

AGENDA

California Avocado Commission Production Research Committee Meeting

Meeting Information

Date: Wednesday, December 8, 2021

Time: 9:00 a.m.

Location: Web/Teleconference

Web Conference URL:

https://californiaavocado.zoom.us/j/5375836823?pwd=aURBZ3BEL29tclBRS1ZRY3QrMkhZQT09

Conference Call Number: 669-900-6833

Meeting ID: 537 583 6823

Passcode: 348652

Meeting materials will be posted online at least 24 hours prior to the meeting at:

https://www.californiaavocadogrowers.com/commission/meeting-agendas-minutes

Committee Member Attendance

As of Wednesday, December 1, 2021, the following individuals have advised the Commission they will participate in this meeting via web/teleconference:

- Leo McGuire, PRC Chairman
- John Burr
- Jim Davis
- Dan Grant
- Darren Haver
- Catherine Keeling
- Ryan Larkan
- Tom Roberts
- Ryan Rochefort
- Rob Grether, CAC Chairman/PRC Ex-Officio

Time	Item	
9:00 a.m.	1.	Call to Order a. Roll Call/Quorum
9:05 a.m.	2.	Opportunity for Public Comment Any person may address the Committee at this time on any subject within the jurisdiction of the California Avocado Commission.
9:10 a.m.	3.	Approval of Minutes a. Consider approval of Production Research Committee Meeting Minutes of August 4, 2021
9:15 a.m.	4.	Research Program Directors Report
		a. Departure of Monique Rivera from UC Riverside
		b. Grower economic survey results
9:40 a.m.	5.	Discussion Items
		a. Presentation from Dr. Ali Montazar, Irrigation and Water Management Farm Advisor, University of California Cooperative Extension, USDA Grant Funding for "Improving Avocado Resource-Use Efficiency through Updated Crop Water Use Information and Irrigation Management Strategies"
10:00 a.m.	6.	Action Items
		a. Consider approval of the research proposal "Understanding the Effects of Soil Microbial Community Enhancement on Avocado Stress Tolerance"
		b. Consider approval of the research proposal "Development of Chloride Mitigation Strategies for Californian Avocado Growers: Technology Review and Treatment Prediction"
		c. Consider approval of the research proposal on mitigation of cadmium in

11:00 a.m. 7. Adjourn Meeting

avocado groves

Disclosures

The times listed for each agenda item are estimated and subject to change. It is possible that some of the agenda items may not be able to be discussed prior to adjournment. Consequently, those items will be rescheduled to appear on a subsequent agenda. All meetings of the California Avocado Commission are open to the public and subject to the Bagley-Keene Open Meeting Act.

All agenda items are subject to discussion and possible action. For more information, or to make a request regarding a disability-related modification or accommodation for the meeting, please contact Emailed from CAC 12-1-2021

Page 2

April Aymami at 949-341-1955, California Avocado Commission, 12 Mauchly, Suite L, Irvine, CA 92618, or via email at aaymami@avocado.org. Requests for disability-related modification or accommodation for the meeting should be made at least 48 hours prior to the meeting time. For individuals with sensory disabilities, this document is available in Braille, large print, audiocassette or computer disk. This meeting schedule notice and agenda is available on the internet at https://www.californiaavocadogrowers.com/commission/meeting-agendas-minutes and https://it.cdfa.ca.gov/igov/postings/detail.aspx?type=Notices.

If you have questions on the above agenda, please contact Tim Spann at <u>tim@spannag.org</u> or 423-609-3451.

Summary Definition of Conflict of Interest

It is each member's and alternate's responsibility to determine whether they have a conflict of interest and whether they should excuse themselves from a particular discussion or vote during a meeting. To assist you in this evaluation, the following *Summary Definition of Conflict of Interest* may be helpful.

A Commission *member or employee* has a conflict of interest in a decision of the Commission if it is reasonably foreseeable that the decision will have a material effect, financial or otherwise, on the member or employee or a member of his or her immediate family that is distinguishable from its effect on all persons subject to the Commission's jurisdiction.

No Commission member or employee shall make, or participate in making, any decision in which he or she knows or should know he or she has a conflict of interest.

No Commission member or employee shall, in any way, use his or her position to influence any decision in which he or she knows or should know he or she has a conflict of interest.

PRODUCTION RESEARCH COMMITTEE MEETING MINUTES

August 4, 2021

A web/teleconference meeting of the Production Research Committee (PRC) of the California Avocado Commission (CAC) was held on Wednesday August 4, 2021 with the following people participating:

MEMBERS PARTICIPATING CAC STAFF PARTICIPATING:

VIA TELECONFERENCE:Tom BellamoreBryce BannatyneKen MelbanJohn BurrApril Aymami

Jason Cole
Dan Grant
Darren Haver

OFFICIALLY PARTICIPATING:
Dr. Tim Spann, Spann Ag Research &

Catherine Keeling Consulting Ed McFadden

Leo McGuire <u>GUESTS PARTICIPATING:</u>
Tom Roberts (9:05) Consuelo Fernandez, Brokaw Nursery

Ryan Rochefort Chuck Bandy, McMillan Farm

Robert Grether (ex officio; 9:04) Management

Bob Schaar, Simpatica

CALL TO ORDER

Leo McGuire, Production Research Committee (PRC) Chairman, called the meeting to order at 9:00 a.m. with a quorum present.

OPPORTUNITY FOR PUBLIC COMMENT

Dan Grant introduced Consuelo Fernandez who manages research for Brokaw Nursery.

<u>APPROVAL OF MINUTES OF JUNE 29, 2021 PRODUCTION RESEARCH</u> COMMITTEE MEETING

MOTION

To approve the minutes of the June 29, 2021 Production Research Committee meeting.

(Burr/Cole) MSC Unanimous

Motion 21-8-4-1

DISCUSSION ITEMS

A. Consider approval of the research proposal "Understanding the effects of soil microbial community enhancement on avocado stress tolerance"

Dr. Spann reminded the Committee that this proposal was presented at the June 29, 2021, PRC meeting, but discussion and a recommendation on the project was tabled until this meeting so the proposal could be reviewed in context of the other proposals being considered for fiscal year 2021-22. The discussion focused on the complex nature of the proposal topic – the soil microbiome. There was general agreement that this could be a powerful tool for addressing a number of issues facing the avocado industry; however, there was uncertainty that this was the best proposal to begin exploring this complex topic.

Mr. Chuck Bandy asked if he could address the Committee and Chairman McGuire agreed. Mr. Bandy stated that he has been using the Great Crops products for about a year and has seen good results. He is seeing reduced phytophthora root rot (PRR) pressure on replants and mature trees affected by the disease are also responding positively. He stated that mature trees with PRR, which historically have small leaves and thin canopies, have put on a good leaf flush resulting in reduced sunburn of the fruit.

Discussion continued and focused on the experimental design and project cost. There was general agreement that the project cost was above what the Committee was comfortable recommending for funding. However, the committee agreed that a tool to help growers manage PRR is needed, and perhaps the proposal could be reworked to focus more on yield benefits of using the Great Crops products, with reduced emphasis on the soil microbiome components, while costing substantially less. Dr. Spann agreed to share this feedback with Great Crops and ask for a revised proposal that could be reviewed at a future meeting.

B. Consider approval of the research proposal "Phenology and ecology of avocado lace bug in Southern California"

Dr. Spann explained to the Committee that this proposal was unsolicited but arose following a recent UC Ag Experts seminar by Dr. Mark Hoddle's in which he was unable to answer basic questions from pest control advisors about the avocado lace bug (ALB). Dr. Hoddle has a new masters degree student starting who would work on the project if funded. Discussion ensued and the Committee questioned the significance of this pest and whether this funding was necessary. Member Rochefort told the Committee that he had several groves he was considering spraying in 2020 due to ALB infestation, but the pest's population crashed following summer heatwaves and has not rebounded in 2021. Other members indicated that they believed this was a pest that could be significant for

California Avocado Commission Production Research Committee Minutes August 4, 2021

the industry and it would be wise to have a better understanding of the pest in case it spreads, and action is required. It was also mentioned that there are currently no approved organic control products for this pest. There was general agreement that this would be money well spent to develop a sound understanding of a pest that poses a significant risk to the industry.

MOTION

To recommend funding the proposal "Phenology and ecology of avocado lace bug in Southern California" as submitted.

(McFadden/Burr) MSC 9 yea, 1 nay

Motion 21-8-4-2

C. Consider approval of the research proposal "Development of chloride mitigation strategies for California avocado groves: Technology review and treatment prediction"

Dr. Spann reminded the Committee that the genesis of this proposal was a meeting between Dr. Haizhou Liu, chemical engineering professor at UC Riverside, and Leo McGuire, John Burr and Dr. Spann to discuss potential technologies for mitigating chlorides in avocado irrigation water. The proposal before the Committee would develop a white paper reviewing chloride mitigation technologies and conduct initial feasibility studies of these technologies to develop a list of technologies that could potentially be field tested in the future.

The Committee's discussion focused on the cost of this initial work. The question was asked whether running the project through the UC instead of Dr. Liu's private company would reduce the cost by eliminating his salary expenses. The need for such a study was not questioned and everyone agreed that chlorides are an industry-wide problem that need to be addressed. The Committee asked Dr. Spann to talk with Dr. Liu about running the project through the UC system and to try to reduce the costs, with the intent of reviewing a revised proposal at a future meeting.

D. GEM avocado scarring trial year 2 of data collection

Dr. Spann reminded the Committee that they recommended and the Board approved funding a 2-year study on the cause of scarring of GEM fruit for fiscal years 2019-20 and 2020-21. The first year of the trial was completed, but the second year did happen go forward due to poor bloom and fruit set on the trees at the two trial sites. Dr. Spann told the Committee that the question before them was whether to fund the second year of data collection in 2021-22 or were the first-year data sufficient. Discussion ensued and there was general agreement that the first-year data were good and sufficiently demonstrated that the cause of the fruit scarring is from wind and a second year of data were unnecessary.

California Avocado Commission Production Research Committee Minutes August 4, 2021

ADJOURN MEETING

Timothy Spann

Leo McGuire, Production Research Committee (PRC) Chairman, adjourned the meeting at 10:59 a.m.
Respectfully submitted,

EXHIBITS ATTACHED TO THE PERMANENT COPY OF THESE MINUTES

EXHIBIT A August 4, 2021 Production Research Committee AB 2720 Roll Call Vote Tally Summary



CALIFORNIA AVOCADO COMMISSION

Production Research Committee AB 2720 Roll Call Vote Tally Summary

To be attached to the Meeting Minutes

Meeting Name:	Meeting Location:	Meeting Date:
California Avocado Commission	Teleconference	August 4, 2021
Production Research Committee		
Meeting		

Attendees Who Voted	<u>MOTION</u> <u>21-8-4-1</u>	<u>MOTION</u> <u>21-8-4-2</u>
Leo McGuire, Chair	Yea	Yea
Bryce Bannatyne	Yea	Yea
John Burr	Yea	Yea
Jason Cole	Yea	Yea
Dan Grant	Yea	Nay
Darren Haver	Yea	Yea
Catherine Keeling	Yea	Yea
Ed McFadden	Yea	Yea
Tom Roberts	Yea	Yea
Ryan Rochefort	Yea	Yea
Outcome	Unanimous	9 Yea 1 Nay

Understanding the Effects of Soil Microbial Community Enhancement on Avocado Stress Tolerance

Tomas Aguayo, Agronomist
Great Crops
1175 Calle Cordoniz Rd
Los Osos, CA 93402
tomas@greatcrops.com
(805) 415-5570

BACKGROUND

The avocado industry in California is facing many challenges such as biotic and abiotic stressors, labor availability, water price and quality, and soil borne diseases, just to mention some. The most important factors that determine fruit yield and quality, are factors that directly affect the performance of the crop mostly for the nature and sensitivity of the avocado itself. Factors such as soil and water salinity, wind, heat, and drought, can and may influence others directly or indirectly — soil chemical and physical properties, soil moisture holding capacity, water infiltration/run-off, nutrient cycling and availability — are all variables that can and must be addressed to stay competitive and profitable.

If we can solve all or part of the above, the crop performance will improve immediately. In the meantime, the trees are struggling to survive, and are in a weakened state, making them more susceptible to existing and new diseases such as Phytophthora root rot.

In search of a true and complete solution for farmers and the farming industry, Great Crops has developed over 12-years of field and lab research in many crops in the U.S. and internationally, a simple nutritional/soil conditioners/soil microbiological program. Through six water applied materials, the program provides a combination of essential nutrients in their proper ratios for key physiological stages of crop development.

These materials with high concentrations of organic carbon from five different sources, other soil conditioners, and highly bio diversified beneficial microbiology, provide protection against biotic and abiotic stressors, while at the same time the necessary nutrients and soil regenerative properties that aim to improve soil structure that will provide an immediate better environment for the soil microbiome by decreasing soil moisture and temperature variations. The program also will improve water infiltration rates, decreasing water and nutrients

run-off; will increase microorganism species richness and biodiversity, providing initial organic food sources and in a short-term improving plant photosynthesis, nutrient cycling and plant performance and resistance.

Current commercial field research has already demonstrated decrease of Phytophthora and Pythium pressure in treated trees by increasing soil biodiversity, while at the same time improving nutrient cycling and availability.

It has also been observed in the last decade through field trials that soil moisture and soil temperatures at different depths (0 to 24 inches), have stayed more stable and decrease oscillation respectively which has helped us to explain the positive changes in soil microbiology and tree performance regardless of soil taxonomy and/or water quality. Some of the crops where the Great Crops technology has been validated include wine grapes, olives, strawberries, almonds, citrus, asparagus, and avocados among others.

For avocados and olives, these projects also included monitoring disease pressure with incredible findings showing significant decrease of soil borne pathogens for avocados as mentioned above and also of *Xylella fastidiosa* in three different olive varieties for oil production.

Further and initial findings are also showing a positive correlation in avocado trees between soil micro biome and plant endophytes, suggesting that the Great Crops program may activate new or metabolic pathways that had stayed "turned off."

OBJECTIVE(S)

The primary goal is to demonstrate that balanced nutrition, soil conditioners, and carbon as a food source for microorganism development, results in an improved soil microbiome thereby increasing soil aggregation, reducing disease pressure and increasing fruit yield and quality.

Specific objectives:

- Compared tree performance, soil health/regeneration improvement
- Carbon sequestration potential (Carbon credits)
- Monitor water holding capacity at different depths (0 to 24 inches every 4")
- Monitor soil temperature

 Demonstrate a possible equal or higher performance of the trees when standard fertility program is reduced in 50% NPK supply alone versus an enhanced biological/carbon-based treatment

RESEARCH PLAN (see also Appendix)

A commercial avocado ranch in the Fallbrook growing area will be selected to test the performance of soil applied liquid materials across full irrigation zones/blocks. Treated (TRT) blocks will be compared with an untreated block that will receive grower standard practices (GSP). Under TRT section, a reduction of GSP will be suggested depending on soil & irrigation water present chemical-physical conditions.

Within each irrigation zone/block of each treatment (TRT & GSP), trees must be the same age, rootstock, variety and ideally grown with similar gradients when on hill sides, making sure they are all under the same exposure (N, S, E or W), and under the same soil taxa.

A minimum of seven to eight tress in each section will be selected as the replicates; from these, all measurements and samples will be taken (soil, tissue, etc.).

Initial samples from trees that are "randomly" selected within each treatment will be collected before the initiation of the project (T0), during November of 2021. Following the initial sampling the sampling frequency during each growing season will be discussed.

Measurements, tools and sampling will be able to demonstrate the following:

- Increase tree performance (yield and fruit quality),
- Provide soil regenerative properties
- More efficient nutrient cycling,
- Provide trees with antibiotic compounds and balance hormone production,
- Decrease of soil borne diseases pressure,
- Increase effectiveness tree photosynthesis,
- Improvement of soil Carbon cycle,
- Improve water infiltration rates (reducing run-off),
- Reduce fruit drop when facing abiotic stressors (wing/heat/cold),

- Decrease water stress and or needs,
- Decrease usage of fertilizers and pesticides.

Avocado Grower Standard Practices (GSP) will be compared to the addition of the Great Crops program (GC). GC program consists in applying via irrigation six specially manufactured and nutritionally balanced materials for key physiological stages of tree development such as root flushes, flowering, vegetative growth, etc. Except for one of the six materials, all others can be combined with standard NPK fertilizers; when GC program is applied, all soil conditioners such as humic acids, seaweed, mycorrhizae, beneficial microbes, organic C, amino acids, and micronutrients, can be taken out of the standard program since they are being provided via the Great Crops program.

