



POTATOES & PESTS PAPAS

Actionable Science Against Nematodes



Figure 1. Testing extracts effects on RKN galling of tomato roots. photo by K. Chandler

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Introduction to PAPAS Objective 3

Novel chemistries for nematode management

The plant *Solanum sisymbriifolium* (also known as litchi tomato) is distant relative to potato. It is resistant to both root-knot and cyst nematodes. Studies of these plants found that these nematodes rarely even enter the plant roots to feed, and those that do, cannot complete their lifecycle. Growing litchi tomato in contaminated field significantly reduces the number of nematodes in the field. Overall, the broad-spectrum resistance presents an invaluable opportunity for investigating new strategies in nematode management, especially as the efficacy of traditional nematicides dwindles due to environmental concerns.

Initial observations of litchi tomato plants hinted at their potential as nematicides, as their exudates



Figure 2. Litchi Tomato flowering. photo by L. Schulz



Figure 3. Litchi Tomato in the greenhouse.
photo by L. Schulz

exhibited a lethal effect on cyst nematode eggs. Research at the University of Idaho uncovered a rich array of biologically active compounds within *S. sisymbriifolium*, including glycoalkaloids, flavonoids, steroids, and glycosides. Despite this wealth of knowledge, the precise mechanisms underlying its nematicidal properties remain elusive. Within the PAPAS research community, there's a concerted effort to unravel the mysteries within *S. sisymbriifolium*. Scientists are working to identify novel bioactive compounds with potent nematicidal effects against both root-knot and cyst nematodes. This pursuit not only advances our understanding of *S. sisymbriifolium* resistance, but also lays the groundwork for potential commercialization in the future.

Methods of Making Extracts

Work by researchers in PAPAS is being

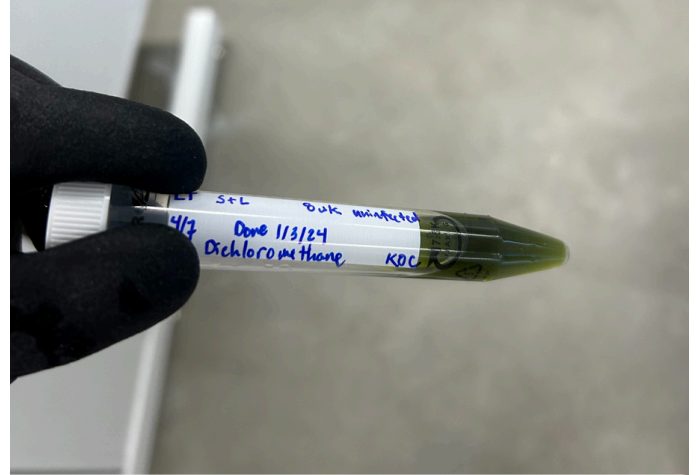


Figure 4. Stem+leaf plant material extracted with dichloromethane. photo by K. Chandler

performed at many institutions to determine the effects of litchi tomato extracts on root-knot and cyst nematodes. Extracts are made by growing litchi tomato plants, freeze-drying them, and grinding up the plant material. The plant material is either litchi tomato roots (R) or the stems and leaves (SL). The powdered plant material then goes through an extraction process that selects different types of compounds at each step. We started with four different solvents to perform our extraction: Hexane, Dichloromethane, Ethyl Acetate, and n-butanol. Each solvent has a different polarity that helps it to extract different components from the litchi tomato plant material. As a result, we have four extractions from the bulked litchi tomato root material, and each extraction contains different mixtures of unknown compounds. (A brief video demonstrating this process can be found at: <https://youtu.be/1uHXkBRKQY8>).

Our first goal is to determine which of the four extracts kills nematode eggs. The different extracts have been utilized in assays on root-knot and cyst nematodes to determine which extracts have toxic effects on the eggs. We are currently at a step where we are doing preliminary testing on extracts, each of which contains a broad range of compounds. From there, we can further fractionate those extracts, parsing out the compounds that are toxic from those that have no effect.

The main experiment performed on both root-knot and cyst nematodes is the effect of the extracts on egg hatching. Both root knot and cyst nematode hatching is severely affected by certain litchi tomato extracts.

