# Mycoremediation Applications for Stormwater Management

**Design Challenge:** Airport Environmental Interactions; Innovative methods for stormwater management at airports

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# Abbreviations

ACRP – Airport Cooperative Research Program
ADF – Aircraft Deicing Fluid
BMP – Best Management Practice
CDPHE – Colorado Department of Public Health and Environment
DIA – Denver International Airport
EMS – Environmental Management System
EPA – Environmental Protection Agency
LiP – Lignin Peroxidase
MCDA – Multiple Criteria Decision Analysis
MnP – Manganese Peroxidase
NPDES – National Pollutant Discharge Elimination System Stormwater Program
PAH – Polycyclic Aromatic Hydrocarbons
PCB – Polychlorinated Biphenyls
SMS – Spent Mushroom Substrate
SSC – Suspended Sediment Concentration
$S^3$ – Sustainable Stormwater Solutions
SWMP – Stormwater Management Plan
TPH – Total Petroleum Hydrocarbons
TSS – Total Suspended Solids
TWG – Targeted Watershed Grant
WSU – Washington State University

# Executive Summary

Sustainable Stormwater Solutions ($S^3$) is a group of six undergraduate engineering students from the University of Colorado, Boulder. In order to participate in the Airport Cooperative Research Program (ACRP) University Design Competition for Addressing Airport Needs, $S^3$ partnered with Denver International Airport (DIA) to design an innovative solution to address a challenge in stormwater management as identified by DIA. The main challenge identified was the recurrence of elevated pH levels in stormwater runoff. DIA linked the elevated pH levels to concrete cutting activities or a new concrete curing agent; precipitation events transport the wet concrete slurry produced by the cutting activities into the stormwater conveyance systems.

As part of a rigorous design process, $S^3$ submitted a proposal with potential treatment options based on a preliminary literature review. The treatment options were developed further in the alternatives analysis phase, where each treatment option was evaluated based on key criteria that were finalized through communication with contacts at DIA. The alternatives were compared against each other, and a mycoremediation/fungal bioremediation alternative was chosen to develop for the final design to submit to the competition. Detailed plans for various phases of implementation, cost estimates, operations and maintenance plans, safety assessments, and social/environmental impacts were developed as part of the final design process.

Finally, mycoremediation was evaluated for its versatility and potential to treat a number of different contaminants that are prevalent at airports. The low cost, easy implementation, and easy maintenance of mycoremediation solutions act as indicators that mycoremediation may be a new, environmentally sustainable, and economically feasible solution for many remediation applications at airports around the world.

# 1.0 Problem Statement and Background

Airports, like any other industrial operation, produce waste materials and chemicals that must be managed in order to protect public and environmental health. Contaminants such as aircraft deicing fluid (ADF), fuel, or solvents can be transported through various mechanisms (e.g. surface runoff or leaks) [1]. Being able to properly manage potential contaminants by keeping them away from soils and groundwater, and being able to promptly treat any contamination is imperative to the safe operation of any airport.

Sustainable Stormwater Solutions ($S^3$) partnered with Denver International Airport (DIA) to participate in the Airport Cooperative Research Program (ACRP) University Design Competition, focusing on innovative methods for stormwater management at airports. DIA operates on a property area of 53 square miles, with six 12,000+ ft. runways, 103 gates, and over 50 million annual travelers [2]. In order to continue the improvement of their operations, DIA has identified on-site issues related to stormwater management that require engineering solutions. The main issue that DIA identified was elevated pH levels in stormwater runoff. This phenomenon is linked to concrete cutting activities; during maintenance of roadways and runways, concrete cutting generates a wet concrete slurry with elevated pH. During precipitation events, uncollected remnants of this slurry are carried off by stormwater, into the drainage system. During a number of high flow events, the contamination from the concrete slurry resulted in pH levels above 9.0, exceeding the allowable levels for DIA's stormwater permit. DIA requested $S^3$ to investigate and design a pilot-scale test for the main affected site, the Pond 927/Outfall A area shown in Figure 1.


*Figure 1: View of Pond 927/Outfall A, with Outfall A in the orange rectangle. Proposed pilot-test location is at two inlets into Pond 927, shown in the green circle.*

Runway maintenance and concrete cutting are both necessary and common occurrences at any airport. A picture of concrete cutting is shown in Figure 2. Other airports may observe elevated pH in stormwater due to the necessity of concrete work. However, variations in pH can also be linked to many sources. For example, elevated pH was measured in certain detention ponds at Seattle Tacoma Airport in 2014-2015 and attributed to algal growth [3]. The main goal of $S^3$ was to find an effective yet affordable solution to mitigate the effects of elevated pH levels in stormwater at DIA in hopes that other airports with similar issues may be able to adopt the solution as well.


*Figure 2: Photo of concrete cutting on a runway. [32]*

## 2.0 Problem Solving and Design Approach

The design process of this project included three phases: proposal, alternatives analysis, and design development. In the proposal phase, preliminary ideas of solutions, project scope, project schedule, and expected deliverables were presented to the project principal and DIA. Preliminary ideas were developed through literature review of the problem and existing solutions. $S^3$ proposed three options to develop in the alternatives analysis phase: bioretention, chemical addition, and mycoremediation/fungal bioremediation.