In this ranch there will be 3 treatments:

- 1. GSP
- 2. GSP + GC
- 3. 50% GSP + GC

Each one of the replicates (7 to 8) of these treatments (3) will have the following tools and samplings:

- Sentek Soil Probes (2 feet) providing measurements of Soil Moisture, Soil Temperature and Soil Salinity every 4" from 0 to 24".
- Weather station
- 3 sampling points, from T0 to T2, each sampling to occur at key physiological stages:
 - After fall root flush (November)
 - During flower bud development (March)
 - During fruit growth and summer vegetative flush
- Samples will be:
 - Soil (0-8") for specific microbiome analysis and disease (i.e., Phytophthora) presence and pressure; also, chemical and physical analysis
 - Tissue for nutritional content that will include standard and SAP analysis

Yearly Budget

Avocado with 8 Reps						
	Price/unit	Samplings/year	# of treatments	# od rep	Tot # of samples	Total
Microbiome+SOIL+HANEY	240	3	3	8	72	17,280.00
SAP	40	3	3	8	72	2,880.00
Tissue	30	3	3	8	72	2,160.00
Phythopthora	100	3	3	8	72	7,200.00
						29,520.00

Avocado with 7 Reps							
	Price/unit	Samplings/year	# of treatments	# od rep	Tot # of samples	Total	
Microbiome+SOIL+HANEY	240	3	3	7	63	15,120.00	
SAP	40	3	3	7	63	2,520.00	
Tissue	30	3	3	7	63	1,890.00	
Phythopthora	100	3	3	7	63	6,300.00	
						25,830.00	

SCHEDULE

November 2021

Selection of ranches, blocks and trees for the study. First soil and tissue samples will be taken, and this will be our TO.

Installation of soil probes and weather stations will take place during this month as well.

Depending on late fall weather and overall conditions of each farm, the first application might be scheduled before the end of the year.

February 2022 to August/September 2022 Application of full program

Repeat above for 2023 and 2024

APPENDIX

- 1. Microbiological analysis of some of the materials to be used are also included below (Great Radix and Great Forti).
- 2. Soil microbiome example report
- 3. Technical sheets and description of the Great Crops materials are included.
- 4. Labels of the products approved by CDFA are also included.

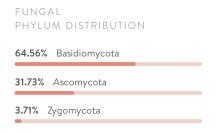




PARCEL VARIETY DATE

MICROBIAL POPULATION

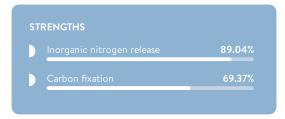
All the information shown in this microbial report is based on the detected presence of 632 different species.



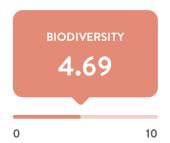


BACT PHYLUM DISTRIB	
Firmicutes	49.35%
Proteobacteria	24.54%
Bacteroidetes	12.56%
Cyanobacteria	6.85%
Actinobacteria	2.09%

CONCLUSIONS



BIOSUSTAINABILITY



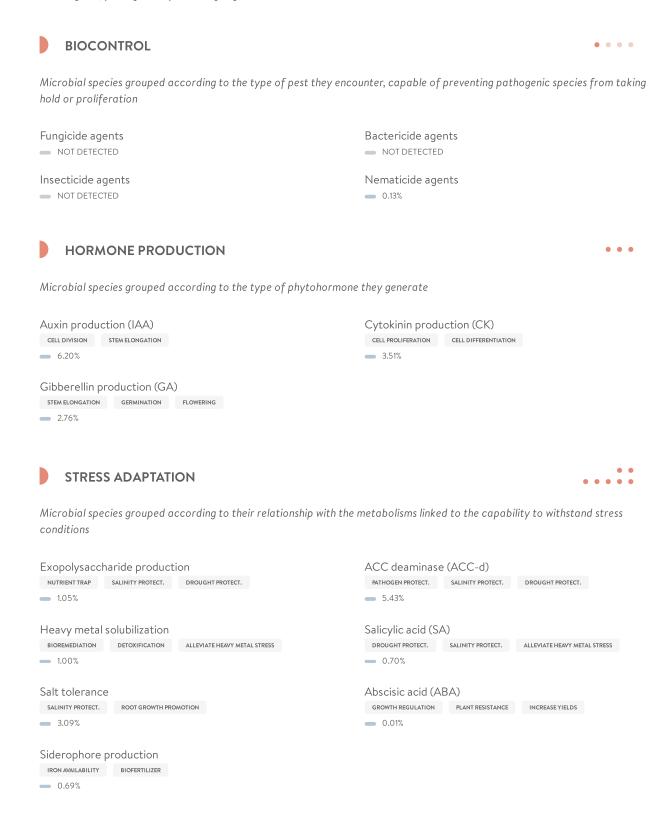
Richness, evenness and equilibrium of microbial species



Capability of soil microbial communities to perform multiple functions

PLANT HEALTH IMPROVEMENT

Biocontrol agents, plant growth promoting organisms

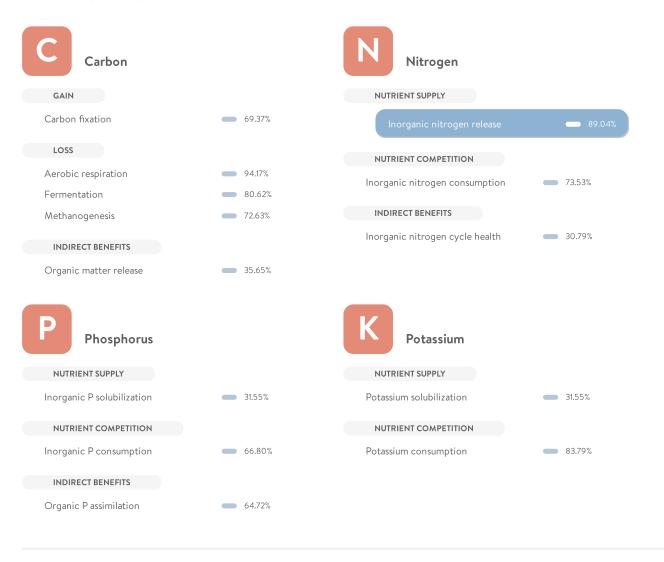


Item 6-a-9 of 5

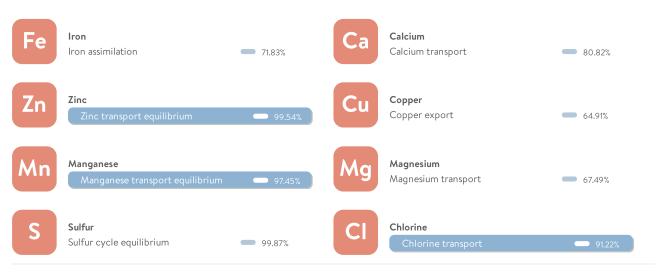
NUTRITION

Nutritional status based on the microbial mobilization of certain compounds

MAJOR COMPOUNDS



MINOR COMPOUNDS



APPENDIX

BACTERIAL QUANTIFICATION

METHODOLOGY

The absolute quantification of bacterial communities using **Next-Gen sequencing** enables the cell number measurements and, thus, the knowledge of the total microbial loads among a sample

The present analysis relies on the application of a **spike-in of exogenous bacterial** with known microbial composition into crude soil amendment samples, under the ZymoBiomics Spike-in Control I technology. After sequencing and data processing, the relative abundance of the exogenous bacterial strains and the known Spike-in cell input were used to transform relative abundances of all bacteria strains in the sample to absolute abundance.

GR2 control

Data in this report correspond to the average absolute abundance obtained from three technical replicates.

1	Nitrososphaera sp.	1.36e+8
2	Rubrobacter sp.	3.94e+7
3	Gemmatimonas sp.	2.65e+7
4	Solirubrobacter sp.	1.86e+7
5	Gaiella sp.	8.82e+6
6	Phycisphaera sp.	7.59e+6
7	lamia sp.	6.91e+6
8	Flavisolibacter sp.	6.71e+6
9	Rhodoplanes sp.	5.99e+6
10	Blastocatella sp.	5.47e+6
11	Chthoniobacter sp.	5.42e+6
12	Gemmata sp.	5.03e+6
13	Patulibacter sp.	4.69e+6
14	Steroidobacter sp.	4.59e+6
15	Bryobacter sp.	4.24e+6

16	Kaistobacter sp.	4.19e+6
17	Nitrospira sp.	3.93e+6
18	Nocardioides sp.	3.84e+6
19	Pedosphaera sp.	3.79e+6
20	Escherichia sp.	3.69e+6
21	Variovorax sp.	3.62e+6
22	Novosphingobium sp.	3.59e+6
23	Pirellula sp.	3.42e+6
24	Lactobacillus sp.	3.21e+6
25	Alysiosphaera sp.	3.20e+6
26	Bacteroides coprosuis	3.18e+6
27	Balneimonas sp.	2.69e+6
28	Lactobacillus acetotolerans	2.68e+6
29	Arthrobacter cereus	2.67e+6
30	Achromobacter sp.	2.58e+6





31	Haliangium sp.	2.42e+6
32	llumatobacter sp.	2.42e+6
33	Skermanella sp.	2.27e+6
34	Planctomyces sp.	2.24e+6
35	Thermomonas sp.	2.13e+6
36	Phenylobacterium sp.	2.05e+6
37	Adhaeribacter sp.	2.04e+6
38	Sphingomonas sp.	1.88e+6
39	Intrasporangium sp.	1.85e+6
40	Mesorhizobium sp.	1.75e+6
41	Ohtaekwangia sp.	1.72e+6
42	Chryseolinea sp.	1.68e+6
43	Marmoricola sp.	1.67e+6
44	Hydrogenophaga pseudoflava	1.61e+6
45	Blastococcus sp.	1.58e+6
46	Opitutus sp.	1.56e+6
47	Ornatilinea sp.	1.56e+6
48	Nitrosococcus sp.	1.50e+6
49	Stackebrandtia sp.	1.48e+6
50	Bosea sp.	1.45e+6
51	Pedobacter sp.	1.34e+6

52	Xiphinematobacter sp.	1.33e+6
53	Lactobacillus oris	1.26e+6
54	Singulisphaera sp.	1.26e+6
55	Ferruginibacter sp.	1.24e+6
56	Azospira sp.	1.20e+6
57	Mycobacterium lentiflavum	1.18e+6
58	Arenimonas sp.	1.12e+6
59	Entotheonella sp.	1.09e+6
60	Reyranella sp.	1.09e+6
61	Microlunatus sp.	1.07e+6
62	Arthrobacter sp.	1.06e+6
63	Conexibacter sp.	1.04e+6
64	Bacillus sp.	9.82e+5
65	Wohlfahrtiimonas sp.	9.45e+5
66	Peptoniphilus sp.	9.37e+5
67	Lactobacillus kimchicus	9.31e+5
68	Altererythrobacter sp.	9.03e+5
69	Pediococcus ethanolidurans	8.53e+5
70	Roseiflexus sp.	8.36e+5
71	Rubellimicrobium sp.	8.32e+5
72	Leptolyngbya boryana	8.28e+5





73	Rhodopirellula sp.		8.00e+5
74	Mycobacterium sp.		7.94e+5
75	Sandaracinus sp.		7.82e+5
76	Aquicella sp.		7.70e+5
77	Acidovorax sp.		7.35e+5
78	Pseudonocardia halophobica		7.15e+5
79	Alcanivorax sp.		6.87e+5
80	Brucella sp.		6.30e+5
81	Luteimonas mephitis		6.24e+5
82	Solibacter sp.		5.94e+5
83	Myroides sp.		5.79e+5
84	Bacillus coahuilensis		5.72e+5
85	Methyloversatilis sp.		5.70e+5
86	Microbacterium arthrosphaerae		5.64e+5
87	Hyphomicrobium sp.		5.45e+5
88	Coprothermobacter sp.		5.39e+5
00			
89	Paenalcaligenes faecalis		5.37e+5
90	Paenalcaligenes faecalis Lysobacter sp.		5.37e+5 5.33e+5
90	Lysobacter sp.	-	5.33e+5

94	Pedomicrobium sp.		4.85e+5
95	Microvirga sp.		4.70e+5
96	Bacteroides sp.		4.61e+5
97	Microthrix sp.		4.48e+5
98	Rheinheimera sp.		4.14e+5
99	Bradyrhizobium sp.	_	4.12e+5
100	Paenochrobactrum glaciei		3.96e+5
101	Devosia insulae		3.94e+5
102	Aciditerrimonas sp.		3.82e+5
103	Leucobacter sp.		3.70e+5
104	Pontibacter sp.		3.67e+5
105	Clostridium sp.	_	3.62e+5
106	Craurococcus sp.		3.61e+5
107	Acinetobacter baumannii	_	3.56e+5
108	Agromyces sp.		3.51e+5
109	Sphingopyxis macrogoltabida		3.51e+5
110	Isosphaera sp.		3.43e+5
111	Zoogloea sp.		3.43e+5
112	Singulisphaera limicola		3.39e+5
113	Filomicrobium sp.		3.33e+5
114	Koribacter sp.		3.27e+5



Great Crops



115	Anaeromyxobacter sp.	3.21e+5
116	Nannocystis sp.	3.15e+5
117	Pedosphaera parvula	3.03e+5
118	Fluviicola sp.	2.91e+5
119	Phaselicystis sp.	2.87e+5
120	Legionella sp.	2.79e+5
121	Sufflavibacter sp.	2.79e+5
122	Cellulomonas sp.	2.59e+5
123	Sorangium sp.	2.58e+5
124	Vogesella sp.	2.58e+5
125	Roseomonas sp.	2.57e+5
126	Ilumatobacter fluminis	2.55e+5
127	Labrys sp.	2.55e+5
128	Peredibacter sp.	2.48e+5
129	Sporosarcina sp.	2.46e+5
130	Treponema sp.	2.36e+5
131	Pseudomonas sp.	2.32e+5
132	Fimbriimonas sp.	2.30e+5
133	Nordella sp.	2.30e+5
134	Afifella sp.	2.26e+5
135	Dysgonomonas sp.	2.24e+5

136	Methylomonas sp.		2.14e+5
137	Bacillus aryabhattai	_	2.09e+5
138	Catellatospora sp.		2.09e+5
139	Chitinophaga sp.		2.06e+5
140	Truepera sp.		2.06e+5
141	Paenibacillus sp.	_	1.95e+5
142	Segetibacter sp.		1.94e+5
143	Brevundimonas diminuta	_	1.88e+5
144	Rhodobium sp.		1.86e+5
145	Desulfomicrobium sp.		1.82e+5
146	Devosia sp.		1.82e+5
147	Ensifer sp.		1.82e+5
148	Pusillimonas sp.		1.82e+5
149	Bauldia sp.		1.78e+5
150	Anaerosalibacter sp.		1.72e+5
151	Caldilinea sp.		1.70e+5
152	Erythrobacter sp.		1.70e+5
153	Methylobacterium sp.		1.70e+5
154	Luteolibacter sp.		1.66e+5
155	Myroides [odoratimimus]		1.63e+5
156	Smithella sp.		1.58e+5





157	Rhodococcus sp.	-	1.55e+5
158	Agromyces flavus		1.51e+5
159	Aquimonas sp.		1.51e+5
160	Methanoregula sp.		1.45e+5
161	Telmatobacter sp.		1.45e+5
162	Lautropia sp.		1.43e+5
163	Nakamurella sp.		1.41e+5
164	Ochrobactrum sp.		1.39e+5
165	Massilia sp.		1.38e+5
166	Tepidimicrobium sp.		1.37e+5
167	Geobacillus sp.	-	1.35e+5
168	Blastococcus aggregatus		1.33e+5
169	Methylorosula sp.		1.33e+5
170	Woodsholea sp.		1.33e+5
171	Pelagibius sp.		1.29e+5
172	Acinetobacter rudis		1.26e+5
173	Oceanobacillus sp.		1.26e+5
174	Acidicaldus sp.		1.21e+5
175	Acidiferrobacter sp.		1.21e+5
176	Aeromicrobium sp.		1.21e+5
177	Hymenobacter sp.		1.21e+5

178	Mariprofundus sp.	1.21e+5
179	Saccharimonas sp.	1.21e+5
180	Thiomonas sp.	1.21e+5
181	Cloacamonas sp.	1.15e+5
182	Euzebya sp.	1.15e+5
183	Gelria sp.	1.13e+5
184	Methanosarcina sp.	1.13e+5
185	Paludibacter sp.	1.13e+5
186	Cellulosimicrobium cellulans	1.09e+5
187	Protochlamydia sp.	1.09e+5
188	Tissierella sp.	1.04e+5
189	Ardenscatena sp.	1.03e+5
190	Blastopirellula sp.	1.03e+5
191	Granulosicoccus sp.	1.02e+5
192	Taibaiella sp.	1.02e+5
193	Asanoa sp.	9.70e+4
194	Coprococcus sp.	9.70e+4
195	Desulfovibrio sp.	9.70e+4
196	Hirschia sp.	9.70e+4
197	Polaromonas sp.	9.70e+4
198	Rhizomicrobium sp.	9.70e+4





199	Oceanobacillus litoralis	9.52e+4
200	Parasegetibacter sp.	9.45e+4
201	Bacillus aeris	9.33e+4
202	Haloferula sp.	9.29e+4
203	Prosthecobacter sp.	9.29e+4
204	Actinotalea sp.	9.09e+4
205	Giesbergeria sinuosa	8.89e+4
206	Alterococcus sp.	8.48e+4
207	Nitrobacter sp.	8.48e+4
208	Nitrolancea sp.	8.48e+4
209	Nocardioides albus	8.48e+4
210	Bosea eneae	7.88e+4
211	Rhodanobacter sp.	7.88e+4
212	Arcobacter cibarius	7.68e+4
213	Arcticibacter sp.	7.57e+4
214	Longispora sp.	7.57e+4
215	Agrobacterium sp.	7.27e+4
216	Anaerolinea sp.	7.27e+4
217	Chloroflexus sp.	7.27e+4
218	Glycomyces harbinensis	7.27e+4
219	Peptostreptococcus sp.	7.27e+4