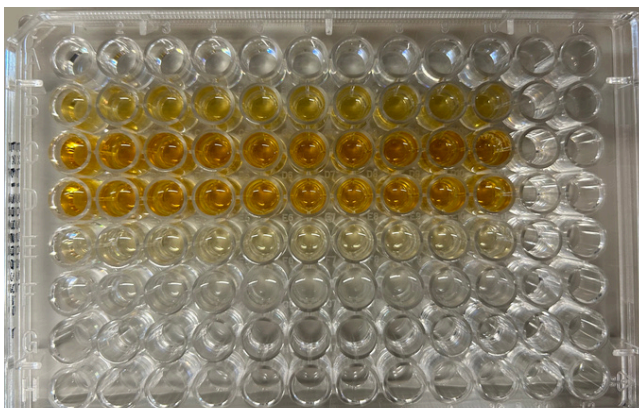


Figure 5. 96-well plate for testing extracts on egg hatching.

Experimental Results

To illustrate this point, we can look at the work of the master's student Koy Chandler on root-knot nematodes. He set up experiments in which the eggs were placed in a 96-well plate with the different extracts. Then the number of J2s that

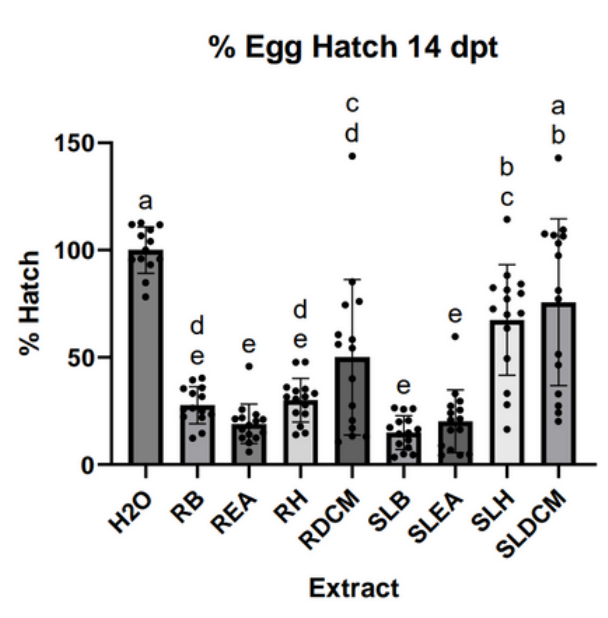


Figure 6. Effects of Extracts on RKN Egg Hatch. R=Roots, SL=Stem+Leaf, B=n-butanol, EA=Ethyl Acetate, H=Hexane, DCM=Dichloromethane.

hatched from these eggs were counted at 14 days after treatment. A control treatment of the eggs in water is compared to the eggs treated with different extracts. Values were calculated by: (eggs at day 1/J2s hatched at day 14) x 100 = % hatch. In the root-knot nematode (RKN), butanol and ethyl acetate extracts from both stem and leaf and root extracts were the most significantly affecting egg hatch. The stem and leaf dichloromethane extract was the only one to not show significance when compared to the control. Interestingly, in pale cyst nematodes (PCN), butanol and hexane extracts from stem and leaf were the only significant extracts to negatively affect egg hatch.

In addition, we have also been testing ability of root-knot nematode *Meloidogyne chitwoodi* eggs that have

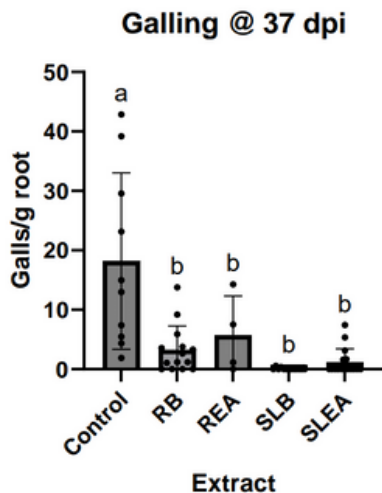


Figure 7. Effects of extracts on galling in tomato plants.

been treated with extracts to hatch and infect tomatoes. Nematode eggs were treated with the different extracts for 5 days and then placed on tomato plants. Galling was counted 27 days after inoculation. In this experiment, only some of the extracts were tested, but ethyl acetate and butanol extracts both significantly reduced galling on roots. From this, it can be assumed that these extracts contain toxic compounds that impact egg hatching, which subsequently prevented root galling.

