 are very smooth relative to the current varieties, 2: noticeable ribs, 3: prominent ribs. Fusarium infected plants tend to have a high degree of ribbiness, due to stunted growth.

Color: This scale is based on the relative shade of green coloring exhibited by the celery variety with a rating scale as follows; 1: light green, 2: medium green, 3: dark green.

Suckering: This scale rates the number of auxiliary shoots that form at the base of the stalk or within the auxiliary buds between each petiole. If suckers occur within the petiole bunch then outer petioles may have to be stripped off, ultimately reducing commercial yields. Plant were rated as follows; 0: No suckers, 1: Very few, 2: Moderate, 3: Many.

Height: Plant height in inches from the plant butt to approximately 1 inch above the average length to the first petiole node.

Results

Data analysis was conducted using Gyllings Agricultural Research Management version 9. Mean comparisons were made using Duncan's New MRT at a 10 percent significance level. It must be kept in mind that the trial has a mix of hybrid and open-pollinated varieties that mature at different rates. While data collection occurs on the same dates for all varieties, it generally is not optimum for either of the variety types.

Santa Maria summer trial site: Fusarium Yellows pressure at the trial site was once again very high. This is the 2nd year in a row that there was a higher degree of variability in Fusarium scores within the same varieties by replicate than has been generally observed in past trials. A10345, TZ4053, Challenger, Merengo, and TZ4068 statistically had the lowest Fusarium scores and thus highest degree of resistance. Statistically the tallest variety was Tax327. Numerically, the highest yielding variety was Merengo

Acknowledgments

I'd like to thank the California Celery Research Advisory Board for the financial funding to continue this long-term study and the following individuals and their companies for their assistance during the course of the trial: Mike Leedom and Plantel in Santa Maria; Tom Minetti and Betteravia Farms in Santa Maria, and all the participating seed companies. Without their cooperation and donation of time and space this study would not have been possible.

Figure 1. Santa Maria trial location field setup.

Figure 2. Discoloration in roots of susceptible celery varieties.

Figure 3. Varietal growth differences observed mid-season.

Figure 4. Field evaluations of commercially important growth characteristics.

Table 3. Santa Maria 2019 trial summary of all data:

Variety	Fusarium Score (0-5)		Height (Inches)		Weight (lb/plant)		Ribbiness (1-3)		Suckers (0-3)		Color (1-3)	
A10345	1.1	k	11.2	c-g	1.6	bc	1.3	ef	1.0	g	2.3	cde
TZ4053	1.2	k	11.7	cd	1.6	abc	1.5	def	1.3	efg	2.2	def
Challenger	1.3	jk	11.5	cde	1.4	bcd	1.3	ef	1.2	fg	3.0	a
Merengo	1.4	jk	11.0	c-h	1.8	a	1.3	ef	1.3	efg	2.5	bcd
TZ4068	1.5	ijk	11.3	c-g	1.5	bc	1.7	cde	1.0	g	2.3	cde
TZ6010	1.8	hij	13.3	b	1.7	ab	1.3	ef	1.8	c-f	2.2	def
AG121767	1.9	ghi	11.4	c-f	1.2	de	2.2	b	1.7	c-g	1.8	f
Balada	2.0	f-i	10.9	c-i	1.4	cd	1.7	cde	1.7	c-g	1.8	f
1001	2.2	e-h	11.8	c	1.3	de	1.8	bcd	2.2	bcd	2.8	ab
Samba	2.3	e-h	10.3	g-m	1.4	cd	1.7	cde	1.8	c-f	1.8	f
Hudson	2.4	efg	11.3	c-g	1.5	bcd	1.7	cde	1.3	efg	2.5	bcd
A10322	2.4	efg	10.7	d-j	1.7	ab	1.3	ef	1.5	d-g	2.5	bcd
TZ3537	2.5	efg	11.9	c	1.6	bc	2.0	bc	1.7	c-g	2.0	ef
TZ5107	2.6	ef	11.2	c-g	1.6	abc	1.8	bcd	2.0	b-e	1.8	f
Tax327	2.7	de	15.5	a	1.6	bc	2.0	bc	1.5	d-g	2.5	bcd
Tax116B	3.1	cd	10.3	g-m	0.8	ghi	2.0	bc	1.2	fg	2.3	cde
Enterprise	3.1	cd	9.8	i-m	1.0	fg	3.0	a	2.7	ab	3.0	a
Command	3.2	cd	9.5	k-n	0.8	gh	2.7	a	2.3	abc	2.0	ef
Celex147	3.5	bc	9.3	mn	1.2	de	2.2	b	2.0	b-e	2.7	abc
Frevo	3.7	b	10.6	e-k	1.2	de	1.2	f	2.2	bcd	2.0	ef
A10324	3.8	b	9.7	j-n	0.9	fg	2.0	bc	2.2	bcd	2.3	cde
Hadrian	3.9	b	10.4	f-l	1.1	ef	2.0	bc	2.2	bcd	2.3	cde
Rivary	3.9	b	9.9	h-m	1.1	ef	1.7	cde	1.3	efg	2.5	bcd
A10331	3.9	b	9.4	lmn	1.2	de	1.3	ef	1.8	c-f	2.7	abc
Bachata	4.0	b	8.7	n	0.7	ghi	2.8	a	2.0	b-e	2.0	ef
Sequoia	4.5	a	11.8	c	0.9	fg	1.8	bcd	2.7	ab	2.3	cde
Mission	4.5	a	5.9	o	0.5	ij	3.0	a	2.0	b-e	3.0	a
Sonora	4.6	a	5.5	op	0.5	ij	3.0	a	2.7	ab	3.0	a
Conquistador	4.7	a	5.4	op	0.6	hi	3.0	a	3.0	a	3.0	a
1004	5.0	a	4.7	p	0.3	j	3.0	a	2.0	b-e	3.0	a
1002	5.0	a	dead	q	dead	k	dead		dead		dead	
1003	5.0	a	dead	q	dead	k	dead		dead		dead	
1005	5.0	a	dead	q	dead	k	dead		dead		dead	
1006	5.0	a	dead	q	dead	k	dead		dead		dead	
Tax112	5.0	a	dead	q	dead	k	dead		dead		dead	

