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# Deploying microbes as a seed treatment for protection against soil-borne plant pathogens Final report submitted to the Organic Farming Research Foundation Rick Carr, primary investigator

## **1. Project Summary**

Plant diseases, especially those caused by soil-borne seed infecting pathogens are a serious constraint to both greenhouse and field production of many agricultural crops. Conventional farming operations often use fumigants and chemical seed treatments for controlling plant pathogens. However, these materials can be harmful to human health and the environment. The use of many of these materials is also strictly prohibited in organic agriculture, limiting the options for plant disease control. Organic amendments such as compost and vermicompost are used as alternatives to synthetic control methods due, in part, to their success in controlling plant pathogens. Previous studies have confirmed consistent disease suppression using solid and liquid forms of organic amendments and the working hypothesis is that microbes are closely associated with disease suppression. Furthermore, only a subset of microbes from the bulk material that colonize the seed coat are responsible for disease suppression. So if the microbes found in composted substrates are deployed as a seed treatment, can we still achieve protection from soilborne plant pathogens? In addition, can this seed treatment be developed for organic production as an effective tool for disease management? The goal of this project was to establish a proof-ofconcept that compost microbes can be deployed on the surface of seeds before sowing to protect against soil-borne plant pathogens, particularly Pythium aphanidermatum causing damping-off, a common and ubiquitous soil-borne plant pathogen. Liquid compost extracts were produced from solid materials, freeze-dried to a powder form, and then applied to the seed coat using standard seed-coating techniques. Average recovery from freeze-drying was 1.05 mg ml<sup>-1</sup> ( $\pm$  0.63). Freeze-dried compost extracts (FDCE) were coated on cucumber seeds using a rotary pan coater at two different application rates, 1.5% and 6% by weight. Plant disease bioassays were conducted to examine the ability of seeds treated with FDCE in suppressing disease. Treated seeds were sown in sterile sand and then inoculated with P. aphanidermatum zoospores; nontreated seeds were used as a control. Three replicate bioassays were completed. Seeds treated with 1.5% FDCE were unable to suppress disease in all bioassays as measured by seedling dry weight and percent seedling emergence. Seeds treated with 6% FDCE in one bioassay showed signs of disease suppression as seedling emergence rose to 42%, still significantly lower than non-inoculated FDCE-treated seeds; however, this could not be repeated in subsequent bioassays. While 42% seedling emergence in any production system is unacceptable, these results show a dose response in the rate of FDCE application on the seed coat - the greater the amount of FDCE application, the greater the level of suppression. Continuing the progress of this project and exploring higher rates of seed treatment application would discover the full extent to which FDCE can suppress infections caused by P. aphanidermatum and provide a potentially promising tool for plant disease suppression in organic agriculture.

#### 2. Introduction to Topic

Each year state extension offices, university agricultural programs, federal and nongovernmental organizations conduct regional or national surveys regarding organic agriculture production, yields, farmer restraints and concerns, and other farmer activities. In addition, it's not uncommon for regional and national conferences to hold breakout sessions in order to identify stakeholder goals, needs and issues. For example, during the 2015 Organic Agriculture Research Symposium, seed producers and researchers had listed plant diseases as a serious constraint to crop seed systems [1, 2]. Members of the Organic Seed Alliance, a 501 (c)(3) organization that "advances the ethical development and stewardship of the genetic resources of agricultural seed", agreed that expanding the options for controlling seed and seedling pathogens would strengthen organic seed production and direct-seeding practices (J. Zystro, personal communication, 2015). Responses from national organic farmer surveys also can be used to illustrate grower needs and issues. In the Fourth National Organic Farmers' Survey (2004) conducted by the Organic Farming Research Foundation (OFRF), production losses due to pests or diseases was among the top eight responses when respondents were asked to identify problems by various production, marketing and regulatory conditions [3]. More recently, the OFRF released their Impacts from OFRF grants: 2006-2014, which illustrated critical stakeholder needs as indicated by OFRF-funded projects [4]. Between 2006 and 2014, among eleven different research commodity types, the most number of OFRF-funded projects were on vegetables, with disease management among the top five funded research topics [4]. These facts and figures represent a specific stakeholder need within the organic agricultural industry.

Plant losses due to the lack of control of soil-borne plant pathogens in organic greenhouse and field crop production systems can be substantial in a given year. For instance, the 2010 USDA

Risk Management Agency Report cites total indemnity for insured crop losses on certified organic farms at \$69 million (2004-2009)ratio of and a loss 0.90 (indemnity compensation/premiums paid) for the same period [5]. This compares to a loss ratio of 0.58 for conventional agriculture in the same counties where certified organic is grown for the same period (i.e., lower indemnity as a percent of premiums paid) [5]. According to the Organic Trade Association, US organic sales reached \$39 billion in 2014, growing at 10% over 2013. It is likely that these losses will increase with increased demand for organic products and subsequent increased acreage under production. Seed and seedling infecting pathogens in the genera Pythium, Phytophthora, Rhizoctonia, and Fusarium species causing damping-off are particularly devastating in vegetable operations or where direct-seeding is practiced (Figure 1). Effective control of soil-borne plant pathogens is often accomplished through the use of fumigants or chemical seed treatments. However, these materials can be potentially harmful to other organisms and the environment [6-9]. Currently, there are few effective products for certified organic agriculture to combat seed and seedling diseases.

**Figure 1**. Damping-off observed in two varieties of chard grown under greenhouse conditions. Photo credit R. Carr.



Public demand for organically-grown food has stimulated research into the use of organic amendments such as compost for disease management [10]. Many scientists and growers alike have long recognized the importance of maintaining healthy soil for producing healthy plants. In fact, for over 60 years, researchers and outreach educators from the Rodale Institute in Kutztown, PA have been working with J.I. Rodale's philosophy (founder of the Rodale Institute) that, "healthy soil = healthy food = healthy people." Compost and other similar organic soil amendments are commonly used for improving the nutrient and microbial diversity of agricultural soils. In general, these materials can improve the health of the soil by increasing organic matter, plant nutrients, and microbial biomass, all of which complement soil processes such as nutrient cycling. The use of these materials in agriculture to suppress plant diseases has been well documented [11-15]. However, their mixed effectiveness in laboratory and field applications [11, 15-17] has stifled the wide-spread adoption of biocontrol materials suitable for organic agriculture. Continued research that focuses on compost-mediated disease suppression will only increase our understanding and the efficacy of organic amendments as effective tools against plant diseases in organic agriculture.

