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Enzymatic treatment effects on dewaterability of anaerobically digested biosolids-I: performance evaluations

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Abstract

This paper reports on the ability of an enzymatic pre-treatment to significantly improve the conditioning of wastewater sludges (biosolids) significantly using by a polymeric flocculant. Experiments used anaerobically digested biosolids samples from two different municipal wastewater treatment facilities, and a formulation containing several different hydrolytic enzymes. Samples were incubated at 35 °C for 16 h then conditioned with the cationic polymer solution. A laboratory scale mechanical dewatering unit, which simulates full-scale belt filter presses, was used in dewatering of the biosolids. Dewaterability was evaluated using capillary suction time (CST), solid content of final cake product, filtrate turbidity, and suspended solids analysis. The enzyme additions enhanced dewaterability of the polymer conditioned samples in terms of CST and solids content of final product, demonstrating the possibility of significant volume reductions using enzymatic treatment at full-scale. The biosolids structures with the additions of enzyme, polymer, and both additives were determined using field emission scanning electron microscopy.

Keywords: Biosolids; Anaerobic digestion; Dewaterability; Enzymes; Drainability; Filterability; SEM

1. Introduction

Anaerobic digestion is one of the more commonly used stabilization processes in sludge management, providing effective pathogen destruction, reduction of volatile solids and odour potential and an energy source in the form of biogas. Although, it has these advantages, the stabilization process may lead to poorer dewatering characteristics, which means higher chemical requirements in biosolids conditioning, lower quality of final processed biosolids, and higher operation and disposal costs.

Microorganisms and microbial-environment conditions are crucial in this process as in other biological treatment processes. During the anaerobic digestion of biosolids, the first stage in the degradation of particulate organic matter is the solubilization and enhanced hydrolysis of complex polymeric organic carbon structures.

Bacteria are usually contained within a flocculated matrix of exopolymeric substances. When biopolymers are released from this floc structure as biocolloids, their properties hinder effective dewatering of the biosolids and are responsible for a significant portion of the polymer demand in the chemical conditioning prior to dewatering processes [1].

The exopolymeric substances also act as a network that confines extracellular enzymes exhibiting hydrolytic activity [2]. Jain et al. [3] showed that both the concentration of hydrolytic enzymes and the contact between these enzymes and their substrates, were very important in their modeling studies of anaerobic digestion of complex particulate substrates.

Novak et al. [1] studied the mechanisms of floc destruction during anaerobic and aerobic digestion and their effects on conditioning and dewatering of biosolids in laboratory scale studies. They showed that enzyme activity declines during both anaerobic and aerobic digestion. For aerobic digestion, glucosidase activity, indicating polysaccharide degradation potential, decreased to zero by day 10 of digestion time. The loss of the activity explained why polysaccharide accumulates during aerobic digestion, while most of the protein is degraded. Under anaerobic conditions,

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enzymatic activity for both protein and polysaccharide degradation declined, but remained higher than during aerobic digestion.

Given the above observations, it would seem logical to supplement the active enzymatic systems in these biosolids. If this serves to enhance the degradation of extracellular polymeric substances, then the resistance to dewatering might be lessened and the amount of polymer required for conditioning might be reduced.

An enzymic supplement for this purpose would not necessarily require an expensive, purified product. Although relatively pure enzyme extracts are required for some applications (such as in clinical diagnosis and in food processing), many useful industrial enzyme preparations are not highly purified. Such enzyme mixtures may include a variety of enzymes capable of numerous catalytic functions, and thus, are useful for very heterogeneous substrates in many industrial applications [4].

This possibility has been explored in one previous study. Thomas et al. [5] added an enzyme product (Degomma 7083®, containing carbohydrase, lipase and proteinase activities) to digested biosolids, which were then incubated approximately 16 h. Laboratory tests showed improved dewaterability, as indicated by CST reductions of 50%. These results are somewhat uncertain because the control sample (with no enzyme addition) was also fairly dewaterable, with a CST of 29 s. However, drainage volumes from sieving tests were increased by up to 43%. Full-scale tests with dewatering by belt filter press showed cake solids improvements of +2.2 to +4.3%, representing potential decreases in sludge mass of 8.4–13.3%. This paper mentioned that polymer was required following the enzyme treatment, and also that comparable results were obtained with primary sludge and waste activated sludge. However, no subsequent reports from these authors could be located, and the use of enzymes for improved dewatering is not known to have been proven in any subsequent studies.

