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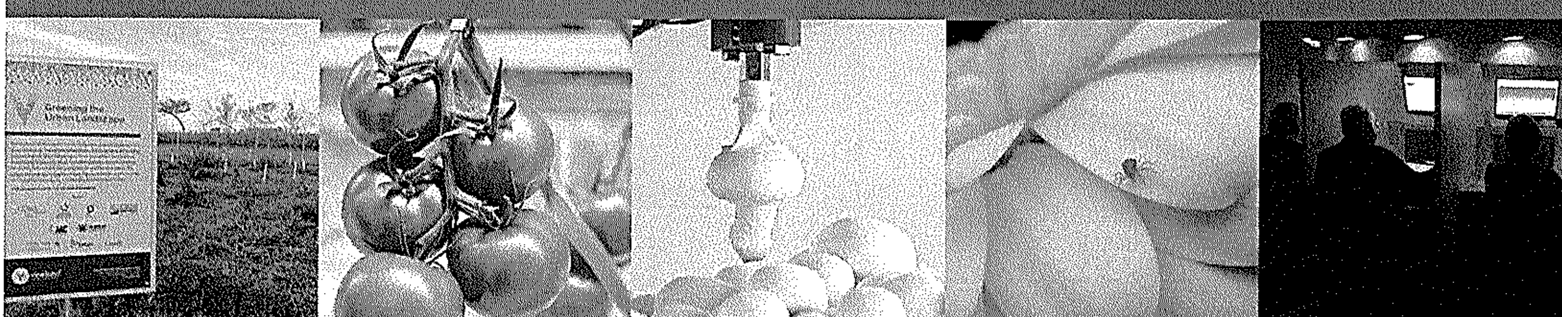

Flume Repurposing Research Project

Exploring the application of flume repurposing in vegetable processing

[INTERNAL] Interim Report delivered to the Ontario Tomato Research Institute

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Executive Summary

Vegetable processing companies generate large volumes of soil sediment through the washing and preparation of field grown vegetables harvested from multiple farms. The resulting sediment is generally landfilled at a cost to processors given that it is classified as industrial waste. With its current classification, the aforementioned sediment or 'flume' can not be reused without a Non-Agricultural Source Materials (NASM) application and corresponding approval. Where a single 150,000 square meter processing facility can generate over 2 million kilograms of flume per year, there is a need to identify alternate pathways for the reuse and application of this material, so as to reduce cost to processors and create opportunities for a more sustainable and environmentally responsible practice at an industry level.

In May 2023, Vineland Research and Innovation Centre (Vineland) entered into an agreement with the Ontario Processing Vegetable Growers, Tomato Research Institute to evaluate the prospective re-use and application of flume sediment derived through vegetable processing. The objective of this project is to evaluate the physical, chemical, biological, biochemical and hydrological properties of flume material for the purpose of reducing odor and clarifying potential reuse opportunities, with particular focus on horticultural and landscape applications.

This report covers activities related to characterization, testing and analysis of flume material conducted by Vineland between May and November 2023. Vineland prepared, processed and analyzed flume samples to present the current interim report outlining progress and key findings derived through a range of project activities.

The current report includes:

- An overview of Vineland's comprehensive literature review, highlighting key findings and resources used to inform the material testing and evaluation and biochemical characterization of flume
- An update on Vineland's progress as it pertains to material testing and evaluation of the physical, chemical, biological and hydrological properties of flume
- A preliminary characterization of the biochemical properties of flume, including high level recommendations for controlling flume odor using additives

Project Description

Although there is precedent for the reuse and application of flume as a feedstock or base material in the development and production of soil products, opportunities to divert flume to soil suppliers interested in using the sediment to develop soil materials are currently limited by the various approvals processes upheld by the Ontario Ministries of the Environment, Conversation and Parks and Agriculture, Food and Rural Affairs. This particular reuse application requires that an Environmental Compliance Approval (ECA) be granted to permit the transfer of flume between vegetable processors and soil suppliers. Additional research is required to identify novel opportunities for the reuse and application of flume at an industry level, as well as to clarify the processes and requirements for preparing and submitting NASM and ECA applications as it relates specifically to the flume generated by the vegetable processing industry. Additional research is also required to support the development and implementation of comprehensive strategies for reducing the severity of flume odor through the use of additives, which may contribute toward minimizing or eliminating the often harsh, unpleasant smell associated with flume derived through vegetable processing.