In the alternatives analysis, these initial ideas were refined by a thorough and focused literature review, contacting experts within the field, visits to the proposed site location, and preliminary identification of design parameters. $S^3$ evaluated current and future conditions at the site, regulatory requirements, and site constraints. Each alternative was developed by investigating case studies/history, potential challenges/limitations, and technical evaluations were provided with preliminary sizing and layout as applicable. Alternatives were quantitatively rated using a multiple criteria decision analysis (MCDA) and cost estimates were calculated for each alternative. As shown in Table 1, the MCDA framework was created with criteria that fell into three categories: technological, environmental, and social. Evaluating the criterion within the Environmental category involved completing a list of questions selected from the ENVISION checklist. The ENVISION checklist is used to determine how sustainable a project will be over its lifecycle [4]. Shown in Figure 3, MCDA scores were graphed against costs, and the alternative that combined a high MCDA score with a low cost estimate was considered to have the best value and selected for recommendation.

*Table 1: MCDA template used to rate alternatives. Maximum/best scores are shown in this table.*

| Category | Category Weight | Criteria | Weight | Score (1-10) | Weighted Score | Category Score | Weighted Category Score |
|---|---|---|---|---|---|---|---|
| Technological | 0.75 | Technical Maturity | 0.34 | 10 | 3.40 | 10.00 | 75 |
| | | Operational Impact/Safety | 0.33 | 10 | 3.30 | | |
| | | Lifespan/Maintenance | 0.33 | 10 | 3.30 | | |
| Environmental | 0.2 | ENVISION Score/Local Ecosystem Impact | 1.00 | 10 | 10.00 | 10.00 | 20 |
| Social | 0.05 | Social Acceptability | 1.00 | 10 | 10.00 | 10.00 | 5 |
| | | | | | | **Total Score:** | **100** |

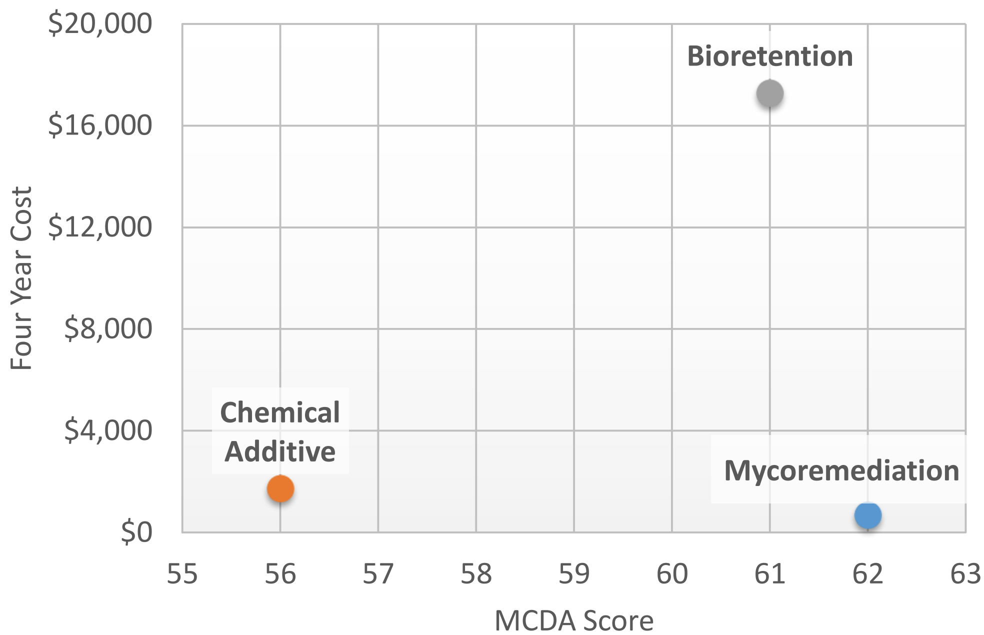


*Figure 3: Cost versus MCDA Score for each alternative.*

Mycoremediation was selected as the alternative to bring to the final design for the competition. The level of technical maturity of mycoremediation called for the final design to include a bench-scale treatability study in addition to a pilot-scale study to investigate the treatment potential of mycoremediation. Further literature reviews and discussions with experts in the field were necessary to develop the final design for the mycoremediation alternative.

# 3.0 Summaries of Literature Review

A literature review for each alternative was conducted, and relevant studies were selected to support the viability of the alternative for this project. Relevant case studies were evaluated by applicability, and limitations were noted and factored in to preliminary design considerations.

## 3.1 Bioretention

### 3.1.1 Background

Bioretention, also known as a biofilter or rain garden, is currently the most widely used stormwater best management practice (BMP) in the United States [5]. Biofilters are considered low impact and are best used in water sensitive urban environments. Rain gardens are small, aesthetically pleasing, and help solve several stormwater management objectives, including the reduction of stormwater peak flow, runoff volume, stormwater pollution, maintenance of groundwater recharge, and stream base flow. The water quality performance of bioretention systems has mainly been assessed in experimental settings [5]. These tests have shown that bioretention systems possess the ability to reduce sediments, heavy metals, and nutrients from synthetic stormwater.

One of the most common goals for stormwater management includes the reduction and removal of pollutants in stormwater runoff. The different mechanisms a rain garden can use to reduce specific types of pollutants in stormwater runoff are shown in Table 2 [6].

*Table 2: Specific pollutants and the removal mechanisms [25]*

| Pollutant Removal Mechanism | Pollutants |
|---|---|
| Absorption to Soil Particles and Plant Uptake | Dissolved metals, soluble phosphorus |
| Microbial Processes | Organics, pathogens |
| Exposure to Sunlight Dryness | Pathogens |
| Infiltration to Runoff | Nutrients |
| Sedimentation and Filtration | Total Suspended Solids (TSS), floating debris, trash, soil bound phosphorus, some soil bound pathogens |

Sedimentation and filtration are the most effective mechanisms for pollutant removal. Bioretention/rain garden systems most effectively remove TSS. A typical design schematic of a cross-sectional view of a bioretention/rain garden system is shown in Figure 4 [6].