In addition, there have been no reports on whether enzymatic pre-treatment alters the requirement for flocculant polymer. In evaluating the feasibility of using an enzyme, this is likely to be an important cost consideration.

2. Objectives

The objectives of the study were therefore:

- to determine whether previously reported enzymic treatment effects could be reproduced or improved, based on both the standard laboratory tests of dewaterability and on the performance of a bench-scale dewatering process;
- to determine whether enzyme pre-treatment changes the amount of polymer required for conditioning, and if so, how the combined used of enzymes and polymer can be optimized;
- to determine whether enzymatic treatment changes the physical or chemical properties of conditioned biosolids; and
- to determine whether enzyme pre-treatment can be optimized with polymer dosage to provide a feasible means of improving biosolids dewatering.

The overall objective of this project was to determine the feasibility of combining enzyme use with polymer conditioning, based on possible improvements in the cost and performance of biosolids dewatering. A companion paper reports on additional work required to complete this determination.

3. Materials and methods

3.1. Materials

Anaerobically digested biosolids were collected from two municipal wastewater treatment facilities, the Newtown Creek Wastewater Treatment Facility in New York City, NY and the Wilmington Wastewater Treatment Facility in Wilmington, DE. These are designated as NYC and WIL samples, respectively.

Polymer conditioning tests were conducted using Percol 757, a high molecular weight, cationic copolymer consisting of 65% AMD (acrylamide monomer) and 35% AETAC (acryloyloxyethyl trimethylammonium chloride) by mole percent, with the monomer structures shown in Fig. 1. Polymer stock solutions were prepared at a 0.5% w/v concentration according to Dentel et al. [6].

A commercial product (Enviro-Zyme 216, Winston Company, Inc., Tulsa, USA) was used for enzymic pre-treatment, which contains protease, lipidase, anaerobic bacteria, *Aspergillus oryzae*, and an enzyme complex mixture (other hydrolytic enzymes). This is a dry powder product. Enzyme stock solution (2%, w/v) was prepared using warm water.

3.2. Experimental procedures

The dry solid contents of NYC and WIL biosolids were found to be 3.2 and 2.6%, respectively. The samples without enzyme and with different enzyme concentrations were first incubated at 35 °C for 16 h. For full-scale application, this could be accomplished using either a secondary digester or a post-settler.

These samples were then conditioned using different volumes of Percol 757 polymer stock solution. The polymer was added to 500 mL volumes of the biosolids samples contained in standard 1 L beakers. Intensive mixing was provided with a household-blending mixer (Braun Multipractic) for 15 s.

To evaluate the filterability of these samples, capillary suction time (CST) analyses were then performed using a

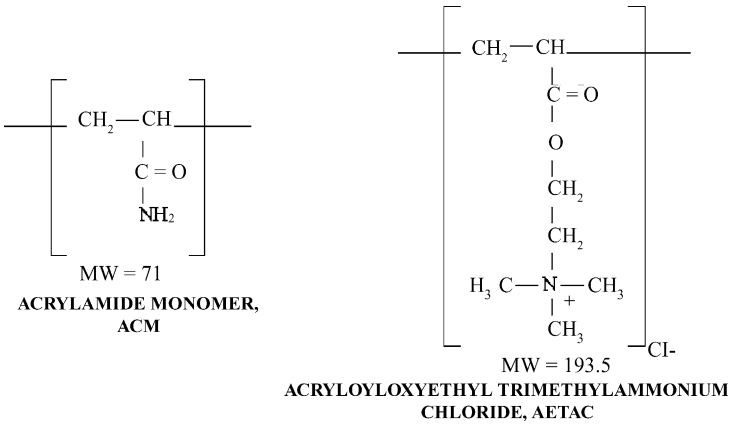

Fig. 1. The structure of polymer used Percol 757, 80% cationic copolymer (ACM/AETAC), molecular mass $\sim 1.8 \times 10^6$.

