Evaluation of grower practices to reduce potential risks associated with the use of bsaaos in leafy greens

Summary

Studies have shown a direct link between the industry practices of using Biological Soil Amendments of Animal Origin (BSAAOs) and increased pathogen prevalence in the farm environment. If not managed properly, bacteria can survive in the farm environment and may contaminate fresh produce, posing a potential food safety risk. This project aims to improve how these amendments are tested, stored, and applied to reduce contamination risks. Over the course the project, we aim to assess crop inputs from both cattle and poultry suppliers for indicator and pathogenic bacteria; evaluate storage and grower application practices to see how they may impact bacterial persistence and survival; analyze weather effects like temperature and rainfall to understand their role in bacterial growth and persistence; and finally investigate factors that may drive bacteria to become activated under certain environmental conditions. By integrating these efforts, the study will evaluate post-delivery grower management practices and environmental conditions to determine risk reduction strategies that support industry decision-making. This work aims to equip leafy green producers with science-based best practices for the use and application of BSAAOs at the field scale, reducing food safety risks and fostering confidence in fresh produce safety.

Objectives

1. Objective 1. Work with producers to conduct assessments of finished materials at appropriate volumes to assess pathogen/indicator loading.
2. Objective 2. Evaluate industry practices at field-scale (stockpiling, temperature/moisture, timing of incorporation/irrigation, etc.) that may cause pathogen/indicator persistence or re-growth.
3. Objective 3. Evaluate the impact of weather conditions on the persistence, re-growth, and movement of pathogens/indicators within the growing environment.
4. Objective 4. Assess composting, generation, and BSAAO's storage measures that may induce or resuscitate pathogen's VBNC physiological state.

Methods

The research team is working with crop input suppliers to obtain products that are currently being used by fresh produce growers in the Southwest growing region. A total of four suppliers, two cattle compost and two heat treated poultry pellet suppliers are being evaluated. Initial samples are currently being screened for microbiological indicators (generic *Escherichia coli [E. coli]*, Total Coliform bacteria, Fecal Coliform bacteria, *Enterobacter*) as well as pathogens (*E. coli* O157:H7, *Salmonella*, STEC) at sufficient volumes (weight). Samples are being screened by both cultural and molecular methods for qualitative (presence/absence) and quantitative (number) data. Additional information has been obtained from each supplier related to time of production, production process, raw material and ingredient information, transport time/temperature, and other relevant characteristics important to the study. The research team is collecting inputs at a total of four different times over the two-year project to assess seasonal influences on delivered input quality (winter, spring, summer, fall). Microbiological indicator organisms including generic *E. coli*, Total Coliform bacteria, Fecal Coliform bacteria, and *Enterobacter* will also be evaluated from each BSAAO input utilizing modified US EPA Method 1680/1681 designed to assess fecal coliform bacteria. Similar methods are also be followed to assess *E. coli* O157:H7 and STEC utilizing the MPN based format and larger equivalent sample volume weights of 30 g from seven samples following methods from Baker et al. (1999) using selective enrichment for *E. coli* O157:H7 and quantitative PCR screening for stx1 and stx2.

Results to Date

Currently the research and extension team are coordinating with product suppliers to obtain raw product samples for microbiological assessment. Additionally, the team has secured University of Arizona land for time/temperature trials as well as storage experiments. Background HTPPs samples have been secured for the research team to conduct initial evaluations and refine method recoveries for each organism. The project team is also currently collecting information of current industry practices related to BSAAO use including stockpiling, timing of incorporation as well as timing of irrigation that may cause pathogen/indicator persistence or re-growth. Once final practices are confirmed with industry, the research team will implement full field scale assessment. Initial methodological assessments of increased sample weights (≥30 g) indicate that variable recovery efficiency and that once irrigation water is applied to finished products, incidence of pathogens can increase from non-detect per 30 grams to 49% presumptive positive per 30 grams after 7 days.

Benefits to the Industry

Overall, this project will improve understanding of the impact of industry practices on organisms that may commonly be present in crop inputs and strategies to reduce those risks. Measures of success will include the assessment of BSAAOs and HTTPs commonly used by the produce industry as well as evaluation of what specific industry practices are associated with enhancing (or diminishing) the presence, persistence, and potential transmission of pathogens to produce. This project will also aid in our understanding of the impact weather events have on pathogen presence, persistence, and potential transmission. Finally, we will identify composting and storage conditions associated with pathogen conversion to the VBNC physiological state, and what storage conditions increase the chance of resuscitation to normal state.

Description	Test Substance	Test Organisms
Input	1. Heat Treated Poultry Pellets - HTPP (n=2); 2. Cattle Compost (n=2)	gEC/FC/TC, *Salmonella*, STEC, O157:H7, and *Enterobacter*
Storage	1. Large Volume Stockpile; 2. Field-Stacking	gEC/FC/TC, TVS353, and MS2
Grower BMPs	1. Immediate Incorporation; 2. Delayed Incorporation; 3. Irrigation with 6hrs; and 4. Delayed irrigation 48hrs	gEC/FC/TC, TVS353, and MS2

Table 1:
Outline of sample description, test substance, and analyte (organism)

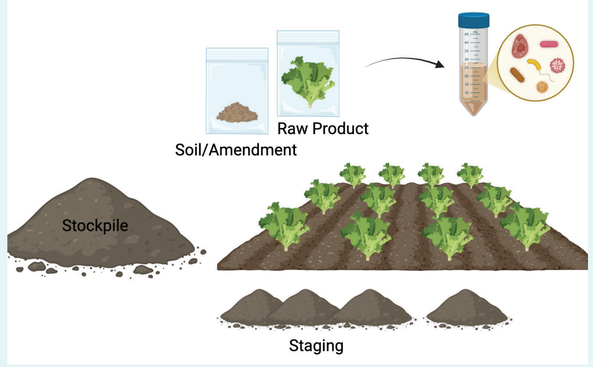

Figure 2:
Schematic of Project Design for Stockpiling and Staging

Figure 3:
Land Application of Heat-Treated Poultry Pellets (HTPPs) Prior to Incorporation

Contact

Channah Rock
University of Arizona
channah@cals.arizona.edu

Authors

Kerry Cooper and Erika Austhof, University of Arizona

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