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A *Bacillus*-Based Bioaugmentation Product Effectively Reduces Sludge In An Analysis Of Eleven Wastewater Lagoons

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Abstract

Managing sludge accumulation in lagoons accounts for a large portion of wastewater treatment budgets. One sludge removal strategy that is both effective and more affordable than mechanical removal methods is bioaugmentation. Treating a lagoon with beneficial microbes, such as *Bacillus*, enhances the microbial community already present in the environment. This leads to more efficient microbial sludge digestion and results in sludge reduction. Here we examine the effect of treatment with *Bacillus*-based Biotifx® products in eleven lagoon systems representing different industries, locations, and sizes. Biotifx® treatment resulted in sludge removal in every system analyzed with an average reduction of 56% (SD = 33%) across a wide array of systems. These reductions were also evident in lagoons with high loading volumes and sludge surface coverage. Bioaugmentation with Biotifx® products is an effective strategy for managing lagoon sludge.

Introduction

More than 50% of industrial wastewater treatment facilities in the US employ lagoons in their treatment process (4). Sludge is an integral part of a lagoon system, but excess amounts can negatively impact performance. Accumulation of sludge alters the holding capacity of the lagoon and topography of the bottom (5). These changes can result in decreased hydraulic retention time and subsequently reduce treatment efficacy. Sludge management in wastewater typically amounts to 40-60% of total plant costs, but charges relating to sludge disposal, such as transportation and landfill costs, have been trending upward, and regulations for land application are becoming stricter (7). One solution to help mitigate these rising disposal costs is the implementation of a biological treatment. Organic sludge components can be effectively digested with a biological treatment, reducing the frequency and cost of mechanical removal events, and making bioaugmentation an attractive sludge management option.

Bioaugmentation aims to increase sludge digestion by enhancing the biology that naturally exists in the environment through the addition of bacterial species and/or nutrients that support microbial growth. In wastewater, the goal of employing bioaugmentation in the form of a bacterial species, such as *Bacillus*, is to augment the native population and attain a community that more effectively digests sludge (2). Sludge composition varies greatly based on the influent sources, but studies have found that municipal sludge is mostly organic with carbohydrates, proteins, and lipids accounting for over 50% of this organic portion (1, 8). The addition of a bacterial species like *Bacillus* helps to enhance the natural sludge digestion process by breaking down complex organic matter in the sludge to create intermediate metabolites that they and other bacteria in the wastewater environment can consume. Biodegradation of sludge is a multi-step process involving the whole bacterial community with a variety of organisms that possess different digestive abilities. As a result of the bacterial activity in the system, the components of sludge are either incorporated into new microbial biomass or are ultimately converted to water and carbon dioxide (2, 3). The amount of carbon dioxide produced due to these processes is minimal especially in comparison to the level produced by alternative mechanical removal methods. Laboratory testing demonstrates an increase in bacterial activity in wastewater treated with Biotifx® *Bacillus*-based products when compared to an untreated sample of the same wastewater (Figure 1). The presence of additional active organisms working to digest more material in their environment, results in measurable differences in lagoon sludge volume over time.