CST instrument (Venture Innovations, Lafayette, Louisiana). However, to simulate full-scale dewatering, such as by belt filter press, a bench-scale dewatering unit was also used, consisting of two stages:

(1) a gravity drainage plough simulator kit, which was a modified apparatus similar to that suggested by Severin et al. [7], used for drainage tests; followed by
(2) a mechanical press commercially available as a Crown Press™ (Neogen Corporation, Lansing MI), used for the dewaterability tests. This press allowed controlled periods of mechanical pressure to be applied to drained biosolids between filter cloths, in the same manner occurring during cake compression in full-scale belt filter press operation [7].

Details on mechanical descriptions of gravity drainage kit, the press, and drainage-filterability tests are given in the companion paper.

To evaluate dewatering performance, dry solid contents of the dewatered samples were analyzed. Filtrate samples after the press zone were also collected, for turbidity and suspended solid (SS) analyses. Turbidity measurements were performed using a Hach 2100P Turbidimeter.

Electron microscopy was used to characterize the physical effects of enzyme treatment. For this, the samples were imaged on a Hitachi 4700 FESEM (field emission scanning electron microscopy) with a Gatan Alto cryostage. Chemical effects of enzyme treatment were determined by measuring the changes in protein and polysaccharide concentrations resulting from incubation with and without enzyme additions.

3.3. Analytical procedures

Solids content, suspended solids, CST and turbidity analyses were carried out according to standard methods [8].

Extracellular polymeric substances (ECP) were extracted from the samples using the heat extraction technique originated by Goodwin and Forster [9]. A 50-mL biosolids sample was first centrifuged at 3000 rpm for 15 min; the supernatant was then decanted and the samples resuspended to their original volume in a buffer solution as described by Frolund et al. [10]. This step is intended to remove the various dissolved solids and extracellular ''slime'' in the biosolids samples. Samples were analyzed for protein content using protein assay kits (Procedure No. P 5656, Sigma Diagnostic®). Polysaccharides in the ECP samples were determined using a phenol-sulphuric acid method developed by Dubois et al. [11].

Biosolids samples with and without enzyme additions and final cake samples for FESEM imaging were mounted dropwise onto an aluminum stub, plunged into a liquid nitrogen slush, transferred under vacuum to the Gatan specimen preparation chamber, and warmed to $-95\ ^\circ C$ for 15 min. By this procedure, surface water was removed by sublimation to reveal the underlying structure in a frozen hydrated state. Etched samples were transferred to the cryostage and viewed at $-120\ ^\circ C$ and 0.7 kV (current 10 μA) with a working distance of 6–7 mm.

4. Results and discussion

CST results for conditioned NYC and WIL samples are given in Figs. 2 and 3, respectively. The CSTs of both were quite high in the absence of any chemical treatment, with the NYC value (8180 s) still higher than that of WIL (2150 s). As is generally observed, polymer additions caused the CST values to decrease until an optimal polymer dose was reached, after which the CST values increased. This pattern was seen both with and without enzyme addition, but the enzyme doses gave significantly lower CST values for both the NYC and WIL results. Thus, based on CST measurements, enzyme additions decreased the polymer demand.

The CST results also indicated an optimum enzyme addition, beyond which the CST showed little further improvement or—in the case of the NYC samples—actually increased. Thomas et al. [5] reported a similar effect, hypothesizing that excessive enzyme activity led to the

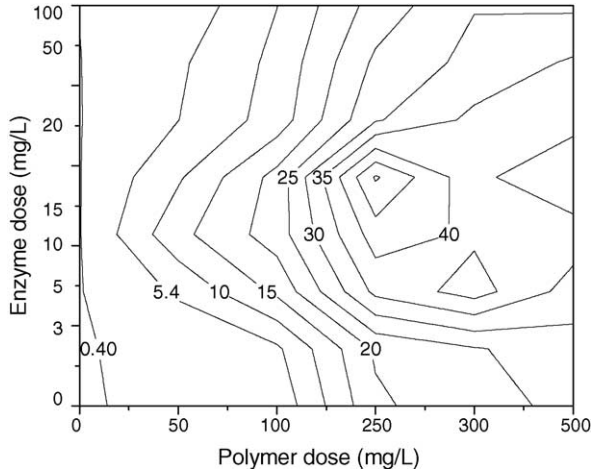

Fig. 2. 1/CST × 10^3 (1/s) values of NYC as a function of polymer and enzyme doses.