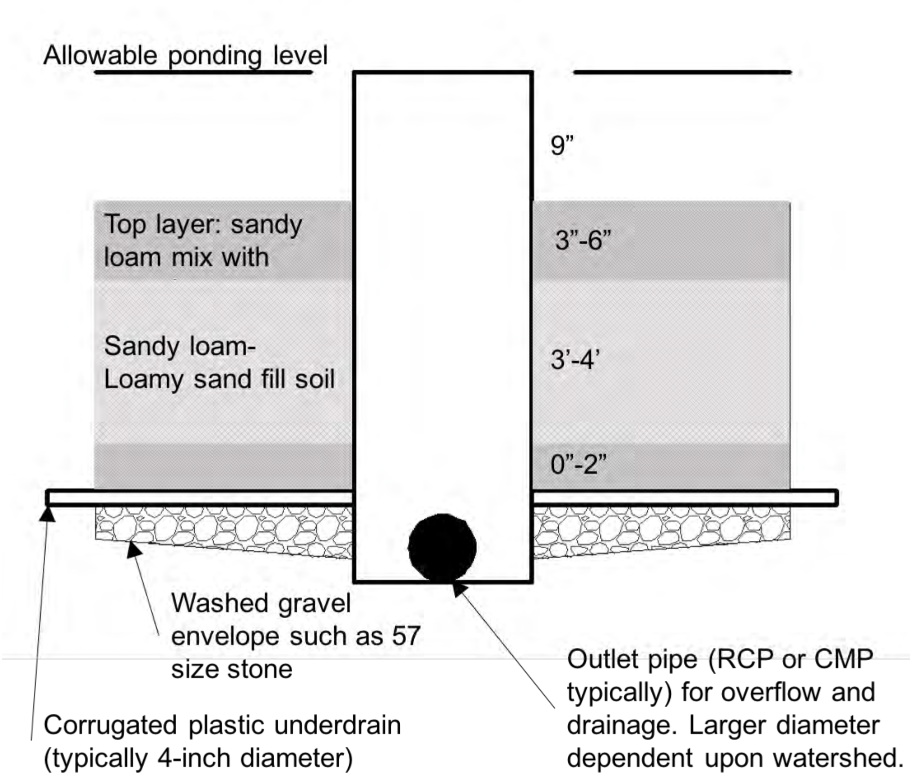


*Figure 4: Cross-sectional diagram of a detention system.* [6]

### 3.1.2 Case Studies

A study conducted in Auckland, New Zealand investigated the removal of the following pollutants using standard laboratory methods: TSS, total Cu, Pb, and Zn. Although these contaminants are not causing the elevated pH levels in DIA’s stormwater, the effectiveness of this bioretention system to reduce TSS and these metals can support the ability of bioretention to remove the concrete slurry particulates found in DIA’s stormwater. The system was effective at reducing TSS in the outflow concentration by a factor of 10 between the inflow and outflow measurement points. The changes in outflow concentrations of the specific pollutants measured are shown in Figure 5 [5]. The biofilter system worked extremely well in reducing TSS, Pb, and both dissolved and total Zn. The data indicates that the system was able to efficiently reduce both dissolved contaminants and particulates that were in the stormwater through plant uptake and soil particle absorption.

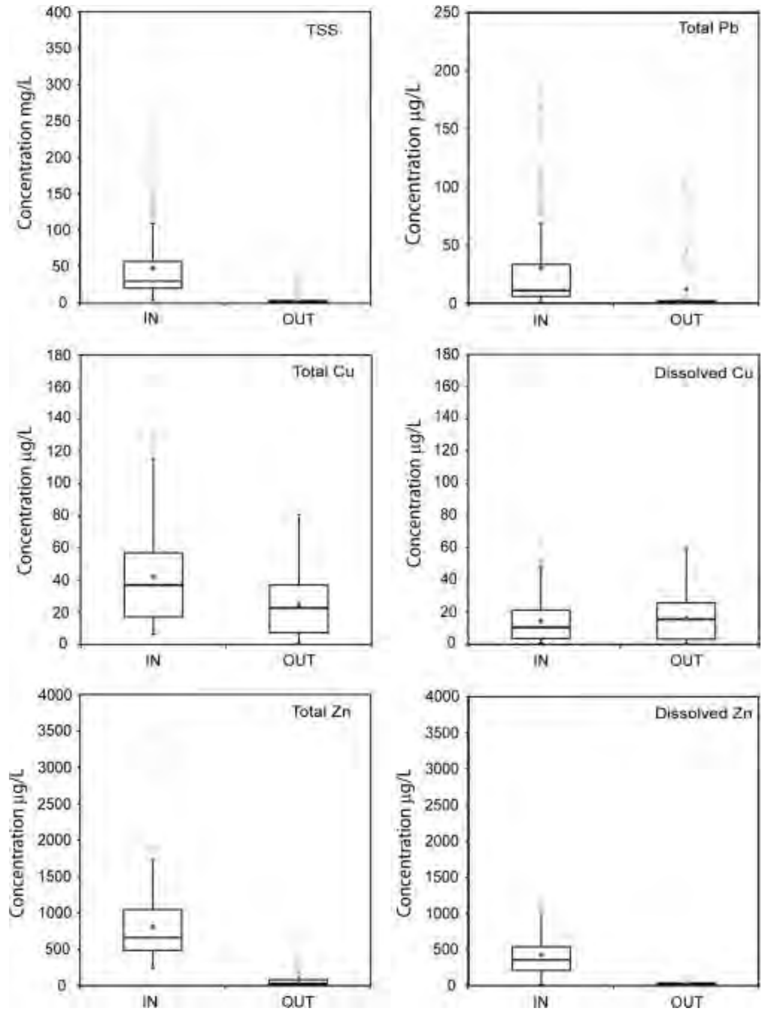

*Figure 5: Concentration changes from inflow to outflow of specific pollutants.* [5]

For bioretention systems to adequately treat water, they must be efficient in collecting runoff during storm events. Designing these systems based on the hydrology of the area is important to ensure correct ponding depth and surface area for efficient capture. A total of 12 rainfall events were monitored and data of the event duration, rainfall, peak inflow, and flow volumes were all recorded. The amount of bypassed volume versus the amount of rainfall that occurs for each event is shown in Figure 6 [5].