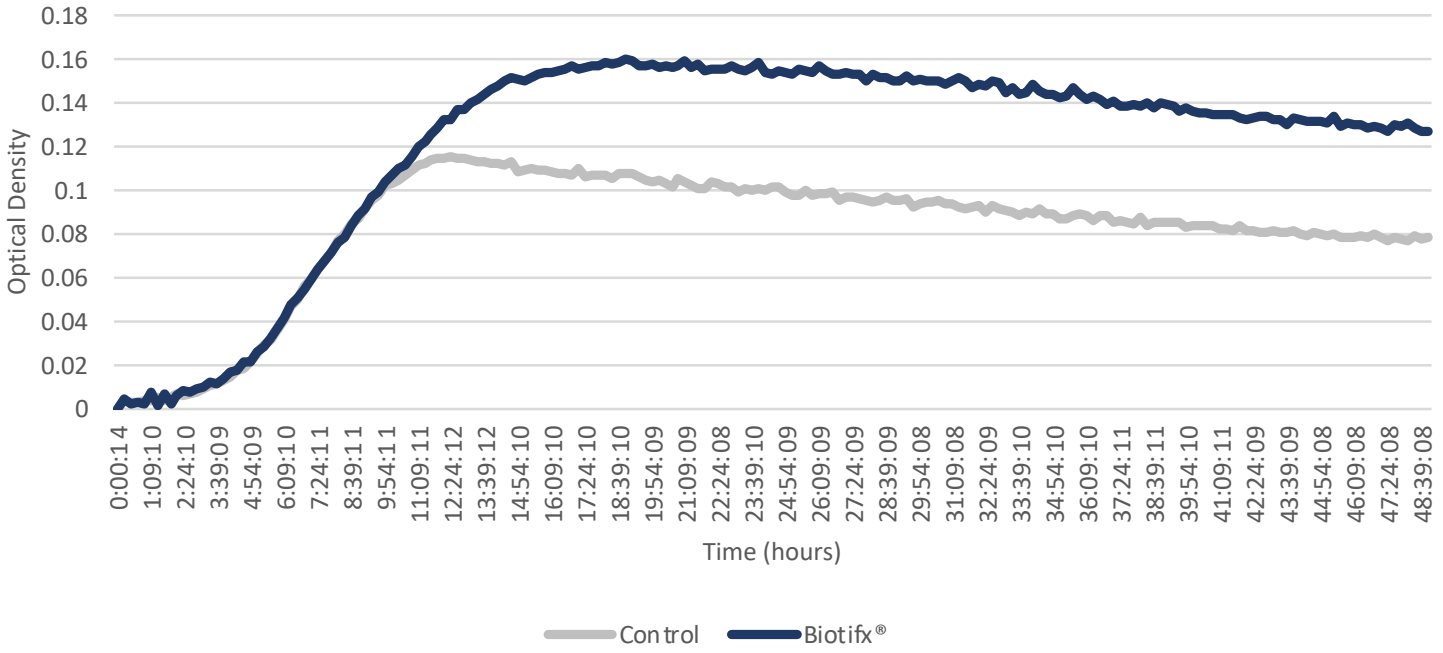

Figure 1. Bacterial growth as indicated by an increase in optical density was measured continuously for 48 hours in an untreated control wastewater sample and the same sample treated with Biotifx®. The sample treated with Biotifx® attains a higher level of bacterial activity overall compared to the untreated sample.

Bacillus bacteria are an ideal candidate for incorporation into wastewater bioaugmentation products. These organisms can enhance the bacterial community as described above and are highly adaptable to an array of environments due to their ability to metabolize a wide variety of substrates. Substances common in the sludge environment such as ammonia, carbohydrates, or fatty acids are taken up by *Bacillus* cells through membrane transports for use in cellular processes, thus clearing them from the system (6). More complex substances can also be digested thanks to the multitude of enzymes different *Bacillus* strains can produce such as protease, cellulase, lipase, amylase, keratinase, lignin peroxidase and many others (6, 9, 11). *Bacillus* can adjust enzyme production and metabolic processes to take advantage of whatever food source is most readily available. These organisms can even break down some of the more difficult to degrade complex carbon sources such as hydrocarbons or residual dyes from production processes that may otherwise persist undigested through wastewater treatment (10). Analysis of the whole genome sequences of Biotifx® *Bacillus* strains demonstrates that they are equipped to metabolize a wide variety of substrates that could be encountered in a sludge environment (Figure 2). Each *Bacillus* strain is unique, so a product designed to incorporate several complimentary strains is a very effective approach to wastewater sludge treatment (2).

