MITIGATING PATHOGEN CONCENTRATION AND REGROWTH: MANAGING SEPTAGE FOR LAND APPLICATION USING GEOTEXTILE BAGS

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ABSTRACT

The land application of hauled sewage (septage) and biosolids is dependent on the quality of the material; it must be ensured that any potential environmental impacts are mitigated. Sludge and wastewater residuals that are high in pathogen content pose a higher risk due to their susceptibility to runoff and subsequent contamination of ground and surface waters. Research has indicated that mechanical methods of dewatering digested municipal sewage biosolids can sometimes lead to increases in certain types of pathogen indicator bacteria. This likely happens as a result of shear forces causing the release of reactivation compounds, which in turn encourages the regrowth of bacteria during storage. This research was focused on the microbial/pathogenic content of passively-dewatered hauled sewage. This study's objectives were to determine if passive dewatering methods may be preferential to limit the sudden increase and regrowth of pathogen indicator organisms by minimizing shear forces during dewatering. A geotextile dewatering bag was used to dewater and consolidate hauled sewage. Counts of three indicator organisms (*E. Coli*, *F. streptococcus*, and *P. Aeruginosa)* were enumerated in the geotextile bag cake over a period of 36 days. It was observed that concentrations of all three indicator organisms decreased considerably from the initial concentrations, with total reductions of 91%, 72%, and 66%, respectively. The results indicate that a passive method of solids dewatering and consolidation, such as a geotextile dewatering bag, can be used for treatment of septage prior to land application, and may help reduce the incidence of sudden increase and regrowth of indicator organisms and pathogens.

INTRODUCTION

Land application of hauled sewage (septage) and biosolids on farmland is a common practice across North America. Management of domestic wastewater sludge via land application is an attractive option due to its ability to act as a nutrient-dense fertilizer or soil amendment for growing crops [Clarke *et al.* (2017), Walker and Mascaro (2018)]. In addition, land application can help alleviate the pumping and trucking costs associated with sending septage to an approved treatment facility. However, sustainable septage application requires haulers to meet a number of requirements, one of the most important being the reduction of pathogens. It is crucial to mitigate pathogen content in septage and biosolids to below regional regulations to prevent downstream contamination of waterways and for the protection of public safety [Thomas *et al.* (2016), Walker and Mascaro (2018)]. To meet the regulatory requirements for land application, wastewater residuals must undergo treatments to reduce pathogens and attractiveness to vectors. Some examples of treatment include digestion (aerobic or anaerobic), thickening, composting, pH

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attenuation using lime (alkaline stabilization), thermal drying, and dewatering [Higgins *et al.* (2007), Jiang *et al.* (2012)].

Sludge thickening followed by dewatering is a common process that treatment plants employ to treat wastewater residuals prior to disposal of solids and/or land application. However, in recent years, there has been an observed phenomenon where certain combinations of digestion and mechanical dewatering of biosolids has resulted in the significant increase of pathogen indicator organism counts (fecal coliforms or *Escherichia Coli*) directly after dewatering, followed by increases during cake storage. This has implications for land application, as the increases observed post-treatment can lead to levels of indicator microorganisms that do not comply with the standards required for land application ("Class A" or "Class B" Biosolids classifications/standards).

For example, Higgins *et al.* (2007) collected samples from two thermophilic digestion processes. The average fecal coliform densities in the digester influent, digester effluent, and centrifuge cake are shown in Table 1.

Table 1. Average FC densities measured in samples taken from thermophilic digestion processes [11].

	Digester Influent	Digester Effluent	Centrifuge Cake
Average fecal coliforms (CFU/gram of dry solids)	1.26×10^8	1.00×10^2	3.16×10^6

From Table 1, it can be seen that the fecal coliform density measured in the influent was typical for digester influent [Higgins *et al.* (2007)]. The effluent from the digesters showed a six log reduction in fecal coliform density. However, immediately after centrifugation, the density of fecal coliforms increased by four orders of magnitude in a relatively short period of time.