Summary

Current research from labs at the University of Idaho, Washington State University, Cornell University, University of Wisconsin, and USDA-ARS are focused on testing compounds from litchi tomato using different solvents (hexane, dichloromethane, ethyl acetate, n-butanol). The goal is to isolate the specific bioactive compounds responsible for the nematocidal effects.

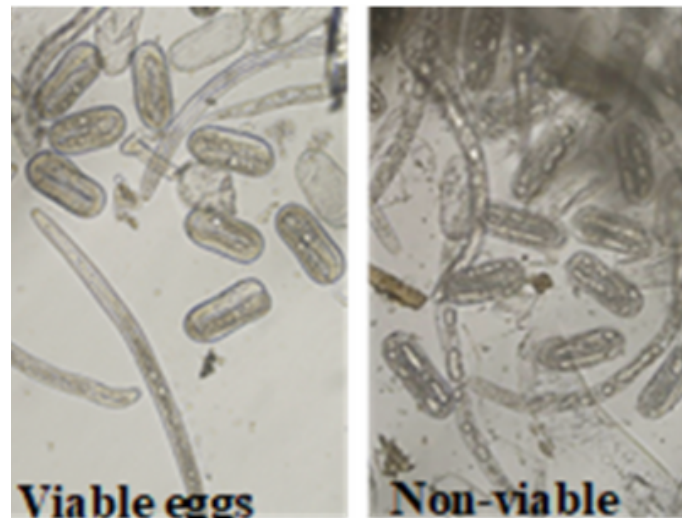


Figure 8. PCN exposed to potato root diffusate (left) or *S. sisymbriifolium* root extract (right). Eggs exposed to root extract do not hatch. (University of Idaho).

Initial findings suggest that extracts from litchi tomato roots and stems/leaves significantly affect nematode egg hatching. Extracts, particularly from ethyl acetate and butanol, have shown significant impacts on nematode egg hatching in both root-knot and cyst nematodes. This effect has been observed through assays measuring egg hatching and subsequent infection rates on host plants like tomatoes. Ongoing research involves further fractionation of these extracts to isolate and identify the exact compounds responsible for nematocidal activity. This includes detailed chemical analysis and testing to understand the mechanisms of action against nematodes. The ultimate goal is to identify novel nematocidal compounds from litchi tomato that could serve as alternatives to traditional chemical nematicides. This is particularly crucial as environmental concerns and regulatory

pressures increase against conventional pesticide use. The research is collaborative, involving multiple institutions and researchers within the PAPAS community, aiming to advance both scientific understanding and practical applications in agriculture. In summary, the study of litchi tomato for nematode management represents a promising frontier in agricultural research, leveraging natural plant defenses and bioactive compounds to develop sustainable solutions for nematode control in crops like potatoes.



Figure 9. Leaf of Litchi Tomato. photo by L. Schulz

Meet The Graduate Student: Koy Chandler



Koy Chandler was born and raised in Southern Idaho where he was surrounded by agriculture. He had the joy of moving plenty of irrigation hand lines during his time growing up, but it did not chase him away from the industry. He did his undergraduate studies in biology from Brigham Young University-Idaho in Rexburg, ID. While attending BYU-I, he helped Dr. Jeff Miller at Miller Research, working on the research farm there. This is where he found his love for plant pathology and chose to pursue further studies at Washington State University.

He is a current M.S. student in the Plant Pathology Program and is advised by Dr. Cynthia Gleason. His research is currently focused on the effects of Litchi Tomato on the root-knot nematode, *M. chitwoodi*. The hypothesis he is testing is that Litchi Tomato (LT) produces nematocidal compounds. His work with collaborators at other universities want to test if there are novel compounds found in Litchi Tomato that can be used as nematicides in the future. They are investigating the effects on several of the most devastating nematodes that infect potatoes. Over the summer, he is analyzing fractions from butanol extracts of LT to help narrow down the compounds responsible for the nematocidal effects.

He is participating in the outreach efforts of the PAPAS group in addition to his research to help reach out and connect with growers. He has contributed in gathering information, making fact sheets, and making videos to showcase the research being performed.

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