

Bioaugmentation Is an Effective Solution for Pulp and Paper Wastewater Treatment

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Abstract

Pulp and paper wastewater treatment facilities produce large amounts of waste rich in hard-to-degrade plant fibers that require frequent mechanical intervention for removal and disposal. Mechanical dredging and disposal by transportation to a landfill and/or incineration are costly to the facility and have a large carbon footprint. *Bacillus* organisms have been shown to produce multiple lignocellulolytic enzymes that break down pulp and paper waste naturally. Therefore, bioaugmentation with *Bacillus* is a more cost effective and environmentally sustainable alternative for wastewater treatment and sludge removal at pulp and paper facilities. In this study, *Bacillus* strains demonstrating ligninolytic activity and the ability to produce β -mannanase, cellulase, and xylanase enzymes were combined with micronutrients to form the Biotifx[®] product. Laboratory testing using wastewater and sludge samples from a pulp and paper lagoon showed that *Bacillus* treatment resulted in improved overall bacterial growth, leading to solids and color reduction. The same lagoon was then treated daily with Biotifx[®] for 124 days to confirm that the efficacy observed in the laboratory would translate when applied in the field. Biotifx[®] treatment reduced sludge depth in the lagoon by 7.9 inches and removed 708 tons of dry solids with a 40% cost reduction compared to dredging and disposal of this amount of sludge. Bioaugmentation with *Bacillus*-based products was found to be an effective and sustainable option for treatment of pulp and paper wastewater.

Introduction

Pulp and paper facilities worldwide manufacture an estimated 400 million tons of board and paper, resulting in approximately 18 billion kilograms of dry sludge waste each year.^{1,2} These large amounts of sludge can limit system capacity and lead to inefficient wastewater treatment.³ The manufacture of pulp and paper products generates wastewater containing high amounts of plant fibers such as cellulose and lignin.^{4,5,6} The high concentration of plant fibers yields dark wastewater comprised of complex organic matter that accelerates sludge accumulation.^{1,6} High sludge volume paired with difficult-to-degrade organic material requires frequent mechanical removal to keep pulp and paper facilities operational.

Excess sludge is removed from the system using mechanical dredging followed by a dewatering process.¹ The remaining dry material is then disposed of either by transportation to a landfill or incineration. Costs for these disposal methods have been estimated at \$309 per ton for landfill and \$332–\$441 per ton for incineration.⁷ Landfill disposal costs are likely to rise as legislation attempts to reduce the amount of material entering landfills.¹

Additionally, these disposal methods have a large carbon footprint because they employ the use of heavy machinery for removal and transportation. Incineration also carries the potential to release harmful byproducts such as sulfur dioxide, nitrogen oxides, and possible chlorinated materials into the atmosphere.^{1,8} Pulp and paper facilities may face additional fines if the wastewater effluent remains too dark after treatment due to its high plant fiber content.^{5,8} Because of these challenges, the costs incurred for disposal of sludge and discharge of wastewater can add up quickly for pulp and paper manufacturers.

A more cost effective and environmentally safer alternative to traditional sludge removal and wastewater treatment methods is bioaugmentation. Bioaugmentation is the process of enhancing the microbial community that naturally exists in an environment through the addition of

bacterial species and/or nutrients that support microbial growth. One way *Bacillus*-based bioaugmentation products work is by producing enzymes that aid in the digestion of the difficult-to-degrade plant fibers found in pulp and paper sludge. Pulp and paper facilities present a unique challenge to the natural microbial community in their wastewater because of the high concentration of plant fibers such as cellulose, xylan, and lignin. These substances are large and complex, which means there are few bacterial species that can effectively degrade them (Figure 1).^{6,8,9} *Bacillus* species are known to produce a wide range of enzymes, including some of those necessary to break down plant fibers.^{8,9} However, not all *Bacillus* strains possess these capabilities. Bioaugmentation using a blend of *Bacillus* strains that can produce the specific enzymes necessary for treating high concentrations of plant fibers will be the most effective choice for improving water quality while reducing sludge and color in pulp and paper wastewater.