Qi *et al*. (2007) performed a similar study, which resulted in comparable findings as per the results from the Higgins *et al*. (2007) study, where the fecal coliform counts immediately after dewatering were much greater compared to just prior to dewatering [Thomas *et al.* (2016)]. However, Qi *et al.* also studied the effect of time on coliform increases, and found that fecal coliform counts (reported in Most Probable Number, MPN, per gram of dry solids) showed a greater increase in dewatered biosolids after 24h of incubation at 25^{o}C than in biosolids that had been digested only. Table 2 summarizes these results.

Table 2. Fecal coliform population of samples from WWTP I after incubation at 25^{o}C for 24 h. MPNs were determined using A-1 media.(Qi et al., 2007)

	Fecal coliform counts *before* incubation (MPN/gram of dry solids)	Fecal coliform counts *after* incubation (MPN/gram of dry solids)
Digested biosolids	5.0×10^3	1.6×10^4

Dewatered biosolids	1.6×10^5	1.0×10^8

The results from Higgens *et al.* (2007) and Qi *et al.* (2007) have been similarly reported by others [Cheung *et al.* (2003), Lu *et al.* (2012)]. These phenomena have been termed "sudden increase" and "regrowth", respectively, and has been observed in a number of studies, primarily after anaerobic/thermophilic digestion and centrifuge dewatering; although a study by Chen *et al.* (2011) found that different combinations of digestion and dewatering have also resulted in the sudden increase and regrowth of certain indicator organisms [Chen *et al.* (2011)]. For example, it was found that mesophilic digestion followed by centrifugation at a prepasturization plant also led to the sudden increase and regrowth of indicator organisms.

The leading theories explaining the cause of sudden increase and regrowth are mainly centered around the dewatering process. From studies by Cheung *et al.* (2003), Monteleone *et al.* (2004) and Qi *et al.* (2007), evidence has shown that mechanical and centrifugal dewatering methods may lead to regrowth of pathogen indicators due to the shear forces imparted during these processes, which leads to break up of the floc. As a result of this floc shear and breakup, increases in pathogen indicator may be observed due to released bacterial growth factors (or lysis of growth inhibitors), exposure to favourable growth conditions (sunlight, temperature, oxygen), and/or increased enumeration efficiency (Figure 1). Regrowth of bacteria typically occurs in the cake over several days after dewatering because of the readily available substrates and nutrients released from floc perturbance.

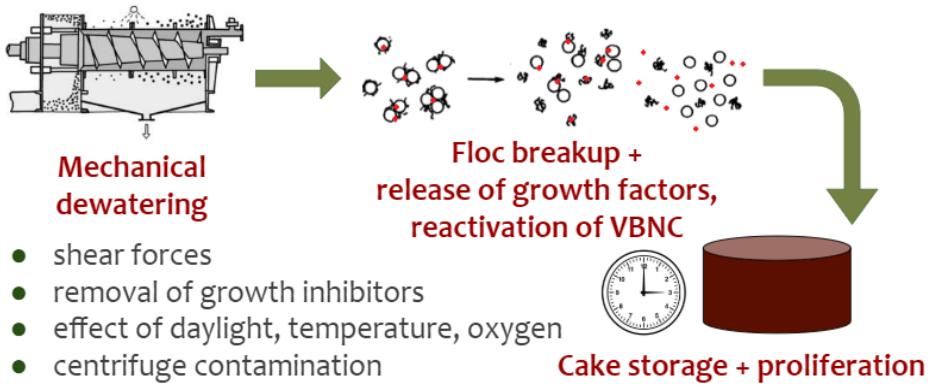

Figure 1. One hypothesis regarding sudden increase and regrowth involves the effect of mechanical dewatering on floc breakup and the resulting release of growth factors and exposure to growth-favouring conditions. Figure by author (Gan).