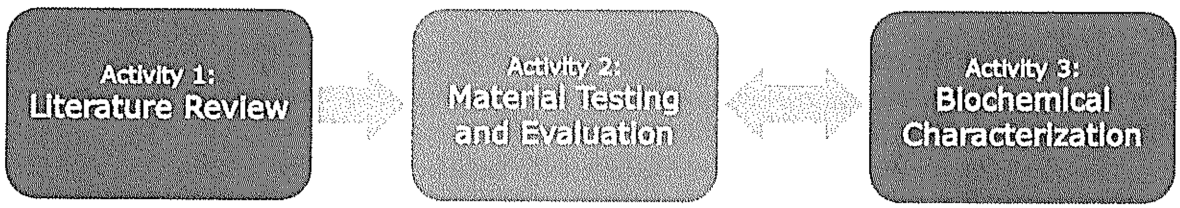


Figure 1. Overview of project process used to evaluate flume for the purpose of reducing odor and clarifying potential reuse opportunities, with particular focus on horticultural and landscape applications. Note that Activities 2 and 3 were conducted concurrently by Vineland's Plant Responses and the Environment and Biochemistry teams, respectively.

The Flume Repurposing Research Project consists of three activities, namely (i) Literature Review (ii) Material Testing and Evaluation and (iii) Biochemical Characterization.

1. **Activity 1, *Literature Review*** involves the comprehensive review of existing literature to identify empirical, evidence-based research that can be used to inform the Materials Testing and Evaluation (Activity 2) and Biochemical Characterization (Activity 3) of flume derived through vegetable processing.
2. **Activity 2, *Materials Testing and Evaluation*** involves the preparation and comprehensive testing of key physical, chemical, biological and hydrological properties of flume. Testing and evaluation will work to establish an empirically derived evidence-base that can be directly used to inform the potential reuse and application of flume at an industry level.
3. **Activity 3, *Biochemical Characterization*** involves the testing and evaluation of the baseline biochemical properties of flume to support the effective characterization of prospective odor mitigating additives. Biochemical characterization will be used to inform the management of flume material, particularly as it relates to controlling flume odor.

Preliminary findings corresponding to each of the aforementioned activities are presented in the current report. Full reporting, including all data, analysis and recommendations will be provided at a later date, in Vineland's final report scheduled to be delivered in January 2024.

Activity 1: Reviewing Alternative Options for Flume Repurposing

Milestone Deliverable 1: Literature Review

- Overview of key findings identified in the literature, with references to the primary information sources
- Interpretation and relevant commentary on published methodologies and results
- List of opportunities and challenges related to the potential re-use of flume, accounting for current regulatory frameworks and market opportunities

Process

In July 2023, Vineland initiated a comprehensive review of existing academic and technical literature to identify resources that could be used to inform the reuse of flume derived through vegetable processing for horticultural applications in agricultural and horticultural production environments. Vineland focused the literature search on resources published between 1995 and present. The literature review surveyed peer-reviewed, scientific literature obtained using academic resource databases, as well as technical articles such as theses and trials that are not peer reviewed. The search was conducted by identifying and inputting key phrases and sorting the resulting resources by relevance until no new relevant articles emerged.

Keywords used for the search are outlined below:
Keyword List:

- "Flume"
- "Tomato washwater"
- "Fruit and Vegetable"
- "Washing sediment"
- "Agricultural waste"
- "Mud Waste"

AND

- "Reuse"
- "Repurposing"
- "Processing"
- "Management"
- "Regulations"

Project Timeline

June 2023 — Finalized the coding structure for the literature review to generate a template with instructions including available resource libraries, keywords, and the appropriate date range of publications.