Supplementing *Bacillus*-based products with a micronutrient blend can further improve the effectiveness of a bioaugmentation treatment program. The high plant fiber content in pulp and paper waste leads to carbon-rich wastewater that can be deficient in other nutrients.⁵ In order to digest carbon, microbes utilize a balance of nitrogen, phosphorous, and other micronutrients; however, they may quickly use up these other available nutrients in carbon-rich water, leading to decreased bacterial growth and activity.³ Treatment with a bioaugmentation product that includes micronutrients helps to correct this nutrient imbalance and benefits the whole bacterial community. The presence of *Bacillus* working to digest fibrous material, along with increased availability of micronutrients necessary for bacterial activity, results in a measurable decrease in sludge volume over time.

In this study, we examined the efficacy of Biotifx[®], a *Bacillus*-based bioaugmentation product that includes micronutrients in pulp and paper wastewater treatment. *Bacillus* strains were first screened for the production of enzymes that target lignocellulosic material, and those with the best range of activity were blended to create the Biotifx[®] product. In the laboratory, this product effectively reduced color and solids in wastewater and sludge obtained from a pulp and paper lagoon. These laboratory findings were then

confirmed by field testing at the same pulp and paper lagoon. In this trial, treatment with Biotifx[®] removed 708 dry tons of sludge after four months. Bioaugmentation with a *Bacillus*-based product was found to be an economical and environmentally friendlier option for treatment of the large volumes of wastewater and sludge rich in difficult-to-degrade plant fibers that result from pulp and paper production.

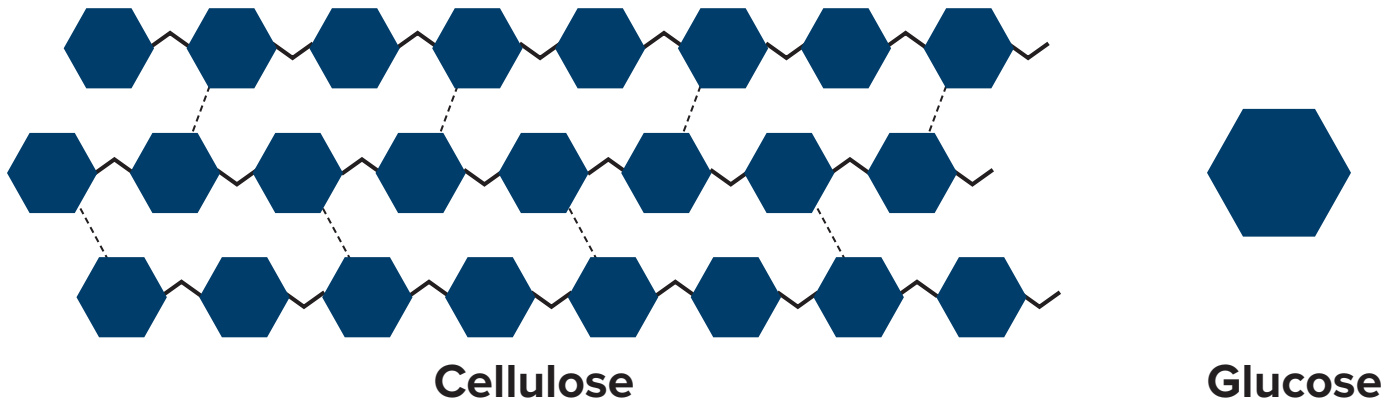


Figure 1. A simplified illustration of cellulose and glucose. Cellulose is a large, complex plant fiber with many more chemical bonds compared to a simple sugar like glucose. Glucose can be broken down by almost any bacterial species thanks to its simple composition, while complex plant fibers like cellulose can only be broken down by bacteria with specific enzymes.