'Means followed by same letter or symbol do not significantly differ (P=.10, Duncan's New MRT).

'Mean comparisons performed only when AOV Treatment P(F) is significant at mean comparison OSL.

TRUMBLE

Management and Economics of Leafminer and Beet Armyworm Control on Celery

John Trumble, University of California, Riverside

Immediate Objectives:

I. Studies to minimize the negative effects of FQPA: Identifying control 'gaps' and finding solutions. Immediate goals are to screen new insecticides for insect control and resistance management programs on celery.

II. Continue economic analyses of IPM approaches developed through the support of the CCRAB using new insecticide chemistries. A comparison between the IPM and a "standardized" chemical approach has been evaluated.

Management and Economics of Leafminer and Beet Armyworm Control on Celery
Annual Report for 2018-2019

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Objective I. Methods:

"Challenger" celery was transplanted on 24 Oct 2018 at Agricultural Operations, University of California, Riverside, California. The plots were drip irrigated, with replicates three beds wide (two rows per bed on 60" centers) by 40 feet. Four replicates of each treatment were organized in a randomized complete block design. The low input treatment had four applications using Exirel 100 SE, Beleaf 50 SG, Radiant 1.0 SC, Sequoia 1.0 SC, and Dipel DF. VST-006340 plus Bioprotec DF was applied six times as a foliar spray with LI-700 used as a surfactant. Bioprotec DF was also applied as a separate treatment using LI-700 as a surfactant. Sequoia 1.0 SC was applied six times as an individual treatment. Radiant 1.0 SC plus Sequoia 1.0 SC rotated with Vydate L was a treatment. The chemical standard treatment consisted of Asana XL, which was applied six times as a foliar spray. Treatments and spray dates are listed in Table 1.

All chemicals were applied by tractor using a tractor-mounted boom sprayer operated at 100 PSI and 100 gal/Ac except the VST treatments, which were applied at 50 gal/Ac. Four, disc-type cone nozzles per bed incorporated D3 orifice disks, #25 cores, and 50 mesh screens.

Field Count Procedures:

The field was surveyed on 23 January for Lepidopteran pests (primarily *S. exigua and T. ni*), leafminer (*L. trifolii*), aphids (*Brevicoryne brassicae*), Lygus (*Lygus Hesperus*) and greenhouse whitefly (*T. vaporariorum)*. Five plants from each plot were counted for a total of twenty plants per treatment.