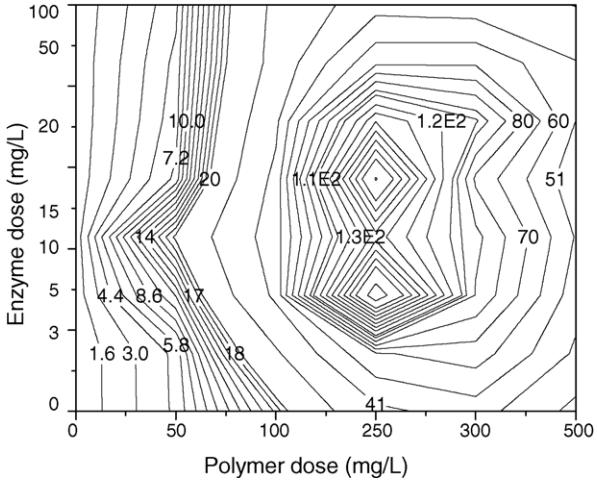

Fig. 3. 1/CST × 10^3 (1/s) values of WIL as a function of polymer and enzyme doses.

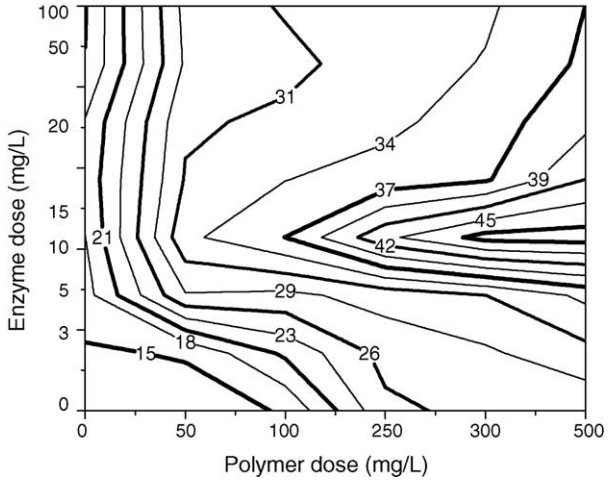

Fig. 4. Solid content (%) of NYC final cake products as a function of polymer and enzyme doses.

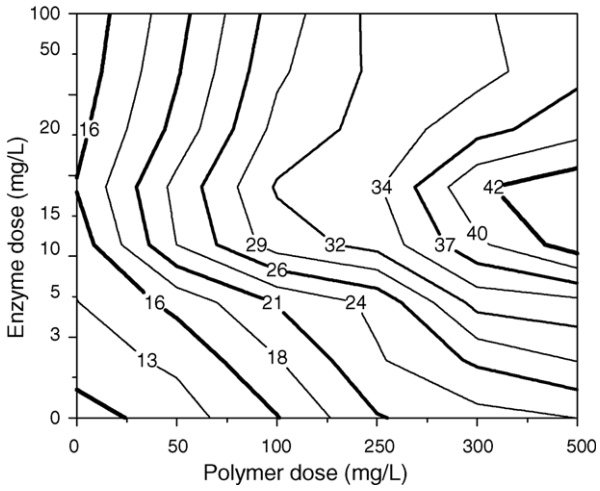

Fig. 5. Solid content (%) of WIL final cake products as a function of polymer and enzyme doses.

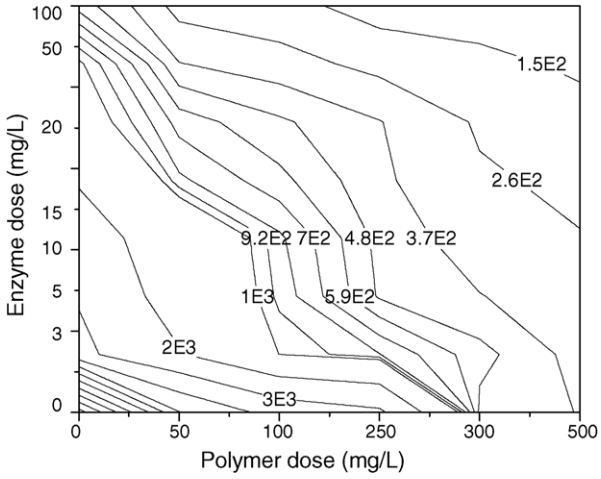

Fig. 6. Turbidity values (NTU) of NYC as a function of polymer and enzyme doses.