Methods

Strain Characterization

A spot plate method was used to test *Bacillus* strains for the production of β -mannanase, cellulase, and xylanase, enzymes important for degradation of pulp and paper materials. Further analysis of ligninolytic capability was conducted by testing the *Bacillus* strains for the ability to degrade reactive black 5 dye (RB5). The same enzymes that degrade RB5 also degrade lignin; therefore, the ability to degrade RB5 would indicate the presence of ligninolytic enzyme activity.

Wastewater and Sludge Laboratory Testing

Wastewater and sludge samples were collected from three locations at a pulp and paper lagoon in order to assess if the Biotifx[®] product would be effective in treating pulp and paper wastewater. Water samples were collected at the surface from near the influent, at the middle of the lagoon, and near the effluent. Sludge samples were collected in the same three locations using a sludge judge. Wastewater treatment efficacy was evaluated by treating each water sample with Biotifx[®] for 72 hours. Higher optical density (OD) indicated a greater amount of bacterial growth. The efficacy of Biotifx[®] treatment in the sludge was tested in lab by measuring reduction of solids. Sludge samples were treated with the microbial product for five days and total solids were measured at Day 0 and Day 5. The OD of the sludge samples with and without Biotifx[®] was also measured to assess color reduction on Day 5. Statistical analysis was performed on the wastewater test results and the sludge sample OD results using a T-test with a 95% confidence interval.

Field Testing

The same pulp and paper lagoon from which samples were collected for lab testing was treated with Biotifx[®] to demonstrate efficacy in the field. The 25-million-gallon lagoon with a 32-million-gallon per day flow was treated daily with 0.1 ppm of Biotifx[®] for 124 days. Depth to sludge was measured prior to beginning treatment and was monitored monthly throughout treatment using a remote control sonar boat and confirmed using a sludge judge at multiple locations throughout the lagoon. The lagoon water level was measured each month to account for the effect of surface-level changes on depth-to-sludge measurements. Total suspended solids (TSS) in the influent and effluent were measured daily, and this data was used to calculate the solids loaded into the system during treatment as well as the water quality of the effluent. Average sludge level changes and solids loading data were used to calculate the dry tons of sludge removed over the course of treatment.

Results

Strain Characterization

Bacillus strains were analyzed individually to characterize their ability to degrade the plant fibers commonly found in pulp and paper wastewater. Each strain of *Bacillus* was screened for the production of ligninolytic enzymes. The strains selected to make up the Biotifx® product produced a range of the enzymes tested, resulting in a blend that was able to produce β-mannanase, cellulase, and xylanase

(Table 1). Additionally, all *Bacillus* strains in the product demonstrated the ability to degrade RB5, which is observed by a color reduction (Figure 2). The same enzymes that degrade RB5 also degrade lignin; therefore, the ability to degrade RB5 indicated ligninolytic enzyme activity. Based on the enzyme production characterized in the Biotifx® *Bacillus* strains, it was expected to perform well in pulp and paper wastewater treatment.

	β-mannanase	Cellulase	Xylanase
BIOTIFX®	✓	✓	✓

Table 1. The *Bacillus* blend that made up the Biotifx® product was formulated to include strains which produce the fiber-degrading enzymes β-mannanase, cellulase, and xylanase.