Higgins *et al.* (2007) has suggested that during thermophilic digestion processes, bacteria can enter a viable but non-culturable (VBNC) state. This would mean that some bacteria within the dewatered solids can still be present and viable, but unable to be enumerated via standard culturing methods. As a result of physical/shear forces during mechanical dewatering, Higgins *et al.* (2007) hypothesized that the VBNC bacteria are "reactivated" to a culturable state, resulting in an apparent sudden increase of indicator organism count post-dewatering. However, while this may explain the sudden increase phenomenon, Chen *et al.* (2011) found that fecal coliform regrowth during storage was shown to occur even when accounting for VBNC bacteria using a quantitative reverse-transcriptase PCR enumeration (RT-qPCR) method. Using RT-qPCR methodology, viable

but non-culturable bacterial pathogens can be detected and quantified [Jiang *et al.* (2012)]. As such, regrowth of pathogenic indicators during centrifuge-dewatered cake storage happens despite accounting for VBNC bacteria.

Studies by Qi *et al.* (2008) and Higgins *et al.* (2007) also confirmed that regrowth of indicator organisms are independent of reactivation, although biosolids can experience both reactivation and regrowth.

The findings of these studies suggested that methods of dewatering that impart shear forces onto sludge likely releases food substrates and bacterial reactivation precursors, resulting in observed fecal coliform and indicator organism increase and regrowth. This implies that dewatering methods that attempt to prevent the release and/or reduce the availability of growth precursors and food substrates can likely control the regrowth of pathogens.

Due to the implications of dewatering process methodology on the possible reactivation and/or regrowth of bacteria within dewatered cakes, this study was developed based on the hypothesis that a passive (non-mechanical) dewatering method may be effective in preventing significant floc shear, which could in turn prevent the sudden increase and regrowth phenomena that has been observed post-mechanical dewatering. With reduced floc shear, the possibility of lysis of bacterial growth inhibitors and the release of growth-supporting proteins may be lessened.

The passive dewatering method chosen was dewatering using geotextile filtration bags. Geotextile bags are comprised of woven or nonwoven fabrics which allow solids to be captured consolidated within the bag as liquid exits through the pores as filtrate (i.e. dewaters). Geotextile bags as a dewatering method is currently employed in full-scale across North America and can be a cost-effective, low-energy alternative to traditional mechanical methods of dewatering [Lawwson (2008)]. As such, passive dewatering using geotextile bags was tested on hauled sewage as a viable alternative to mechanical dewatering processes for aid in the reduction and regrowth prevention of indicator and pathogenic organisms.

MATERIALS AND METHODS

In this limited study, septage was collected from a local sewage hauler in Eganville, Ontario, Canada. The raw septage was a composite of septic tank wastewater collected from septic tanks across the region. Raw septage was chosen as the material to be dewatered to limit the possibility of dewatering sludge containing indicator organisms that had undergone VBNC inactivation. The raw septage (hauled sewage) had not undergone any digestion, stabilization, or treatment processes and it is unlikely that any bacteria would have been induced into a VBNC state. Therefore, it has been assumed that all indicator organisms detected in the raw sample were representative of the entire enumerable population of indicator organisms. The characteristics of the raw septage utilized in this study are shown in Table 3.

Table 3. Characteristics of raw septage from Eganville, Ontario.

pH	7.74
Percent solids	0.22%

E. coli concentration	6.6 x 10^6 CFU/100mL
F. streptococcus concentration	220 x 10^3 CFU/100mL
P. aeruginosa concentration	43.5 x 10^3 CFU/100mL

To achieve optimal dewatering of the septage using a geotextile bag, chemical conditioning to floc the solid material should be performed beforehand to ensure minimal passage of solids through the filter material. As such, samples of the raw material were tested using various polymers to determine the appropriate chemical conditioning to achieve the most stable floc. A high-charge cationic polymer was used to flocculate the material. Once flocculated, approximately 100-L of the septage was poured into two Geotextile Dewatering Test (GDT) bags (Figure 2) and allowed to dewater. A GDT bag is a small-scale/pilot version of a full-scale geotextile dewatering bag, used to determine achievable percent solids, filtrate release, and filtrate quality prior to implementation of a full-scale geotextile bag dewatering project.