220	Pseudoxanthomonas sp.		7.27e+4
221	Terrabacter sp.		7.27e+4
222	Olivibacter sp.		6.97e+4
223	Pseudonocardia sp.		6.97e+4
224	Pseudospirillum sp.		6.97e+4
225	Acidothermus sp.		6.87e+4
226	Caulobacter sp.		6.67e+4
227	Ruminococcus sp.		6.67e+4
228	Planifilum sp.		6.51e+4
229	Adhaeribacter aerolatus		6.46e+4
230	Streptomyces sp.	_	6.36e+4
231	Defluviicoccus sp.		6.06e+4
232	Rhodomicrobium sp.		6.06e+4
233	Rubricoccus sp.		6.06e+4
234	Ureibacillus sp.		5.93e+4
235	Lactobacillus acidipiscis		5.86e+4
236	Peribacillus muralis		5.66e+4
237	Rhodocytophaga sp.		5.66e+4
238	lgnatzschineria sp.		5.54e+4
239	Brevibacillus agri		5.45e+4
240	Brevundimonas sp.		5.45e+4





241	Catelliglobosispora sp.	5.45e+4
242	Desulfotomaculum sp.	5.45e+4
243	Dyella sp.	5.45e+4
244	Streptosporangium roseum	5.45e+4
245	Kaistia granuli	5.25e+4
246	Syntrophomonas sp.	5.25e+4
247	Byssovorax sp.	5.15e+4
248	Actinomadura bangladeshensis	4.85e+4
249	Couchioplanes sp.	4.85e+4
250	Coxiella sp.	4.85e+4
251	Gemmobacter sp.	4.85e+4
252	Hamadaea yuxiensis	4.85e+4
253	Nitrososphaera gargensis	4.85e+4
254	Oryzihumus leptocrescens	4.85e+4
255	Parvibaculum sp.	4.85e+4
256	Pimelobacter simplex	4.85e+4
257	Xanthomonas sp.	4.85e+4
258	Blautia sp.	4.61e+4
259	Thermobacillus sp.	4.61e+4
260	Alkaliphilus sp.	4.58e+4
261	Virgisporangium ochraceum	4.54e+4

262	Amaricoccus sp.		4.44e+4
263	Hydrogenoanaerobacterium saccharovorans		4.44e+4
264	Phaselicystis flava		4.44e+4
265	Rhodobacter sp.	_	4.44e+4
266	Roseimicrobium sp.		4.44e+4
267	Pseudomonas fluorescens		4.36e+4
268	Bacillus oleronius		4.24e+4
269	Desulfovibrio mexicanus		4.24e+4
270	Lysinibacillus sp.	_	4.24e+4
271	Pilimelia sp.		4.24e+4
272	Sphaerobacter sp.		4.24e+4
273	Cryptosporangium sp.		4.04e+4
274	Anaerovorax sp.		3.98e+4
275	Corynebacterium sp.	_	3.94e+4
276	Brevibacillus laterosporus		3.84e+4
277	Symbiobacterium sp.		3.84e+4
278	Ignatzschineria larvae		3.81e+4
279	Actinobaculum sp.		3.64e+4
280	Anaerotruncus sp.		3.64e+4
281	Angustibacter sp.		3.64e+4
282	Arthrospira sp.		3.64e+4





283	Bacillus alkalitelluris	-	3.64e+4
284	Blastomonas sp.		3.64e+4
285	Cryocola sp.		3.64e+4
286	Metachlamydia lacustris		3.64e+4
287	Niastella sp.		3.64e+4
288	Proteiniphilum sp.		3.64e+4
289	Rhodococcus caviae	_	3.64e+4
290	Stella sp.		3.64e+4
291	Janthinobacterium lividum		3.48e+4
292	Shimazuella sp.		3.48e+4
293	Tumebacillus sp.		3.43e+4
294	Limnobacter sp.		3.33e+4
295	Sedimentibacter sp.		3.29e+4
296	Algoriphagus terrigena		3.23e+4
297	Dongia sp.		3.23e+4
298	Oscillibacter sp.		3.23e+4
299	Epulopiscium sp.		3.15e+4
300	Nonomuraea sp.		3.15e+4
301	Aquamicrobium sp.		3.03e+4
302	Ornithinicoccus sp.		3.03e+4
303	Gracilibacter sp.		2.94e+4

304	Caloramator sp.	2.91e+4
305	Agaricicola sp.	2.83e+4
306	Oxobacter sp.	2.83e+4
307	Cohnella arctica	2.80e+4
308	Bacillus fordii	2.79e+4
309	Actinoplanes sp.	2.73e+4
310	Arthrobacter alkaliphilus	2.67e+4
311	Azospirillum sp.	2.56e+4
312	Agrococcus jenensis	2.42e+4
313	Aminobacter aminovorans	2.42e+4
314	Arcobacter sp.	2.42e+4
315	Cytophaga sp.	2.42e+4
316	Dechloromonas sp.	2.42e+4
317	Herpetosiphon sp.	2.42e+4
318	Hyphomonas sp.	2.42e+4
319	Ignavibacterium sp.	2.42e+4
320	Jatrophihabitans sp.	2.42e+4
321	Legionella pneumophila	2.42e+4
322	Litorilinea aerophila	2.42e+4
323	Luedemannella sp.	2.42e+4
324	Methanoculleus sp.	2.42e+4





325	Methylotenera sp.		2.42e+4
326	Parasegetibacter luojiensis		2.42e+4
327	Pelagibacterium sp.		2.42e+4
328	Virgisporangium sp.		2.42e+4
329	Wandonia sp.		2.42e+4
330	Aneurinibacillus aneurinilyticus		2.28e+4
331	Desulfotomaculum aeronauticum		2.27e+4
332	Sphingobacterium sp.		2.25e+4
333	Lacibacter sp.		2.18e+4
334	Rhodococcus erythropolis	_	2.18e+4
335	Thermobacillus composti		2.18e+4
336	Acidocella sp.		2.12e+4
337	Asanoa hainanensis		2.12e+4
338	Dactylosporangium matsuzakiense		2.12e+4
339	Garciella sp.		2.12e+4
340	Prauseria sp.		2.12e+4
341	Saccharopolyspora sp.		2.12e+4
342	Saccharothrix sp.		2.12e+4
343	Chelatococcus sp.		2.02e+4
344	Fodinicola sp.		2.02e+4
345	Meniscus sp.		2.02e+4
	<u>'</u>		

346	Methylobacillus sp.		2.02e+4
347	Nocardia coubleae		2.02e+4
347	Nocarala coubleae		2.026+4
348	Paenibacillus lentimorbus	_	1.96e+4
349	Flavisolibacter ginsengisoli		1.94e+4
350	Flavobacterium saliperosum	_	1.94e+4
351	Lactobacillus halophilus	_	1.94e+4
352	Actinopolymorpha cephalotaxi		1.82e+4
353	Alteribacillus persepolensis		1.82e+4
354	Belnapia sp.		1.82e+4
355	Desulfobulbus sp.		1.82e+4
356	Dokdonella sp.		1.82e+4
357	Fastidiosipila sp.		1.82e+4
358	Glycomyces sp.		1.82e+4
359	Kribbella sp.		1.82e+4
360	Luteimonas composti		1.82e+4
361	Magnetospirillum magneticum		1.82e+4
362	Micromonospora sp.		1.82e+4
363	Parapedobacter sp.		1.82e+4
364	Spirochaeta sp.		1.82e+4
365	Stenotrophomonas maltophilia		1.82e+4
366	Tuberibacillus sp.		1.82e+4





367	Sediminibacterium sp.	1.70e+4
368	Actinophytocola sp.	1.62e+4
369	Dichotomicrobium sp.	1.62e+4
370	Dyadobacter sp.	1.62e+4
371	Haliea sp.	1.62e+4
372	Martelella sp.	1.62e+4
373	Methanolinea sp.	1.62e+4
374	Mycoplana sp.	1.62e+4
375	Paludibacter propionicigenes	1.62e+4
376	Pseudochrobactrum sp.	1.62e+4
377	Paucisalibacillus sp.	1.58e+4
378	Caldicoprobacter sp.	1.56e+4
379	Dysgonomonas capnocytophagoides	1.51e+4
380	Macellibacteroides sp.	1.51e+4
381	Roseococcus sp.	1.51e+4
382	Roseomonas aerilata	1.51e+4
383	Methylobacterium adhaesivum	1.45e+4
384	Tumebacillus ginsengisoli	1.41e+4
385	Paenibacillus polymyxa	1.40e+4
386	Ammoniphilus sp.	1.31e+4
387	Acholeplasma sp.	1.21e+4

388	Actinomadura sp.	1.21e+4
389	Aequorivita sp.	1.21e+4
390	Alicyclobacillus herbarius	1.21e+4
391	Aliihoeflea aestuarii	1.21e+4
392	Alkaliphilus oremlandii	1.21e+4
393	Aminobacter sp.	1.21e+4
394	Bythopirellula goksoyri	1.21e+4
395	Constrictibacter sp.	1.21e+4
396	Elusimicrobium sp.	1.21e+4
397	Erysipelothrix inopinata	1.21e+4
398	Erysipelothrix sp.	1.21e+4
399	Flavihumibacter sp.	1.21e+4
400	Fulvimarina sp.	1.21e+4
401	Hyalangium sp.	1.21e+4
402	Hydrogenoanaerobacterium sp.	1.21e+4
403	Latescibacter sp.	1.21e+4
404	Litorilinea sp.	1.21e+4
405	Magnetospirillum sp.	1.21e+4
406	Methanobacterium sp.	1.21e+4
407	Methanomassiliicoccus sp.	1.21e+4
408	Methylocaldum sp.	1.21e+4





409	Nocardioides koreensis	1.21e+4
410	Pseudoclavibacter sp.	1.21e+4
411	Pseudoxanthomonas dokdonensis	1.21e+4
412	Rhabdochlamydia porcellionis	1.21e+4
413	Rubritepida sp.	1.21e+4
414	Rugosimonospora acidiphila	1.21e+4
415	Runella zeae	1.21e+4
416	Salana sp.	1.21e+4
417	Sneathiella sp.	1.21e+4
418	Sphingomonas dokdonensis	1.21e+4
419	Thermaerobacter marianensis	1.21e+4
420	Thiovirga sp.	1.21e+4
421	Virgibacillus halophilus	1.21e+4
422	Rummeliibacillus sp.	1.11e+4
423	Azotobacter sp.	1.01e+4
424	Cryptanaerobacter sp.	1.01e+4
425	Desulfurispora sp.	1.01e+4
426	Streptomyces lannensis	1.01e+4
427	Pricia sp.	9.70e+3
428	Proteiniclasticum sp.	9.43e+3
429	Actinocorallia longicatena	9.09e+3

430	Allocatelliglobosispora scoriae		9.09e+3
431	Longispora fulva		9.09e+3
432	Rhodococcus fascians	_	9.09e+3
433	Sphingobacterium composti		9.09e+3
434	Stenotrophomonas sp.		9.09e+3
435	Streptomyces lateritius	_	9.09e+3
436	Actinomyces sp.		8.08e+3
437	Allocatelliglobosispora sp.		8.08e+3
438	Arenibacter sp.		8.08e+3
439	Caenimonas sp.		8.08e+3
440	Desulfocapsa sp.		8.08e+3
441	Desulfosporosinus sp.		8.08e+3
442	Elstera sp.		8.08e+3
443	Lentibacillus sp.		8.08e+3
444	Nannocystis exedens		8.08e+3
445	Oscillospira sp.		8.08e+3
446	Phaeospirillum fulvum		8.08e+3
447	Plesiocystis sp.		8.08e+3
448	Sporocytophaga sp.		8.08e+3
449	Sulfuricurvum kujiense		8.08e+3
450	Thermincola sp.		8.08e+3





451	Thermopolyspora sp.		8.08e+3
452	Mesobacillus foraminis		7.71e+3
453	Caldalkalibacillus sp.		7.27e+3
454	Dorea sp.		7.27e+3
455	Paenibacillus motobuensis	_	7.27e+3
456	Aeribacillus barengoltzii		6.93e+3
457	Lutispora sp.		6.93e+3
458	Clostridium bowmanii	_	6.73e+3
459	Actinoallomurus sp.		6.06e+3
460	Actinocorallia sp.		6.06e+3
461	Altererythrobacter aestuarii		6.06e+3
462	Armatimonas sp.		6.06e+3
463	Brevibacillus sp.	_	6.06e+3
464	Brevibacterium album		6.06e+3
465	Castellaniella sp.		6.06e+3
466	Cellvibrio sp.		6.06e+3
467	Chryseobacterium daejeonense		6.06e+3
468	Chryseobacterium sp.		6.06e+3
469	Chthonomonas sp.		6.06e+3
470	Dictyoglomus sp.		6.06e+3
471	Geobacter sp.		6.06e+3

472	Haloplasma sp.	6.06e+3
473	Hydrogenophaga sp.	6.06e+3
474	Leucobacter komagatae	6.06e+3
475	Longilinea sp.	6.06e+3
476	Mangroviflexus xylanolytica	6.06e+3
477	Microbispora sp.	6.06e+3
478	Neochlamydia sp.	6.06e+3
479	Niabella sp.	6.06e+3
480	Nitratireductor sp.	6.06e+3
481	Owenweeksia sp.	6.06e+3
482	Paracraurococcus ruber	6.06e+3
483	Petrimonas sp.	6.06e+3
484	Silanimonas sp.	6.06e+3
485	Singulisphaera acidiphila	6.06e+3
486	Solimonas flavus	6.06e+3
487	Thiobacillus sp.	6.06e+3
488	Woodsholea limnia	6.06e+3
489	Xanthobacter sp.	6.06e+3
490	Paenibacillus agarexedens	5.59e+3
491	Paenibacillus koleovorans	5.59e+3
492	Sporolactobacillus laevolacticus	5.19e+3





493	Thermoactinomyces sp.		5.19e+3
494	Aeribacillus sp.		4.85e+3
495	Aeromonas sp.		4.85e+3
496	Atopostipes sp.		4.85e+3
497	Cupriavidus oxalaticus		4.85e+3
498	Nevskia sp.		4.85e+3
499	Paenibacillus edaphicus	_	4.66e+3
500	Acidovorax delafieldii		4.04e+3
501	Alicyclobacillus sp.		4.04e+3
502	Clostridium intestinale	_	4.04e+3
503	Comamonas sp.		4.04e+3
504	Dehalobacter sp.		4.04e+3
505	Demequina sp.		4.04e+3
506	Desulfatiferula sp.		4.04e+3
507	Elioraea tepidiphila		4.04e+3
508	Ethanoligenens sp.		4.04e+3
509	Persicitalea sp.		4.04e+3
510	Pleomorphomonas sp.		4.04e+3
511	Prosthecobacter debontii		4.04e+3
512	Shinella sp.		4.04e+3
513	Simplicispira sp.		4.04e+3

514	Staphylococcus sp.	4.04e+3
515	Sulfurospirillum sp.	4.04e+3
516	Syntrophaceticus sp.	4.04e+3
517	Turicibacter sp.	4.04e+3
518	Calditerricola sp.	3.64e+3
519	Alkalihalobacillus clausii	3.46e+3
520	Catenibacterium sp.	3.46e+3
521	Aneurinibacillus sp.	3.31e+3
522	Alkalibacter sp.	3.03e+3
523	Catellatospora citrea	3.03e+3
524	Janthinobacterium sp.	3.03e+3
525	Kineosporia rhamnosa	3.03e+3
526	Mangroviflexus sp.	3.03e+3
527	Melghirimyces thermohalophilus	3.03e+3
528	Pandoraea sp.	3.03e+3
529	Pasteuria sp.	3.03e+3
530	Rhodococcus corynebacterioides	3.03e+3
531	Sphingobacterium paucimobilis	3.03e+3
532	Thermobispora bispora	3.03e+3
533	Thermoflavimicrobium sp.	3.03e+3



535	Paenibacillus harenae	_	2.80e+3
536	Azospirillum genomospecies	_	2.69e+3
537	Acetobacterium sp.		2.42e+3
538	Bacillus coagulans		2.42e+3
539	Bacillus thermolactis	_	2.42e+3
540	Bacteroides ovatus		2.42e+3
541	Caldibacillus sp.		2.42e+3
542	Chlorothrix sp.		2.42e+3
543	Filimonas sp.		2.42e+3
544	Flavobacterium group	_	2.42e+3
545	Lactobacillus harbinensis	_	2.42e+3
546	Natranaerovirga hydrolytica		2.42e+3
547	Desulfosporosinus meridiei		2.20e+3
548	Bacillus funiculus	_	2.02e+3
549	Desulfitibacter sp.		2.02e+3
550	Halomonas sp.		2.02e+3
551	Sporacetigenium sp.		2.02e+3
552	Sporomusa sp.		2.02e+3
553	Coprobacillus sp.		1.73e+3
554	Desulfohalotomaculum peckii		1.73e+3
555	Proteiniborus sp.		1.73e+3

556	Tepidimicrobium ferriphilum	1.73e+3
557	Alkalihalobacillus rhizosphaerae	1.51e+3
558	Alkaliphilus transvaalensis	1.35e+3
559	Fonticella sp.	1.35e+3
560	Myroides marinus	1.35e+3
561	Heliobacterium gestii	1.21e+3
562	Laceyella sacchari	1.01e+3

NOTES

Species belonging to a genus present in the CDFA Approved Microorganisms List

Species present in the CDFA Approved Microorganisms List

APPENDIX

RELATIVE FUNGAL ABUNDANCE

The relative abundance of the fungal communities is reported in %. Data in this report correspond to the average relative abundance obtained from three technical replicates. The absolute quantification of fungal communities using amplicon sequencing has not yet been validated at Biome Makers, Inc.