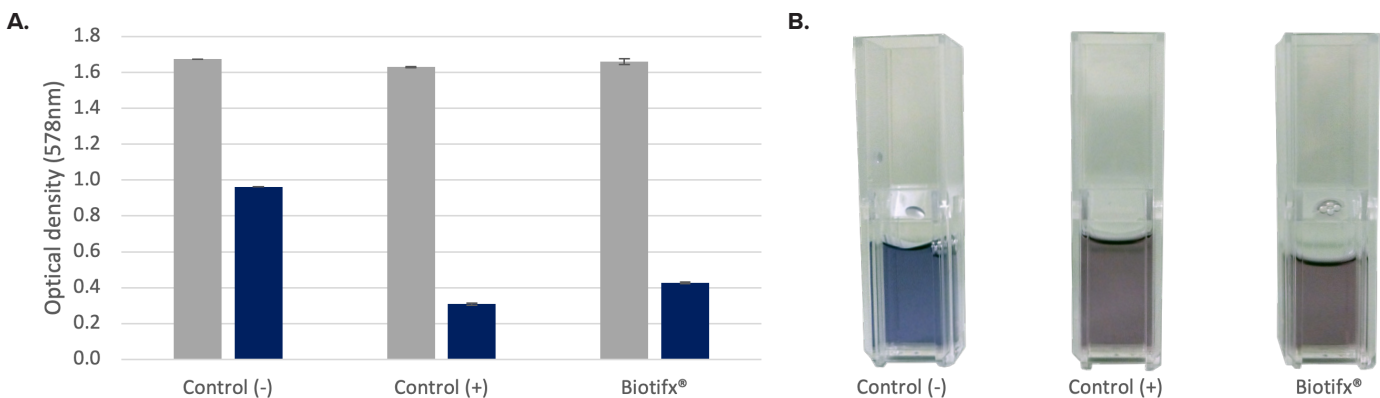


Figure 2. Degradation of RB5 dye was used as an indicator of ligninolytic activity. Results for Biotifx® were compared to an untreated negative control and a known positive strain. The decrease in optical density after three days indicated that Biotifx® *Bacillus* strains were capable of ligninolytic enzyme activity [A]. There was also a visual difference in the color of RB5 when treated with the product [B].

Wastewater and Sludge Laboratory Testing

Laboratory testing was conducted to determine if Biotifx®, a product formulated with a blend of *Bacillus* strains and micronutrients, could effectively treat pulp and paper wastewater. Bacterial growth in pulp and paper wastewater samples increased significantly (p-value <0.05) when treated with Biotifx® compared to the untreated control after 72 hours (Figure 3). The OD of the sludge samples treated with Biotifx® was significantly (p-value <0.05) lower, resulting in a visual color reduction (Figure 4). Treatment with Biotifx® was effective in laboratory testing, resulting in increased bacterial activity and color reduction

in pulp and paper lagoon samples. This indicated that dense plant fibers were being degraded, which led to a reduction in biological oxygen demand, TSS, and color in the wastewater.

The effect of *Bacillus* treatment on pulp and paper solids was also studied using sludge obtained from the lagoon. There was an improvement in solids reduction in the samples treated with Biotifx® compared to the untreated control (Figure 5). This solids reduction observed in lab suggests Biotifx® would reduce sludge levels in a pulp and paper lagoon.