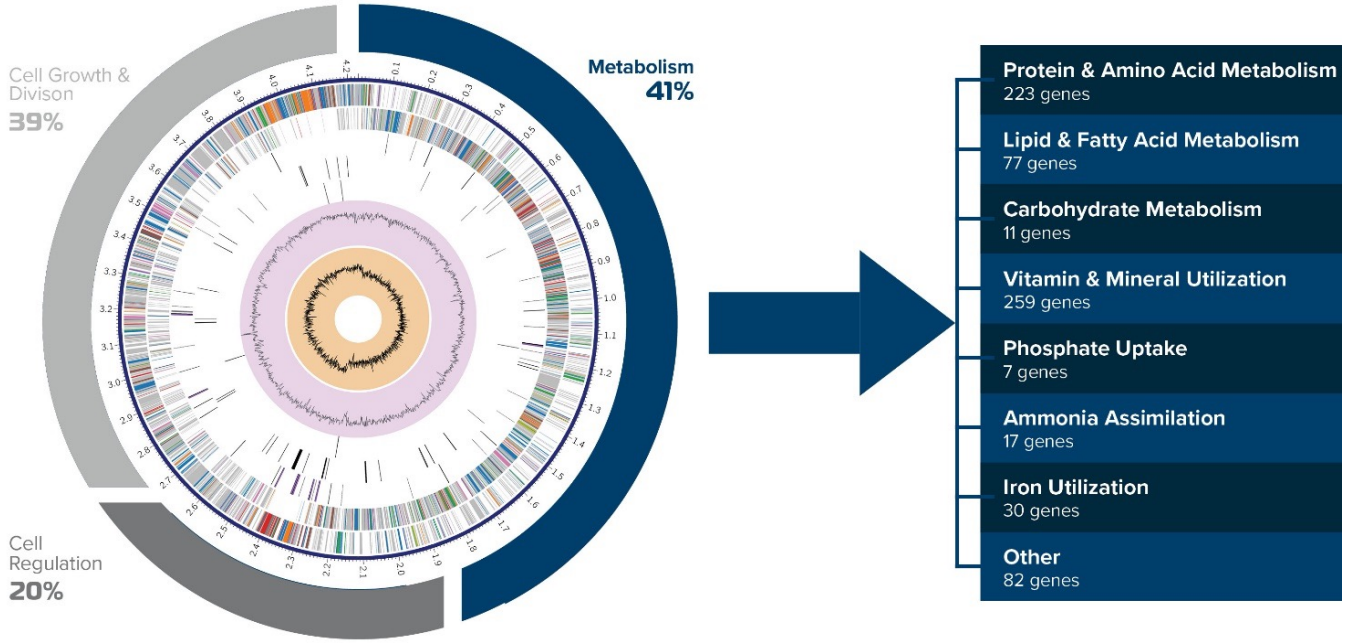

Figure 2. The genome of one of the *Bacillus* strains included in Biotifx® products is presented in a graphical representation. Each color in the ring represents a different subsystem or set of proteins that work in unison to execute a certain biological process that fall into the categories of metabolism, cell growth and division, and cell regulation. We then zoom in on the number of genes this strain has dedicated to metabolic processes. This data demonstrates the wide variety of substrates this organism has the genetic ability to metabolize.

Sludge accumulation is an important factor to consider in the health and efficacy of wastewater treatment lagoons. Bioaugmentation with *Bacillus*-based products is a reliable solution to reduce the need for more costly mechanical sludge removal methods. In this study, we have analyzed data in eleven diverse lagoon systems treated with Biotifx® and found that sludge was reduced by an average of 56% (SD = 33%) within the first year of treatment.

Methods

The goal of this study was to analyze sludge data from a range of different lagoons treated with Biotifx® products. The eleven lagoon systems studied were located across 8 different US states (n=9) and two additional countries (n=2) and spanned a wide range of industries and lagoon sizes (Table 1).