Harvest Evaluation Procedures:

Lepidoptera: Evaluation was based on the number of damaged plants found in 25 plants per replicate (4 replicates/treatment) from the center rows of each replicate at harvest 5 March. Beet armyworm, *Spodoptera exigua* (Hübner), pressure was high and made up most of the Lepidoptera population; black cutworm, *Agrotis ipsilon*, populations were low

this year.

Aphids/Lygus: The plants that were inspected for Lepidoptera damage were also inspected for aphids and Lygus damage. Cabbage aphid (*Brevicoryne brassicae*) and *Lygus Hesperus* were present in the field and there were significant differences between the treatments.

Treatments

Table 1. IPM Materials Under Trial:

Compound [b]	Formulation	Rate/Ac	MoA [a]	Supplier
1- Control-Untreated	--	--	--	--
2- Low Input				
a-Exirel	100 SE	13.5 oz	28	Dupont
+ Beleaf	50 SG	2.8 oz	9C	FMC
b-Radiant	1.0 SC	8.0 oz	5	Dow
+ Beleaf	50 SG	2.8 oz	9C	FMC
c-Exirel	100 SE	13.5 oz	28	Dupont
+ Sequoia	1.0 SC	5.0 oz	4	Dow
d-Dipel	DF	1 lb	11	Valent
(a=12/19,b=1/10, c=1/30,d=2/13)				
3- VST-006340+	--	16 fl oz	--	Vestaron
Bioprotec CAF	Btk	16 fl oz	11	AEF Global
LI-700	--	0.125%		
(12/19, 1/4, 1/10, 1/24, 1/30, 2/13)				
4- VST-006340+	--	32 fl oz	--	Vestaron
Bioprotec CAF	Btk	16 fl oz	11	AEF Global
LI-700	--	0.125%		
(12/19, 1/4, 1/10, 1/24, 1/30, 2/13)				
5- Bioprotec CAF	Btk	24.0 oz	11	AEF Global
LI-700		0.125%		
(12/19, 1/4, 1/10, 1/24, 1/30, 2/13)				
6- Sequoia	1.0 SC	4.5 oz	--	Dow
(12/19, 1/4, 1/10, 1/24, 1/30, 2/13)		0.125%		
7- a,c,e-Radiant +	1.0 SC	7.0 oz	5	Dow
Sequoia	1.0 SC	4.5 oz	4	Dow
b,d,f-Vydate	L	2 pt	1A	Dupont
(a=12/19, b=1/4, c=1/10, d=1/24, e=1/30, f=2/13)				
8- Asana	XL	9.0 oz	1	Dupont
(12/19, 1/4, 1/10, 1/24, 1/30, 2/13)				

[a] MoA – Mode of Action for insecticide resistance management
[b] Spray dates are included.

IRAC

Using the guidelines established by the Insecticide Resistance Action Committee (IRAC) we are classifying the compounds used in the field trial based on their mode of action (MoA) (Table 1). The purpose is to assist growers to develop insecticide resistance management (IRM) strategies, which will potentially improve the effectiveness of IPM programs, slow resistance development, and extend the life of the pesticides. Additional information and MoA classifications can be found in the IRAC handout or online at www.irac-online.org.

> "Resistance to insecticides may be defined as '*a heritable change in the sensitivity of a pest population that is reflected in the repeated failure of a product to achieve the expected level of control when used according to the label recommendation for that pest species*' (IRAC). This definition differs slightly from others in the literature, but IRAC believes it represents the most accurate, practical definition of relevance to farmers and growers. Resistance arises through the over-use or mis-use of an insecticide or acaricide against a pest species and results in the selection of resistant forms of the pest and the consequent evolution of populations that are resistant to that insecticide or acaricide.
>
> In the majority of cases, not only does resistance render the selecting compound ineffective but it often also confers cross-resistance to other chemically related compounds. This is because compounds within a specific chemical group usually share a common target site within the pest, and thus share a common mode of action (MoA). It is common for resistance to develop that is based on a genetic modification of this target site. When this happens, the interaction of the selecting compound with its target site is impaired and the compound loses its pesticidal efficacy. Because all compounds within the chemical sub-group share a common MoA, there is a high risk that the resistance that has developed will automatically confer cross-resistance to all the compounds in the same sub-group. It is this concept of cross-resistance within chemically related insecticides or acaricides that is the basis of the IRAC mode of action classification".