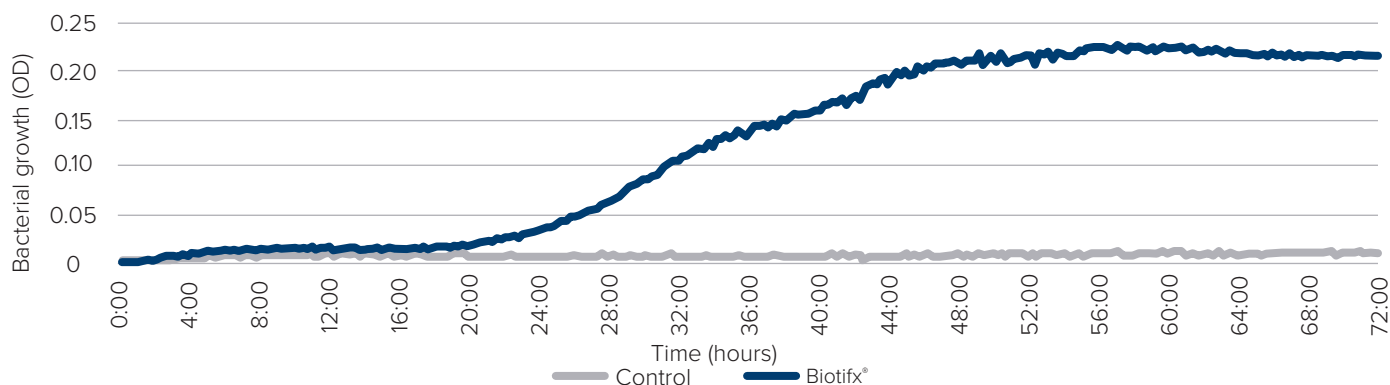


Figure 3. Bacterial growth was measured by optical density (OD) in water samples collected from a pulp and paper lagoon prior to treatment with Biotifx®. Bacterial growth was significantly (p-value <0.05) higher after 72 hours when treated with Biotifx® compared to untreated control water samples.

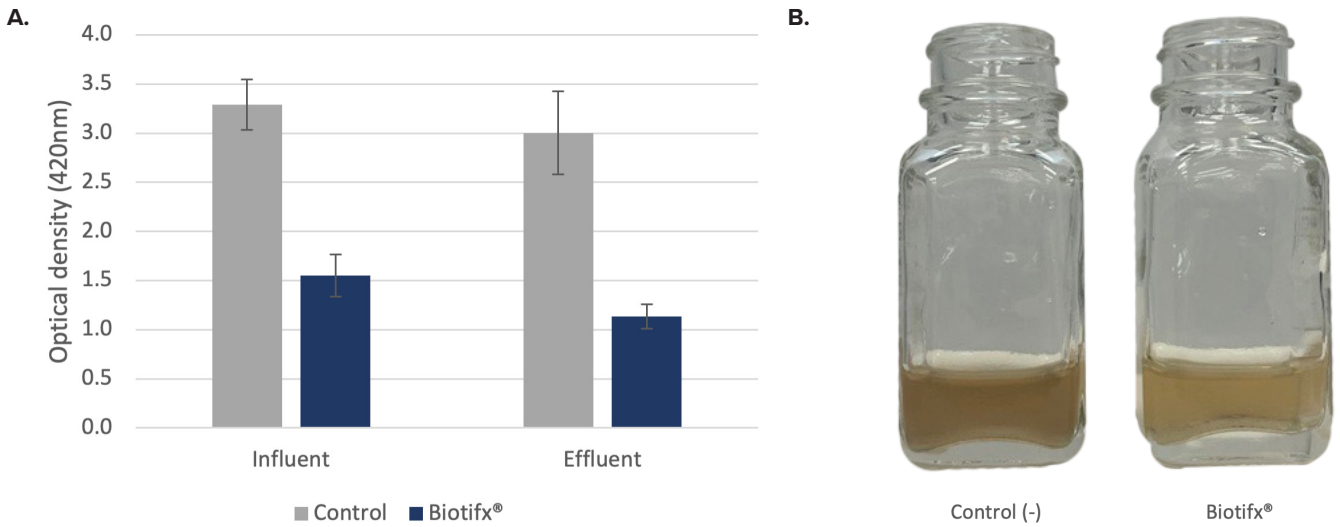


Figure 4. Color reduction was analyzed in sludge samples treated with Biotifx[®] collected from the pulp and paper lagoon influent and effluent compared to untreated control samples. This bioaugmentation treatment significantly (p -value <0.05) decreased the optical density of influent and effluent samples after five days [A], and there was a visual color reduction after treatment with Biotifx[®] [B].