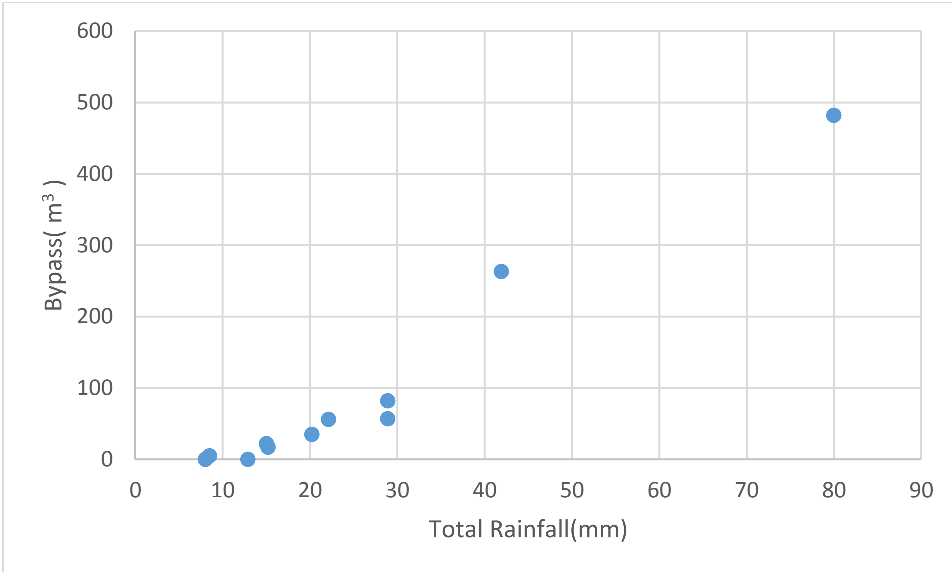


*Figure 6: Graph of bypass volume vs amount of rainfall for each precipitation event.* [5]

The trend in Figure 6 shows that increasing amounts of rainfall during a storm event causes a higher bypass volume. Optimizing the design factors permeability and surface area with the hydrology of DIA will minimize bypass and overflow volume. The ability of the bioretention system to collect runoff effectively will be important in maintaining the neutralized pH and water quality downstream.

All of DIA's monitoring of pH and TSS is performed during the colder winter season from October to May. During these months stormwater runoff has different contaminants and hydrological characteristics. Typically, DIA has higher sediment loads and chloride concentrations in stormwater runoff from road de-icing. Hydrologically there is a lower intensity of runoff rates because of the slow release of water from snowmelt. The cold climate has the ability to change both the hydrological and water quality performance of the system. The city of Calgary conducted a study on the effects of cold weather conditions on bioretention [7]. From the study, cold conditions generally saw higher peak flows, lower amount of volume of runoff stored (higher effluent volumes), and initial soil moistures did not differ. The differences of soil moisture and flow rates compared to warm weather conditions are shown in Figure 7 [7].