Figure 2 Left: GDT experimental setup. Right: Storage and sampling of cakes from the GDT.

The GDT geotextile bags are made from GT-500 fabric, which is a "Specifically Engineered Dewatering Textile" manufactured from high tenacity polypropylene multifilament and monofilament yarns [Tencate Geosynthetics (2019)]. This fabric allows flocculated solids to be retained and consolidated within the sealed bag while allowing filtrate to escape through the pores. As the solids consolidate within the geotextile bag, the moisture content of the dewatered GDT solids (“cake’) decreases over time, and percent solids increase.

Composite solids samples from the GDT cake were taken immediately after dewatering, as well as every seven days over a period of 36 days. This was to observe the long-term effects of cake storage on pathogen indicator content within a geotextile dewatering bag. In real-world applications, the capacity of the geotextile bags often exceed the volume of biosolids/sludge to be dewatered during one pumping event. Therefore, geotextile bags can remain onsite for a number of days prior to land application of the internal cake. In addition, as the solids within the geotextile bag continue to dewater over a period of time, the cake volume within the bag decreases (moisture content decreases), increasing the capacity within the bag available for the addition of more sludge.

The GDT geotextile bags were stored indoors at room temperature for the duration of the experiment. Solids samples were collected from resealable ports on the top of each GDT geotextile bag and stored in sterile, airtight containers prior to delivery to the MECP for the analysis of the three key indicator organisms--*Escherichia Coli, Fecal streptococcus*, and *Pseudomonas aeruginosa*. The indicator organisms chosen have traditionally been used as indicators of bacterial pathogen presence and general wastewater contamination, and methods to enumerate these organisms are widely employed to replace the costly and inefficient processes of direct pathogen enumeration [Thomas *et al.* (2016)]. Counts of these indicator organisms, reported in colony-forming units (CFU) per gram of dry weight solids, were enumerated using the methods outlined in Table 4.

Table 4. Indicator organism enumeration methods.

Indicator organism	**Method**
Escherichia Coli	● Membrane Filtration Using DC Agar Isolation ● Detection, Enumeration in Solids and Semi-Solids by MF Methods E3371 & E3433 of MECP LaSB
Fecal Streptococci	
Pseudomonas Aeruginosa	● Detection and quantification by MPN assay (PSEUDALERT®TM - QUANTI-TRAY/2000®TM) Method E3560 of MECP LaSB

RESULTS AND DISCUSSION

It was observed that concentrations of all three indicator organisms decreased significantly from the initial concentrations, with total reductions of 91%, 72%, and 66% for *Escherichia Coli, Fecal streptococcus*, and *Pseudomonas aeruginosa*, respectively (Figure 3, Figure 4, Figure 5). Table 5 below summarizes these results and compares them to post-dewatering coliform counts as observed in the studies cited.

Table 5. Summary of results.

Study	Indicator organism	Initial concentration	Post-dewatering concentration	% Reduction

Gan *et al.* (2018)	*Escherichia Coli*	5.1 x 10^6 CFU/g dry solids	4.5 x 10^5 CFU/g dry solids	91%
	Fecal Streptococci	2.3 x 10^4 CFU/g wet solids	6.4 x 10^3 CFU/g wet solids	72%
	Pseudomonas Aeruginosa	6.1 x 10^4 MPN/g wet solids	2.1 x 10^4 MPN/g wet solids	66%
Qi *et al.* (2007)	Fecal coliforms	5.0 x 10^3 CFU/g dry solids	1.6 x 10^5 CFU/g dry solids	N/A (net increase of 1.55 x 10^5 CFU/g dry solids)
Higgins *et al.* (2007)	Fecal coliforms	1.00 x 10^2 CFU/g dry solids	3.2 x 10^6 CFU/g dry solids	N/A (net increase of 3.16 x 10^5 CFU/g dry solids)