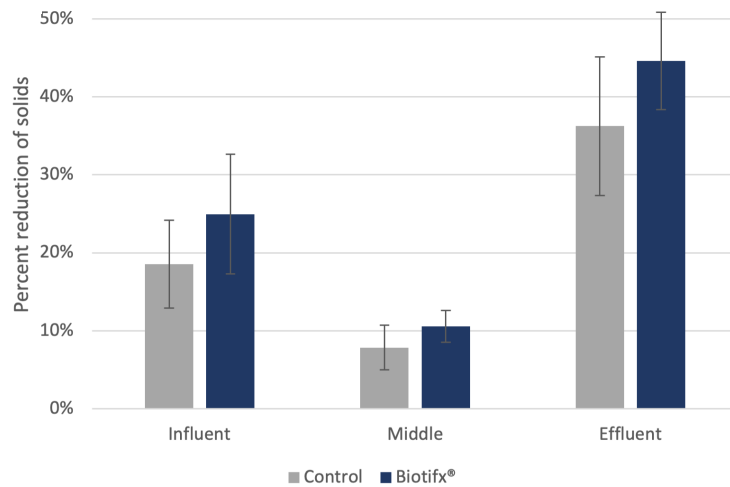


Figure 5. Treatment with Biotifx[®] resulted in a numerical increase in the reduction of solids in pulp and paper lagoon sludge samples after five days compared to untreated control samples.

Field Testing

The efficacy of Biotifx[®] treatment was then evaluated in field testing at the pulp and paper lagoon from which samples were previously collected for the laboratory testing. Evaluation of the lagoon through sonar and survey prior to treatment showed solids were visible above the water level (Figure 6A and 7A). After 124 days of treatment with Biotifx[®], the average sludge depth decreased by 7.9 inches and solids were no longer visible above the water (Figures 6B, 7B, and 8B). While treating the lagoon, there was a period between August 22 and October 1 where TSS loaded into the lagoon

was 41% above the upper control limit (UCL) (Figure 8A). Despite the high levels of solids coming into the lagoon, the effluent quality improved during treatment when the average TSS removal rate within the lagoon increased by 28% compared to pre-treatment baseline levels (Figure 9). Treatment with Biotifx[®] proved effective in field testing, resulting in the removal of 708 dry tons of solids in 124 days despite the period of above-average solids loading while also improving the quality of the effluent.