Figure 2. Representative 7-day		Non-inoculated	Inoculated
old cucumber seedlings showing the difference in disease symptoms between sand and 40% (v:v) vermicompost (VC)- amended sand [18]. All inoculated treatments received equal densities of <i>Pythium</i> <i>aphanidermatum</i> zoospore inoculum. Vermicompost batches 1 (2006), 2 (2007) and 3 (2008). Figure courtesy of Dr. Allison Jack.	Sand	THUS	and there a
	Sterile VC Batch 3	<b>VIII</b>	Tomar
	VC Batch 1	TOST	ADD
	VC Batch 2	WIPT	WW
	VC Batch 3	SUDE	TITA

It has been well documented that the microorganisms associated with organic amendments are responsible for much of the observed disease suppression [14, 19, 20] and research from Cornell University is consistent with these results (**Figures 2-3**). Studies from Nelson and colleagues [18, 21-23] have elucidated some of the mechanisms responsible for suppression of Pythium damping-off using vermicompost and municipal biosolids compost (**Table 1**). These mechanisms continue to be explored; however, one important finding has been that suppression is biologically mediated, meaning that the microbes found in compost and vermicompost are responsible for the observed levels of disease suppression. Furthermore, these data also indicate that a subset of microbes from the bulk material that colonize the seed surface are responsible for suppression (**Figure 4**) [24, 25]. Recent research also has revealed a biochemical nature of

disease suppression by which anti-microbial toxins are produced while affecting pathogen development and plant infection [22].



**Table 1**. Mechanisms of biologically-based disease suppression in the cucumber-*P*. *aphanidermatum* pathosystem researched previously by Cornell University.

- 1) Microbes disrupt chemotaxis of *P. aphanidermatum* zoospores and thus prevent seed infection [18, 21],
- 2) Microbes alter pathogen development decreasing the number of infective foci [22], and
- 3) Anti-microbial toxins are produced by certain microbes that inhibit pathogen development and infection [22].

There are several different approaches for the application of composted substrates. Solid materials can be amended to soil or potting media [26], or added to water where microbes and soluble plant nutrients can be extracted from the solid material [27] that later can be used as soil and foliar applications, commonly referred to as compost extracts or teas. Both solid and liquid materials have been shown to suppress plant diseases [14, 20, 27-30]. One advantage of using liquid extracts is that they maintain similar characteristics in disease suppression compared to the solid material [18] but have added dispersal methods.



**Figure 4.** Schematic of the spermosphere, setting for the interaction between the plant, pathogen and beneficial microorganisms. Figure courtesy of Dr. Allison Jack.

Although compost extracts can be used as soil drenches or foliar applications, transporting large volumes of water can be costly; additional costs are incurred to modify or outfit irrigation systems to handle liquids with fine particulate matter. Despite these obstacles, compost extracts are routinely applied in organic greenhouse and field production systems for nutrient and disease management. However, a previous research agenda from Cornell University and one of their industry collaborators (Worm Power, Avon, NY) was to explore a potentially promising application approach for liquid vermicompost extracts that involved freeze-drying the liquid into a fine, dry powder. When needed, the powder, representing a complex community of microorganisms, could be reconstituted back into its original liquid form at the desired concentration. The preliminary studies showed that freeze-dried extracts retained disease suppressive properties similar to that of liquid extracts (**Figure 3**); with the benefit of being more easily transported.

The use of a complex community of microorganisms deployed on the seed surface to control plant pathogens has been attempted previously with mixed results. Soaking seeds in compost extracts for hours to days at a time appears to be the most common method for "biopriming" seeds with beneficial microbes from compost; although anecdotally, this technique has been known to have negative impacts on seed germination. Kasselaki and colleagues [31] were successful in disinfecting tomato seeds infected with bacterial canker after soaking seeds in compost extracts overnight; while other researchers had first isolated potentially antagonistic bacteria from compost and vermicompost and then treated the seeds with individual bacterial strains with mixed success in plant growth promotion [32]. In order for a biologically-based seed treatment application for disease control to become adopted by the organic agricultural industry the treatment must demonstrate two essential components: consistency and predictability. Over the last several decades researchers have accomplished this goal with some success with single-strain biological control agents such as *Trichoderma* spp. and *Bacillus* spp. [31, 33]; however, as mentioned previously, these products have not been able to fill grower's needs for controlling soil-borne plant pathogens in organic agriculture.

Given the findings from preliminary studies and that seed companies commonly use chemical seed coatings for plant disease control, the Nelson Lab at Cornell University conceived the idea that the microbial fraction produced from freeze-dried compost extracts (FDCE) could be applied as a seed coating for a more directed disease control application. Since then, researchers from Rodale Institute and Cornell University have received funding from OFRF in 2014 to explore the use of FDCE as a novel technology for controlling Pythium damping-off in cucumber. The project has developed gradually and has received considerable attention – it was listed among the "top ten most visited project web pages from December 2013 to September 2015" in the Impact of OFRF Grants report [4].

Deploying microbes from freeze-dried compost extracts as a seed treatment presents a novel application approach towards biological control as well as advancing compost science and utilization. The working hypothesis for exploring FDCE technology has developed over years of research on biologically-based disease suppression (**Figure 5**). An overwhelming number of stakeholders have expressed an interest in exploring the use of FDCE for controlling seed and seedling damping-off. Throughout the course of the project, the short-term goal was to evaluate

the efficacy of FDCE in controlling *P. aphanidermatum* causing damping-off. Accomplishing this goal required identifying the optimal FDCE seed treatment application rate for suppressing disease. Once a successful treatment has been identified, the long-term goal is to commercialize the technology and make it available for the entire organic agricultural industry.

Figure 5. Working hypothesis and rationale for exploring the use of freeze-dried compost extracts for controlling seed rotting pathogens.
If solid vermicompost suppresses plant disease,
↓ Liquid vermicompost extract suppresses plant disease,
↓ Liquid extract can be freeze-dried into a powder,
↓ Freeze-dried extract can be reconstituted back to liquid form, and
↓ Reconstituted extract suppresses plant disease, then...
Can the freeze-dried extract be deployed as a microbial seed treatment and still achieve plant protection from soil-borne plant pathogens?
Can the seed treatment application be commercialized for organic production as an effective tool for plant disease management?