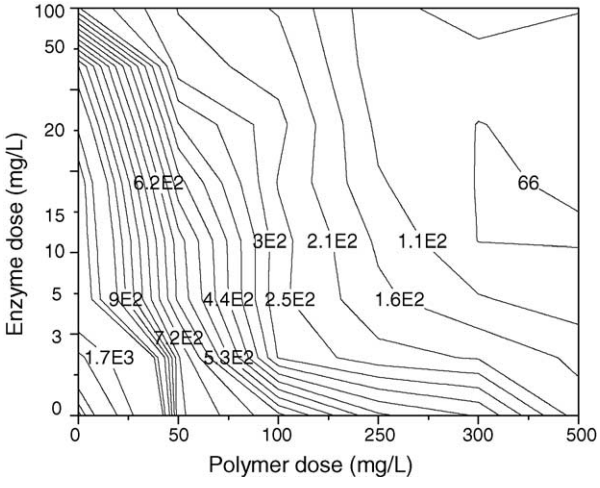

Fig. 7. Turbidity values (NTU) of WIL as a function of polymer and enzyme doses.

Table 1
Effect of enzyme treatment on dry solids (DS)

Biosolids ID	DS (%), without enzyme	DS (%), with enzyme	DS reduction (%)	Mass reduction (%)
NYC-0	12.99 (0), 26.57 (500)	–	–	–
NYC-3	12.99 (0), 26.57 (500)	14.97 (0), 30.48 (500)	1.98, 3.91	13.23, 12.83
NYC-5	12.99 (0), 26.57 (500)	16.95 (0), 34.62 (500)	3.96, 8.05	23.38, 23.24
NYC-10	12.99 (0), 26.57 (500)	17.95 (0), 48.54 (500)	4.96, 21.96	27.65, 45.25
NYC-15	12.99 (0), 26.57 (500)	18.81 (0), 42.05 (500)	5.83, 15.47	30.97, 36.80
NYC-20	12.99 (0), 26.57 (500)	18.19 (0), 38.59 (500)	5.21, 12.01	28.61, 31.13
NYC-50	12.99 (0), 26.57 (500)	14.53 (0), 37.12 (500)	2.39, 10.54	15.53,28.40
NYC-100	12.99 (0), 26.57 (500)	15.25 (0), 36.66 (500)	2.26, 10.08	14.81, 27.51
WIL-0	9.03 (0), 25.98 (500)	–	–	–
WIL-3	9.03 (0), 23.66 (500)	11.71 (0), 29.13 (500)	2.69, 5.83 3.90, 7.53	22.93, 18.76 30.16, 29.40
WIL-10	9.03 (0), 23.66 (500)	14.01 (0), 43.89 (500)	4.99, 10.22	35.58, 46.09
WIL-15	9.03 (0), 23.66 (500)	15.82 (0), 44.11 (500)	6.79, 10.22	42.93, 46.37
WIL-20	9.03 (0), 23.66 (500)	14.83 (0), 38.68 (500)	5.81,12.70	39.14,38.83
WIL-50	9.03 (0), 23.66 (500)	13.98 (0), 36.29 (500)	4.96, 17.90	35.45, 34.80
WIL-100	9.03 (0), 23.66 (500)	13.52 (0), 36.76 (500)	4.49, 18.12	33.22, 35.64

Biosolids ID indicates sample origin and enzyme dose in mg/L. DS values given for 0 and 500 mg/L polymer doses, with and without enzyme additions. Final columns given change in %DS and percent reduction in final cake mass after press zone.