GR2 control

#	Genus & Species		Percentage
1	Trechispora sp.		61.2759%
2	Monascus purpureus		15.9043%
3	Diutina rugosa		5.1512%
4	Mortierella sp.		3.5310%
5	Pseudeurotium hygrophilum		3.2877%
6	Arachnomyces pilosus		1.8883%
7	Coprinellus sp.		1.8658%
8	Cortinarius sp.		1.2709%
9	Suhomyces xylopsoci		0.6309%
10	Fusarium equiseti		0.5926%
11	Aspergillus terreus		0.4822%
12	Mycothermus thermophilus		0.4372%
13	Scedosporium dehoogii		0.3132%
14	Pichia manshurica		0.2974%
15	Aspergillus flavus	_	0.2051%
16	Aspergillus piperis	_	0.1960%
17	Chrysosporium lobatum		0.1915%
18	Stachybotrys chartarum		0.1758%
19	Penicillium parvum	_	0.1713%

#	Genus & Species	Percentage
20	Sodiomyces alcalophilus	0.1667%
21	Talaromyces minioluteus	0.1577%
22	Cephaliophora sp.	0.1487%
23	Alternaria atra	0.1329%
24	Mortierella ambigua	0.1239%
25	Aphanoascus mephitalis	0.1127%
26	Stemphylium vesicarium	0.1059%
27	Talaromyces purpureogenus	0.0924%
28	Schwanniomyces occidentalis	0.0879%
29	Preussia sp.	0.0834%
30	Solicoccozyma aeria	0.0631%
31	Penicillium simplicissimum	0.0608%
32	Rhodotorula toruloides	0.0563%
33	Torula sp.	0.0496%
34	Chrysosporium sp.	0.0473%
35	Mucor circinelloides	0.0473%
36	Pichia fermentans	0.0473%





RELATIVE FUNGAL ABUNDANCE

The relative abundance of the fungal communities is reported in %. Data in this report correspond to the average relative abundance obtained from three technical replicates. The absolute quantification of fungal communities using amplicon sequencing has not yet been validated at Biome Makers, Inc.

#	Genus & Species	Percentage
37	Pyrenochaetopsis leptospora	0.0473%
38	Scutellinia torrentis	0.0473%
39	Candida stellimalicola	0.0428%
40	Lophotrichus fimeti	0.0428%
41	Curvularia buchloes	0.0406%
42	Acremonium sp.	0.0315%
43	Aspergillus neoniveus	0.0293%
44	Lectera colletotrichoides	0.0293%
45	Lomentospora prolificans	0.0293%
46	Acremonium fusidioides	0.0270%
47	Fusarium sporotrichioides	0.0248%
48	Amandinea punctata	0.0203%
49	Apiotrichum mycotoxinovorans	0.0203%
50	Cladosporium macrocarpum	0.0203%
51	Emmonsia crescens	0.0158%
52	Talaromyces rugulosus	0.0113%
53	Zopfiella sp.	0.0090%
54	Debaryomyces nepalensis	0.0068%
55	Aspergillus sydowii	0.0045%

#	Genus & Species	Percentage
56	Candida parapsilosis	0.0045%
57	Clavispora fructus	0.0045%
58	Cystobasidium slooffiae	0.0045%
59	Kazachstania humilis	0.0045%
60	Mortierella clonocystis	0.0045%
61	Trichosporon asahii	0.0045%
62	Acremonium rutilum	0.0023%
63	Aspergillus niger	0.0023%
64	Aspergillus wentii	0.0023%
65	Cirrenalia macrocephala	0.0023%
66	Coniochaeta cateniformis	0.0023%
67	Paecilomyces sp.	0.0023%
68	Penicillium madriti	0.0023%
69	Sparassis sp.	0.0023%
70	Starmerella apicola	0.0023%

COPYRIGHT ® 2020 BIOME MAKERS INC. ALL RIGHTS RESERVED.

For research purposes only. Reference herein to any specific commercial product, process, or service by its trade name, trademark, manufacturer, or otherwise, does not necessarily constitute or imply its endorsement, recommendation, or favoring by the company. Results relate only to the sample as received and information provided by the client. Biome Makers accepts no responsibility or liability for the origin, traceability or quality of samples prior to receipt by the laboratory, either of which may compromise the quality or accuracy of the results generated. No representation or warranty is given by Biome Makers, or any member or employee as to the accuracy of any test method or test results. Results are an interpretation of the potential function or potential impact of tested sample in the soil based on the characterization of the sample microbiome. Likewise, the information provided should not be considered as indicative of any future results.

You must not rely on the information in the report as an alternative to agronomic advice from an appropriately qualified professional. Always consult with your trusted agronomist before making any application of information, processing, application of products or any other actions, without limitation, that may be applied after receiving this information. Any action you take upon the information on this report is strictly at your own risk, Biome Makers will not be liable for the use of guidance in the report in respect of any business losses, including without limitation loss of or damage to profits, income, revenue, use, production, anticipated savings, business, contracts, commercial opportunities or goodwill. No part of this report may be reproduced or use for commercial or public use without permission in writing from Biome Makers.



Any further enquiry concerning this report will be attended at

+1 (415) 795 7469 info@biomemakers.com

BIOMEMAKERS

Microbiome Analysis Report



SUMMARY



CONCLUSIONS

- You have a Very low biodiversity value. Aggressive management can be affecting your soil biosustainability.
- Carbon nutrition value is Low.
- The Nitrogen and Potassium nutrition values are low.

BIOSUSTAINABILITY



Richness, evenness and equilibrium of microbial species



Capability of soil microbial communities to perform multiple functions



Ability of communities or populations to remain unchanged when stressed by disturbance



HEALTH

HEALTHINESS

Medium

Disease
Risks found



Crop health according to the pathogens detected

SLIGHT RISK DETECTED



VERTICILLIUM WILT



MEDIUM Risk level

BLACK MOLD ROT

NOT DETECTED

ALTERNARIA STEM CANKER • ANTHRACNOSE • BACTERIAL CANKER • BACTERIAL LEAF BLIGHT • BACTERIAL SOFT ROT • BACTERIAL SOUR ROT • BACTERIAL SPECK • BACTERIAL SPOT • BACTERIAL STEM ROT • BLACK MOLD • BLACK ROOT ROT • BUCKEYE ROT • CHARCOAL ROT • CORKY ROOT ROT • FUSARIUM CROWN AND ROOT ROT • FUSARIUM WILT • GRAY LEAF SPOT • GRAY MOLD • GRAY MOLD ROT (BOTRYTIS FRUIT ROT) • PITH NECROSIS • PYTHIUM DAMPING-OFF AND STEM ROT • PYTHIUM FRUIT ROT • ROOT MAT • SEPTORIA LEAF SPOT • SOUR ROT • SYRINGAE BLIGHT AND LEAF SPOT • TARGET SPOT • WHITE MOLD







Low

Microbial species grouped according to the type of pest they encounter, capable of preventing pathogenic species from taking hold or proliferation

Fungicide agents

LOW

Bactericide agents

NOT DETECTED

Nematicide agents

LOW

MEDIUM

HORMONE PRODUCTION

⊣igh

Microbial species grouped according to the type of phytohormone they generate



STRESS ADAPTATION

Very High

Microbial species grouped according to their relationship with the metabolisms linked to the capability to withstand stress conditions

Exopolysaccharide production			ACC deaminase	e (ACC-d)	
NUTRIENT TRAP	SALINITY PROTECT.	DROUGHT PROTECT.	PATHOGEN PROTECT.	SALINITY PROTECT.	DROUGHT PROTECT.
- HIGH			VERY HIGH		
Heavy metal s	solubilization		Salicylic acid (SA	4)	
BIOREMEDIATION	DETOXIFICATION	ALLEVIATE HEAVY METAL STRESS	DROUGHT PROTECT.	SALINITY PROTECT.	ALLEVIATE HEAVY METAL STRE
- HIGH			VERY HIGH		
Salt tolerance	:		Abscisic acid (A	ВА)	
SALINITY PROTECT.	ROOT GROWTH PRO	MOTION	GROWTH REGULATION	PLANT RESISTANCE	INCREASE YIELDS
VERY HIGH			VERY HIGH		
Siderophore p	roduction				
IRON AVAILABILITY	BIOFERTILIZER				





NUTRITION

Nutritional status based on the microbial mobilization of certain compounds

MAJOR COMPOUNDS



Nitrogen LOW

CARBON PATHWAYS











PHOSPHORUS PATHWAYS



POTASSIUM PATHWAYS

NUTRIENT SUPPLY	NUTRIENT COMPETITION
Potassium solubilization	Potassium consumption
MEDIUM	VERY HIGH

MINOR COMPOUNDS



















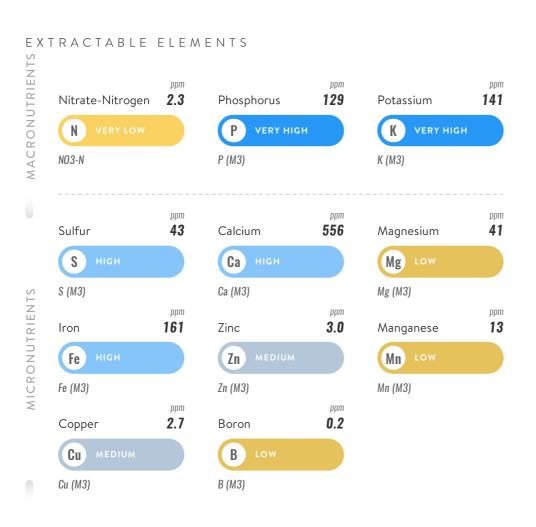


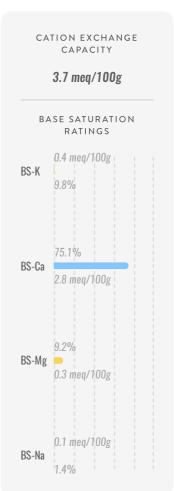
CHEMICAL FERTILITY RATINGS

GENERAL INSIGHTS









DETRIMENTAL ELEMENTS

Sodium 12
Na VERY LOW
Na (M3)



Item 6.a-32

COPYRIGHT ® 2020 BIOME MAKERS INC. ALL RIGHTS RESERVED.

For research purposes only, the information is not advice, and should not be treated as such. Results relate only to the sample as received and information provided by the client. Biome Makers accepts no responsibility or liability for the origin, traceability or quality of samples prior to receipt by the laboratory, either of which may compromise the quality or accuracy of the results generated. No representation or warranty is given by Biome Makers, or any member or employee as to the accuracy of any test method or test results.

You must not rely on the information in the report as an alternative to agronomic advice from an appropriately qualified professional. Any action you take upon the information on this report is strictly at your own risk, Biome Makers will not be liable for the use of guidance in the report in respect of any business losses, including without limitation loss of or damage to profits, income, revenue, use, production, anticipated savings, business, contracts, commercial opportunities or goodwill. No part of this report may be reproduced or use for commercial or public use without permission in writing from Biome Makers.



Any further enquiry concerning this report will be attended at

+1 (415) 795 7469 info@biomemakers.com

BIOMEMAKERS

The Great Crops line of products has been carefully manufactured after over a decade of field and lab research, period in which only the highest quality ingredients have been chosen.

The Great Crops line was developed to facilitate the work load of the farmer while making his Crops Great.

More than a program, the Great Crops line consists of eight products where each one has been specially designed to supply a balanced concentration of amino acid chelated and complexed nutrients, soil microflora and microflora food source, and organic key compounds, to trigger and support specific demands during the five most important physiological stages of any crop. In chronological order, these stages are:

- Soil and plant stimulator for a strong initial development,
 GREAT SOLUM™
- 2. Boosting root development to maximize a strong crop foundation, GREAT RADIX™
- 3. Balanced plant vegetative growth to support a strong load, GREAT FORTI ™
- 4. Nutrients and compounds to promote flowering and polinization, GREAT FLOS™
- 5. Enhancing high quality and yielding harvesting material, GREAT MAXIMUS™

The other three products that complete the Great Crops line are materials that have been manufactured with the intention to support the five key stages mentioned above when soil and environmental conditions make their use critical; these conditions are:

6. Sodic & calcareous soils, high Mn & B, and high heat and drought, GREAT MOTUS™

Although the best possible results are achieved when the Great Crops products are applied during their respective stages, the Great Crops line will also work when used individually.

Each product is described individually and in order of application in the next slides.



Great Solum

1.0 - 0.6 - 0.4

Soil & Plant Stimulator

Great Solum is a unique product designed to be "6 products in one":

- 1. Soil Conditioner,
- 2. Food source for microbes,
- 3. Provides key nutrients to the soil and plant for initial stages of crop establishment,
- 4. Enzyme promoter for utilization of carbohydrates,
- 5. Promote beneficial soil microbiology,
- 6. Starter fertilizer enriched with nitrogen, calcium, zinc and copper

Great Solum is a soil and root stimulator, improving soil structure, water infiltration and holding capacity, nutrient cycling and availability, and improve soil conditions of alkaline and saline soils.

All of the above is achieved by the key ingredients and ratio of nutrients to stimulate both, a balanced soil environment, and plant nutrient availability.

Contains among others:

- · Enzyme precursors and stimulants
- Microbial metabolites
- Natural plant growth regulators
- Carbohydrates
- Organic acids
- Organic Carbon
- · Humic and fulvic acids
- Saponins
- All essential micronutrients plus Nickel
- Low Salt Index (8)
- Positive redox potential
- pH of 2.9

Application rate of 4 to 7 gal/ac via irrigation before or during crop establishment or during initial stages of development of permanent crops.





Root Development

Great Radix is a unique product specifically designed to enhance initial root development and root flushes. Its formulation provides all essential nutrients with high available P in the form of orthophosphate; all of its micronutrients are 100% chelated to alleviate most deficiencies.

Great Radix has an important amount of readily available Magnesium and Iron which is key for initial stages of photosynthesis.

All of the complete range of micronutrients are chelated with natural organic acids, amino acids and carbohydrates that are readily biodegradable supplying energy to the plant and food source for the soil microflora.

Enzyme precursors in this blend enhance utilization and translocation of nutrients by buffering, chelating and complexing natural soil fertility keeping them in solution.

Great Radix is a very effective water buffering agent and it contains many key ingredients that aim in soil regenerative properties; among them are:

- Natural plant growth regulators
- Carbohydrates
- Organic acids
- Organic Carbon
- · Humic and fulvic acids
- Saponins
- · All essential micronutrients plus Cobalt
- Low Salt Index (8)
- Positive redox potential
- pH of 3.4

Application rate of 4 to 7 gal/ac via irrigation before or during root development and/or root flushes.



Great Forti

1.0 - 1.6 - 0.9

Vegetative Growth

Great Forti is a unique product specifically designed to promote balanced vegetative growth which will support flowers and fruit loads by increasing photosynthetic rates through a complete supply of nutrients with a great NPK ratio for this stage of growth. It contains P as orthophosphate together with Calcium in true solution. Most deficiencies in this stage translates into seed or fruit physiological disorders that lead into fruit, seed and or plant diseases.

Great Forti highly available Potassium is key in the activation of more than 40 enzymatic reactions, crucial for plant metabolism. Among these reactions are the opening and closing of stomata, water relations within the plant, and membrane integrity.

Nitrogen is in its most available form, Nitrate, to promote its uptake and translocation so it can efficiently be used in this critical growth stage for building complex metabolites such as proteins, nucleic acids and porphyrins.

Great Forti contains complexed micronutrients and Magnesium using natural organic and amino acids.

It is also an enzyme precursor that enhances the utilization and translocation of nutrients by buffering, chelating and complexing natural soil fertility and keeping them in solution.

Great Forti is a very effective watering buffering agent containing key ingredients necessary to achieve a very effective plant response while replenishing organic materials to the soil. among these ingredients are:

- Natural plant growth regulators, carbohydrates, saponins
- Organic acids and high organic carbon to support high rates of plant growth
- · Humic and fulvic acids
- All essential micronutrients
- Low Salt Index (8)
- Positive redox potential
- pH of 3.3
- Fish protein & fremented grain

Application rate of 4 to 7 gal/ac via irrigation during high vegetative growth.



1.5 - 2.4 - 0.9

Budding & Flowering

Great Flos is a unique product specifically designed to promote and support the flowering stage providing all essential nutrients at specific ratios during this high nutritional demand period.