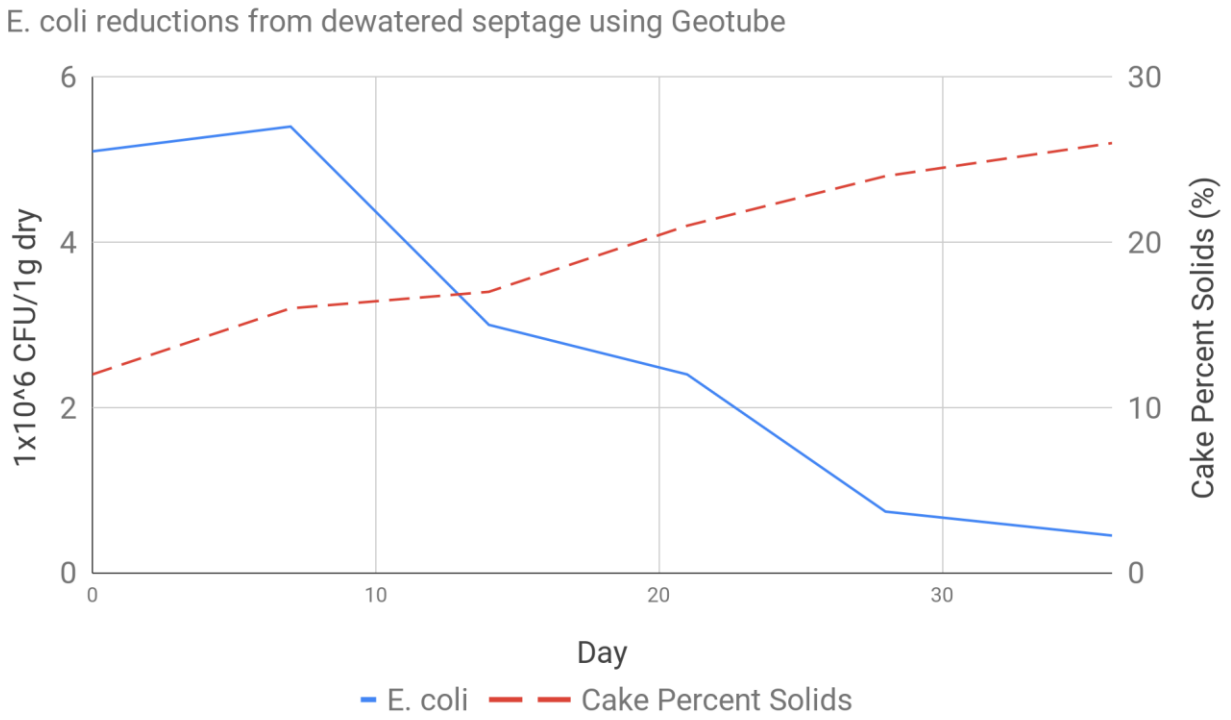

Figure 3. Reductions of *E. coli* from dewatered septage using a geotextile bag dewatering test (GDT). Percent solids of the consolidated cake is shown on the secondary axis. Initial concentration of *E. coli* in the raw septage (pre-dewatering) was 6.6 x 10^6 CFU/100mL. Percent solids of the raw septage was 0.22%.