Before beginning treatment, each lagoon was surveyed to create an appropriate treatment plan. Basic lagoon statistics such as surface area, volume, and daily flow, along with water and sludge samples were collected. Sludge measurements were collected using a sludge judge at multiple locations throughout the lagoon proportional to lagoon size. These measurements were then used to calculate the average sludge depth of the lagoon. For Lagoon 7 and Lagoon 8 sludge loading was estimated using the facilities' daily loading data. In cases where sludge was so built up that it prevented proper sampling, the percent of the surface area with sludge up to or above the water surface of the lagoon was calculated before and after treatment.

Length of treatment with Biotifx® varied by location from 2 months to 1 year. Product was applied daily, weekly, or monthly depending on several factors including size of the system and daily flow rates (hydraulic retention time), severity of the sludge accumulation, and ease of application at the site.

Significance between pre- and post-treatment measurements was determined by performing a one sample T-test comparing the average percent difference between pre- and post-treatment to the hypothesized mean of 0 with a 95% confidence interval calculated using Minitab 17.

LAGOON SYSTEM	INDUSTRY	LOCATION	LAGOON SYSTEM VOLUME (GALLONS)	LAGOON SYSTEM FLOW (GALLONS/ DAY)
LAGOON 1	Food/Beverage Processing	US - Central	6,000,000	Unknown
LAGOON 2	Meat Processing	US - Midwest	5,000,000	300,000
LAGOON 3	Municipality	US - South	12,000,000	300,000
LAGOON 4	Dairy	US - Midwest	40,000,000	150,000
LAGOON 5	Meat Processing	US - Midwest	22,000,000	50,000
LAGOON 6	Food/Beverage Processing	International	55,000	4,500
LAGOON 7	Municipality	International	35,000,000	2,000,000
LAGOON 8	Pulp and Paper	US - Midwest	238,000,000	35,000,000
LAGOON 9	Municipality	US - Pacific	24,000,000	300,000
LAGOON 10	Meat Processing	US - Northwest	25,000,000	800,000
LAGOON 11	Meat Processing	US - Midwest	10,000,000	1,200,000

Table 1. Eleven lagoons analyzed in this study represent a variety of different locations, sizes, and industries. Location describes region within the United States (US), or international if located outside of the US. The lagoon volumes are rounded to the nearest 1,000,000 gallons (with the exception of Lagoon 6).

Results

Across all eleven lagoons there was a significant (p-value <0.05) reduction in sludge resulting from treatment with Biotifx® products, with an average of 56% (SD = 33%) sludge reduction. A wide range of lagoon sizes and sludge conditions pre-treatment contributed to high variability in results between systems, however there was measurable and valuable reduction in sludge in eleven of eleven systems.

The sludge reduction in each lagoon where sludge volume could be calculated based on sludge judge measurements (n=8) is reported in Figure 3. Lagoons 1-6 (Figure 3A) were smaller in size compared to Lagoons 7 and 8 (Figure 3B). Sludge reductions were observed in all eight of these lagoons following treatment with Biotifx®.

In addition to reducing the existing sludge in these lagoons, Biotifx® also digested incoming sludge, lowering the rate of sludge accumlation. When analyzing only the volume of sludge in the two largest systems pre- and post-treatment, there is a 2% and 8% reduction in sludge volumes for Lagoons 7 and 8 respecitvely (Figure 3B). However, these systems processed more than 2,000,000 gallons of flow per day resulting in large amounts of new sludge entering the lagoon during the treatment period on top of the high levels of sludge already present. Total sludge reduction, accounting for both the sludge volume that was in the lagoon and the amount of sludge loaded during the course of treatment, was actually 14% and 20% for Lagoons 7 and 8 (Figure 4).

There were three systems where sludge coverage on the surface of the lagoon restricted boat access and sludge judge measurements were not possible. Performance in these systems was evaluated based on the difference in the surface area covered by sludge pre- and post-treatment. Biotifx® treatment was able to reduce sludge coverage by an average of 92% (Table 2). In every lagoon system analyzed, regardless of industry, location, or size, treatment with Biotifx® resulted in a reduction in sludge.