With the high P demand in this stage, it is very important to provide the plant/tree with highly available P, formulation that Great Flos provides in the form of polyorthophosphate.

Great Flos provides most of essential micronutrients but also it provides Zinc and Copper in their ionic form for fast absorption in utilization providing these and other nutrients in their proper ratio to support this key stage while increasing the activity rate of the crop. Its high humic and fulvic acids concentration plus the high organic carbon content, makes Great Flos a unique alternative to decrease negative responses due to abiotic stressors in this stage.

Great Flos highly available Zinc is key in the activation of more than 70 enzymatic reactions, with one of them being related to the activation of an enzyme that participates in the evapotranspiration and gas exchange process between the atmosphere and the leaves of the plant. It is also an enzyme precursor that enhances the utilization and translocation of nutrients by buffering, chelating and complexing natural soil fertility and keeping them in solution.

Great Flos is an effective water buffering agent containing besides other key ingredients necessary to achieve a very effective plant response while replenishing organic materials to the soil; among these ingredients are:

- Natural plant growth regulators & carbohydrates
- Organic acids and high organic carbon & saponins
- Very high concentration of Humic and fulvic acids
- All essential micronutrients plus Molybdenum
- Low Salt Index (9)
- Positive redox potential
- pH of 3.6

Application rate of 4 to 7 gal/ac via irrigation and/or 1 to 2 gal/ac as a foliar spray before flowering stage.



Great Maximus

0.9 - 1.2 - 1.3

Yield & Quality

Great Maximus is a unique product developed to provide proper NPK and micronutrient ratios, as well as a stress releaser during the later stages of crop and during fruit-seed-tuber development.

Great Maximus has low N with high P and K plus a complete range of micronutrients with Cobalt and Molybdenum.

One of its key ingredients is the brown algae Ascophyllum nodosum which provides more resistance under abiotic stressors plus enhancing balanced sizing, growth and weight of the final harvested product.

Calcium and Boron in their chelated form are also in the proper ratio to support cell division and expansion.

All of the above factors combined give the result of a high yielding and prime quality final product.

Another advantage of Great Maximus is enhancing root growth to support later stages of plant develoment.

Other Great Maximus ingredients include:

- Enzyme precursors and stimulants
- Natural plant growth regulators & carbohydrates
- Organic acids and high organic carbon to support fruit growth
- High humic and fulvic acids
- Saponins
- Low Salt Index (9)
- Positive redox potential
- pH of 3.5

Application rate of 4 to 7 gal/ac via irrigation and/or 1 to 2 gal/ac via foliar spray during fruit-seed-tuber development.



Great Motus 0.2 - 0.0 - 0.1

Stress Relief

Great Motus is a unique product that has been designed to reinforce the crop performance under stressful soil and environmental conditions such as calcareous and sodic soils, high heat, drought, etc.

Its readily available Silicon provides many benefits to soils and crops:

- 1. Reduced water loss by cuticular transpiration
- 2. Improvement of SAP circulation
- 3. Greater resistance to fungal attacks
- 4. Increase plant tolerance to high levels of Boron and Manganese
- 5. Increase resistance to lodging and pests
 - 6. Structure rigidity and fruit storage life
 - 7. Increase availability of soil P
 - 8. Aids in proper N concentration in plants under deficient N soil conditions

Great Motus contains sea kelp.

It is a great addition to any nutritional program specially when crops are grown under the stressful conditions mentioned above. THIS IS A STAND ALONE PRODUCT!

Other Great Motus ingredients include:

- Mircoalgae & Isolate Strains
- Enzyme precursors and stimulants
- · Natural plant growth regulators
- Carbohydrates
- Organic acids
- · Humic and fulvic acids
- Lowest Salt Index (2)
- Positive redox potential
- pH of 11.0

Application rate of 3 to 7 gal/ac via irrigation when crops are grown under extreme soil and weather conditions.





GREAT FLOS

1.5 - 2.4 - 0.9

Great Flos is marketed as a unique product whose nutrients are derived from food waste compost and earth worm castings.

Great Flos is formulated to provide the growers with a high quality material suitable for use on a variety of cops.

Great Flos is recommended to promote and support the high nutrient needs for crops previous or during flowering stage in soils for conventional agriculture.

DIRECTIONS FOR USE

Application rates: 5 to 7 gallons per acre
For Soil: Apply via irrigation to soil one to three weeks
before flowering or at flower bud break

GUARANTEED ANALYSIS

Total Nitroge (N)	1.5%
0.9% Ammoniacal Nitrogen	William Control
0.5% Other Water Soluble Nitrogen	
0.1% Water Insoluble Nitrogen	AL PROPERTY.
Available Phosphate (P2O5)	2.4%
Soluble Potash (K2O)	0.9%

Derived from: earth worm castings and food waste compost

Guaranteed by: GREAT CROPS 575 Saint Mary Avenue Cayucos, CA 93430



Net contents:



GREAT FORTI

1.0 - 1.6 - 0.8

Great Fortis is marketed as a unique product whose nutrients are derived from food waste compost and earth worm castings.

Great Fortis is formulated to provide the growers with a high quality material suitable for use on a variety of cops.

Great Fortis is recommended to support growth during the stage of vegetative development for conventional agriculture.

DIRECTIONS FOR USE

Application rates: 5 to 7 gallons per acre For Soil: Apply via irrigation to soil to support the vegetative growth stage

GUARANTEED ANALYSIS

Total Nitrogen (N)	1.0%
0.3% Ammoniacal Nitrogen	
0.6% Other Water Soluble Nitrogen	
0.1% Water Insoluble Nitrogen	
Available Phosphate (P2O5)	1.6%
Soluble Potash (K2O)	0.8%

Derived from: earth worm castings and food waste compost

Guaranteed by: GREAT CROPS 575 Saint Mary Avenue Cayucos, CA 93430



Net contents:



GREAT MAXIMUS

0.9 - 1.2 - 1.3

Great Maximus is marketed as a unique product whose nutrients are derived from food waste compost and earth worm castings.

Great Maximus is formulated to provide the growers with a high quality material suitable for use on a variety of cops.

Great Maximus is recommended to promote fruit/nut/ tuber growth and sizing during this key stage in soils for conventional agriculture.

DIRECTIONS FOR USE

Application rates: 5 to 7 gallons per acre For Soil: Apply via irrigation to soil during fruit/nut/tuber development

GUARANTEED ANALYSIS

Total Nitrogen (N)	0.9%
0.1% Ammoniacal Nitrogen	
0.6% Other Water Soluble Nitrogen	
0.2% Water Insoluble Nitrogen	
Available Phosphate (P2O5)	1.2%
Soluble Potash (K2O)	1.3%

Derived from: earth worm castings and food waste compost

Guaranteed by: GREAT CROPS 575 Saint Mary Avenue Cayucos, CA 93430



Net contents:



GREAT MOTUS

0.2 - 0 - 0.07

Great Motus is marketed as a unique product whose nutrients are derived from earth worm castings.

Great Motus is formulated to provide the growers with a high quality material suitable for use on a variety of cops.

The use of Great Motus is recommended when crops grown during hot summer days in soil with low organic matter content in soils for conventional agriculture.

DIRECTIONS FOR USE

Application rates: 5 to 7 gallons per acre
For Soil: Apply via irrigation to soil before and/or during
extreme weather events such as heat, drought, wind, freeze.

GUARANTEED ANALYSIS

Total Nitrogen (N)	0.2%
0.1% Water Soluble Nitrogen	*
0.1% Water Insoluble Nitrogen	
Soluble Potash (K2O)	0.07%

Derived from: earth worm castings

Guaranteed by: GREAT CROPS 575 Saint Mary Avenue Cayucos, CA 93430



Net contents:



GREAT RADIX

1.2 - 1.0 - 0.8

Great Radix is marketed as a unique product whose nutrients are derived from food waste compost and earth worm castings.

Great Radix is formulated to provide the growers with a high quality material suitable for use on a variety of cops.

Great Radix is recommended to promote root mass during root development in soils for conventional agriculture.

DIRECTIONS FOR USE

Application rates: 5 to 7 gallons per acre
For Soil: Apply via irrigation to soil during root development
and/or root flushes

GUARANTEED ANALYSIS

Total Nitrogen (N)	1.2%
0.6% Ammoniacal Nitrogen	The state of the s
0.5% Other Water Soluble Nitrogen	
0.1% Water Insoluble Nitrogen	
Available Phosphate (P2O5)	1.0%
Soluble Potash (K2O)	0.8%

Derived from: earth worm castings and food waste compost

Guaranteed by: GREAT CROPS 575 Saint Mary Avenue Cayucos, CA 93430



Net contents:



GREAT SOLUM

0 - 0.6 - 0.4

Great Solum is marketed as a unique product whose nutrients are derived from food waste compost and earth worm castings.

Great Solum is formulated to provide the growers with a high quality material suitable for use on a variety of cops.

Great Solum is recommended to enhance a good soil environment that acts as a soil conditioner and to promote and sustain microbial activity in soils for conventional agriculture.

DIRECTIONS FOR USE

Application rates: 5 to 7 gallons per acre For Soil: Apply via irrigation to soil between 1 to 2 weeks before planting or when soil temperatures supports root activity for permanent crops.

GUARANTEED ANALYSIS

Total Nitrogen (N)	1.0%
0.3% Ammoniacal Nitrogen	
0.3% Other Water Soluble Nitrogen	
0.4% Water Insoluble Nitrogen	
Available Phosphate (P2O5)	0.6%
Soluble Potash (K2O)	0.4%

Derived from: earth worm castings and food waste compost

Guaranteed by: GREAT CROPS 575 Saint Mary Avenue Cayucos, CA 93430



Net contents:



GREAT FLOS

1.5 - 2.4 - 0.9

Great Flos is marketed as a unique product whose nutrients are derived from food waste compost and earth worm castings.

Great Flos is formulated to provide the growers with a high quality material suitable for use on a variety of crops.

Great Flos is recommended to promote and support the high nutrient needs for crops previous or during flowering stage in soils for conventional agriculture.

DIRECTIONS FOR USE

Application rates: 5 to 7 gallons per acre For Soil: Apply via irrigation to soil one to three weeks before flowering or at flower bud break

Derived from: earth worm castings and food waste compost

GUARANTEED ANALYSIS

Total Nitroge (N)	1.5%
0.9% Ammoniacal Nitrogen	
0.5% Other Water Soluble Nitrogen	
0.1% Water Insoluble Nitrogen	
Available Phosphate (P2O5)	2.4%
Soluble Potash (K2O)	0.9%

Guaranteed by: GREAT CROPS 575 Saint Mary Avenue Cayucos, CA 93430



Net contents:

Development of Chloride Mitigation Strategies for Californian Avocado Groves: Technology Review and Treatment Prediction

Project Director:

Haizhou Liu, Ph.D. Chief Technology Officer Water Illumination Inc. 240 Desert Bloom Irvine, CA 92618

Tel: (206) 214-7162

E-mail: haizhou@waterillumination.com

Table of Content

1.	Executive Summary	1
2.	Background and Introduction	2
3.	Overall Project Objectives and Tasks	3
4.	Technical Approach	4
5.	Research Work Plan and Schedule	7
6.	Qualification of the Research Team	7
7.	Project Budget	9
R	Literature Cited	10

1. Executive Summary

The goal of this research project is to conduct a phase-one feasibility study to develop chloride mitigation technologies from irrigation water at Californian avocado groves. The elevated level of chloride in irrigation water is one of the greatest threats to avocado productivity in California. The development of efficient, cost-effective on-site water desalination technologies to selectively remove chloride from the irrigation water for Californian avocado groves will significantly increase the yield of avocado, provide reliably high-quality irrigation water, and consequently increase the profits and competitiveness of Californian avocado groves. The proposed project has *four major tasks*: (1) understand chloride ion activity and chemistry in irrigation water at Californian groves by conducting a comprehensive chemical modeling; (2) screen viable chloride removal technologies uniquely applicable to avocado industry by conducting an extensive literature review; (3) predict the treatment efficiency and economic cost of the most prioritized chloride removal technologies; and (4) map out the next-phase experimental investigation of candidate technologies.

The *outcome* of the phase-one project will generate a white paper that critically reviews chloride mitigation options, identify top candidate technologies, reach recommendations for the next phase study, and potentially form an interdisciplinary team for future phases of the study. The project team will actively collaborate with California Avocado Commission and stakeholders. This proposal requests \$100,000 beginning November 1, 2021 over 12 months. Funds will be used primarily for the costs of research personnel and research activities. The project will benefit Californian avocado industry by addressing the most challenging issues on chloride management, water security and fruit yield.

2. Background and Introduction

The **overarching objective** of this project is to conduct a phase-one feasibility study to develop chloride mitigation technologies from irrigation water at Californian avocado groves. The elevated level of chloride in irrigation water is one of the greatest threats to avocado productivity for many growers in California. The development of efficient, cost-effective on-site water desalination technologies to selectively remove chloride from the irrigation water for Californian avocado groves will significantly increase the yield of avocado trees, provide reliably high-quality irrigation water, and consequently increase the profits and competitiveness of Californian avocado groves.

Chloride toxicity to avocado. Avocado is one of the most salinity sensitive horticultural crops, but is commonly grown in areas having saline irrigation water, especially with a chloride level higher than 100 mg/L. Problems associated with high soil salinity and chloride toxicity are well documented, including a significant reductions in fruit yield and tree size, lowered leaf chlorophyll content, decreased photosynthesis, poor root growth, and leaf scorching.¹ In recent years, salinity problems with avocado groves have become increasingly common as the cost for irrigation water has gone up and the availability of low salinity water for agriculture has diminished. The project team's recent conversation with California Avocado Commission (CAC) has learned that 100 mg/L of chloride in the irrigation water can result in more than 50% yield reduction from avocado trees. This has resulted in requests for information on what mitigation strategies are available to remove chloride from the irrigation water so that growers can better address salinity problems in source water and increase the long-term fruit yield.

Elevated chloride in irrigation water. In California, the high level of chloride in the irrigation water poses constant challenges to the avocado industry. As climate impact, population growth and water scarcity severely limit the access to high-quality fresh water, Californian groves are facing the challenges of dealing with saline irrigation water that is the only option to them, including saline groundwater or surface water diverted from elsewhere by public utilities, local reclaimed water and other impaired water resources. As a result, chloride accumulates in the soil pores after regular irrigation. In addition, rainfall and irrigation events creates strong capillary forces around the avocado root zone that transport chloride from the soil to the trees, thus inducing chloride toxicity constantly.

Lack of chloride water treatment tailored for avocado industry. Current salinity management strategies for the avocado industry largely focus on the manipulation of plant physiology and biology (e.g., creating more salt-tolerant rootstocks) or the optimization of irrigation practice (e.g., changing irrigation scheduling and flushing practice). Although these strategies can be effective, they do not directly tackle the problem of chloride removal at its source and often require very labor-intensive management skills. Meanwhile, existing mature desalination technologies are mostly designed for extremely high-salinity water at a centralized large-scale treatment facility (e.g., seawater or brackish water desalination). These existing desalination technologies

depends on the use of high pressure-driven reverse osmosis (RO) membranes, and requires an easy access to brine disposal, either along the coast or into deep underground wells. Unfortunately, these disposal options are not available to Californian groves. Furthermore, the membrane-based technologies are non-selective in removing salinity, and thus also removing other beneficial ions from the irrigation water in addition to chloride. This would be undesirable for application in the avocado industry. Until now, there lacks an efficient water treatment technology that can best suits for onsite chloride removal from the irrigation water.

Significance to California Avocado Commission. Considering the urgency, relevance, importance and promise of chloride removal from irrigation water, the development of efficient water treatment technologies to selectively remove chloride can become a game-changer for the Californian avocado industry to increase its profit and enhance its global competitiveness. The proposed research will be the first of its kind to directly address chloride issue at the source water, and take the first step aiming to develop a cost-effective and efficient chloride removal technology. Specifically, it will achieve a detailed understanding of chloride chemistry in irrigation water, a comprehensive screening of available desalination technologies, and a roadmap recommendation to prioritize next-step technology development and collaboration, with an ultimate goal to benefit the California avocado industry. This transformative research topic is timely. The topic of engineering chloride mitigation strategy is an important area that has not been traditionally supported by the California Avocado Commission (CAC), and therefore possessing a strategic significance to the industry and its stakeholders.

3. Overall Project Objectives and Tasks

The research plan was developed to conduct a phase-one feasibility study on the development of chloride mitigation technologies from irrigation water at Californian avocado groves. This 12-month research project includes four major project activities:

- 1. Understand chloride ion activity and chemistry in irrigation water at Californian groves (Months 1-4). We will conduct a comprehensive chemical modeling simulation to understand the interactions between chloride ion and other chemical constituents in irrigation water. The chemical modeling results will provide a solid theoretical framework for a tailored design of chloride removal technology.
- 2. Screen viable chloride removal technologies uniquely applicable to avocado industry (Months 3-8). We will conduct an extensive literature review to evaluate the applicability of both existing and emerging water desalination technologies that can selectively remove chloride for onsite water treatment.
- 3. Predict the treatment efficiency and economic cost of the most prioritized chloride removal technologies (Months 6-12). We will identify the top candidate technologies and predict their treatment efficiencies at the lab scale and full scale onsite. In addition, we will conduct a cost analysis to compare the top prioritized technologies.