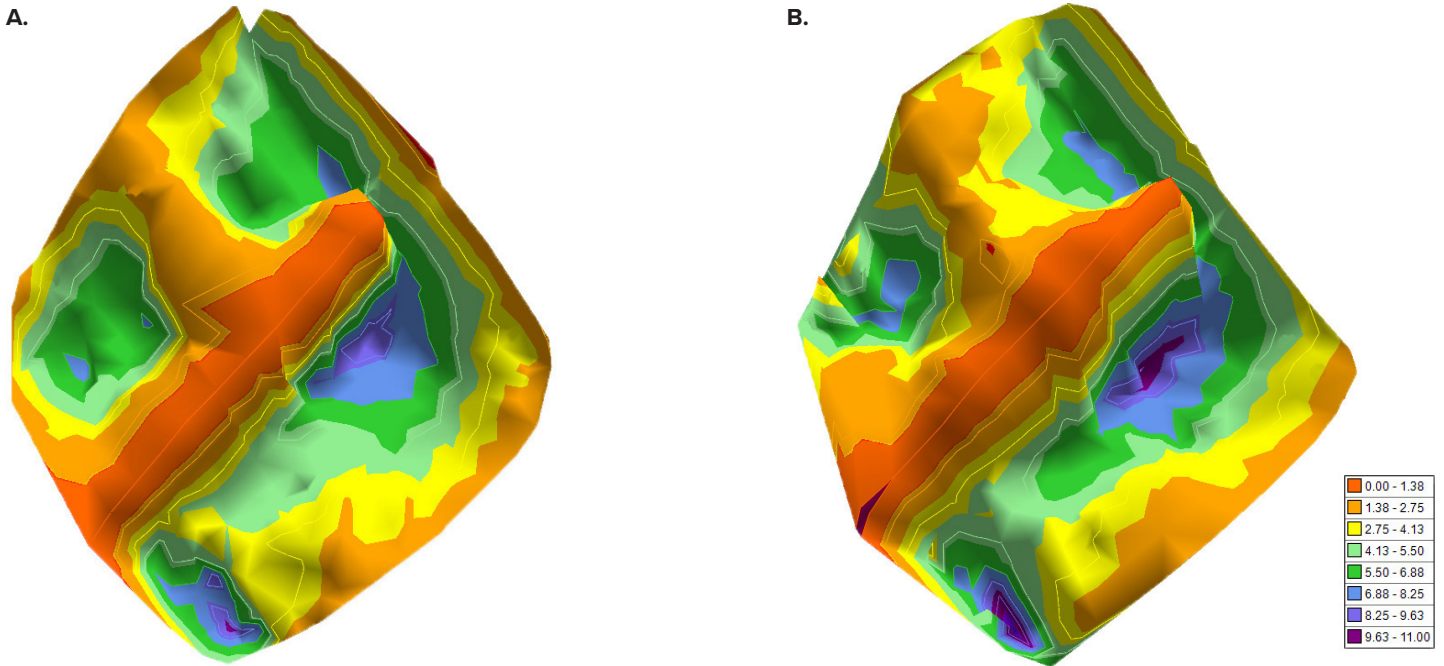


Figure 6. Sonar images before [A] and after [B] treatment. Sonar testing displays shallower areas in the orange, yellow, and light green color range, while deeper areas appear in the dark green, blue, and purple color range. Many areas of the lagoon deepened as a result of Biotifx® treatment, as illustrated by the increase in blue and purple areas after treatment.



Figure 7. Lagoon images before [A] and after [B] treatment. The image prior to treatment shows that solids had accumulated above the water level [A]. After Biotifx® treatment, solids no longer appeared above water level and the sludge level decreased by an average of 7.9 inches [B].

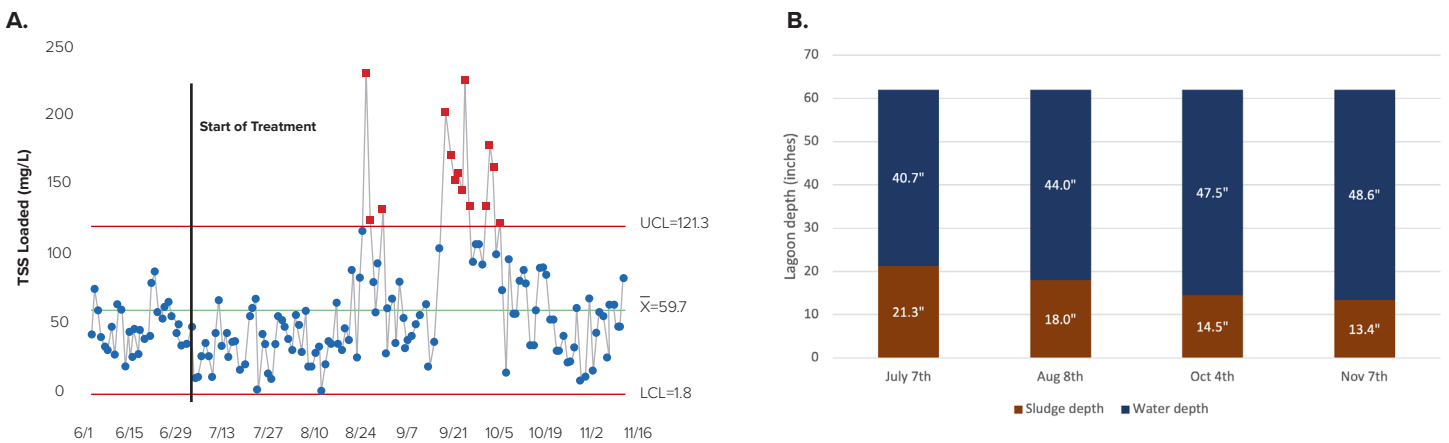


Figure 8. TSS loaded into the lagoon was measured and recorded daily prior to beginning and throughout the duration of treatment. TSS was above the UCL at times between August 22 and October 1, indicating high loading [A]. During treatment, the average sludge depth was measured monthly, and an overall reduction of 7.9 inches was observed despite the periods of above-average loading [B].