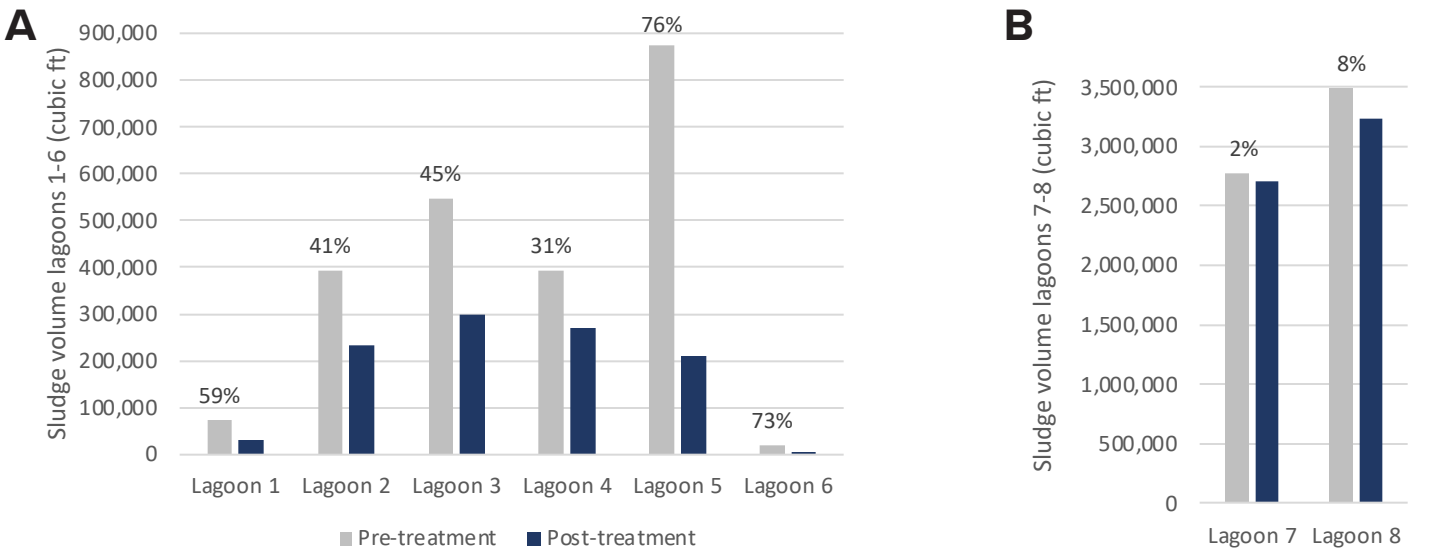

Figure 3. Reduction in sludge volume in lagoon systems treated with Biotifx®. Lagoon systems 1-6 had under 1,000,000 cubic feet of sludge accumulation (A) while lagoon systems 7 and 8 had over 2,000,000 cubic feet of sludge (B). Length of treatment ranged from 2 months to 1 year.