July 2023 — Due to the lack of research on the reuse of flume sediment from tomato processing two related fields were identified as applicable to the current review, namely **(i) fruit and vegetable washwater quality** and **(ii) sediment reuse in agriculture**.

August 2023 Collected a range of academic and technical literature to formulate an evidence-based database.

September-October 2023 Reviewed literature to identify key findings resources integral to effectively evaluating opportunities related to the reuse and application of flume sediment in agricultural and horticultural production.

Key Findings

1. **Requirements for field application of agricultural washwater.**
Wash-water from fruit and vegetable processing is regulated by the Ontario Ministry of the Environment, Conversation and Parks (MECP) as it has the same degree of impact on the environment as industrial wastewater. This is due to the high content of soil, organic matter, and pathogens in washwaters that can negatively impact aquatic ecosystems, wildlife, and humans when released into water bodies untreated. A common example of these impacts is eutrophication of water bodies, where algae grows in the water due to the excessive concentrations of nitrogen and phosphorus. In addition, washwaters with high levels of organic loads can consume oxygen from water bodies, negatively affecting the aquatic ecosystem and its fish species. To mitigate these effects, washwaters must be treated to meet regulatory standards before being released into the environment. The direct field-application of tomato washwater will require an Environmental Compliance Approval (ECA) from the MECP or a Non- Agricultural Source Material Plan (NASM) from Ontario Ministry of Agriculture, Food, and Rural Affairs (OMAFRA). To meet these standards water quality parameters should be monitored for environmental and food safety purposes including water clarity, nutrient content, organic matter, dissolved oxygen, pH and microbiological levels.

2. **Baseline properties of fruit and vegetables washwater.**
Samples of raw sewage were collected from two facilities located in the Podlasie province. One facility processed fruit and vegetables, producing mainly juice of fruit and vegetable purée, and the other processed dairy products,mainly hard cheese, yogurt and cottage cheese. The properties of the wastewater samples were characterized and compared to municipal wastewater. It was determined that fruit and vegetable waste had higher values of BOD5 and CODCr and comparable levels of nitrogen and phosphorus.

Table 1. The average monthly values for raw sewage (Puchlik & Struk-Sokolowska, 2017).

Parameter	Unit	Raw wastewater					
		Fruits and vegetables processing			Dairy		
		V	VI	VII	V	VI	VII
BOD	mgO_2/dm^3	860	2 200	3 200	1 220	2 800	3 900
COD	mgO_2/dm^3	919	3 350	3 700	1 680	3 300	5 420
$N_{tot.}$	mgN/dm^3	40.0	48.0	60.0	76.0	69.0	87.0
$P_{tot.}$	mgP/dm^3	9.4	14.8	16.0	12.6	16.0	22.0
TSS	mg/dm^3	249	356	420	200	245	286
pH	-	5.5-7.2	4.3-7.1	4.6-7.9	6.1-9.3	6.6-9.0	6.8-9.4

Puchlik, M., & Struk-Sokołowska, J. (2017). Comparison of the composition of wastewater from fruit and vegetables as well as dairy industry. *E3S Web of Conferences*, *17*, 77-. https://doi.org/10.1051/e3sconf/20171700077

Potential applications of fruit and vegetable washwater.
There are multiple opportunities identified within the literature for washing sediment re-use in agriculture, forestry, and horticulture. One opportunity identified to have high potential is the use of this sediment in the production of manufactured topsoil and plant-growing media for application in nurseries, landscaping, parks, sport pitches, wetland construction, brownfield redevelopment, and restoration of mining sites. More specifically, the reuse of washwater as a fertilizer or soil amendment has had increasing interest as it can improve the fertility of soils under conventional farming practices and in the recycling of nutrients in crop production.