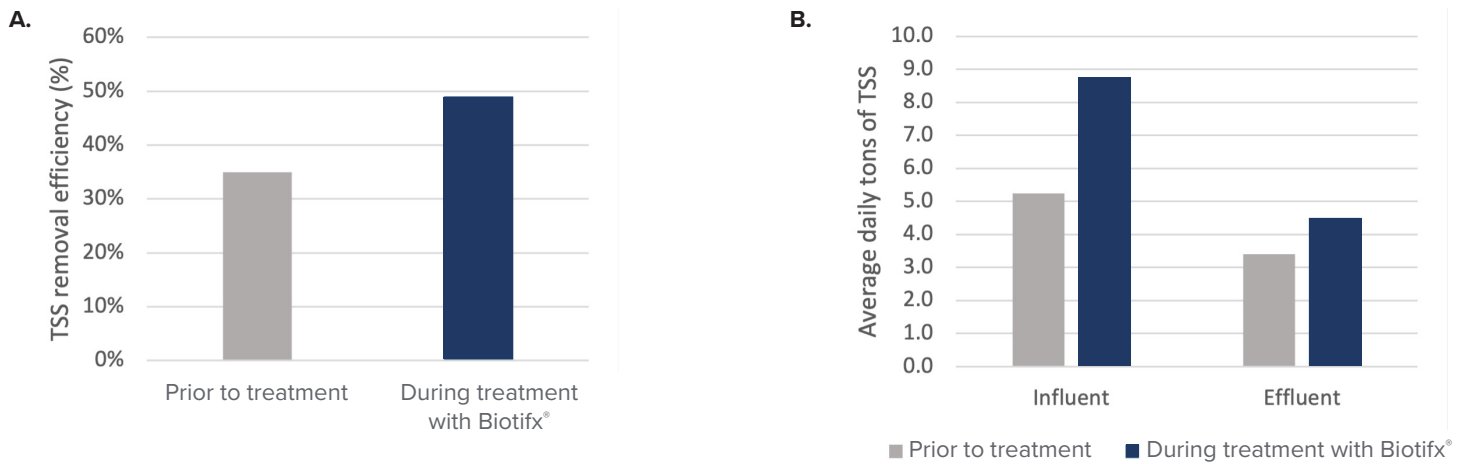


Figure 9. Daily tons of TSS prior to treatment (127 days) and daily tons of TSS during treatment with Biotifx® (124 days) were measured in the influent and effluent [A] and used to calculate the efficiency of TSS removal in the lagoon. Treatment increased the efficiency of TSS removal from 35% to 49% [A] despite the increase in daily tons of influent TSS during the treatment period [B].

Conclusions

Bioaugmentation with Biotifx® was an effective treatment for pulp and paper wastewater. Laboratory testing demonstrated that the *Bacillus* strains in the product have ligninolytic enzyme activity, which is one of the key components for success when treating pulp and paper sludge containing large amounts of complex plant fibers. Wastewater and sludge treatment with Biotifx® in the laboratory was effective, resulting in increased bacterial growth, color reduction, and solids digestion. Laboratory results were confirmed in the field study, where Biotifx® treatment resulted in an average reduction in sludge depth of 7.9 inches and the removal of 708 tons of dry solids after 124 days.

Prior to treatment, the facility was adding 4.5 inches of sludge and 1,000 tons of dry solids annually, which needed to be mechanically removed, dewatered, and transported to a landfill. Despite an increase in solids loading during the treatment period, Biotifx® was able to lower the overall sludge level while improving effluent quality without mechanical intervention. This biological sludge removal required no additional labor or equipment for the facility, decreased its carbon footprint, and equated to a 40% cost reduction compared to dredging and disposal of the same amount of sludge. Bioaugmentation with *Bacillus*-based products is an effective and more sustainable option for treatment of the wastewater and large sludge accumulation that results from pulp and paper production.

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