Celery IPM Trial Results

Harvest assessment results showed the efficacy of chemicals against lepidopterous pests and has been presented in Table 2. Populations of beet armyworm were high in the study; the control suffered 34% damage. This allowed statistical separation of the various treatments. Cutworm (*Agrotis ipsilon*) pressure was not measurable due to very low pressure this season. Leafminers (*L. trifolii*) have not been included because of low pest pressure and a very rainy growing season. Whitefly pressure was also very low. Adults were seen in the field but nymphal populations were very low and no separation was possible. Cabbage aphid (*Brevicoryne brassicae*) was present in the field this year along with *Lygus Hesperus* and there were differences between treatments. No phytotoxicity was observed in any of the treatments.

Field count results are shown in Figure 1. The Lygus damage on the plants was significant and there were aphids present throughout the field. The lepidopteran field

counts were not significant, but the harvest assessment numbers showed a high level of worm damage and significant differences between treatments.

Figure 1. Field counts level of infestation

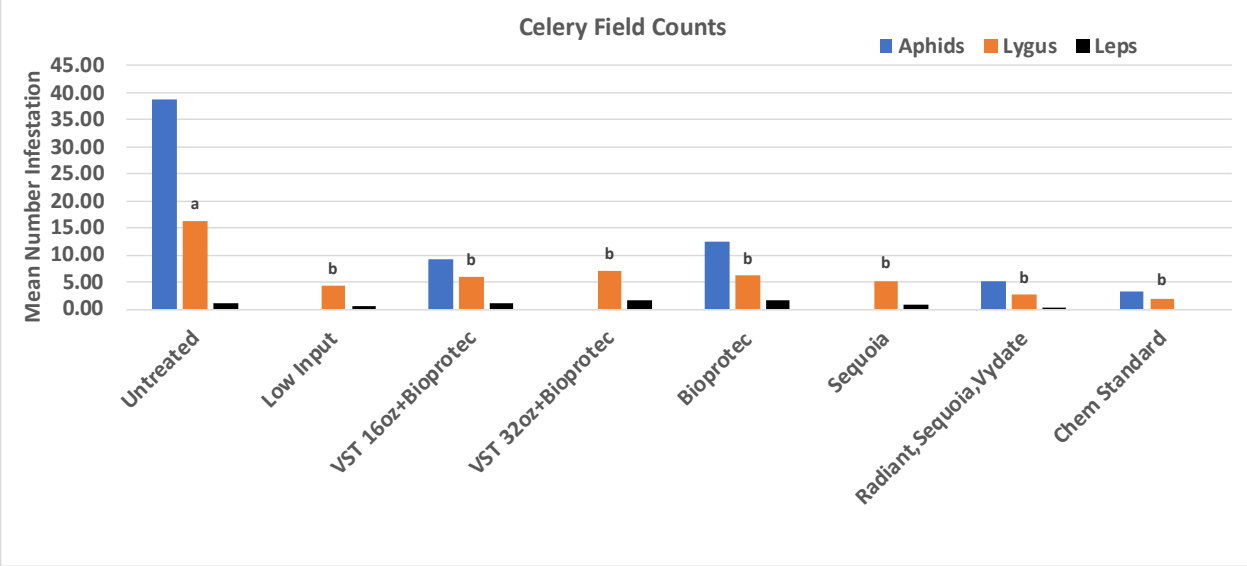


TABLE 2. MEAN NUMBER OF PLANTS DAMAGED BY LEPIDOPTERA

		Mean Number of Plants Damaged by [a]		
Treatment	**Rate**	**Cabbage Aphid**	**Beet Armyworm**	**Lygus**
1- Control-Untreated	--	7.00 a	8.75 a	14.25 a
2- Low Input a-Exirel + Beleaf b-Radiant + Beleaf c-Exirel + Sequoia d-Dipel (a=12/19,b=1/10, c=1/30,d=2/13)	 13.5 oz 2.8 oz 8.0 oz 2.8 oz 13.5 oz 5.0 oz 1 lb	0.75 c	0.50 c	1.75 b