Highlighted Resources

1. Bertoldi, B., Bardsley, C. A., Baker, C. A., Pabst, C. R., Gutierrez, A., De, J., Luo, Y., & Schneider, K. R. (2021). Determining Bacterial Load and Water Quality Parameters of Chlorinated Tomato Flume Tanks in Florida Packinghouses. *Journal of Food Protection, 84*(10), 1784–1792. https://doi.org/10.4315/JFP-21-100

- Flume tank water samples were collected from three packing houses in Florida and analyzed for pH, total dissolved solids (TDS), free chlorine, chemical oxygen demand (COD), oxidation-reduction potential, and turbidity.
- Additional flume water samples were collected and analyzed for total aerobic plate count (APC), total coliforms (TC), and *Escherichia coli*.

- Fresh tomatoes were collected before and after washing and tested for total aerobic plate count (APC), total coliforms (TC), and *Escherichia coli*.

2. Rushing, J. W. (Clemson U., Cook, W. P., & Spell, L. (1995). Accumulation of pesticides in tomato packinghouse wastewater and the influence of integrated pest management on reducing residues. *HortTechnology (Alexandria, Va.)*, *5*(3), 243–247. https://doi.org/10.21273/horttech.5.3.243

- The analysis of water from six commercial tomato packinghouse dump tanks in South Carolina identified that metal and pesticide residues accumulate in the dump-tank water during daily operation.
- The amount of metal and pesticide residues varied: Asana (esfenvalerate), 0.3 to 13.8 ppb; Bravo (chlorothalonil), 0.1 to 2.7 ppm; copper, 2.0 to 7.3 ppm; and manganese, 0.1 to 2.5 ppm.
- Contamination decreased when growers implemented integrated pest management (IPM) in their production practices.

3. Lucia, C., Pampinella, D., Palazzolo, E., Badalucco, L., & Laudicina, V. A. (2023). From Waste to Resources: Sewage Sludges from the Citrus Processing Industry to Improve Soil Fertility and Performance of Lettuce (Lactuca sativa L.). *Agriculture (Basel)*, *13*(4), 913-. https://doi.org/10.3390/agriculture13040913

- To assess the effect of Citrus Sewage Sludge (CSS) applied at different concentrations (2.5, 5, 10 t ha−1) on soil fertility and lettuce performance.
- The CSS amendment improved soil fertility by increasing total organic C, and, at the highest dose, P availability and microbial biomass C.
- The resulting increase in soil fertility and soil microorganisms, lead to an increase in lettuce biomass.

4. Vidal-Beaudet, L., Charpentier, S., & Rossignol, J. P. (2009). Physical and mechanical properties of washed sediment mixed with organic matter. *Soil Use and Management*, *25*(2), 141–151. https://doi.org/10.1111/j.1475-2743.2009.00209.

- The physical and mechanical properties of the non-structured washed soil and the effectiveness of organic matter amendments (peat and green waste compost) were evaluated.
- When compressed, large pores (radius >1500 lm) disappeared in the washed soil mixtures but were still observed in the control and were maintained by aggregate stability.
- The additions of organic matter improved all the washed soil properties by increasing structural porosity and vertical stress resistance.

5. Kiani, M., Raave, H., Simojoki, A., Tammeorg, O., & Tammeorg, P. (2021). Recycling lake sediment to agriculture: Effects on plant growth, nutrient availability, and leaching. *The Science of the Total Environment*, *753*,

141984–141984. https://doi.org/10.1016/j.scitotenv.2020.141984

- Plant growth conditions were improved from the addition of lake sediment as treatments had nearly double the phosphorus uptake of ryegrass compared to the control soil.
- To improve plant yield and soil nutrient status without increasing phosphorus and nitrogen leaching from the soil a 75-cm thick layer of sediments was applied on the agricultural sandy loam soils surrounding the lake.
- In addition, a 2-cm layer of biochar between the sediment and soil reduced P and N leaching by 50%.