# 3. Objectives Statement

The current project set out to address a critical need for developing new tools for controlling seed and seedling infecting pathogens, particularly those causing damping-off. Specific stakeholder groups (**Table 1**) were engaged to outline the project goal and objectives (**Table 2**). Funding support from OFRF has been instrumental in initiating the project and accomplishing the objectives.

project.		
<u>Stakeholder</u>	Involvement	Role
• Organic vegetable producers	Field and greenhouse trials	Research and extension
• Seed industry collaborator	Providing high-quality seeds, access to controlled growth trials using varied growing conditions	Research and extension
• Cornell University	Seed treatment applications, plant disease bioassays	Research
• Kutztown University	Access to students, faculty and staff	Research and education
Rodale Institute	Compost and extract production, freeze-drying, plant disease bioassays, outreach and education	Research, education and extension

 Table 1. Stakeholders groups having direct involvement in the development of the current project.

Table 2. Ou	tline of the project goal and objectives.
<u>Goal</u> :	Develop additional tools for controlling soil-borne, seed and seedling infection pathogens in organic agriculture
Objectives:	<ol> <li>Develop a reliable seed coating procedure for treating seeds with freeze-dried compost extracts (FDCE)</li> </ol>
	(2) Access the viability of compost microbes through the freeze-drying and seed- coating procedures.
	(3) Determine the efficacy of seed-coated FDCE on cucumber for protection against <i>Pythium aphanidermatum</i> .

# 4. Materials and Methods

**Figure 6.** Generalized approach for exploring the use of freeze-dried compost extracts as a seed treatment application for controlling soil-borne plant pathogens. Steps a-e were addressed in the 2013 OFRF-funded project. Steps f-i are being proposed for a new phase of the work.

a. Prepare compost		
↓ I		
b. Prepare compost extracts		
c. Freeze-dry compost extracts (FDCE) into a powder		
A Ample EDCE ante conde et sur inc. rates		
d. Apply FDCE onto seeds at varying rates		
e. Evaluate disease suppression using rapid bioassays under laboratory conditions		
f. Evaluate FDCE performance under greenhouse and field conditions		
g. Disseminate results through research, education and extension opportunities		
h. Evaluate commercialization of FDCE seed treatment application		
i. Proceed toward FDCE commercialization		

a. <u>Compost preparation</u>

Thermophilic windrow compost was prepared outside at Rodale Institute's large-scale composting facility. Compost feedstocks included hog bed-pack from Rodale Institute and mixed leaves from the City of Allentown. The feedstocks were mixed at a ratio of 1:4 (v:v) bed-pack: leaves and put into a windrow that was 52.4 x 4.6 x 1.6 m (L x W x H). Active composting occurred between June 5 and September 21, 2014 (107 days), at which point the pile was cured for 42 days. The windrow was turned using a 3.0 x 2.1 m (W x H) Sittler Windrow Turner (model #1014) and John Deere 7530 tractor (**Figure 7**). Pile temperature was measured as

described in **Figure 8**. Initially, the compost pile was managed to achieve **USDA** NOP time/temperature certification after but accomplishing certification the pile was managed using the following qualitative indicators: temperature, moisture, particle size, odor and texture. The pile dimensions were measured after the last turn. Temperature measurements ceased after 136 days of composting. Sample collection began 150 days after composting. Compost dimensions were collected 167 days after composting in order to determine the percent decrease in volume. On April 13, 2015, bulk density was measured and on May 5, 2015 three 1  $m^3$  samples were

**Figure 7**. Thermophilic windrow compost production showing the Sittler turner and John Deer 7530 tractor at Rodale Institute, Kutztown, PA. Photo credit J. Carr.



collected in plastic bulk bags. The bags were placed inside a covered facility without temperature control. On August 31, 2015 three samples were harvested from stored compost, screened to 6.35 mm and then submitted to the Penn State Agriculture Analytical Services Lab (University Park, PA) for nutrient analysis.



#### b. Compost extract preparation

The apparatus for consistently preparing compost extracts was modified from a previously developed system [18] (Figure 9). Non-aerated compost extracts (NCE) were prepared in 227-

liter, food-grade plastic vessels at a ratio of 1:25 (v:v) compost:water. Compost was screened to 6.35 mm, added to approximately 180 L of dechlorinated water and then the total volume was adjusted to 189.2 L. Soluble nutrients and microorganisms were extracted from compost in 189.2-liter batches for 7 days at  $15^{\circ}C (\pm 3^{\circ})$  while recirculating the solution every 12 h for 30 min using a timer and submersible pump. After extraction, 10 samples of NCE, 300 ml each were collected after recirculating the solution for 1 minute. The samples were collected using the sump pump, which was located 10 cm above the bottom of the vessel. All samples were screened to 500 µm and then frozen in sealed freeze-dry glassware at an angle to create the greatest amount of liquid surface contact to the air. A total of six replicate batches of NCE were prepared; batches 2-4 were prepared to generate the freeze-dried material

**Figure 9**. Compost extract production vessels at Rodale Institute, Kutztown, PA. Photo credit R. Carr.



needed for conducting plant disease bioassays (see below) and batches 1, 5-6 were prepared for additional analyses. The bacteria colony forming units (cfu) was measured for batch 6.

#### c. Freeze-dry compost extracts

Compost extracts frozen in freeze-dry glassware were freeze-dried using a Labconco Freeze Dry System (**Figure 10**). Samples were freeze-dried at temperatures below  $-50^{\circ}$ C and a vacuum pressure near 450 x  $10^{-3}$  mBar. Dried samples were removed from the apparatus, weighed and then stored at  $-20^{\circ}$ C in sealable glass containers. The rate of recovery from three batches of NCE used for generated material for seed treatment was compared using Tukey-Kramer HSD test, JMP Pro 12.0 (SAS). The rate of recovery between three batches of NCE used for generating freeze-dried material for seed application and three batches of freeze-dried material not used for seed application was compared using Student's t-test, JMP Pro 12.0 (SAS).

**Figure 10**. Compost extract frozen in freeze-dry glassware, ready to be placed on the freeze-dry apparatus (left). Labconco Freeze-Dry System showing barrel manifold, vacuum pump, and refrigeration unit (middle) and freeze-dried compost extract in bottom of the glassware (right). Photo credits R. Carr.



d. Apply FDCE onto seeds

Organic cucumber (*Cucumis sativus*) seeds (Marketmore 76, seedlot OMG18-18-44539, High Mowing Organic Seeds,) were seed encrusted (seed coated) [34] in a laboratory-scale, rotary pan coater (model R-6, University Coating Systems, Independence, OR) (**Figure 11**). A minimum of five grams of seed was used during each batch of seed encrusting. Seeds were placed on the rotary pan; while spinning, the seeds were wetted with an aqueous dispersion of an OMRI-approved binder (SOI053, Incotec, Salinas, CA). The FDCE was combined with the binder as a slurry. During coating, filler was applied in small amounts into the barrel and dispersed by rotation onto the moistened seeds. The liquid binder was applied alternately with filler to affix FDCE to the seed surface until the desired amount of freeze-dried extract was reached. Two rates of FDCE were applied to the seed surface, 1.5% and 6% by weight. All seeds were stored at room temperature until further use.