4. Recommend a phase-two study on experimental investigation of candidate technologies (Months 10-12). We will reach recommendations on selective technologies for the next phase lab-scale validation study, and potentially form an interdisciplinary team for future comprehensive development of chloride mitigation strategies.

4. Technical Approach

Four research tasks address each of the four objectives, respectively. The project team brings unique strengths to the phase-one study: (1) A combination of more than 20 years' experience in water engineering for agricultural applications and desalination technologies; (2) A team of wellrecognized water engineers and practicing chemists; (3) A partnership to address chloride and salinity issues with California Avocado Commission and its members. Figure 1 shows the schematics of the research tools and core components. Specifically, the experimental investigation will utilize both cutting-edge chemical modeling techniques and critical literature review processes to examine the unique chloride chemistry in irrigation water and evaluate the best water treatment technologies for chloride removal.

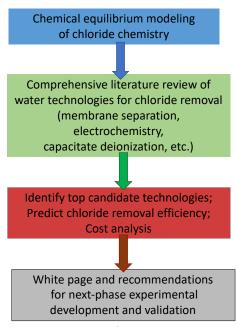


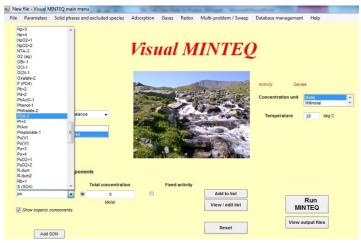
Figure 1 Research core components.

Objective 1: Understand chloride ion activity and chemistry in irrigation water at Californian groves.

Task 1: Conduct a comprehensive chemical modeling simulation to understand the interactions of chloride ion with other chemical constituents in irrigation water. In order to develop the best possible water treatment technologies to removal chloride from the irrigation water, it is important to first understand the forms of chloride ions existing in irrigation water. Saline irrigation water has a very complex water chemistry and many chemical components in addition to chloride, including sodium, sulfate, bicarbonate (also known as alkalinity), protons and hydroxides (expressed as water pH), calcium and magnesium (contributing to water hardness), residual nutrients (including nitrate and phosphate) iron and manganese (especially for groundwater sources), dissolved organic carbon (soil humic substances), and chlorine/chloramines (residual disinfectants in treated potable water). For example, chloride can form solid in the presence of calcium and phosphate. Chloride can also be oxidized into chlorine under a desirable redox condition. These chemical components can interact with chloride, generate secondary derivatized chloride-containing molecules, and affect the chemical reactivity of chloride, thus affecting the choice of most desirable technology to remove it.²

We will utilize a fundamental chemical equilibrium modeling approach to understand the speciation and reactivity of chloride ion in the complex chemistry of irrigation water. The

chemical modeling results will provide solid theoretical framework for a tailored design of chloride removal technology. The modeling prediction conducted using the Visual Minteg software, which will take account into a variety of chemical equilibrium and complexation reaction thermodynamics data (Figure 2). The chemical components and their concentrations will act as input



parameters of the chemical model. Figure 2 The interface of the chemical modeling software.

The detailed chemistry of the irrigation water will be obtained from water quality reports available from specific avocado groves. In cases the water quality report is not available, the project team can collect irrigation water samples for chemical analysis.

Objective 2: Screen viable chloride removal technologies uniquely applicable to avocado industry.

<u>Task 2: Conduct an extensive literature review on selective chloride removal technologies from irrigation water.</u> In this task, we will conduct a critical literature review to evaluate water existing and emerging water treatment technologies that are applicable to on-site irrigation water treatment at avocado groves. The screening of candidate technologies will utilize the chloride chemistry modeling data obtained from Task 1, and identify technologies that can be a good fit to specific irrigation water at different sites. The initial groups of candidate technologies are listed in **Table 1** on Page 6, and the list will be expanded as the literature review progresses.

Specifically, traditional RO membrane technologies are unlikely to best suit for the irrigation water treatment, due to the generation of RO brine that is challenging to dispose of and the energy-intensive operation. We will review RO brine treatment options onsite that can reduce the volume of the brine. In contrast, nanofiltration (NF) can be a potential physical separation technology. NF requires a much lower energy input and pressure, but can still achieve a significant percentage of chloride removal. There are different NF membrane materials that are selective towards different ions. The best NF membranes will be evaluated based on available literature.

In addition, electrochemical technologies will be evaluated, including electrodialysis and electrolysis. In both cases, the saline irrigation water will subject to the application of different electric voltage using submerged electrodes. During electrodialysis, chloride ions will migrate towards the anode via ion exchange membranes, thus separating it from the water. During electrolysis, a higher voltage will be applied at the electrodes, and chloride will participate in redox reaction and potentially transform into chlorine gas, which

can be then stripped out from the irrigation water. However, these electrochemical technologies have not been critically reviewed for feed water with a chloride level in the 100-200 mg/L range, and this task will look closely into the technology applicability.

Furthermore, several emerging technologies will be evaluated, including capacitive deionization with carbon electrodes, which can be more energy saving and chloride selective by choosing the desirable electrodes; chemical precipitation method, which will take advantage of chloride-containing solid precipitation by harnessing the irrigation water chemical matrix; solvent-based low-gradient thermal extraction method, which is based on the affinity of chloride with specific organic solvents that will vary depending on temperature and thermal cycles. The outcome of this task is to screen all candidate water technologies for chloride removal, and develop viable options for avocado industry.

Table 1 A list of candidate chloride removal technologies from irrigation water for a comprehensive literature review.

Chloride removal Technology	Treatment Category	Focus of literature review
Reverse osmosis (RO)	Membrane separation	Propose brine treatment technologies to reduce the volume of RO brine waste.
Nanofiltration (NF)	Membrane separation	Identify candidate NF membranes that are selective in removing chloride, while requiring a low pressure and maintaining a high flow rate.
Electrodialysis	Electrochemical separation	Evaluate the choice of electrode materials and the ion selective membranes for irrigation water application.
Electrolysis	Electrochemical oxidation	Evaluate the possibility of oxidizing chloride to free chlorine gas and subsequent removal of the gas product.
Capacitive deionization	Electrochemical adsorption	Review the choices of carbon electrodes and the process design for selective chloride removal and avoiding organic fouling on the electrode.
Ultraviolet- based advanced oxidation	Photochemical degradation	Evaluate the removal of chloride via energy-saving UV LED light-based degradation process.
Solid formation	Chemical precipitation	Review the feasibility of removing chloride via precipitating it out as calcium- or other metal-based minerals.
Solvent-based selective removal	Thermal extraction	Review the feasibility of using organic solvents for low-gradient thermal extraction of chloride from water.

Objective 3: Identify the top candidate chloride removal technologies

Task 3: Predict the treatment efficiency and economic cost of the most prioritized chloride removal technologies. Based on the candidate list generated from Task 2, we will identify the top candidate technologies, predict their treatment efficiencies, and conduct a cost analysis. The ranking of the top candidate technologies will consider the flowing factors: predicted treatment efficiency; cost estimate; treatment capacity; risks for secondary waste water generation; and the easiness to operate and maintain. Data will be obtained from a comprehensive literature review.

Objective 4: Write a white page and recommend the phase-two study

Task 4: Generate a white page summary and a future roadmap on experimental investigation of candidate technologies (Months 10-12). This task will integrate findings from the previous tasks. We will reach recommendations on selective technologies for the next phase lab-scale validation study, and potentially form an interdisciplinary team with other complimentary expertise (*e.g.*, plant physiology, soil physics) for future comprehensive development of chloride mitigation strategies.

5. Research Work Plan and Schedule

The research work plan of individual tasks and significant milestones is developed as below.

Table 2 Proposed research work plan and schedule.

Individual Task	Timeline	Significant Milestone
Task 1	Months 1-4	Understand the interactions of chloride ion with other chemical constituents in irrigation water.
Task 2	Months 3-8	Critically compare both existing and emerging water desalination technologies that can selectively remove chloride for onsite water treatment.
Task 3	Months 6-12	Identify the top candidate technologies and predict their treatment efficiencies at the lab scale and full scale onsite.
Task 4	Months 10-12	Optimize the treatment efficiency towards different scale-forming components.
Final white paper	Month 12	Summarize research findings and reach recommendations on selective technologies for the next phase lab-scale validation study.

6. Qualification of the Research Team

The research team has adequate qualifications, capabilities, and experience. Dr. Haizhou Liu, co-founder and Chief Technology Officer of Water Illumination Inc., will serve as the Principal investigator. Water Illumination Inc. is technology company founded in 2021 and

aims to develop innovative water treatment technologies. The company is spined off from Dr. Liu's research at the University of California, Riverside, where he is also holding a tenured professor position since 2013, in the Department of Chemical and Environmental Engineering. Dr. Liu has been Principal Investigator or Co-Principal Investigator on 30 projects worth more than \$10.5 million in funding since 2013 sponsored by Federal, State/Local, and Industry organizations. The project sponsors include U.S. Department of Agriculture, National Science Foundation, Department of Interior, Department of Energy, Water Research Foundation and California Water Resource Center. He has active collaborations with faculty in Engineering, Chemistry, Environmental Sciences and Toxicology, as well as industrial and international collaborations to address interdisciplinary and global environmental and agricultural challenges. A short resume of Dr. Liu is attached at the end of this application package.

Dr. Liu's has extensive research expertise in water treatment, water reuse, desalination and environmental remediation. He has more than 20 years' research and engineering application experience in water system engineering, agriculture sustainability, water reuse and desalination technology – these are areas of research closely tied with the expertise needed to complete the proposed project. His research group has developed a variety of innovative and sustainable water treatment technologies for agricultural and municipal purposes. Selective examples include sustainable chemical treatment that converts toxic elements in agricultural drainage water to benign end products, innovative desalination processes that prevents membrane scaling and harvests valuable minerals, and novel photochemical treatment processes that degrade contaminants and recover fresh water from deteriorated water resources.

In recognition of his significant contribution to water reuse and desalination, Dr. Liu receives numerous national and international awards. He is an Emerging Investigator in Water Engineering and Technology awarded by the Royal Society of Chemistry in 2016, a prestigious National Science Foundation Faculty Early Career Development (CAREER) Award in 2017, and a Professional of the Year Award from International UV Association in 2019. His recent research is featured in journal front cover images and selected as best paper by multiple leading journals including *Environmental Science & Technology*, *Environmental Science: Water Research & Technology*, and *Environmental Science: Process & Impact.* Professor Liu is a member of several professional societies and currently serving on the International Water Association's specialist committee on metals and toxic substances.

The researchers involved in the project will have the requisite office space. Computers and the necessary chemical modeling software are available in the office of Water Illumination Inc. Other research team members on this project will be provided computers access and necessary codes and software application training. The PI has ample office space for himself. In addition, in case that additional water quality analysis is needed, the PI's team will outsource with the University of California, Riverside and have access to the start-of-the-art analytical equipment there.

To foster a productive collaboration, the project team will communicate regularly via emails, phone calls and meetings with the CAC Production Research Committee to update research progress, share results, and discuss near-term experimental plans.

7. Project Budget

Introduction

This proposal requests \$74,600 over 12 months beginning November 1, 2021. The funding will be used to support the Principal Investigator, one researcher, plus travel and literature review materials/publication fees necessary to complete the research as proposed. Details of the budget request are below.

BUDGET ITEM DESCRIPTION	COMPUTATION		QUANTITY	TOTAL COST	
BUDGET HEM DESCRIPTION	\$/Unit	Quantity	TYPE	TOTAL COST	
SALARIES/WAGES					
PI: Haizhou Liu	\$150	160	Hourly	\$24,000	
Team member: Researcher (TBD)	\$70	630	Hourly	\$44,100	
			Subtotal	\$68,100	
TRAVEL					
Meeting with CAC stakeholders	\$100	5	trips	\$500	
			Subtotal	\$500	
SUPPLIES AND MATERIALS					
Chemical modeling software/Literature database subscription/other publication fees	\$6,000	1	LS	\$6,000	
			Subtotal	\$6,000	
TOTAL DIRECT COSTS				\$74,600	
INDIRECT COSTS					
year and type	0.0%		base	\$0	
TOTAL ESTIMATED PROJECT COSTS				\$74,600	

Salaries and Wages

The Principal Investigator, Dr. Haizhou Liu, is eligible to receive salary from extramural contracts and grants. The budget proposes a total of 160 hours with a hourly rate of \$150, based on the equivalent salary and fringe benefit level of experts with similar level of experience.

One Researcher, to be named, will work for 630 hours with an hourly rate of \$70 for this project under the PI's supervision. The amount requested is based on equivalent salary for Postdoctoral Researchers at UC Riverside.

Travel

The budget includes \$500 for travel by the PI and project team to different California avocado groves and discuss the project with grove owners, as well as travel to the CAC headquarter for project meetings. The travel budget will cover gasoline expenditures for 5 day trips.

Materials and Supplies

The budget includes \$6,000 for purchasing chemical modeling software, subscription fees to download publications from literature review database, and publication fees for future results dissimilation.

Indirect Costs

None

Total Cost

Total project cost is \$74,600.

8. Literature Cited

- 1 Mickelbart, M.V.; Melser, S.; Arpaia, M.L. Salinity-induced changes in ion concentrations of 'Hass' avocado trees on three rootstocks. *Journal of Plant Nutrition*. **2007**, 30, 105-122.
- 2 Benjamin, M. M. Water Chemistry. 2015. Waveland Press, Inc.

BIOGRAPHICAL SKETCHES

Haizhou Liu

Co-founder and Chief Technology Officer, Water Illumination Inc., Irvine, CA 92618 Tel: (206) 214-7162; e-mail: haizhou@waterillumination.com

Education and Training

Sichuan University, Chengdu, China	Environmental Engineering	B.S./2006
University of Washington, Seattle	Environmental Engineering	M.S./2007
University of Washington, Seattle	Environmental Engineering	Ph.D./2010
University of California, Berkeley	Environmental Engineering	Postdoc/2010-2012

Research and Professional Experience

2021-Present	Co-founder and Chief Technology Officer, Water Illumination Inc., Irvine, CA 92618
2013-Present	Associate Professor (2018-present), Assistant Professor (2013-2018), Department of Chemical and Environmental Engineering, University of California, Riverside, CA.
2010-2012	Postdoctoral Researcher, Department of Civil and Environmental Engineering, University of California, Berkeley, CA.
2006-2010	Graduate Research Assistant, Department of Civil and Environmental Engineering, University of Washington, Seattle, WA

Honors and Awards

2021	Outstanding Educator A	Award of the Year	r, Orange Count	y Engineering	Council, CA.

- 2019 Distinguished Advisor Award, University of California
- 2019 Academic Senate Research Award, University of California
- 2019 Runner-up Best Paper Award on Environmental Science, Royal Society of Chemistry
- 2019 International Ultraviolet Association Best Research Paper Award
- 2018 Journal of Environmental Science & Technology Excellence in Review Award
- 2018 International Ultraviolet Association Young Professional Award
- 2017 National Science Foundation Faculty Early Career Development (CAREER) Award
- 2017 Regents Faculty Development Award, University of California
- 2017 World Water Forum Innovator Award, Metropolitan Water District of Southern California
- 2016 Emerging Investigator in Water Engineering and Technology, Royal Society of Chemistry
- 2016 Hellman Family Fund Faculty Fellowship, University of California

Selective Grants Received

[1] NSF	(PI)	\$562,320	4/17-3/22	
CAREER: Beyo	ond Conventional	Drinking Water Ma	nagement: Control of Redox-driven in situ	
Release of Accumulated Inorganic Contaminants from Water Distribution Infrastructure				
[2] USDA	(Co-PI)	\$5,112,000	1/17-1/22	

Deployment of a Spectrum of Bactericides to Cure and Prophylactically Treat Citrus Huanglongbing

[3] USDA (PI) \$200,000 4/20-3/22

Application of Nanotechnology to Design Tailored Photocatalyst for Nitrate Removal in Agriculture Impacted Water

[4] NSF (PI) \$479,997 9/16-8/19

GOALI: SusChEM: Experimental Investigation of Chloramine and Persulfate based Aqueous Photochemistry and Development of Efficient UV-Based Water Reuse

[5] NSF (Co-PI) \$439,301 12/18-11/21

D3SC: Data-Driven Modeling and Experimental Investigation for Discovery of Aquatic Chemistry Reaction Kinetics: New Tools for Water Reuse Applications

[6] University of California (Co-PI) \$1,135,304 1/17-12/20

Fighting Drought with Stormwater: from Research to Practice

[7] Department of Interior (PI) \$300,000 1/19-12/20 Innovative Water Reuse Systems Harnessing Chloramine Photochemistry for Potable Water Reuse

[8] Department of Education (PI) \$894,000 9/18-10/21

GAANN: Sustainable Environment and Infrastructure Development Fellows for the Future

Professional Experience

- 1. Broadening the participation of underrepresented groups. I collaborate with community colleges and high schools in the Riverside region, and since 2013 I have successfully recruited 30 undergraduate students (half of them women or underrepresented ethnic minorities) to work in my lab. I have encouraged these students to participate in regional and national meetings as presenters, and three have won student poster awards.
- 2. Contributions to student training and mentoring. I have been actively engaged in mentoring both undergraduate and graduate students at UCR. With my mentorship, the students are becoming independent critical thinkers, have developed technical skills, and have won many awards.
- 3. Service to the scientific community. I served as a conference organizer for 2013 ACS Colloidal Symposium, 2015 ACS Spring Meeting, 2016 ACS Spring Meeting, 2016 Emerging Contaminant Summit, 2016 ACS Fall Meeting and 2016 Gordon Conference on Water. I serve as a journal reviewer regularly for Environmental Science & Technology, Environmental Science & Technology Letter, Water Research, and Applied Biochemistry and Biotechnology. I served as a panelist to review proposals for NSF CBET programs and USDA.
- 4. Service to the professional and industrial community. I am a core committee member of the International Water Association's Specialist Group on Metals and Toxic Substances in Drinking Water.