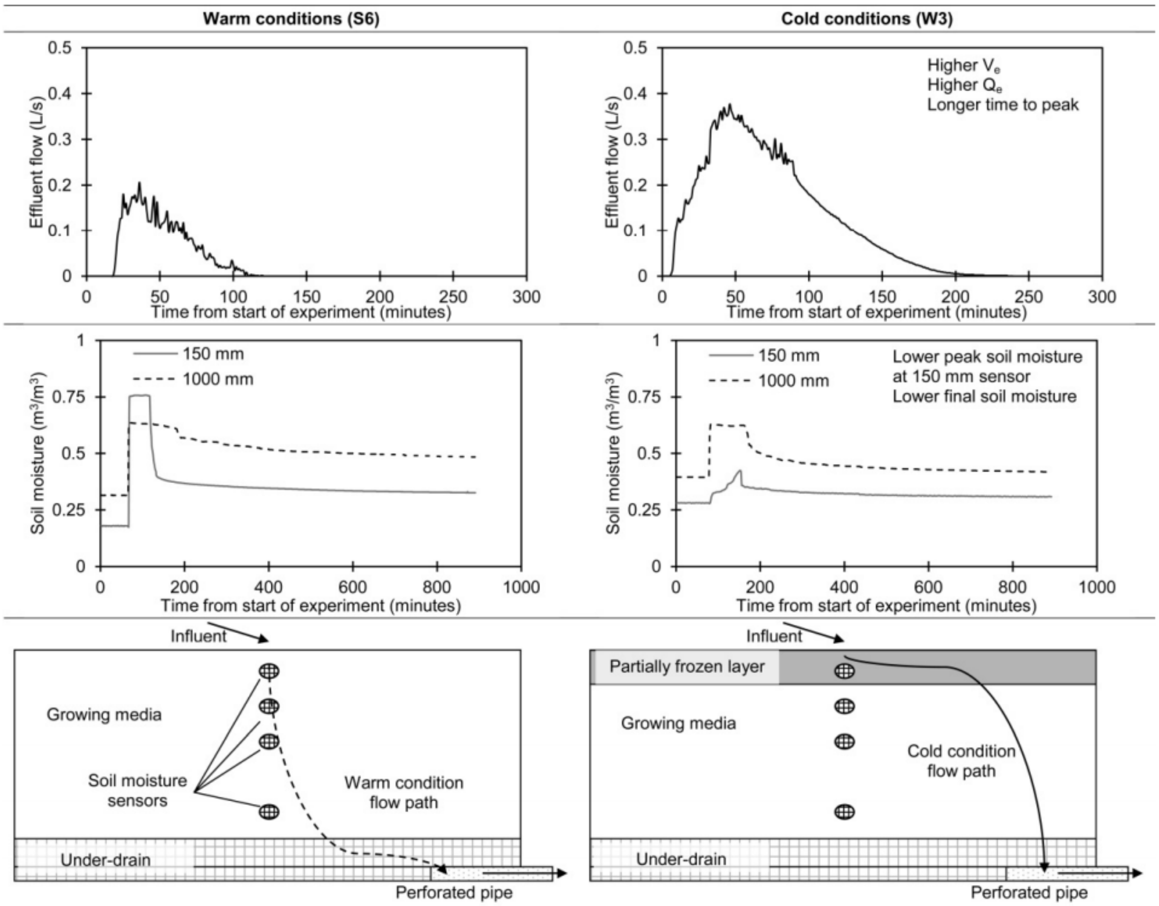


*Figure 7: Warm condition retention times, moisture, and infiltration pattern compared to cold conditions* [7]

Although cold weather can cause a short circuiting effect the performance change did not significantly affect the system's ability for pollutant removal. The change in concentration and mass of specific pollutants in the tested stormwater runoff can be seen in Figure 8. The ability of this system to remove both TSS and suspended sedimentation concentration (SSC) effectively indicates that such a system at DIA, even during colder weather, can perform efficiently in neutralizing pH by removing particulates in stormwater.

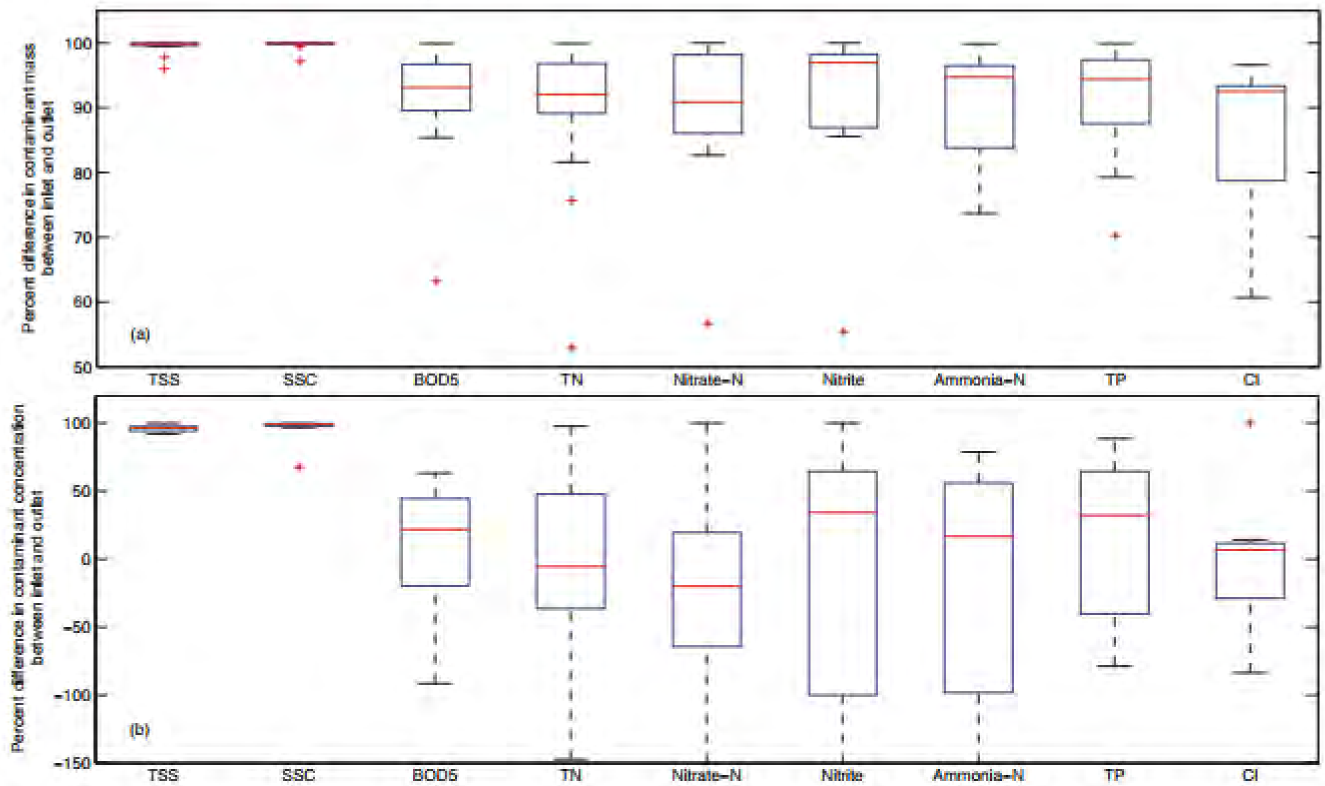


*Figure 8: Concentrations changes from inflow to outflow of specific pollutants (a) in concentration and (b) in mass.*

## 3.2 Chemical Addition

### 3.2.1 Background

The pH of water can be adjusted easily through chemical addition. An elevated pH means that the water is more basic than acidic. The stormwater at DIA has more hydroxide ions than hydrogen ions, which has led to the pH imbalance. In order to lower the pH, hydrogen ions must be produced. Carbon dioxide can be used to lower the pH of water because of its overall chemical reaction when introduced to water. Carbon dioxide reacts with water to produce the carbonic acid ($H_2CO_3$). Carbonic acid then dissociates into a hydrogen atom ($H^+$) and a bicarbonate anion ($HCO_3^-$). This can be shown through the overall chemical reaction below:

$$CO_2 + H_2O \leftrightarrow H_2CO_3 \leftrightarrow HCO_3^- + H^+$$

The production of the proton within the chemical reaction will balance the number of hydroxide ions already present in the water, lowering the pH of the stormwater.