**Figure 11**. R6 model rotary pan coater showing discharge shoot and barrel coating chamber with control unit (left) and interior view of barrel coating chamber showing rotary pan and atomizing wheel for dispersing coating (right). Photo credits A. Taylor.



## e. Plant disease bioassays

Plant disease bioassays were conducted to measure the effect of treating cucumber seeds with FDCE to suppress infections caused by *P. aphanidermatum* (Edson) Fitzp. (Pa68). Bioassays were carried out under controlled laboratory conditions. Seeds were placed in 100 ml beakers containing 40 ml of sterile sand, 10 seeds per beaker, and 5 replicate beakers per treatment. Ten milliliters of sterile sand was then placed on top of the seeds. Inoculated treatments received 10 ml of *P. aphanidermatum* zoospore suspension (5000 zoospores ml<sup>-1</sup>) prepared in sterile water as described previously [18] and non-inoculated treatments received 10 ml sterile water. All treatments were incubated at 25°C with 16 h light for 7 days. All seeds and seedlings were harvested after incubation and percent seedling emergence and dry weight were recorded. Data was analyzed using JMP Pro 12.0 (SAS).

# **5. Project Results**

#### a. Compost analysis

Hog bed-pack and leaves were actively composted for 107 days and then cured for an additional 42 days. Four days after the pile was built and turned for the first time, the average pile temperature was between 55-77°C (**Figure 12**). The pile achieved USDA National Organic Program temperature certification (NOP §205.203) fifteen days later. Five turns were completed during the NOP certification process and then the pile was turned an additional 6 times during the course of active composting. Finished compost had a bulk density of 0.73 g (cm<sup>3</sup>)<sup>-1</sup> (± 0.018). **Table 3** describes the results from three replicate compost nutrient analyses.



2014 to September 21, 2014.

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Lab. Three replicate samples were submitted for analysis.				
Analyte	Results (as is basis)		Results (dry weight basis)	
pH	$8.1 \pm$	0.00	na	
Soluble salts (1:5 w:w)	$2.04 \pm$	0.07 mmhos/cm	na	
Solids	$49.90\%~\pm$	0.01	na	
Moisture	$50.10\% \pm$	0.01	na	
Organic matter	$19.90\%~\pm$	0.00	$39.87\%~\pm$	0.45%
Total Nitrogen (N)	0.84% ±	0.00	$1.68\%~\pm$	0.03%
Organic Nitrogen	0.84% ±	0.00	$1.68\%~\pm$	0.03%
Ammonium N (NH <sub>4</sub> -N)	$2.43 \pm$	0.06 mg/kg	$493.33\%~\pm$	5.77%
	or		or	
	$0.0002\% \pm 0$	).0001%	$0.0005\%~\pm$	0.0000%
Carbon (C)	$12.14\%~\pm$	0.21%	$24.33\%~\pm$	0.21%
Carbon:Nitrogen (C:N) ratio	$14.53 \pm$	0.35	14.53 $\pm$	35.12%
Phosphorus (as $P_2O_5$ ) <sup>2</sup>	$0.51\% \pm 0.51\%$	).0272%	$0.01 \pm$	0.05%
Potassium (as $K_2O$ ) <sup>2</sup>	$0.28\% \pm 0$	).0074%	$0.01 \pm$	0.01%
Calcium (Ca)	$3.70\% \pm 0$	).0850%	$0.07~\pm$	0.22%
Magnesium (Mg)	$0.76\% \pm 0.1\%$	).0485%	$0.02 \pm$	0.08%
Sulfur (S)	$0.13\% \pm 0.13\%$	).0010%	$0.00 \pm$	0.01%
Sodium (Na)	$406.29~\pm$	11.48 mg/kg	$814.33~\pm$	20.98 mg/kg
Aluminum (Al)	$3050.41~\pm$	128.05 mg/kg	$6111.14 \pm$	192.69 mg/kg
Iron (Fe)	$5613.89~\pm$	312.23 mg/kg	$11249.59 \pm$	629.87 mg/kg
Manganese (Mn)	$397.47~\pm$	11.00 mg/kg	796.55 $\pm$	25.97 mg/kg
Copper (Cu)	$23.04~\pm$	1.17 mg/kg	$46.16~\pm$	1.75 mg/kg
Zinc (Zn)	91.47 ±	3.78 mg/kg	183.26 $\pm$	5.76 mg/kg

**Table 3**. Compost nutrient analysis completed by Penn State Agriculture Analytical Services Lab. Three replicate samples were submitted for analysis.

#### b. Compost extract analysis

Three replicate batches of NCE were prepared in time to generate the material needed for freezedrying, seed application and plant disease bioassays. Three additional batches of NCE were prepared for other analyses. The coloration varied qualitatively among the three batches. A bacteria analysis completed on the sixth batch of NCE frozen and then thawed for analysis had a total of 9.6 x  $10^{-4}$  cfu ml<sup>-1</sup>.

## c. Freeze-dried compost extract (FDCE)

A total of six batches of NCE were freeze-dried to make FDCE (**Figure 13**). The average time for freeze-drying 250 ml of NCE was 4 days. The Labconco Freeze-dry system could handle only 2 samples at a time due to the condenser overloading with ice. At times, samples needed to be rotated between valve stems to avoid ice blockage inside the barrel manifold. The average recovery rate of FDCE from NCE batches 2-4 was 0.72 g ml<sup>-1</sup> ( $\pm$  0.52); however, significant differences occurred among those recovery rates (P > 0.05). Additional batches of NCE were

prepared and freeze-dried but not used for seed treatment applications. The recovery rate was significantly different between batches used for seed treatment application (NCE batches 2-4) and those not used for seed treatment application (NCE batches 1, 5-6) (P > 0.05), indicating variability among NCE batches.



**Figure 13**. The mean amount of freeze-dried compost extract (FDCE) recovered per batch after freeze-drying. Batches 2-4 were used for seed treatment and subsequent plant disease bioassays. Bars represent standard deviation. Columns that have different letters are significantly different (P < 0.05) according to Tukey-Kramer honestly significant difference test.