3- VST-006340+ Bioprotec CAF LI-700 (12/19, 1/4, 1/10, 1/24, 1/30, 2/13)	16 fl oz 16 fl oz 0.125%	4.25 b	0.25 c	3.50 b
4- VST-006340+ Bioprotec CAF LI-700 (12/19, 1/4, 1/10, 1/24, 1/30, 2/13)	32 fl oz 16 fl oz 0.125%	3.50 b	1.50 c	3.50 b
5- Bioprotec CAF LI-700 (12/19, 1/4, 1/10, 1/24, 1/30, 2/13)	24.0 oz 0.125%	4.00 b	3.75 b	3.00 b
6- Sequoia (12/19, 1/4, 1/10, 1/24, 1/30, 2/13)	4.5 oz 0.125%	0.00 c	1.75 c	0.50 b
7- a,c,e-Radiant + Sequoia b,d,f-Vydate (a=12/19, b=1/4, c=1/10, d=1/24, e=1/30, f=2/13)	7.0 oz 4.5 oz 2 pt	0.00 c	0.50 c	0.50 b
8- Asana (12/19, 1/4, 1/10, 1/24, 1/30, 2/13)	9.0 oz	0.25 c	0.00 c	0.25 b
ANOVA F_{value}		10.564	19.714	9.895
ANOVA P value		0.001	0.001	0.001

[a] 25 plants examined per replicate, four replicates per treatment; means in columns followed by the same letter are not significantly different ($P < 0.05$ level, Fisher's Protected LSD).

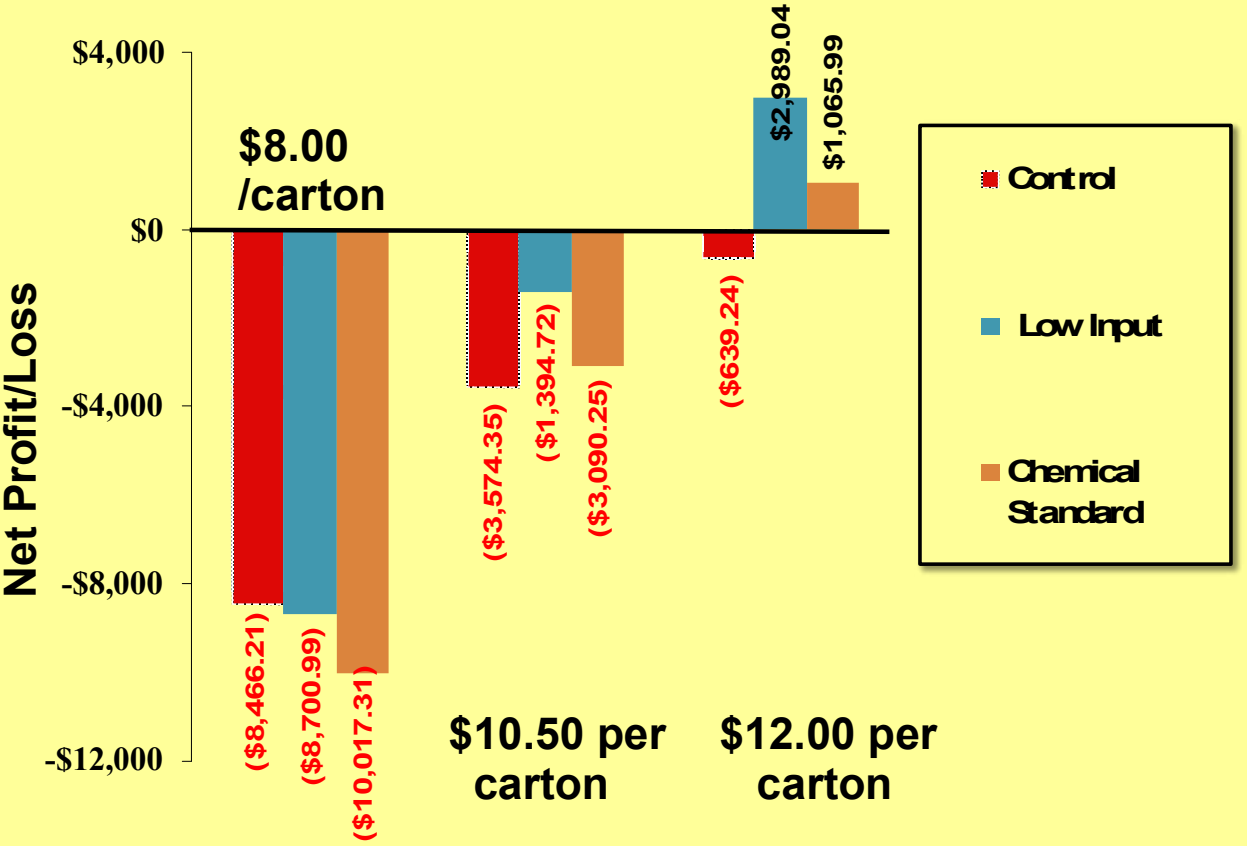
An economic analysis was prepared to compare harvest yields between treatments. Damage to the celery, costs of applications, and material costs were factored into the calculations. Figure 2 shows that the “Low Input” treatment performed the best at $12.00 per carton. The breakeven point for most growers is $10.50 per carton. For this season, we did not factor in the number of hearts for the economic analysis since the low input program had zero damage, and no produced hearts. Because hearts carry a high dollar value in the calculations, the results would have been skewed.