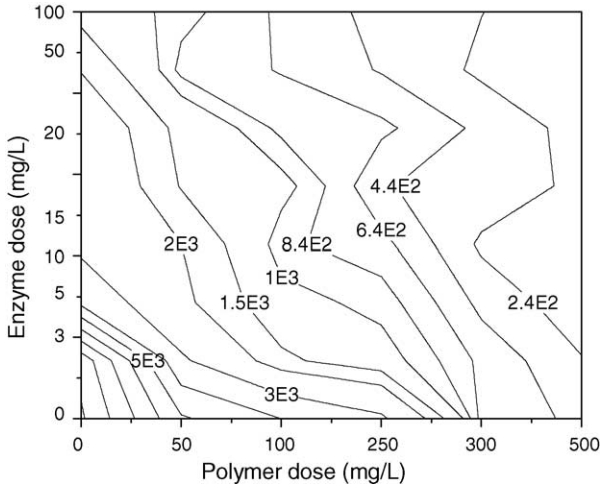

Fig. 8. SS values (mg/L) of NYC as a function of polymer and enzyme doses.

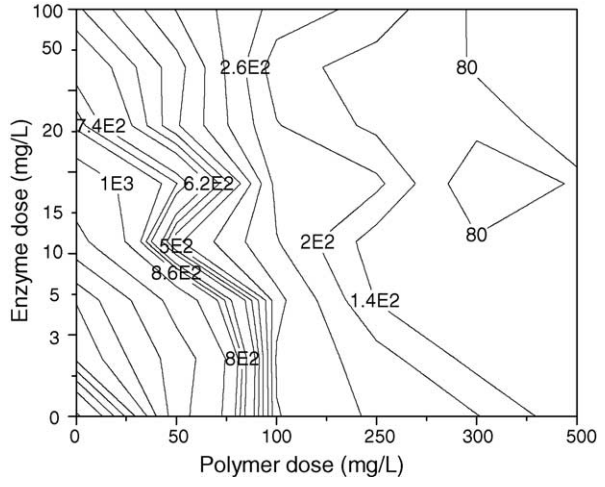

Fig. 9. SS values (mg/L) of WIL as a function of polymer and enzyme doses.

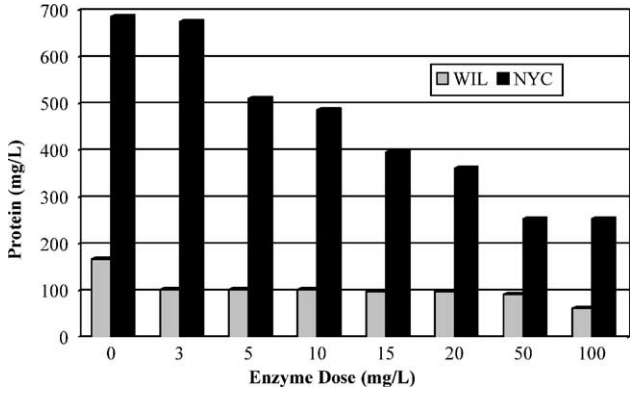

Fig. 10. Remaining protein concentration as a function of enzyme addition. Results for Wilmington (WIL) and New York City (NYC).

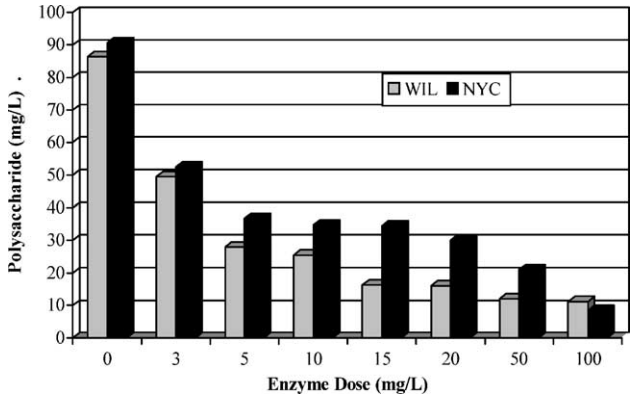

Fig. 11. Remaining polysaccharide concentration as a function of enzyme addition. Results for Wilmington (WIL) and New York City (NYC).