## d. Seed treatment application

Untreated, organic cucumber seeds were treated with either 1.5% or 6% FDCE by weight using a laboratory-scale rotary pan seed coater (**Figure 14**). The treatment coverage was not uniform for either application rate but 6% FDCE appeared to have considerably greater coverage.



**Figure 14**. Cucumber seeds treated with FDCE at two application rates. Untreated cucumber seeds (left) treated with 1.5% (middle) and 6% (right) FDCE by weight.

## e. Plant disease bioassays

Non-treated and FDCE-treated cucumber seeds were exposed to *P. aphanidermatum* zoospore inoculum during plant disease bioassays. The mean seedling emergence and dry weight of the treatment after destructively harvesting each bioassay were recorded. Three replicate bioassays were completed. Bioassay results were not consistent among the three bioassays. During two bioassays, it is likely that disease pressure (i.e., level of inoculum) was too high to observe any differences among treatments, and therefore the FDCE treatment did not have any effect on disease suppression. However, results from one bioassay showed a dose response in the rate of FDCE application on disease suppression (**Figure 15**). These results are discussed below in detail.



**Figure 15**. Representative 7-day old cucumber seedlings from plant disease bioassays. Seeds were treated with two application rates (% by weight) of FDCE. Inoculated treatments received equal quantities of zoospore inoculum. Photo credits: Mary Ann Karp.

Non-inoculated seeds were used as a control and showed no sign of disease. Non-treated, inoculated seeds showed 100% infection. Seedling emergence increased with increasing rate of FDCE application (**Figure 16**). Seedling emergence in 6% FDCE was significantly greater than inoculated, non-treated seeds (P < 0.05, Student's t-test); however, there was no significant difference (P < 0.05) when seedling emergence was analyzed among all inoculated treatments. The mean dry weight increased significantly with increasing FDCE application among inoculated treatments (**Figure 17**). A significant difference in dry weight was measured between 6% FDCE inoculated and non-inoculated treatments.



Figure 16. Mean seedling emergence in response to seed applications of freeze-dried compost extracts (FDCE). Seeds were sown in sterile sand, inoculated with equal densities of P. aphanidermatum zoospore inoculum and then incubated at 25°C for 7 days. Noninoculated treatments received sterile water as a control. The percent FDCE application rate is by weight. Bars represent standard error. \* Indicates significant differences (P < 0.05) between inoculated and non-inoculated to Student's t-test. treatments according Among inoculated treatments, columns that have different letters are significantly different (P < 0.05) according to Tukey-Kramer honestly significant difference test.

Figure 17. Mean dry weight of treatments and seedlings) after destructively (seeds harvesting plant disease bioassays. Seeds were sown in sterile sand, inoculated with equal densities of P. aphanidermatum zoospore inoculum and then incubated at 25°C for 7 Non-inoculated treatments received davs. sterile water as a control. The FDCE seed treatment application rate (%) is by weight. Bars represent standard error. \* Indicates significant differences (P < 0.05) between inoculated and non-inoculated treatments according Student's t-test. to Among inoculated treatments, columns that have different letters are significantly different (P <0.05) according to Tukey-Kramer honestly significant difference test.

#### 6. Conclusions and Discussions

Public demand for organically-grown food has stimulated research into the use of organic amendments such as compost and vermicompost for disease management [10]. Seed and seedling infecting pathogens causing damping-off are particularly devastating because the causal agents commonly have numerous host plants. Vegetable operations or where direct-seeding is practiced are particularly vulnerable to disease outbreaks. Effective control of soil-borne plant pathogens is often accomplished through the use of fumigants or chemical seed treatments. However, these materials can be potentially harmful to other organisms and the environment [6-9]. Currently, there are few effective products for certified organic agriculture to combat seed and seedling diseases. Stakeholders such as organic growers and seed producers have long proclaimed a critical need for new techniques and technologies for disease management. Therefore, the goal of this project was to develop additional tools for managing seed and seedling infecting pathogens causing damping-off.

Compost and other similar organic soil amendments are commonly used for improving the nutrient and microbial diversity of agricultural soils. In general, these materials can improve the health of the soil by increasing organic matter, plant nutrients, and microbial biomass, all of which complement soil processes such as nutrient cycling. The use of compost in agriculture to suppress plant diseases has been well documented [11-15] and an overwhelming body of literature suggests that disease suppression is biologically mediated, meaning that the microbes found in the material are responsible for suppression.

Compost applications, either solid or liquid, are often broadly applied in agriculture for controlling soil-borne plant diseases. Perhaps a more directed application approach for disease control is to provide protection at the site of infection (i.e., the surface of the seed). A number of biologically-based seed treatments have been approved for organic agriculture, usually comprised of single-strain biological control agents. Deploying a complex community of microorganisms has never been attempted with any significant success. This project has been operating under a working hypothesis that if (1) compost can suppress disease, (2) compost extracts can similarly suppress disease, and (3) compost extract can be freeze-dried into a powder and reconstituted back into its original concentration and still maintain disease suppression, then can freeze-dried compost extract (FDCE) be applied as a seed treatment for protection against soil-borne plant pathogens?

Cucumber seeds were treated with two application rates of FDCE and then challenged with *P. aphanidermatum* causing damping-off. Although disease control was not achieved using FDCE, the results from this project have elucidated a dose response in the rate of application of FDCE – the greater the rate of application, the greater the level of disease suppression. Increasing the rate of FDCE application on the seed surface could improve disease suppression to acceptable limits; however, a bottleneck occurred in the production of FDCE, which prohibited exploration of application rates greater than 6% FDCE by weight. Variability in the rate of FDCE production from compost extracts was an additional obstacle during the project that could have influenced downstream results. New techniques have been identified to reduce variability in compost and compost extract as well as new procedures for handling compost extracts before freezing and freeze-drying so that the freeze-drying process can yield greater results.

Disease management in organic agriculture requires an entire tool box full of different management techniques. Some of these techniques could be cultural, some chemical, and others biological such as compost; however, stakeholders across the United States need additional tools for their tool box in order to adapt to changing disease and climate patterns. Increased exploration into the use of freeze-dried compost extracts as a seed treatment would reveal the full potential for suppressing devastating soil-borne plant pathogens and provide another tool for disease management.