Although the stoichiometric analysis justifies why the injection of carbon dioxide into stormwater would lower the pH, the chemical addition technique has not been widely used in stormwater applications. In order to assess this alternative appropriately, a pilot test plan will be produced on site. For this small-scale test, carbon dioxide will be added to the stormwater at the drainage exit, Outfall A, on the land side of DIA. This area has been indicated by DIA as the main area of concern for elevated pH. $CO_2$ in gaseous form will be dissolved into the stormwater in the holding pond through a standard bubbler system. This is similar to an aeration system which mixes the carbon dioxide into the water to carry out the chemical reaction. A sample bubbler system is shown in Figure 9.

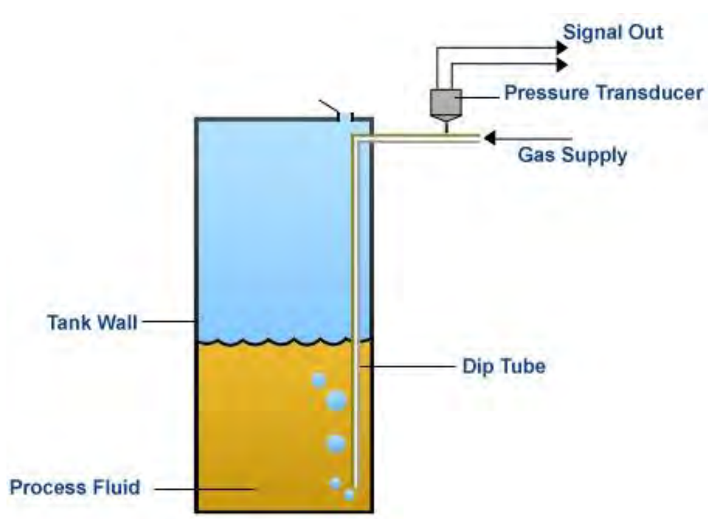


*Figure 9: Standard bubbler system. [33]*

A pH probe would be connected to a signal indicator which will tell the system when the pH reaches an unacceptably high level. This will then start the bubbler system, pumping the stored $CO_2$ down into a tube. The outlet area of the tube, where $CO_2$ is released, will be located at a depth within the stormwater that has collected at Outfall A. This depth is dependent on the amount of stormwater runoff collected in the detention pond. The tube and piping may be laid horizontally at the bottom of the pond area based on the low level of stormwater within this holding pond.

### 3.2.2 Case Studies

The Department of Ecology within the State of Washington has outlined a BMP for the neutralization of stormwater using carbon dioxide. The BMP details the use of solid or compressed carbon dioxide gas in the treatment process. As carbon dioxide is added to water, carbonic acid is formed which dissociates into a proton and bicarbonate anion. The proton produced within this reaction is a weak acid that can help lower the overall pH of the water [8]. The Department of Ecology has cited a number of advantages to $CO_2$ addition, including: the rapid neutralization of high pH water, the cost effective nature of the technique, the levels of safety when compared to handling other acids and chemicals, and the buffering nature of $CO_2$ that stops the pH from dropping to dangerously low levels. There is also a readily available source for all of the required materials, which makes purchasing chemicals for ongoing operations a simple process. When performing this management practice, procedures for pH neutralization typically requires continuous treatment [8].

Carbon dioxide has been increasingly used to lower the pH in water treatment plants, due to its ability to quickly lower the pH of the inlet raw water without taking the pH to undesirable levels. Carbon dioxide is also non-corrosive to pipes and equipment, and requires less maintenance when compared to other chemical alternatives [9].

## 3.3 Mycoremediation

### 3.3.1 Background

The study of mycoremediation started to gain traction when several case studies examined the biodegradation of lignin by white-rot fungi. Lignin is a complex organic compound which typically persists since most organisms do not have the ability to degrade it. *Phanerochaete Chrysosporium*, a species of white-rot fungi, was found to degrade lignin with high efficiency. The fungi first uses extracellular enzymes such as lignin peroxidase (LiP), manganese peroxidase (MnP), and laccases to begin degrading lignin, then uses mineralization for further degradation [10]. One study examined the extracellular enzymes produced to degrade lignin, and proposed a hypothetical pathway for the degradation of lignin using the MnP enzyme as seen in Figure 10 [11]. Of note for this project is the mineralization into carbon dioxide and the production of organic acids. The fungi degrade lignin as an intermediate step in order to obtain the sugars that are located behind the lignin barrier, yet much remains unknown about the biodegradation processes. The results of these studies revealed the need for better understanding of the complex biological processes of fungi and their potential applications to bioremediation.