Objective II. Economic Analyses of an IPM and Chemical Standard Trial.
For this study, we tested new combinations of products designed to 1) reduce costs, 2) manage insecticide resistance, 3) increase harvest/yield, and 4) provide critical information to regulatory agencies on how products fit within an IPM program. The program consisted of a low input rotation of 1) Exirel, Beleaf, Radiant, Sequoia and Dipel versus 2) Asana. In the IPM plots chemicals were applied 4 times over the course of an 18 week season. In the ‘chemical standard’ treatment the materials were applied 6 times.

The net profit/loss information is presented in Figure 2. The data do not include all aspects of harvest. There were differences between yield and net profits when using the IPM program versus the chemical standard treatment at market values of $10.50 and $12.00 per carton. The “Low Input” program provided the highest net profits.

We used very progressive chemical treatments with fewer applications for the low input program. In spite of using chemicals that target specific insects, which in most cases have higher costs per application, we were able to increase net profits for the Low Input IPM program. Other benefits such as environmental effects and worker health and safety aspects are improved in the IPM programs, and are necessary to conform with the FQPA. As reported to the Cal EPA, without some pesticidal control, celery damage by insects will be extensive, and losses would not allow a reasonable profit to be made. Thus, some pesticides must continue to be registered for celery for the crop to remain viable in California.

Figure 2. Net profit or loss in IPM, chemical standard, and control treatments in 2018. Numbers associated with each bar are net profits (if numbers are in parentheses, they represent net losses).



Additional Research

Celery Residue Concerns

I continued to address the concerns of celery being classified in a consumer advocates publication as part of the dirty dozen regarding pesticide residues. The amount of pesticides used in crop protection in California has declined throughout the period of 1997-2009. The goal of current pest management programs is to reduce the amounts of pesticide applied which minimizes environmental, occupational, and consumer exposures.

Lygus hesperus

The field trials this season had significant levels of pressure from Lygus bugs. One of the materials of interest is VST-006340, which is now known as Spear T, has shown some efficacy in the lab with the new liquid formulation as a spray treatment contact insecticide. Capsil is a surfactant, which may increase the efficacy of the material when used on Lygus bugs, and we are looking forward to testing it in the lab and in the field this year. This same material showed good results on the two spotted spider mite at the 5 ppt rate from tests completed this year. We will also be testing Radiant, Sequoia, and Vydate in combinations to control lygus and other pest complexes. Sequoia alone showed good results for Lygus control and this treatment will be repeated the following season to confirm our findings.

Future Research

In our 2019 celery field trials we will be utilizing promising compounds that may be included in an IPM program. We will be testing IPM programs against a chemical standard program to continue to identify the most effective and economical pest control strategies. We would like to test Sequoia for aphid control again and it has shown promising results on sucking insects. We will also try to determine if VST-006340 and Sequoia has continued efficacy against Lygus bugs in the upcoming celery season. VST-006340 is now registered in California for use on celery as Spear-Lep™.