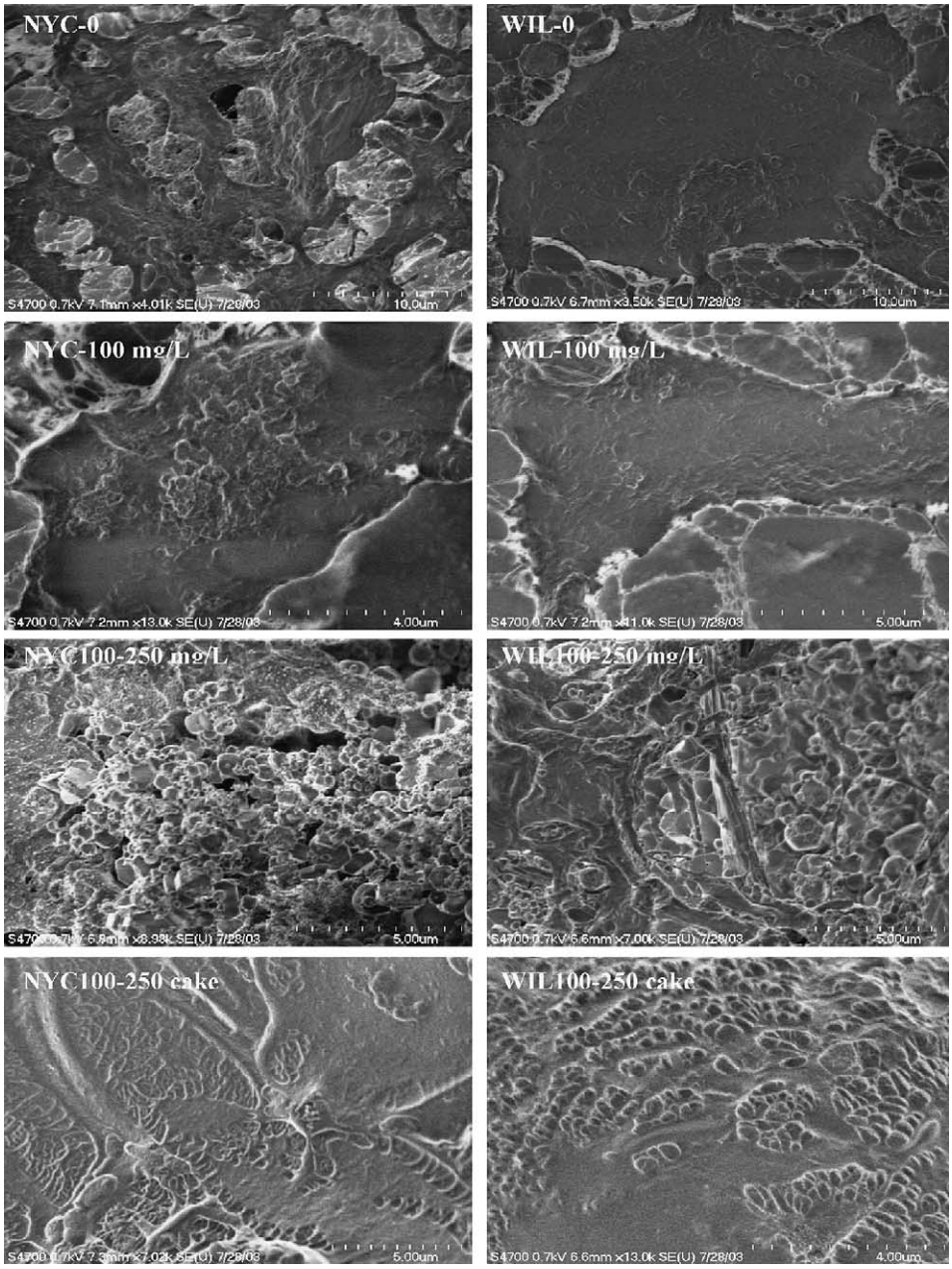

Fig. 12. SEM images for NYC (left) and WIL (right) biosolids samples: (a) raw, (b) enzyme treated (100 mg/L), (c) enzyme treated and polymer flocculated (250 mg/L), and (d) final cake product.

creation of numerous hydrophilic end groups that counteract flocculation.

The CST test indicates the drainability of a sludge, but does not simulate the expression stage of dewatering. The Crown Press™ results may be considered as a better measure of this step, quantified by the final solids content obtained. Figs. 4 and 5 show these data for the NYC and WIL samples, respectively.