List of Selective Publications in Refereed Journals during the Past Four Years

- 1. Parker, E.A.; Grant, S.B.; Cao, Y.; Rippy, M.A.; McGuire, K.; Holden, P.; Feraud, M.; Avasarala, S.; **Liu, H.**; Hung, W.; Rugh, M.; Jay, J.; Peng, J.; Shao, S. Predicting unsteady solute transport through green stormwater infrastructure with transit time distribution theory. *Water Resources Research.* **2021**, *57* (2), e2020WR028579.
- 2. Avasarala, S.; Orta, J.; Schaefer, M.; Abernathy, M.; Ying, S.; Liu, H. Effects of residual disinfectants on the redox speciation of lead (II)/(IV) minerals in drinking water distribution systems. *Environmental Science: Water Research & Technology.* **2021**, *7* (2), 357-366.
- 3. Tan, C.; Avasarala, S.; **Liu, H.** Hexavalent chromium release in drinking water distribution systems: new insights into zerovalent chromium in iron corrosion scales. *Environmental Science & Technology*. **2020**, *54* (20), 13036-13045.
- 4. **Liu, H.**; Yu, X. Hexavalent chromium in drinking water: chemistry, treatment, challenges and future outlook on Sn(II)- and photocatalyst-based treatment. *Frontiers of Environmental Science & Engineering.* **2020**, *14* (5), 88.
- 5. Naddeo, V.; Liu, H. 2019 novel coronavirus (SARS-CoV-2): what is its fate in urban water cycle and how can the water research community respond? *Environmental Science: Water Research & Technology.* **2020**, *6* (5), 1213-1216.
- 6. Chen, G.; **Liu, H.** Photochemical removal of hexavalent chromium and nitrate from ion-exchange brine waste using carbon-centered radicals. *Chemical Engineering Journal.* **2020**, *396*, 125136.
- 7. Matsumoto, M.; **Liu, H.** Mercury speciation and remediation potentials at a historically contaminated hazardous waste site. *Journal of Hazardous Materials*. **2020**, *384*, 121351.
- 8. Mangalgiri, K.; Patton, S.; Wu, L.; Xu, S.; Ishida, K.; **Liu, H.** Optimizing potable water reuse systems: chloramines or hydrogen peroxide for UV/based advanced oxidation process? *Environmental Science & Technology.* **2019**, *53* (22), 13323-13331
- 9. Jain, T.; Sanchez, E.; Owens-Bennett, E.; Trussell, R.; Walker, S.; Liu, H. (2019) Impacts of antiscalants on the formation of calcium solids: implication on scaling potential of brackish desalination concentrate. *Environmental Science: Water Research & Technology*. 5 (7) 1285-1294.
- 10. Wang, Z.; Chen, G.; Patton, S.; Ren, C.; Liu, J.; Liu, H. (2019) Degradation of nitrilotrismethylenephosphonic acid (NTMP) antiscalant via persulfate photolysis: implications on desalination concentrate treatment. *Water Research*. 159, 30-37.
- 11. Korshin, G. V.; **Liu, H.** (2019) Controlling the colloidal stability of PbO₂ and lead release in drinking water distribution systems. *Environmental Science: Water Research & Technology.* 5 (7) 1262-1269.
- 12. Yu, X.; Niksa, D.; Ge, X.; **Liu, H.**; Hille, R.; Mulchandani, A. (2019) Synthesis of formate from CO₂ gas catalyzed by an O₂-tolerant NAD-dependent formate dehydrogenase and glucose dehydrogenase. *ACS Biochemistry*. *58* (14), 1861-1868.
- 13. Henrie, T.; Plummer, S.; Orta, J.; Bigley, S.; Seidel, C.; **Liu, H.** (2019) Full-scale demonstration testing of hexavalent chromium reduction via stannous chloride application. *American Water Works Association Water Science*. e1136. DOI: 10.1002/aws2.1136

- 14. Fraiese, A.; Naddeo, V.; Uyguner-Demirel, C. S.; Prado, M.; Cesaro, A.; Zarra, T.; Liu, H.; Belgiorno, V.; Ballesteros, Jr. F. (2019) Removal of emerging contaminants in wastewater by sonolysis, photocatalysis and ozonation. *Global NEST Journal*. 21 (2), 98-105.
- 15. Chen, G.; Hanukovich, S.; Chebeir, M.; Christopher, P.; **Liu, H.** (2019) Nitrate removal via formate radical-induced homogeneous photochemical denitrification. *Environmental Science & Technology*. 53 (1), 316-324.
- 16. Patton. S.; Li, W.; Romano, M.; Naddeo, V.; Ishida, K. P.; Mezyk, S. P.; Liu, H. (2018) Impacts of chloramines on UV photolysis of hydrogen peroxide: removal of trace organic contaminants for potable reuse. *Environmental Science & Technology*. 52 (20), 11720-11727.
- 17. Li, W.; Xu, E.; Schlenk, D.; **Liu, H.** (2018) Cyto- and geno-toxicity of 1,4-dioxane transformation products during UV/AOP for potable water reuse. *Environmental Science: Water Research & Technology.* 4 (9), 1213-1218.
- 18. Ngo-Duc, T.; Plank, J. M.; Chen, G.; Harrison, R.E.S.; Morikis, D.; **Liu, H.**; Haberer, E. D. (2018) M13 bacteriophage spheroids as scaffolds for directed synthesis of spiky gold nanostructures. *Nanoscale*. *10* (27), 13055-13063.
- 19. Chebeir, M.; **Liu, H.** (2018) Formation of Cr(VI) in drinking water via iron chromite oxidation by chlorine: role of iron, bromide, pH and carbonate. *Environmental Science & Technology*. 52 (14), 7663-7670
- 20. Li, W.; Patton. S.; Ishida, K. P.; Mezyk, S. P.; Liu, H. (2018) Simultaneous UV photolysis of chloramine and persulfate in reserve osmosis permeate for potable water reuse. *Environmental Science & Technology*. 52 (11), 6417-6425.
- 21. Chen, G.; Feng, J.; Wang, W.; Yin, Y.; **Liu, H**. (2017) Synthesis and application of highly reductive TiO₂ nanocrystals to photo-catalytically remove hexavalent chromium. *Water Research*. 108, 383-390.
- 22. Chen, G.; **Liu, H**. (2017) Understanding the kinetics and mechanisms of aqueous vanadium(V) reductive reactions using electrochemical techniques. *Environmental Science & Technology*. 51 (20), 11643-11651.
- 23. Orta, J.; Patton, S.; **Liu, H**. (2017) Bromide-assisted catalytic oxidation of lead(II) solids by chlorine in drinking water distribution systems. *Chemical Communications*. *53* (62), 8695-8698.
- 24. Li, W.; Orozco, R.; Camargos, N.; Liu, H. (2017) Impacts of pH, alkalinity and chloride on heterogeneous persulfate activation for groundwater remediation. *Environmental Science & Technology*. 51 (7), 3948-3959.
- 25. Wordofa, D.N.; Walker, S.L.; **Liu, H**. (2017) Sulfate radical induced disinfection of a pathogenic strain of *Escherichia coli* by iron activated persulfate. *Environmental Science & Technology Letters*. 4 (4), 154-160.
- 26. Patton, S.; Li, W.; Couch, K.; Mezyk, S.; Ishida, K.; Liu, H. (2017) Impact of the UV photolysis of monochloramine on 1,4-dioxane removal: new insights into potable water reuse. *Environmental Science & Technology Letters*. 4 (1), 26-30.
- 27. Pari, S.; Wang, I.; Liu, H.; Wong, B.M. (2017) Assessment of sulfate radical oxidation reactions in aromatic contaminants using density functional theory and high-level quantum chemical methods. *Environmental Science: Process & Impact.* 19 (3), 395-404.

- 28. Li, W.; Jain, T.; Ishida, K.; Remucal, K. C.; **Liu, H**. (2017) Kinetics modeling of the degradation of trace organic contaminants in UV/hydrogen peroxide, UV/persulfate and UV/free chlorine. *Environmental Science: Water Research & Technology. 3* (6), 128-138.
- 29. Duan, W.; Chen, G.; Chen, C.; Sanghvi, R.; Walker, S.; **Liu, H.**; Ronen, A.; Jassby, D. (2017) In-situ hexavalent chromium removal via composite carbon nanotubes electrically conductive ultrafiltration membranes. *Journal of Membrane Science*. *531*, 160-171.
- 30. Prado, M.; Borea, L.; Cesaro, A.; Liu, H.; Naddeo, V.; Belgiorno, V.; Ballesteros, F. (2017) Removal of emerging contaminant and fouling control in membrane bioreactors by combined ozonation and sonolysis. *International Biodeterioration & Biodegradation*. 119, 577-586.

A survey of cadmium concentrations in California avocado orchards: the possible role of rootstock, variety and geographical location

Ben Faber

UC-ANR, Ventura County, Ventura CA; bafaber@ucanr.edu

Joseph Mark Buhl

Avotopia, 46750 Los Gatos Rd., Temecula, CA 92560; mark@avotopia.com

Mary Lu Arpaia

Dept. of Botany and Plant Sciences, University of California, Riverside; mlarpaia@ucanr.edu

Project Duration: 1 to 2 years

Project Budget: \$199,868

Rationale and Significance:

In recent years, the presence of cadmium in avocado fruit has become an issue for fruit destined for export. We believe that we need to address this issue for both domestic and export markets. Recently D. Mendoza, C. Lin and K. Clark from the University of Missouri received a USDA TASC grant to examine potential bioremediation strategies and to understand the contribution of innate gene transcriptional programs in avocado related to uptake and allocation by the plant. This project complements the objectives of the TASC grant and aims to collect data on the potential impact of varietal and rootstock, geographical location and fruit development on uptake and allocation to the fruit which can guide the University of Missouri research team. This data could also assist future efforts in avocado breeding in California.

Cadmium is a heavy metal that can cause liver and kidney damage when ingested, as well as bone demineralization, and lung cancer when inhaled. It is a toxicant that accumulates in the body over decades due to a long biological half-life. Cadmium is a naturally occurring element and avocados and other plants can be affected by natural plant uptake from soil high in cadmium. Contamination often occurs from agronomic practices, such as via applications of phosphate fertilizers. Cadmium is a naturally occurring element that can contaminate phosphate during the mining process. It does not degrade in the environment and once in soil, is not easily removed. Soluble cadmium in soil is taken up by plants, then transferred to human tissue via the bloodstream after the plants are ingested. Crop species and crop varieties all differ in their propensity and ability to take up and accumulate cadmium. Complex interactions between agronomic practices, soil chemistry and plant genetics all drive the potential for cadmium contamination in avocados.

A brief survey of macro- and micronutrient levels in California 'Hass' avocado fruit was conducted by Arpaia and Hofshi (unpublished) as part of the development of the fruit nutrient removal calculator in 1999 for www.avocadsource.com. In this survey, 3 mature, market quality fruit were harvested from 4 cooperator sites in June 1999. One site was in San Diego County and 3 sites were in Ventura County. Macro- and micronutrients for the seed, flesh and peel were calculated separately and then the total amount of the macro- or micronutrient on a fruit basis was entered into the nutrient removal calculator. Cadmium was detected in fruit from all locations. The highest amount of cadmium was found in the fruit

flesh at all sites as compared to the peel and seed. The average cadmium per fruit (on a total fruit basis) ranged from 0.1 - 0.6 ug/g fresh weight with an overall average of 0.37 ug/g fresh weight.

Objective and goals:

Our overall objective in this project is to survey California avocado orchards to determine the range of cadmium that are found in leaf and fruit tissues. The goal is to understand whether there is a correlation between soil, fruit, and leaf concentrations to have a predictive tool for the grower to know if cadmium could be an issue for the upcoming season. Around this idea we have 4 specific objectives that are separate but interconnected. We have listed these objectives in order of importance:

- 1) Test the hypothesis that there are likely genetic differences in uptake and movement into the avocado fruit and leaf tissue as influenced by rootstock and scion variety;
- 2) Sample 27 orchards throughout the California production area for soil and leaf analysis and obtain grower fertilization records;
- 3) Sample the same groves in objectives 1-2 for cadmium levels in mature fruit;
- 4) Conduct a fruit development study, similar to those conducted in South Africa demonstrating calcium uptake, to study the uptake pattern of cadmium into the developing fruit and to test the hypothesis that cadmium levels in developing fruit can be used to predict cadmium levels in mature fruit.

Methodology

Objective 1. Are there genetic based differences in uptake and movement into the avocado plant and fruit as influenced by rootstock and scion variety?

M.L. Arpaia established a scion:rootstock trial in 2012 that has 9 clonal rootstocks and 5 scion varieties. Trees are replicated 10 times in a randomized block design in a commercially operated grove near Saticoy, CA. The trees were planted in 2012 and yield and tree size data has been collected and previously shared with the industry through grower meetings and tours. It is likely, based on UC Davis soil maps for Ventura County developed by R. Arkley in the 1950's that cadmium is present in the soil at this site (Chang et al, 2004). This is also supported by the USGS map of national cadmium soil levels (Smith et al, 2019).

Leaf samples (20 leaves per tree) were harvested in Fall 2021 by the research team. Leaf samples were collected following the protocols developed by Jones and Embleton (1978): mature spring leaf from non-flushing, non-fruiting shoots. The leaf samples will be analyzed to see if cadmium concentration in the leaf tissue is influenced by rootstock or scion variety and to establish the baseline concentration of cadmium in the soil where rooting occurs. We will do a complete leaf analysis so that any relationships with other macro- and micronutrients can be ascertained. There are approximately 420 individual samples since some of the original trees have died in previous years.

Additionally, 10 soil samples taken in a transect across the field were collected. These soil samples represent soil in the top 8" below the mulch layer and represents where most avocado roots are typically found. The soil samples have been sent to the University of Missouri for analysis (see letter of support by K. Clark).

Objective 2. Survey 27 orchards throughout California for the presence of cadmium in leaf tissue and soil samples.

We will identify 'Hass' orchards on the same rootstock (most likely Dusa, Toro Canyon or Duke 7) in the following counties: San Diego (5 sites), Riverside (4 sites), Orange County (3 sites), Ventura (5 sites), Santa Barbara (3 sites), San Luis Obispo (5 sites) and the San Joaquin Valley (2 sites). We will collect 4 replicate samples per site for both the leaf and soil sample. These samples will be analyzed for a standard complete analysis with cadmium added into the analysis. The data collected from this objective will give a better idea on how widespread cadmium is within the growing regions of California. This portion of the study will be conducted in Fall 2022 since we have currently passed the correct timing for sample collection in Fall 2021. We will also obtain nutrient management records from the cooperators in this survey to understand the timing, amount, type of fertilizer and source of the fertilizer.

Objective 3. Collect a mature fruit sample from each grove from objectives 1-2 and have the fruit analyzed for the presence of cadmium in the peel, flesh, and seed tissue.

- 1) From Objective 1: For the Saticoy research site, 10 fruit from each combination of scion:rootstock will be collected in winter/early spring 2022. This translates into approximately 1 fruit per tree for a total of approximately 45 combinations (9 rootstocks x 5 varieties). The peel, flesh and seed will be separated from each other, recording by fresh and dry weight of each portion of fruit. A complete macro- and micronutrient analysis will be conducted on the samples for each rootstock:scion combination. Three replicates will be taken of each tissue type per sample bringing the total to 135 samples for each tissue type.
- 2) From Objective 2: Mature 'Hass' fruit samples will be taken from each site (10 fruit per site of approximately same size) in late Winter/early Spring 2022. Each composite sample will be divided into the peel, flesh and seed as described above and 3 replicate samples per tissue type will be analyzed from each site for a total of 9 samples per site and a total of 246 samples. A complete macro- and micronutrient analysis will be conducted on the samples for each site.
- 3) Since the leaf sampling for Objective 2 is likely not to occur until Fall 2022, we propose that we also repeat the fruit sampling in late Winter/early Spring 2023 as well. We believe it is critically important to conduct this initial sampling in Winter/Spring 2022 to understand the magnitude of the cadmium situation in California and to identify sites which have little or no detectable cadmium in the fruit. By having a follow-up sampling in Winter/Spring 2023 we will be able to examine the influence of year to year variability.

Objective 4. Is there a peak period of cadmium uptake in developing fruit?