Activity 2: Material Testing and Evaluation

Milestone Deliverables 2: Soil Health Reports, Recommendations for Use and Application of Flume, Environmental Compliance Approval (ECA) Application Guidelines for Flume Repurposing, Non-agricultural Source Materials (NASM) Application Guidelines for Flume Repurposing

- Two comprehensive Soil Health Reports detailing physical, chemical, biological and hydrological properties of flume sediment provided by Conagra and Highbury Canco
- Recommendations for the prospective use and application of flume based on its observed properties
- Comprehensive guideline outlining the various steps required to complete an ECA or NASM applications to permit the use of flume in agricultural/horticultural applications

Experimental Design

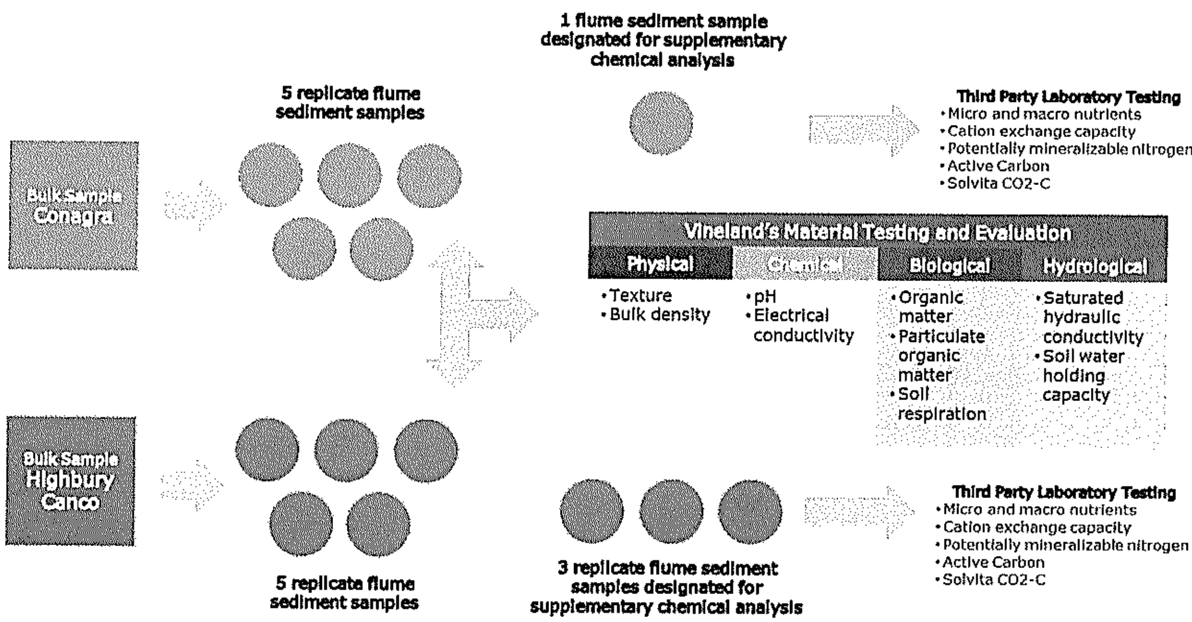


Figure 2. Overview of experimental design used for material testing and evaluation of flume sediment

- 5 replicate samples of material provided by both Conagra and Highbury Canco were used to evaluate the physical, chemical, biological and hydrological properties of flume sediment derived from vegetable processing
- 3 replicate samples of the material provided by Highbury Canco were used to evaluate the supplementary chemical properties of the flume sediment, including Solvita CO_2-C, potentially mineralizable nitrogen, active carbon and macro and micro nutrient content. Only 1 sample of the material provided by Conagra was submitted for supplementary chemical analysis, where

- The data and information derived through the characterization of physical, chemical, biological, hydrological and supplementary chemical properties will be used to inform the prospective use and application of flume sediment, allowing Vineland to provide detailed recommendations for the transfer and management of material as it relates to its use in horticultural and landscape applications
- With an understanding of the inherent properties of flume sediment and its recommended use and application, Vineland will develop a step by step guideline outlining the information required to complete either an ECA or NASM application to permit the use of fume according to its intended reuse application.