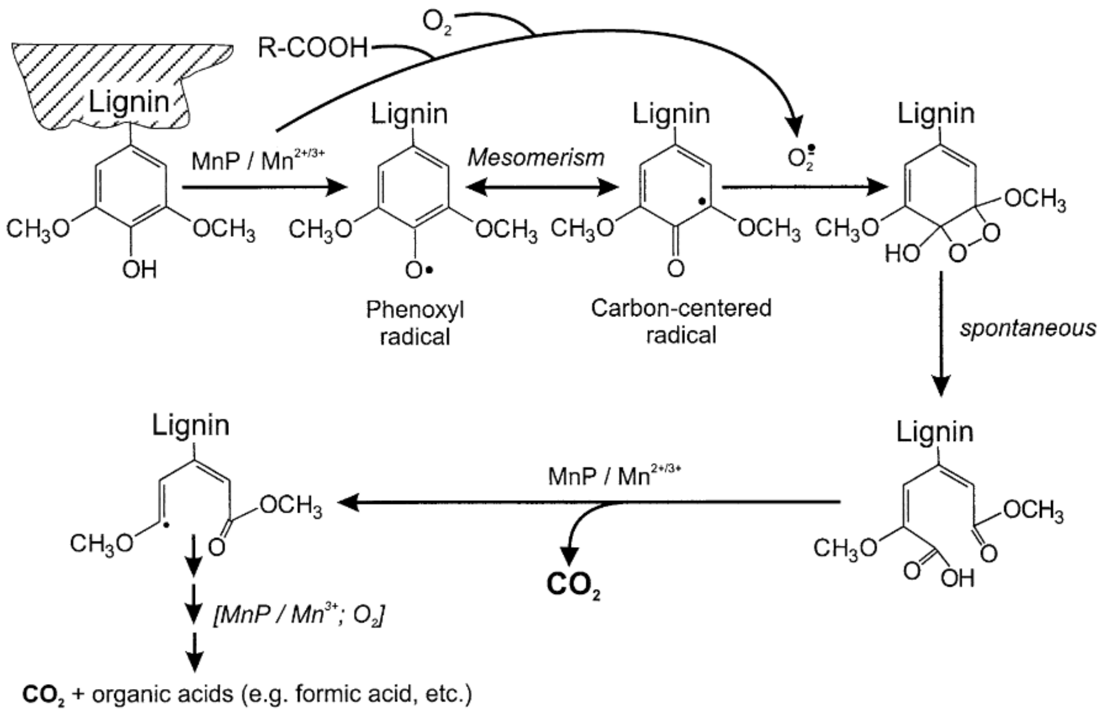


*Figure 10: A proposed pathway for the degradation of lignin using MnP. [11]*

### 3.3.2 Case Studies

In 2012, Fungi Perfecti collaborated with Washington State University (WSU) to investigate the potential of mycoremediation to treat *E. coli* in stormwater [12]. The goal of the project was to develop a method that was durable, resilient, and effective at removing *E. coli* from flowing water. The case study involved growth and resiliency tests, permeability tests, and bacterial removal tests. The results of the study indicated high efficacy of the mycoremediation for removing *E. coli* from stormwater, and also indicated the feasibility of mycoremediation applications for stormwater treatment. One key result from this study was the similarity in performance between the fungi inoculated under optimal environmental conditions, and the fungi that were inoculated and subjected to the extremes of the resiliency tests [12]. For a site like DIA with wide variations in temperature throughout the year, resiliency is a key aspect for success of a mycoremediation solution. Fungi Perfecti provided $S^3$ with experimental pH data shown in Table 3 [13]. The results of this experiment indicate that the *P. Ostreatus* and *S. Rugoso-*

*annulata* species prefer slightly acidic environments, and have the ability to adjust the pH of the surrounding substrate to a more optimal pH for growth. The buffering capabilities of fungi are supported in the literature through the examination of the natural processes of fungi and the pH data provided by Fungi Perfecti, supporting the possibility of mycoremediation applications for this project.

*Table 3: pH measurements for different substrates for a control, Pleurotus Ostreatus (PO), and Stropharia Rugoso-annulata (SRA) after one month. [13]*

| Substrate | No fungus | PO | SRA |
|---|---|---|---|
| **None** | 6.68 | | |
| **Alder chips** | 6.10 | 5.69 | 3.88 |
| **Douglas-fir bark** | 3.41 | 5.77 | 4.81 |
| **Peat** | 4.80 | 4.78 | 3.85 |
| **Alder/Douglas-fir** | 3.94 | 5.55 | 3.95 |
| **Alder/peat** | 5.29 | 4.35 | 3.54 |

Mycoremediation has been tested in larger stormwater applications outside of laboratory settings. The Dungeness River Targeted Watershed Initiative was a multi-phase project funded by the Environmental Protection Agency (EPA) Targeted Watershed Grant (TWG). The project included a microbial source tracking study, innovative BMP demonstrations, effectiveness monitoring studies, and public outreach plans. For one of the innovative best management practice demonstrations, a study was conducted using mycoremediation in combination with bioretention cells to test treatment of *E. coli* [14]. The fecal coliform concentrations were monitored over a period of six months, and the fungal bioretention cell was found to be 24% more effective at removing *E. coli* than the control bioretention cell. Most studies of mycoremediation applications to stormwater have focused on the removal of *E. coli*, and much remains to be explored in terms of treatment potential of mycoremediation for various contaminants in stormwater applications.