# 7. Useful Tools, Information, and Resources for Farmers

Exploring the use of deploying FDCE as a seed treatment for controlling plant disease is still developing. Additional research would expose the full extent to which the seed treatment can suppress soil-borne pathogens causing damping-off. This concept has been presented to numerous audiences during the project with overwhelming support. Once the efficacy of the seed

treatment has been determined, and if shown to be successful at controlling disease, the technology will be shared with the entire organic agricultural industry.

A number of resources have been created to assist farmers with compost activities. These include understanding what compost is and how it can be prepared as well as uses and applications. During the time of the project, a Master Composter of Rodale Institute Training Program was launched to educate a diverse audience on the social, economic and environmental benefits of composting and how it fits into a larger picture of solid waste management. Only two of several fact sheets developed for the program have been included in **12. Photos and other Addenda**. Additional fact sheets are available upon request.

# 8. Outreach

The project idea and information was disseminated using a number of research, education and extension opportunities. Project information will continue to be released to the public as new results are generated. **Table 4** lists all the outreach events and activities that were used to disseminate project information.

**Table 4**. Research, education and extension opportunities used to disseminate project information and results.

Year	Activity/event	Audience	Туре
			Education
2016	Pennsylvania Farm Show	Consumers	
2016	Presentation: Deploying compost microbes as a seed	Composting	Research and
	treatment for protection against soil-borne plant	practitioners	education
	pathogens; USCC Conference, Jacksonville, FL		
2015	Rodale Institute Annual Field Day	Growers	Extension
2015	Presentation: Compost in agriculture; Pennsylvania	Growers	Extension
	Women in Agriculture, Rodale Institute		
2015	BioCycle publication [35]	Composting	Research and
2010		practitioners	extension
2015	Brain Bug Kutztown University interview	Children	Education
2013	Drain Dug Kutztown Oniversity interview	Ciliaren	Luucation
2015	Dedele Institute mentale an more isomnexting	Candanana	Education and
2015	Rodale Institute workshop: vermicomposting	Gardeners	Education and
			extension
2015	Presentation: The science behind compost teas and how	Landscape	Extension
	to use them; Managing Landscapes Sustainably, Cornell	professionals	
	Cooperative Extension, Ronkonkoma, NY		
2015	Rodale Institute workshop: compost teas and extracts	Growers and	Education and
		gardeners	extension
2015	Custom compost presentation for Franklin and Marshall	College	Education
2010	College students	students	20000000
2015	Custom compost presentation for PA Governor's School	High school	Education
2013		students	Laucation
2015	for Engineering and Technology at Lehigh University		D 1
2015	Custom compost presentation for Argentinian	Growers	Research and
	agricultural professionals		extension

2015	Master Composter of Rodale Institute Training Program; six week program, Rodale Institute	Gardeners	Research, education and extension
2015	Presentation: Compost and Soil Health; Collaborative Regional Alliance for Farmer Training (CRAFT), Rodale Institute	Growers	Education and extension
2015	Rodale Institute workshop: backyard composting	Gardeners	Education and extension
2015	Presentation: Deploying microbes as a seed treatment for protection against soil-borne plant pathogens; Organic Agriculture Research Symposium, La Crosse, WI	Scientists	Research
2015	Presentation: Biological control for plant disease management: Understanding the options and their effectiveness; NOFA-NY, Saratoga Springs, NY	Growers	Research, education and extension
2014	Rodale Institute workshop: vermicomposting	Gardeners	Education and extension
2014	Presentation: Compost; Spiral Path Farm Field Day, Loysville, PA	Growers	Education and extension
2014	Rodale Institute Annual Field Day	Growers	Extension
2014	Custom compost and vermicompost presentation, Valley Youth, Rodale Institute	Students	Education
2014	Presentation: Compost and Soil Health, Collaborative Regional Alliance for Farmer Training (CRAFT), Rodale Institute	Growers	Education and extension
2014	Presentation: Backyard composting; Lutron Electronics Sustainability Week, Coopersburg, PA	Consumers	Education
2014	Presentation: Impacts of compost on <i>Pythium</i> <i>aphanidermatum</i> pathogenic development: a cautionary tale for biological control; USCC Conference, Oakland, CA	Composting practitioners	Research, education and extension
2014	Rodale Institute workshop: backyard composting	Gardeners	Education and extension

# 9. Financial Accounting

Rodale Institute Budget v. Actual OFRF-Funded Deploying Microbes as a		
Expense Line Item	BUDGET	ACTUAL EXPENSES
Salaries	\$8,565	\$5,601
Fringe Benefits	1,711	1,452
Travel	\$1,106	\$595
Supplies/Testing	\$3,550	\$7,730
Total Direct Costs	\$14,932	\$15,378
Indirect Costs	\$0	\$0
Indirect Costs	\$0	\$0
Total Direct and Indirect Costs	\$14,932	\$15,378

# **10. Leveraged Resources**

No resources have been leveraged as a result of this project. However, the project idea and preliminary results have received considerable attention by growers and seed producers. This attention is being used to leverage future collaborations and project support for additional grant funding.

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# 12. Photos and Other Addenda

#### a. Compost fact sheet

#### **COMPOST FACTS**

Compost is prepared by managing aerobic decomposition of organic residuals. Compost can be made from a wide variety of starting materials; some of the more common materials among backyard composters are: yard trimming, grass, leaves, kitchen scraps, paper, manures, straw, hay, wood chips and sawdust. A complex community of microorganisms feed on compost inputs, breaking them down and releasing valuable plant nutrients. Compost typically reaches high temperatures (150°F) during peak decomposition. These high temperatures are a result intense microbial activity, particularly bacteria. As food sources become less available, the temperature of the compost pile will decrease to ambient air temperatures. At this point the pile cures and is ready for use as a soil amendment.

**Compost benefits** – there are number of benefits for composting and using compost as a soil amendment. These benefits include:

- Diverting waste from the landfill
- Creating a valuable plant resource from waste material
- Numerous plant nutrients
- Beneficial microbes
- Plant disease suppression
- Bioremediation
- Erosion control
- Increasing water holding capacity in the soil
- Increasing organic matter in the soil

#### Answers to commonly asked questions about your compost

# 1. The disposal of solid waste is a problem that can be dealt with only through municipal government action. False.

Each of us can play our individual part to reduce the volume of solid waste that we generate. Most kitchen and yard wastes can be recycled naturally through the process of composting. Composting in the yard or at a local community garden requires much less energy and is less expensive than bagging, hauling, and processing such waste through municipal landfills.