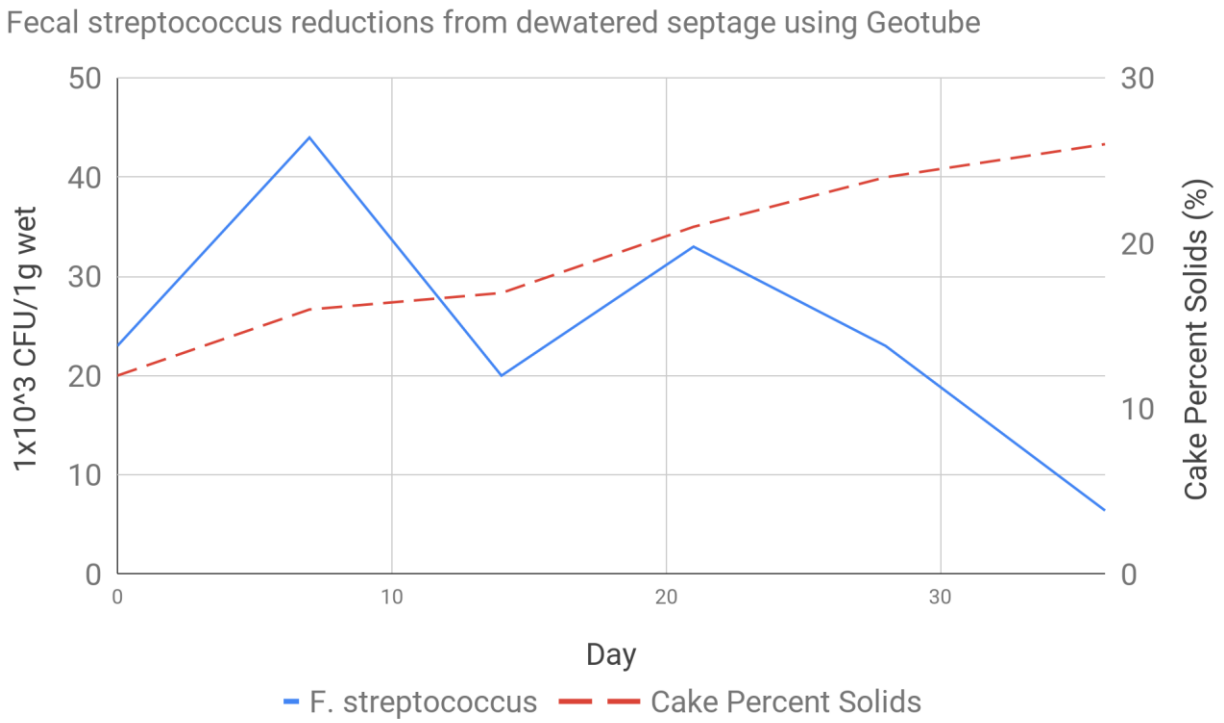

Figure 4. Reductions of *F. streptococcus* from dewatered septage using geotextile bag dewatering test (GDT). Percent solids of the consolidated cake is shown on the secondary axis. Initial concentration of *F. streptococcus* in the raw septage (pre-dewatering) was 220 x 10^3 CFU/100mL. Percent solids of the raw septage was 0.22%.