Key Analysis Methods

The following indicators will be used to characterize the baseline physical, chemical, biological and hydrological properties of flume sediment provided by Conagra and Highbury Canco:

Indicator		Description
Physical	Texture	Refers to the total sand, silt and clay content of soil. Measured as a percent of the total soil mass (%). Known to influence drainage, water-holding capacity, nutrient retention and availability.
	Bulk Density	Refers to the dry weight of soil, divided by its volume. Indicator of soil compaction, negatively correlated with root extension and growth.
Biological	Organic Matter	Refers to the various organic constituents of soils, including living organisms, plant residues, detritus and humus. Known to regulate soil structure, water holding capacity and nutrient content while serving as a vital food source for soil microbial and fungal communities.
	Particulate Organic Matter	Refers to active, unprocessed organic matter typically associated with soil microbial and fungal communities.
	Soil Respiration (respiration over 96 hours)	Refers to the measure of carbon dioxide (CO_2) released through both the decomposition of organic matter and respiration of plant roots and other below ground biota.
	Solvita CO_2-C (respiration over 24 hours)	Refers to the measure of carbon dioxide (CO_2) released through the decomposition of soil organic matter and respiration of plant roots and other soil borne organisms. Measured in milligrams per gram (ppm).
	Potentially Mineralizable	Refers to the fraction of organic or unavailable nitrogen that is converted into the mineral form.

	Nitrogen	Used as an indicator the capacity of a soil's microbial community to convert nitrogen tied up in complex organic residues into the plant available form of ammonium. Measured in microgram nitrogen per gram of soil per week (µg N/ g soil/ week).
	Active Carbon	Refers to the fraction of soil carbon that is readily available to a soil's microbial community. Measured in parts per million (ppm).
Chemical	pH	Refers to the acidity or alkalinity of soil. Indicator of nutrient availability and loss.
	Electrical Conductivity	Refers to the conductivity of soil, used as an indicator of soil nutrient content.
	Macro and Micro Nutrient Content	Refers to the relative measures of vital plant nutrients including phosphorus (P), potassium (K), magnesium (Mg) and calcium (Ca), sodium (Na), sulfur (S), boron (B), copper (Cu), manganese (Mn), iron (Fe), zinc (Zn) and aluminum (Al).
	Cation Exchange Capacity	Refers to a soil's ability to hold and exchange cations. Used as an indicator of soil fertility, where higher CEC coincides with increased availability of Ca, K and Mg in soil. Measured in centimoles of positive charge per kilogram of soil (cmolc/kg).
Hydrological	Saturated Hydraulic Conductivity (KSat)	Refers to the ease with which pores of a saturated soil material transmit water. In-lab measurement is used to characterize the rate at which water moves through materials that are completely saturated. Measured in centimeters per day (cm/day).
	Soil Water Holding Capacity (HYPROP)	Refers to the tenacity with which particles of soil retain water. In-lab measurements are used to generate soil moisture release curves that estimate the total force required to extract water from soil across the entire range of soil moisture. Measured in megapascals (MPa).

Project Timeline

May 2023 Vineland identifies that approximately 20 kg of air dried flume sediment is required to support Activities 2 (Material Testing and Evaluation) and 3 (Biochemical Characterization). Vineland requests the required flume samples from project partners (Conagra and Highbury Canco)

September 21st 2023 20 kg of saturated flume sediment or 5 kg of air dried flume sediment received from Conagra

September 26th 2023 40 kg of air dried flume sediment received from Highbury Canco

September 26th – November 8th 2023 Conagra flume sediment was air dried at room temperature until constant weight was achieved (approximately 6 weeks of drying).

NOTE: The flume sediment provided by Conagra was not air dried prior to delivery. Accordingly, the 20 kg sample of saturated flume sediment was dried at Vineland, delaying Activity 2: Material Testing and Evaluation by approximately 6 weeks. The 20 kg sample of saturated flume sediment equated to approximately 7 kg of air dried material, as a result of which the experimental design has been modified to accommodate reduced sample volume. Given an insufficient amount of flume sediment, Vineland will conduct comprehensive testing using fewer replicates to characterize the properties of the flume sediment provided by Conagra.