Assuming that at one of the sites used in Objectives 1 – 3 we observe 'Hass' fruit and/or leaf tissue high in cadmium level we propose to select one of these sites for detailed fruitlet sampling. We will select trees for this study which are neither "on" or "off" but with moderate fruit set. Fruit samples will be collected from multiple trees adjacent to each other at the research site. We anticipate that we will need for the course of the study 20 trees total. Care will be taken not to collect early fruit that appear ready to abscise but we will collect fruitlets that are dark green and shiny at the time of collection. Commencing roughly 4 to 6 weeks after fruit set (pea size) we will collect fruit and analyze for all macroand micronutrients in addition to cadmium. It is probable that we will not be able to separate the peel, flesh, and seed during the very early stages of development, but we will keep the seed (and seed coat) separate from the flesh/peel portion of the young fruit. Sampling will be weekly with 5 replicate samples

until roughly the end of August 2022. We will then switch to biweekly sampling through December 2022 and then to monthly sampling through approximately March 2023. We expect that we will be able to separate the peel from the developing flesh sometime in July. We will also collect leaves for leaf analysis from the site in fall 2022 and conduct a more intensive soil sampling of the site where the trees being used in this study are located. This data will be used to gauge how cadmium levels in the fruit may change during fruit development.

Milestones:

Objective 1 – Expect completion by June 2022

- 1. Tissue Sampling **completed** through sampling, washing and drying of leaf samples; sampling completed in October 2021. *Analysis of the samples remains to be completed*.
- 2. Soil Sampling **completed** in October 2021. Sample sent to University of Missouri for analysis in November 2021. *Anticipate results to be available early 2022*.
- 3. Analysis of data and interpretation of results to be completed but anticipate with 6 to 8 weeks following receipt of results.

Objective 2 – Expect completion by June 2022:

- 1. Identify sampling sites in January February 2022.
- 2. Soil samples will be collected in Spring 2022 and submitted for analysis. Data analysis and interpretation of results will be completed approximately 6 8 weeks following the receipt of the results.
- 3. Leaf samples will be collected in Fall 2022 and submitted for analysis. It is too late in the current year to collect leaf samples following the current guidelines for leaf collection. Results will be compared to fruit analysis results obtained in late winter/early spring 2022 and to fruit analysis results from the 2023 sampling.

Objective 3 – Expect completion by June 2022 for 3.1 and 3.2 Expect completion by June 2023 for 3.3

- From the rootstock:scion trial in Saticoy, mature fruit samples will be collected in consultation
 with the grove management but expect this will be in late Winter/spring 2022. The results of the
 fruit analysis will be compared to that of the leaf analysis samples from Objective 1 to see any
 relationships between cadmium levels in leaf and fruit samples. Expect completion by June
 2022.
- 2) Fruit from Objective 2 will be collected in later winter/early spring 2022. Expect completion by June 2022.
- 3) Fruit from Objective 2 will also be collected in late winter/early spring 2023. Expect completion by May 2023.

Objective 4 – Expect completion by June 2023

1) Commence sampling approximately in early June 2022 and complete in March 2023. Expect completion of objective by June 2023.

Anticipated results:

Overall, we believe that the results of this study will document the levels of cadmium that could be anticipated to be found in developing fruit, will establish whether there is a relationship between leaf tissue levels and fruit cadmium levels and will give us an idea whether rootstocks and/or varieties differ in their ability to accumulate cadmium. All this information is important to help the California grower begin the process of identifying mitigation strategies for reducing cadmium in mature fruit.

We anticipate that by sampling leaf and fruit samples of the scion:rootstock trial in Saticoy, CA (Objective 1) we will learn if there are any detectable differences in cadmium uptake due to a genetic component. We expect we will see differences due to rootstock since it is already documented in previous literature that macro- and micronutrient uptake is influenced by rootstock (Mickelbart et al, 2007) as well as yield. Since we have the 2021 yield data and will be collecting the 2022 yield data, we will be also to look at the yield dynamic. If we observe a strong rootstock or scion effect on cadmium accumulation in either leaf or fruit tissue, this information will be shared with the University of Missouri group as well as Arpaia's current collaboration with the UC, Irvine on genome mapping of the avocado. This information long-term could lead to a genetic solution to the issue of cadmium uptake by avocado.

The results of objective 2 are expected to highlight whether cadmium uptake by the avocado is a problem just in Ventura or in other production areas. Chang et al (2004) evaluated soil cadmium from 50 benchmark soils throughout California. These samples were initially taken in 1950, then resampled in 1967 and 2001 to assess the impacts of management practices on cadmium and other materials over time (Chang et al 2004). The sampling identified areas of the state with high naturally occurring levels of cadmium and the impact of fertilizer practices on those levels over time. Especially in vegetable fields where high rates of phosphorus fertilizer use had occurred, high levels of Cd were found in the later samplings.

By collecting both leaf samples (Objectives 1, 2), at a point in time which is recommended for general nutritional health management, and mature fruit samples (Objective 3), we will be able to establish whether there is a correlation between leaf tissue levels of cadmium and that of those found in the fruit. If we can establish such a relationship, it is possible that growers could use this as a tool to predict whether they will expect to have levels in their mature fruit that may cause concern.

We anticipate that we will observe a small peak in cadmium uptake in the developing fruit about 8 weeks after fruit set, like calcium since cadmium is taken up by the tree through the transpiration stream. Work by Witney et al (1990) demonstrated this to be the case for calcium uptake. Witney et al also showed that tree vigor also influenced uptake. Developing fruit on vigorous 'Hass' and 'Fuerte' trees (= Off crop) had lower levels of calcium as compared to fruit from non-vigorous trees (= On crop). The authors also noted that Fuerte overall had lower levels of fruit calcium than Hass, suggesting a genetic component for calcium uptake. Researchers in Australia have also hypothesized that rootstocks vary in their ability to take up calcium and this in turn influences disease tolerance (Coates et al, 2011). If our hypothesis proves correct, it is feasible that early fruit development sampling could be used by growers to predict cadmium levels in their fruit. Additionally, if an early peak in cadmium uptake was documented this may guide the timing of mediation treatments.

Outreach and dissemination of data:

The results will be disseminated to the industry through UC ANR Topics in Subtropics newsletter, California Avocado Society quarterly and California Avocado Commission publications as appropriate.

Faber and Arpaia give frequent grower presentations to CAS and CAC as well as other grower groups and we anticipate sharing this data within those venues as well. Additionally, we will share these results with the University of Missouri research team examining the genetic components of uptake and remediation through their USDA-TASC grant. We also anticipate the preparation of at least 1 referred journal article on our findings.

References:

- 1. Chang, A.C, Page, A.L. and Krage, N.J. 2004. Role of fertilizer and micronutrient applications on arsenic, cadmium and lead accumulation in California cropland soils Final Report to CDFA. Pp. 124. https://www.cdfa.ca.gov/is/docs/CDFAFinalReport.pdf.
- 2. Coates, L.M, Dann, E.K., Shuey, L.S., Smith, L.A., Dean, J.R., Cooke, A.W., Pegg, K.G., Hofman, P.J., Marques, R., Stubbings, B. and Whiley, A.W. 2011. Effects of rootstock on avocado fruit quality assessment of postharvest disease, major cations and biochemical traits. Proceedings VII World Avocado Congress 2011 (Actas VII Congreso Mundial del Aguacate 2011). Cairns, Australia. 5 9 September 2011.
- 3. Jones, W.W. and Embleton, T.W. 1978. Leaf analysis as a guide to avocado fertilization. *IN:* Soil and plant tissue testing in California, H.M Reisnaruer ed. UC Bulletin 1879. Pp 14-15.
- 4. Mickelbart, M.V., Bender, G.S., Witney, G.W., Adams, C., and Arpaia, M.L. 2007. Effects of clonal rootstocks on 'Hass' avocado yield components, alternate-bearing, and nutrition. J. Hort. Sci. and Biotech. 82(3): 460-466.
- 5. Smith, D. B., Solano, F., Woodruff, L.G., Cannon, W.F. and Ellefsen, K.J. 2019. Geochemical and mineralogical maps, with interpretation, for soils of the conterminous United States. U.S. Geological Survey Intranet. URL: http://pubs.usgs.gov/sir/2017/5118/sir20175118_element.php. Page Contact Information: Contact USGS. Page Last Modified: November 12 2019 16:32:37. Accessed November 30, 2021.
- 6. Witney, G.W., Hofman, P.J. and Wolstenholme, B.N. 1990. Effect of cultivar, tree vigour and fruit position on calcium accumulation in avocado fruits. Scientia Horticulturae. 44:269-278.

Budget Justification:

General Statement: We have prioritized the activities of this project. Although the research team believes that all project objectives merit funding since they seek different insights into this problem, each objective could be basically funded on its individual merit.

The bulk of the expenses for this project are related to tissue and soil analysis expenses. We have contacted various commercial laboratories in the state and in general the analysis of cadmium is an added expense to a standard analysis for macro- and micronutrients. We have budgeted, without having a firm number from any laboratory yet of \$100 per sample. If the California Avocado Commission can assist in reducing the amount of the analysis there would be a corresponding reduction in the price of the project.

Objective 1. We have already collected the leaf and soil samples from the scion:rootstock trial (October 2021) and prepared (washing and drying) the leaf samples. We are not requesting reimbursement for these activities but rather are requesting funding to complete the leaf analysis (grinding and analysis).

Objective 2. We are requesting support for leaf and soil analysis for the collected samples. We are assuming that for this objective, that we will submit the fresh leaf samples directly to the analytical laboratory which will then do the requisite washing and drying prior to analysis. We are also requesting

travel for PIs (mileage) to the sites for collecting the leaf and soil samples. Mileage rates are calculated using current approved rates (IRS 2021 rate = \$0.56 per mile).

Objective 3. We are requesting support for fruit analysis of the collected samples. We will need to separate the fruit tissues into peel, flesh and seed and collect fresh and dry weight of the tissues. The dried tissue (unground) will be submitted to the analytical laboratory. We are also requesting labor reimbursement for contract worker at UC Kearney Ag Center (currently \$25 per hour) to assist in the preparation of the tissue samples. The third item is travel reimbursement to the sites to collect the mature fruit samples. Mileage rates are calculated using current approved rates (IRS 2021 rate = \$0.56 per mile).

Objective 4. We are requesting support for fruit analysis of the collected samples. The fruit samples will be separated into peel, flesh and seed (including seed coat). Fresh and dry weight of each sample will be collected. The dried tissue (unground) will be submitted to the analytical laboratory. We are also requesting labor reimbursement for contract worker at UC Kearney Ag Center (currently \$25 per hour) to assist in the preparation of the tissue samples. The third item is travel reimbursement to the sites to collect the mature fruit samples. Mileage rates are calculated using current approved rates (IRS 2021 rate = \$0.56 per mile). We are assuming that the experimental site will be in Ventura County and base mileage estimate on this assumption.

Budget Summary

UC Kearney Ag Center Labor (Contract by hour)

Objective 1. Are there genetic based differences in uptake and movement into the avocado plant and fruit as influenced by rootstock and scion variety?	42,000
Objective 2. Survey 27 orchards throughout California for the presence of cadmium in leaf tissue and soil samples.	22,608
Objective 3. Collect a mature fruit sample from each grove from objectives 1 – 2 and have the fruit analyzed for the presence of cadmium in the peel, flesh, and seed tissue.	
Objective 4. Is there a peak period of cadmium uptake in developing fruit?	42,952
TOTAL BUDGET FOR ALL OBJECTIVES	199,868

Budget Detail by Objective		
Objective 1. Are there genetic based difference	es in uptake and movement into the avo	cado plant
and fruit as influenced by rootstock and scion	variety?	-
Services:	•	
Leaf samples	420 samples @ \$100 per sample	42,000
Soil samples	10 samples - will be completed by the	-
	University of Missouri (Mendoza et. al.)	
Total for Objective 1		42,000
Objective 2. Survey 27 orchards throughout Co	alifornia for the presence of cadmium in I	eaf tissue
and soil samples.		
Travel:		
Mileage Roundtrip from UC Kearney Agricultural Center	Estimated mileage = 700 miles with visits to	392
- Riverside/Orange/San Diego Counties	various sites	
Mileage Roundtrip from UC Kearney Agricultural Center	Estimated mileage = 500 miles with visits to	280
- Ventura County/Santa Barbara Counties	various sites	200
Mileage Roundtrip from UC Kearney Agricultural Center - San Luis Obispo County	Estimated mileage = 500 miles with visits to various sites	280
Mileage Roundtrip from UC Kearney Agricultural Center	Estimated mileage = 100 miles with visit to	56
- San Joaquin Valley	various sites	30
	Total Mileage	1,008
Services:	5	,
Leaf samples	27 sites @ \$100 per sample x 4 replicates	10,800
Soil samples	27 sites @\$100 per sample x 4 replicates	10,800
·	Total Services	21,600
Total for Objective 2		22,608
Objective 3. Collect a mature fruit sample from	m each grove from objectives 1 – 2 and ha	•
analyzed for the presence of cadmium in the	•	
Travel:	occi, neon, and occu doode.	
Mileage Roundtrip from UC Kearney Agricultural Center	Estimated mileage = 700 miles with visits to	392
- Riverside/Orange/San Diego Counties	various sites	332
Mileage Roundtrip from UC Kearney Agricultural Center	Estimated mileage = 500 miles with visits to	280
- Ventura County/Santa Barbara Counties	various sites	
Mileage Roundtrip from UC Kearney Agricultural Center	Estimated mileage = 500 miles with visits to	280
- San Luis Obispo County	various sites	
Mileage Roundtrip from UC Kearney Agricultural Center	Estimated mileage = 100 miles with visit to various sites	56
- San Joaquin Valley	Total Mileage	1,008
Services:	i otai iviii cage	1,008
Jei vices.		

1. Rootstock: Scion Trial	405 samples - 40 hours preparation @ \$25 per hour; estimate 80 samples per day can be processed	1,000
2. Fruit samples Winter 2022 from 27 sites	243 samples - 24 hours preparation @ \$25 per hour	600
3. Fruit samples Winter 2023 from 27 sites	243 samples - 24 hours preparation @ \$25 per hour	600
	Total contract labor	2,200
Tissue analysis		
1. Rootstock: Scion Trial	45 combinations x 3 replicates x 3 tissues @ \$100 per sample	40,500
2. Fruit samples Winter 2022 from 27 sites	27 sites x 3 replicates x 3 tissues @ \$100 per sample	24,300
3. Fruit samples Winter 2023 from 27 sites	27 sites x 3 replicates x 3 tissues @ \$100 per sample	24,300
	Total tissue analysis	89,100
	Total Services	91,300
Total for Objective 3		92,308
Objective 4. Is there a peak period of cadmiu	m uptake in developing fruit?	
Travel:		
Mileage Roundtrip to Ventura County	Estimated mileage = 400 miles to collection trip x 23 collection dates	5,152
	Total Mileage	5,152
Services:		
UC Kearney Ag Center Labor (Contract by hour)		
Preparation of samples	345 samples across the 23 sampling dates - 4 hours preparation @ \$25 per hour per sampling date	2,300
	Total contract labor	2,300
Tissue analysis		
Weekly sampling (June - August): 12 collections Biweekly sampling (September - December): 6 collections	23 collections x 5 replicates x 3 tissue types x \$100 per sample	34,500
Monthly sampling (January - May): 5 collections		
Soil sampling for trees (1 sample per tree) bulked into 5 replicates		500
Leaf sampling for trees used for collection study (5 replicate samples)		500
	Total tissue analysis	35,500
	Total Services	37,800
Total for Objective 4		42,952



International Programs

College of Agriculture, Food and Natural Resources

University of Missouri

I-28A Agriculture Building - Columbia, MO 65211
Phone: 660-351-4696 - Fax: 573-882-9931
Email: ClarkK@missouri.edu Web: http://www.cafnr.missouri.edu/cip

November 29, 2021

To: California Avocado Commission

A vital part of the University of Missouri research on cadmium in California avocados will be an understanding of which rootstocks and cultivars are most prone to cadmium uptake. Because our funding is limited, it is important that we make well informed decisions when selecting the plant material that will be used in both our bioremediation and genomics research. There are multiple rootstocks available but it is unknown if there is variability in uptake between them.

The University of California researchers have collected leaf samples across a variety of rootstocks and scions that may be able to show correlation and interaction between cadmium uptake and cultivar. Having this leaf material tested would inform our decisions on choice of rootstock to work with.

We are writing to support funding for the testing of this plant tissue from a variety of rootstocks and cultivars collected in an area of California where cadmium is known to exist in the soil. We do not have funding to test the plant material at MU, but we will be testing for cadmium soil levels where the leaf samples were collected. These five soil samples are being provided to the University of Missouri by the University of California researchers. Both parties agreed to the soil testing at MU so that we could gain insight into the cadmium levels found in affected soil.

This plant tissue testing will help support decision making in our pre-research phase and will provide information for elucidating the relationships between soil cadmium at affected orchards and avocado tree uptake, assimilation and partitioning.

Sincerely,

Kerry Clark

Crop Scientist

Kerry M. Clark

Director, International Agriculture Programs

clarkk@missouri.edu