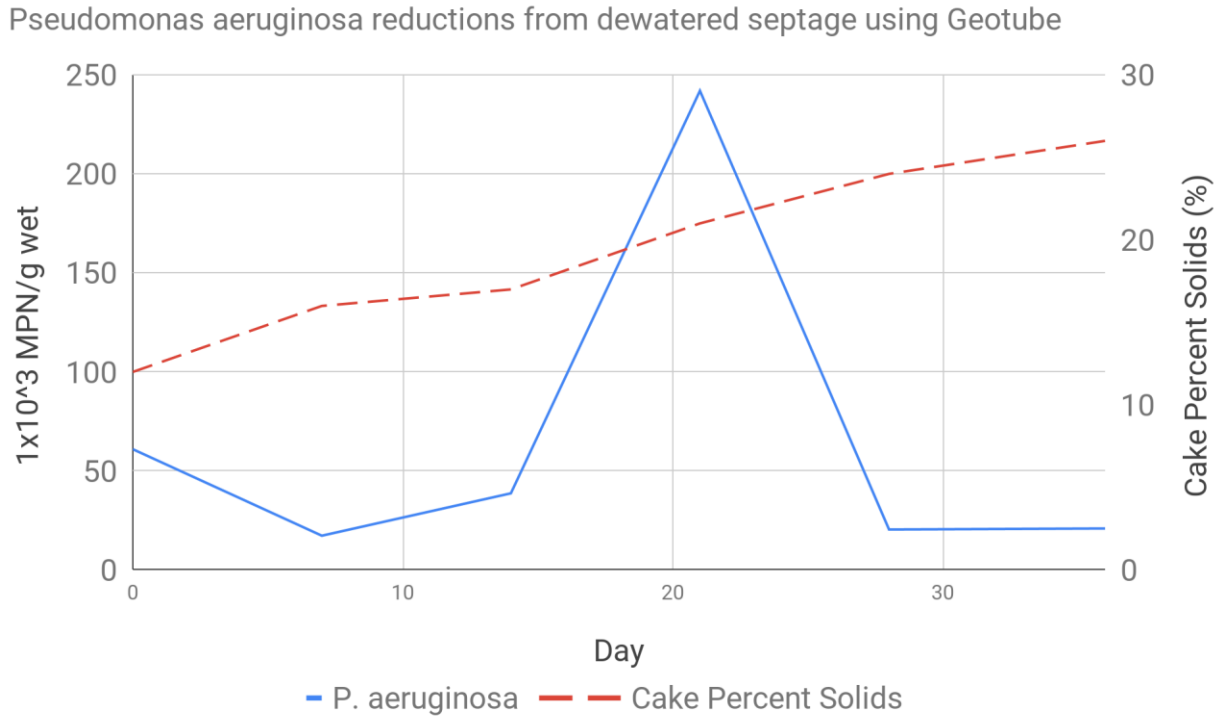

Figure 5. Reductions of *P. aeruginosa* from dewatered septage using geotextile bag dewatering test (GDT). Percent solids of the consolidated cake is shown on the secondary axis. Initial concentration of *P. aeruginosa* in the raw septage (pre-dewatering) was 43.5 x 10^3 CFU/100mL CFU/100mL. Percent solids of the raw septage was 0.22%.

No evidence of sudden increase occurred based on indicator organism count immediately following the dewatering process. Some evidence of limited regrowth in *P. aeruginosa* and *F. Streptococci* was shown on one and two occasions, respectively; however, the overall trend of indicator organism reduction was observed to result in a net decrease of colony forming units (net decrease of 4 x 10^4 MPN/g wet solids in *P. aeruginosa* and net decrease of 1.6 x 10^4 CFU/g wet solids in *F. Streptococci*). It is possible that the isolated observed increases of indicator organisms were a result of cross-contamination due to sampling methodology. As the resealable sampling ports were opened multiple times during the course of the project, solids by the port were exposed to ambient air (oxygen) and environmental conditions, including environmental microorganisms. As noted by Qi *et al.* (2007), exposure to the favorable growth conditions may induce regrowth during storage. Decreases in pathogen indicator organisms after these count spikes may have been due to the reduction of pathogen indicators as a result of subsequent limited exposure to these favorable growth conditions.

CONCLUSION

Overall, results from this experiment have provided evidence that passive dewatering methods may prevent the sudden increase and regrowth of indicator organisms by limiting shear forces and floc breakup, which in turn limits the release of growth factors for pathogenic indicators. It is hypothesized that the geotextile dewatering unit also provided limitations on additional microorganism growth parameters such as sunlight exposure and oxygen diffusion, which aided in the prevention of regrowth. The results have led to the conclusion that a passive method of solids dewatering and consolidation, such as a geotextile dewatering bag, may be advantageous for treating septage and biosolids with high anticipated pathogen content, and that such a method may be used to aid in the reduction of indicator organisms.

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