#### 2. The process of composting requires expensive shredders, containers, and other equipment. False.

Although certain hardware adds efficiency to the composting process, little investment is required. Chopping large pieces of waste with a shovel or large knife hastens the decomposition process. Leaves and stems can be broken into small pieces if processed through a rotary lawn mower. Low cost bins can be constructed from used fencing, lumber or cement blocks. Using a pitchfork facilitates turning and digging the compost.

#### 3. Compost is a valuable soil amendment for use in the garden and landscape plantings. True.

The use of compost improves plant growth by providing soluble plant nutrients and microbes that play numerous beneficial roles in the soil. Adding compost to the soil will also add organic matter, which plays a vital role many in soil activities.

#### 4. Grass clippings should be removed after cutting to improve growth. False.

Save time and money by letting short grass clipping fall back to the lawn rather than bagging and discarding them. Clippings break down rapidly and produce nitrogen. Alternatively, when gathered and added to a compost pile,



clippings can provide the nitrogen needed to break down other more woody wastes. Caution: clippings from lawns treated with pesticides or herbicides contain residues. Some of these residues may persist in the soil or a compost pile and may be harmful to other plants.

#### 5. Practically any plant material can be composted. True.

Leaves are ideal, but pieces of sod, manure, lawn clippings, fine wood chips or sawdust, straw, hay and plant refuge from the garden or the kitchen also can be used. Shredding coarse materials such as mature cornstalks and woody prunings into smaller sized pieces will reduce the length of time needed for them to decompose. Even newspapers can be composted, provided they are finely shredded, mixed with other materials, and supplied with nitrogen.

#### 6. Meat, dairy products and fatty and oily foods can be composted. Yes, but...

These materials will breakdown in most composting systems but can attract pests and fly nuisances if they are not managed properly. Compost these materials only if you are a seasoned composter and feel comfortable doing so. In order to prevent problems, thoroughly cover the materials with leaves, woodchips, sawdust, newspaper or any other carbon-rich material. Make sure that no meat or dairy products are ever showing.

#### 7. Diseased vegetable and flower plants should not be composted. True.

Diseased plants from the garden should not be used for compost if the compost is to be returned to the garden. Most diseases are killed by heating during compost formation, but unless the compost is turned frequently and allowed to remain unused for several years, some of the disease organisms may be returned to the garden with the compost.

# 8. For the composting process to occur most efficiently, special microorganisms, hormones, and activators need to be added to the compost pile. False.

The microorganisms needed to break down wastes into compost are present in great numbers in all garden soils. In fact, there usually are sufficient microorganisms floating around in the air to start the decomposition process. A few handfuls of garden soil added to the compost pile will ensure inoculation, thus eliminating the need to purchase any sort of "compost starter".

#### 9. Weeds heavily laden with seeds should not be composted. True.

Although most plants and their seeds are killed during composting, some can be returned alive to the garden with the compost, thus creating an unnecessary weed problem. Most weeds that have been pulled or cut before developing seeds can be composted. Mature, vigorous-growing perennial weeds such as quackgrass, bindweed, and nutsedge often are sufficiently hardy to survive decomposition and should not be composted.

#### **10.** Moisture is necessary for the composting process to occur. True.

The microorganisms that do most of the decomposition in a compost pile live in the film of moisture on particle surfaces. Material that is too dry will break down very slowly. However, balance is very important. Too much moisture leaves no air spaces in the pile, and insufficient oxygen will lead to bad smells. The best level of moisture leaves the material like a completely damp, wrung-out sponge. When you squeeze the material, only a few drops of water should drop out.

Contact the Rodale Institute or visit their website (<u>www.rodaleinstitute.org</u>) for more information.

b. Compost extract preparation and usage fact sheet



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# **Preparing Aerated and Non-Aerated Compost Teas**

# **Summary**

Compost teas are liquid versions of the solid compost material that are prepared in water in order to extract beneficial microorganisms and soluble macro- and micronutrients from the solid material. Perhaps an alternative name for the liquid material should be 'compost extract'; however, compost 'tea' has become the colloquial name despite also becoming a misnomer. There have been no standards on the production or terminology among practitioners and the composting industry when discussing these materials, which has made it very difficult to synthesize the costs and benefits for plant production. There is one dichotomy within the industry, however, aerated versus non-aerated. For the remainder of this document, compost teas will be referred to as either aerated (ACE) or non-aerated compost extracts (NCE). Aerated compost extracts are actively aerated, full-time using a blower, bubbler or any other device used to force air into liquid. Alternatively, NCEs are not actively aerated but occasionally mixed or stirred to re-suspend solid materials that have settled to the bottom of the extraction vessel. Despite much criticism from the industry, NCEs are not anaerobic extracts (Figure 1) but indeed will have a much lower oxygen content then aerated extracts. However, when the oxygen supply for ACEs is shut off, the bacteria and other aerobic microorganisms consume all available dissolved oxygen within minutes to hours, causing the solution to become anaerobic. The costs and benefits for each method and are outlined in Table 1. General production procedures are below followed by recommendations and warnings.

	<u>Costs</u>	<u>Benefits</u>
Aerated compost extracts	Greater production costs for apparatus	Greater bacterial populations
	Greater energy inputs	Shorter production time
	Lower stability (shelf-life)	(hours-3 days)
	More mechanics, plumbing and	
	equipment	
Non-aerated compost	Lower bacterial populations	Lower production costs for
<u>extracts</u>		apparatus
	Longer production time (7-10	Greater stability (shelf-life)
	days)	Fewer energy inputs

Table 1. Comparing the costs and benefits for producing aerated or non-aerated compost extracts.

Figure 1. Chemical and biological characterization of non-aerated vermicompost (VC) extracts with VC to water ratios from 1:5 to 1:60 (mass:mass (m:m). All extracts were prepared in 50-gallon volumes using water sterilized by reverse osmosis. Extraction period was 7 days, at which point the pH, bacteria colony forming units (CFU), dissolved oxygen (DO, ppm), and electrical conductivity (EC, mmhos cm<sup>-1</sup>). Figure courtesy of Dr. Allison Jack.