These figures show obvious improvements in dewatering with enzyme addition. For both biosolids, all enzyme doses produced higher solids concentrations than were obtained without enzyme addition, and this was the case for all polymer doses used. For NYC, the highest solids content obtainable with polymer alone was 26%, but with 10 mg/L of enzyme at this same polymer dose (300 mg/L), the solids content was increased to 48%. In other words, a small enzyme addition almost doubled the solids concentration, halving the sludge mass that will remain. The result of enzyme addition is potentially to reduce biosolids transportation and disposal costs by almost 50%.

A similar result is seen for the WIL results. With polymer alone, a solids concentration of 24% can be attained, but with 15 mg/L of enzyme, this is increased to 42% solids. The sludge mass will be decreased by over 40% with this use of enzyme.

For both NYC and WIL, there is no clear optimum polymer dose. Increasing polymer led to gradual increases in solids levels obtained. However, an optimum enzyme addition exists in both cases. Above 10 mg/L enzyme for NYC, and 15 mg/L for WIL, no further improvement in the cake solids occurs, and usually, the value goes down substantially.

The potential sludge mass reductions can be similarly calculated for any of the experimental conditions used. Table 1 reports these results as a function of enzyme additions for the zero and 500 mg/L polymer doses. The reductions in sludge mass are more dramatic than those reported by Thomas et al. [5].

It might be hypothesized that hydrolyzing enzymes would reduce the particle size in biosolids, allowing increased release of turbidity or suspended solids in the filtrate. However, this was not the case. Figs. 6 and 7 actually demonstrate that filtrate turbidities were decreased with the use of enzymes with polymer for both NYC and WIL samples. Filtrate suspended solids shown in Figs. 8 and 9 confirm this for both biosolids.

The ECP analyses following sample incubation with enzymes showed lower protein and polysaccharide concentrations with increasing enzyme additions. This was the case for both NYC and WIL samples, as seen in Figs. 10 and 11 (even though the NYC sample did have much higher extracellular protein concentrations than WIL). ECP analyses following polymer additions did not indicate the polymer to be responsible for any further changes in protein or polysaccharide concentrations.

From these findings, it can be said that enzyme product additions improved the degradation of extracellular protein and polysaccharide, which have been claimed responsible for poor dewaterability and high conditioner demand (e.g. Novak et al. [1]).

An extensive gel-like structure was observed in unconditioned anaerobically digested NYC and WIL samples using FESEM (Fig. 12a). Fig. 12b shows some SEM images for 100 mg/L enzyme addition at different magnifications. The SEM images are given Fig. 12c for 250 mg/L polymer added samples. SEM images for final cake products are shown in Fig. 12d. Enzyme additions did not produce any structural changes visible at the colloidal scale, although more bacteria were observed during the image scanning.

Polymer additions apparently replace the biosolid structures with the much larger polymer structure, with a gel-like biocolloidal matrix collapsed onto the surface of the polymer backbone. These images are comparable to those of Poxon [12], who reported similar observations for anaerobically digested biosolids conditioned with a cationic polymer. That study concluded that anaerobically digested biosolid is shown to possess a space-filling, gel-like biocolloidal structure that lead to poor dewaterability. Destruction of the structure significantly improves the dewaterability. Herein, the polymer additions led to this same type of structural changes, which provide a reduction in the volume occupied by the biocolloidal structure. The effect of enzymes was not apparent at this scale, however, and thus, the enzymic effect appears to be at the sub-colloidal level, as shown by the effects on ECP.

5. Conclusions

- Enzyme product additions with polymer conditioning improved the dewaterability of both anaerobically digested biosolid samples based on CST, solid contents of final product, filtrate turbidity and suspended solid measurements.
- Comparison of cake solids measurements indicates that substantial reductions in sludge mass may be obtained with enzyme use.
- Protein and polysaccharide concentrations of the biosolid samples decreased with the increasing enzyme additions, consistent with the hypothesis that the enzymes degrade the chemical constituents responsible for poor dewaterability.
- FESEM images do not reveal structural changes at the colloidal level when enzymes are used, so enzyme effects are evidently at the sub-colloidal scale.

Additional data from enzyme addition experiments are presented in a companion paper, along with more detailed analysis of drainability and filterability results. Ongoing work is investigating the effects of specific, pure enzyme preparations on biosolids dewatering in lab scale and full-scale studies.

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