November 8th 2023 Comprehensive testing and evaluation of the physical, chemical, biological and hydrological properties of flume sediment initiated by the Vineland Team. Sub-samples prepared and sent to a third party laboratory for supplementary chemical analysis.

December 2023 Vineland to complete comprehensive and reduced testing and evaluation of flume sediments. Vineland will use the data and information derived through Material Testing and Analysis to generate (i) soil health reports, (ii) recommendations for use and application of flume, (iii) environmental compliance approval (ECA) application guidelines for flume repurposing and (iv)non-agricultural source materials (NASM) application guidelines for flume repurposing.

January 2024 Vineland to deliver reporting for all activities to Ontario Processing Vegetable Growers, Tomato Research Institute

Key Findings

1. Preliminary handling of flume suggests that opportunities for the r**euse and application of *saturated* flume sediment as a feedstock or base material in the development and production of soil products** is limited in scope.

2. **There is a need for material testing and evaluation guidelines to support the efficient and effective submission of NASM applications**
The reuse and application of flume sediment derived through vegetable processing will benefit from clear, comprehensive guidelines to support the transfer of materials from processors to field producers, who could utilize flume sediment to

supplement loss of mineral soil in field operations. Where NASM approval is required to facilitate such opportunities, Vineland will provide comprehensive guidelines outlining the various steps required to complete a NASM application to support the use of flume in agricultural/horticultural applications.

Activity 3: Biochemical Characterization

Milestone Deliverables 3: Biochemical Characterization, Additive Impact and Recommendation Report

- Detailed characterization of the baseline chemistry of flume sediments provided by Conagra and Highbury Canco using mass spectrometry (GC-MS, LC-MS)
- List of potential strategies for controlling flume odor using additives, including a detailed characterization of chemical changes that might be affected by the incorporation of various additives into flume sediment
- Recommendations as to which additives warrant further evaluation, which may include a detailed characterization of the impacts of said additives on the volatile and non-volatile biochemical components of flume sediment

Experimental Design

- Multiple technical replicates (aliquots) of the bulk samples provided by the two (2) sources were used for characterization samples/replicates
- Mass spectrometry-based chemical analyses were used to characterize similarities and differences between the flume samples from the two sources.
- Chemical components will be identified that might underlie malodorous attributes, or influence potential uses for repurposed flume.

Key Laboratory Methods

The following analytical methods were used to characterize and compare the baseline biochemical compositions of flume sediment provided by Conagra and Highbury Canco:

Gas chromatography–mass spectrometry **GC-MS**	• Odor-causing volatile chemicals emitted from flume samples were trapped and concentrated, and primary chemical components were identified by GC-MS • This provides a comprehensive analysis of organic aroma chemicals that might contribute to odor • This "volatile" chemical fingerprint contributes to understanding the source of odorant chemicals in flume, and ways to mitigate their production
Liquid chromatography–mass spectrometry **LC-MS**	• Water-soluble extracts of flume samples were analyzed using untargeted LC-MS analysis, providing a high-resolution profile of the organic chemicals present • This approach provides a comprehensive profile of thousands of flume-associated chemicals • This fingerprint of non-volatile chemical helps identify potential sources of odorant chemicals identified by GC-MS, as well as other plant-, or microbe-derived chemicals that could affect potential uses in the re-purposing of flume.

Project Timeline

September 21st 2023	20 kg of saturated flume sediment or 5 kg of air dried flume sediment received from Conagra
September 26th 2023	40 kg of air dried flume sediment received from Highbury Canco
October 2023	LC-MS and GC-MS profiling of flume sediment
October - November 2023	Data analysis and reporting of Biochemical Characterization results

Preliminary Results

Volatile profiling identifies toluene as a primary component of wet flume

- The volatile aromatic hydrocarbon toluene was the primary component (>75%) of volatile organic chemicals collected from wet flume samples provided by Source 1 (Figure 3A). Methylphenol (cresol), a microbial metabolite of toluene, was also abundant (~20%) (Figure 3A).
- Flume from Source 2, which was dry, did not release detectable levels of toluene or methylphenol (Figure 3B).