# General procedure for producing 50 gallons of ACE (volumes and ratios can be adjusted accordingly):

- 1. Obtain mature compost and screen to ¼ inch.
- For 50 gallon volumes, fill the brewing vessel (Figure 3) with ~45 gallons of water and let aerate for 24 hours. Set aside an additional 5 gallons for later. This will eliminate issues with chlorine. If using de-chlorinated water, well-water, filtered water or any other water that does not contain chlorine then no need to aerate for 24 hours.
- 3. Measure the desired amount of compost to produce the compost:water ratio. For example, to prepare 50 gallons of 1:25 (v:v) compost:water extract, place 2 gallons of extract in 48 gallons of water. A standard procedure for measuring the volume of compost is as follows: i) fill a one gallon bucket with compost, ii) lift the bucket 6 inches from the ground and drop, repeat 10 times, iii) fill the bucket to the one-gallon mark, iv) repeat steps i-iii 3 times, the last fill should be to the one-gallon mark and no more.
- 4. Place each gallon of compost (2 gallons total) into a 0.5 mm mesh bag.
- 5. Slowly add the mesh bag to the actively aerated, de-chlorinated water.
- 6. Using the 5 gallons of water set aside, fill the vessel to 50 gallons.
- 7. Actively aerate (brew) for 24 hours to 3 days.

8. After extraction, filter the solution to 0.5 mm and use the liquid immediately after brewing. If unable to use the material after the desired extraction period then continue to actively aerate until ready. Discontinuing aeration will degrade the final product.

Figure 3. Vessel options for preparing aerated compost extracts. Top two images are large-scale vessels (ca. 250 gal) while the bottom images are vessels that hold less than 50 gallons. All vessels require plumbing and mechanical devices to force air into the solution. Photos courtesy of Elzinga & Hoeksema Greenhouses in Portage, Michigan (top right) and www.captaincompost.com (bottom left).



# General procedure for producing 50 gallons of NCE (volumes and ratios can be adjusted accordingly):

- 1. Obtain mature compost and screen to ¼ inch.
- 2. For 50 gallon volumes, fill the brewing vessel (Figure 4) with ~45 gallons of water and let sit/recirculate/aerate for 24 hours. Set aside an additional 5 gallons for later. This will eliminate issues with chlorine. If using de-chlorinated water, well water, filtered water or any other water that does not contain chlorine then no need to aerate for 24 hours.
- 3. Measure the desired amount of compost to produce the compost:water ratio. For example, to prepare 50 gallons of 1:25 (v:v) compost:water extract, place 2 gallons of extract in 48 gallons of water. A standard procedure for measuring the volume of compost is as follows: i) fill a one gallon bucket with compost, ii) lift the bucket 6 inches

from the ground and drop, repeat 10 times, iii) fill the bucket to the one-gallon mark, iv) repeat steps i-iii 3 times, the third fill should be to the one-gallon mark and no more.

- 4. Add the measured amount of compost directly to the de-chlorinated water.
- 5. Using the 5 gallons of water set aside, fill the vessel to 50 gallons.
- 6. Brew for 7-10 days.
- 7. Mix and re-suspend particles in the solution one or two times a day. Use a long-handled spoon or any other instrument for mixing. Alternatively, use a submersible pump with a timer. Set the time for mixing and recirculation for 15-30 minutes every 12-24 hours.
- After brewing, apply the material as needed. If unable to use the material after the desired brewing time then continue to recirculate until ready or else discontinue recirculation and let sit with no lid. The liquid material should remain stable, viable and active until ready to use.

Figure 4. Vessel options for preparing non-aerated compost extracts. Top two images are a vessel designed by Rodale Institute that can prepare aerated and non-aerated compost extracts. A submersible pump can be adjusted to recirculate water (non-aerated) on a timer without adding additional oxygen or adjusted to move liquid from the bottom of the vessel and spray onto the surface of the solution, thereby adding oxygen (aerated). The bottom two images are simple vessels, 5-gal (left) and 50-gal (right, photo courtesy of www.tinyfarmblog.com/ <u>a-spot-of-tea</u>) without any mechanical apparatus.



**Recommendations and warnings** 

- Err on the safe side and prepare ratios no stronger than 1:25 compost:water, 1:25-50 will be fine. Strong solutions can burn plant parts and prevent seed germination and seedling growth (Figure 5).
- Until the practitioner is confident on what the liquid can do, never apply more than once a week.
- Organic salts such as magnesium sulfate or potassium chloride (and many others) that are derived from the solid compost can accumulate in the soil causing phytotoxicity. Seeds, seedlings and young plants are most susceptible to salt toxicity. Use caution when applying compost and compost extracts so that salts do not accumulate and damage plants.
- Test your material on a subset of plants before applying widely. There are simple seed germination tests to determine the optimum compost:water ratio, application rate, or spray regime. For testing spray applications, prepare the extract and then spray on a set of plants (at least three separate plants of the same specie). Diluting the extract by 25% or 50% after the extraction period will provide comparison among compst:water ratios. Spray water on a similar group of plants. Testing on seeds or seedlings can provide rapid results. Compare growth one, two and three weeks after application. Observe for spots on leaves, chlorotic lesions, reduced germination, plant vigor etc.
- There is no one right way to make ACE or NCEs just as there is no one right way to make compost. Find a system that is effective, efficient and does not negatively impact plant production. It is more important not to cause damage to plants than it is to increase production. Often, the benefits of compost extracts are too small to observe but they still exist.

Figure 5. Cucumber seed germination after 2 days in different non-aerated vermicompost extract (NCE) to water ratios (m:m). There was no significant difference among seeds germinated in water and those germinated in either 1:30 NVE or 1:60 NVE. Photo courtesy of Dr. Allison Jack



Scale bar = 1 cm

Rodale Institute 50 gallon Compost Extract Brewer

Figure 6. Rodale Institute 50-gal compost extract brewing vessel. The vessel is a foodgrade, high-density plastic donated by Pepsi Cola, which was previously used to store syrup for preparing soda. All tubing is 1" PVC. Most connection are glued together using rubber cement; however, because all parts are contained within the vessel any leaks would fall back into the solution, which reduces the need to glue all connections. The submersible pump is raised above the bottom of the vessel approximately 4" using a perforated metal tube; although 1/4" welded wire has been working best. See Figure 4 for additional images. The direction of flow can be manipulated to allow for aerated or non-aerated or both types of extract production.



# Specifications:

Submersible pump: Wayne Utility Pump, 1/5 HP, 1700 gallons/hour
Timer: Woods Appliance Timer, 24 h, 15 A, 125 v, not digital
Keys (valve openings) = 3
T-valves = 2-3, depending on the need for dual outputs inside the vessel
90° connections = 6
1" to ½" adaptor = 2, for the upper output inside the vessel; this will create equal pressure between the two outputs.
1" screw valves = 4

1.25" to 1" adaptor = 1