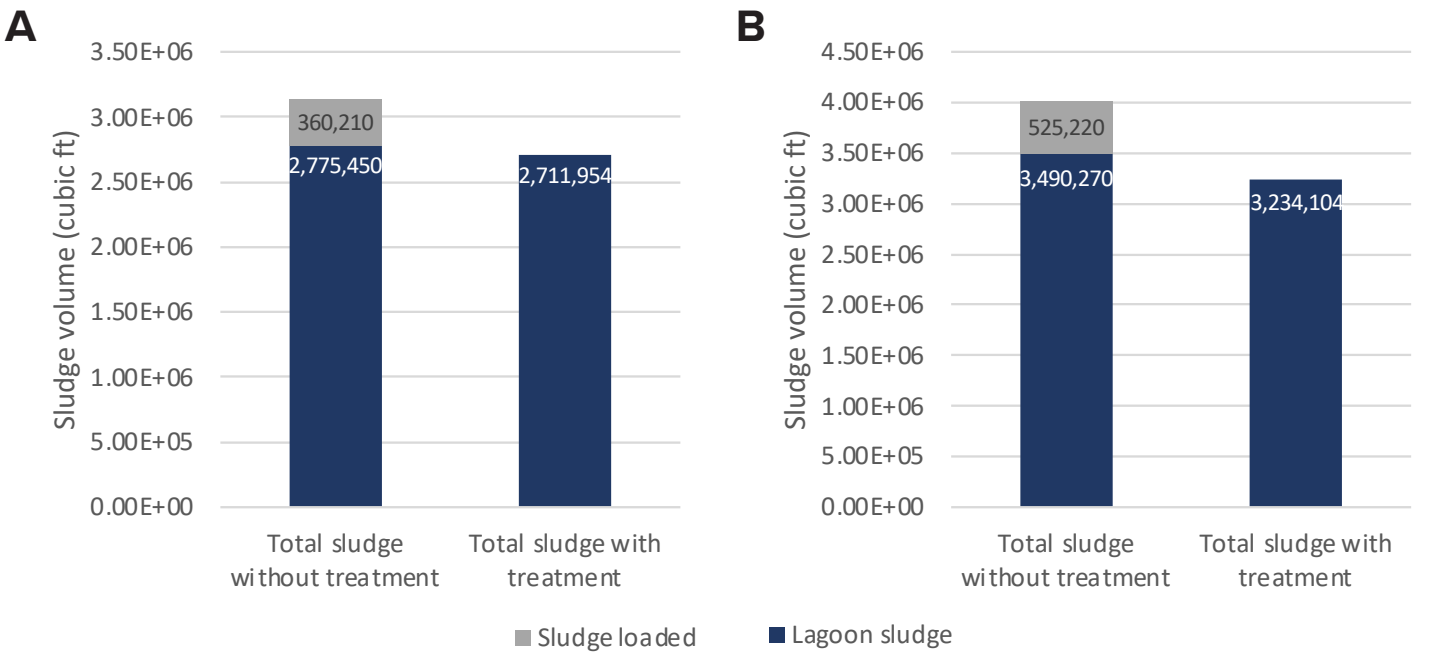

Figure 4. Total sludge reduction for lagoons treated with Biotifx®, including existing sludge and sludge loading volumes. When factoring in the amount of sludge loaded during the treatment period, the percent sludge reduction for Lagoon 7 (A) is 14% (versus 2% difference in sludge volume) and for Lagoon 8 (B) is 20% (versus 8% difference in sludge volume).

LAGOON SYSTEM	PRE-TREATMENT LAGOON COVERAGE (SQ FT)	POST-TREATMENT LAGOON COVERAGE (SQ FT)	PERCENT REDUCTION	LAGOON SYSTEM FLOW (GALLONS/ DAY)
LAGOON 9	110,000	<100	>99%	Unknown
LAGOON 10	27,600	4,600	83%	300,000
LAGOON 11	105,000	7,000	93%	300,000

Table 2. Difference in surface area (square feet) of the lagoon covered with sludge in the form of either floating sludge or sludge islands pre- and post-treatment with Biotifx®.

Conclusions

Biotifx® treatment resulted in sludge reduction in each of the eleven lagoon systems studied. This success was seen in systems of various sizes, across different industries and locations. In total, Biotifx® products were able to remove more than 2,000,000 cubic feet of sludge in the lagoons studied here. This amount of sludge equated to between 5:1 and 9:1 return on investment as reported by the water treatment facility compared to mechanical removal, sludge hauling, and disposal costs. Bioaugmentation using *Bacillus*-based Biotifx® products is an effective sludge management strategy.

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