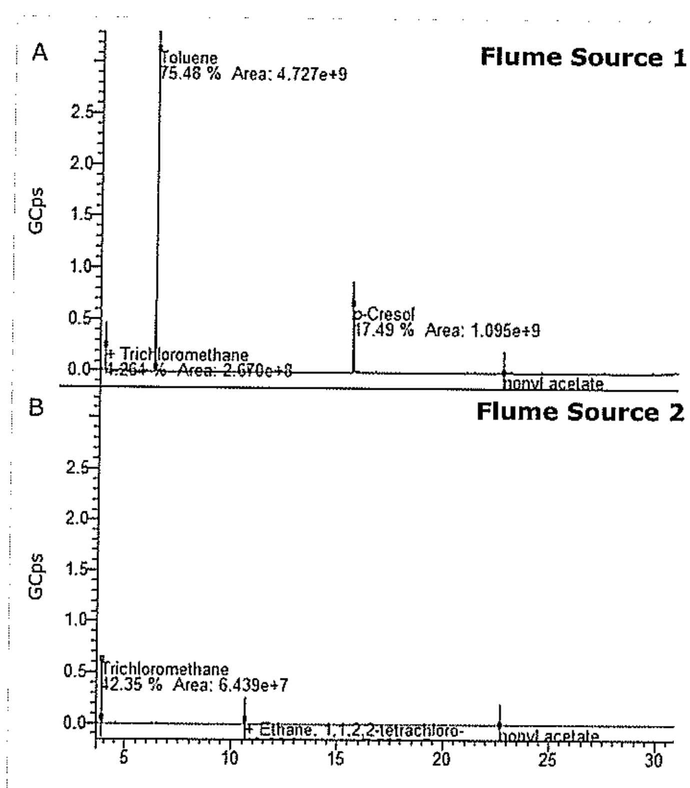


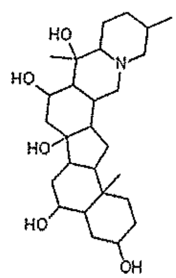
Figure 3. Volatile profiling for flume sources 1 and 2

LC-MS metabolomics reveals substantial steroidal alkaloid content in flume

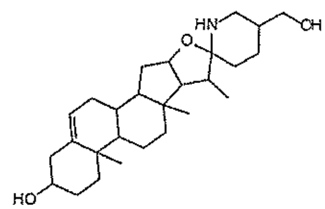
- Three (3) of the most abundant chemicals detected at similar levels in flume samples from both sources belong to a class of tomato-derived defense biochemicals called steroidal alkaloids, known for their antibacterial and antifungal properties (Table 2).

Table 2. Overview of most abundant chemicals detected across both flume sediment samples

Compound	Formula	Mass Error (ppm)	Putative ID	Class
19.33_463.3293n	$C_{27}H_{45}NO_5$	-1.1	Pingpeimine A	steroidal alkaloid
22.38_429.3237n	$C_{27}H_{43}NO_3$	-1.4	Solaparnaine	steroidal alkaloid
21.13_463.3292n	$C_{27}H_{45}NO_5$	-1.2	Pingpeimine A	steroidal alkaloid



Pingpeimine A



Solaparnaine

Key Findings

1. **Toluene was the primary volatile chemical in wet flume from Source 1,** likely a product of microbe-mediated anaerobic fermentation when flume is in storage.
2. **Flume from both sources contained steroidal alkaloids as abundant components,** specialized metabolites from tomato with antibacterial and antifungal properties.
3. **Mitigation of anaerobic fermentation when flume is in storage** might reduce toluene production, and emission of its malodorous metabolites.