## 4.0 Safety Risk Assessment and Regulatory Evaluation

DIA's stormwater management is regulated by the National Pollutant Discharge Elimination System Stormwater Program (NPDES). The Colorado Department of Public Health and Environment (CDPHE) is the state entity of the NPDES appointed by the EPA to enforce the stormwater-related permits and regulations as mandated by the Clean Water Act [15]. DIA has two permits including the municipal stormwater permit, or MS4 Permit (COS-000001), and the Industrial Permit (COS-000008). DIA's stormwater management plan (SWMP) includes BMPs, identification of potential sources and pollutants, and describes practices to reduce pollutants from municipal and industrial stormwater. The MS4 and Industrial stormwater permits are one component of the larger Environmental Management System (EMS) at DIA used to "proactively identify and mitigate all potential impacts to the environment from airport operations" [16]. Based on the permit stipulations, changes within the permits may be necessary to implement new solutions for stormwater management.

S$^3$ performed a thorough review of DIA's MS4 and Industrial stormwater permits in order to evaluate feasibility of implementation of alternatives, potential challenges, and potential risks to public and environmental health and safety. This review indicated that DIA is not permitted to treat stormwater on site, nor are they permitted to directly add any chemicals into the stormwater systems. Based on these limitations, the mycoremediation and chemical additive alternatives will have to be reviewed and approved by the state in order to allow DIA to carry out these treatment techniques. If DIA is unable to comply with any discharge limitations, and regulations are broken, the Division and EPA must be notified. From here, DIA will be responsible for describing steps being taken to reduce and eliminate the issue and what they plan to do in order to prevent future noncomplying discharge.

# 5.0 Technical Description

## 5.1 Bench-Scale Treatability Study

Since much remains unknown about the specific processes of fungi, S$^3$ recommends a bench-scale treatability study to determine the feasibility and efficacy of mycoremediation at lowering elevated pH levels in stormwater. S$^3$ has designed a bench-scale treatability study modeled after the 2012 Fungi Perfecti study of mycoremediation treatment of *E. coli*. There were two parts of the study: a resiliency testing phase conducted at Fungi Perfecti, and a treatment test conducted at WSU. The growth and resiliency test studied six fungal species and five different substrate combinations [12]. After inoculation and incubation, 19 viable batches were subjected to saturation, dehydration, heat treatment, and freezing. Each batch consisted of 17 mycofilters, four of which were controls. The results of the initial resiliency tests are shown in Figure 11. The species with the highest resiliency was *Stropharia Rugoso-annulata*, represented by the "Str" abbreviation in Figure 11. In subsequent tests, the *Irpex* species also showed potential for high resiliency, and both species were used in the treatment studies. The combinations of substrates used in the study that are represented by letters in Figure 11 are listed in Table 4.

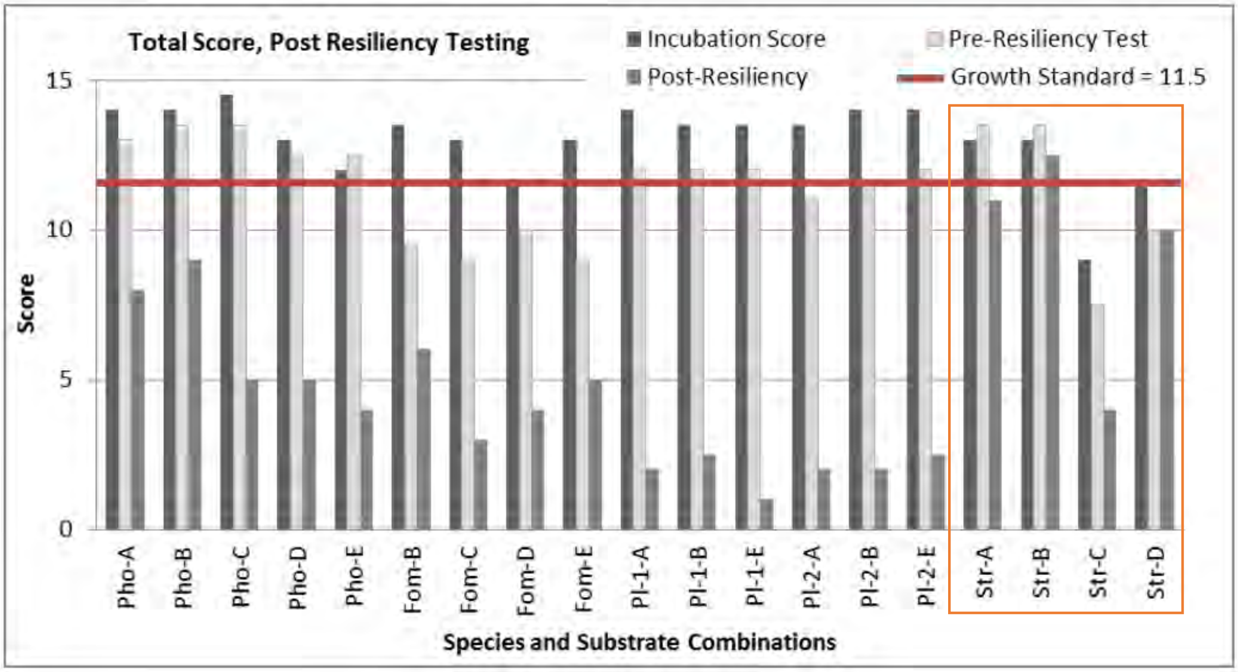


*Figure 11: Results of resiliency testing. The three letter prefixes are abbreviations of fungal species, and the letters represent different substrates. The orange box was added to highlight the most resilient species. [12]*

*Table 4: The different substrate combinations represented by letters in the 2012 study.*

| Letter | Substrate Combination |
|---|---|
| A | 100% Alder Chips |
| B | 50% Alder Chips; 50% Sawdust |
| C | 25% Alder Chips; 50% Straw; 25% Sawdust |
| D | 50% Alder Chips; 25% Straw; 25% Sawdust |
| E | 25% Alder Chips; 25% Straw; 50% Sawdust |

### 5.1.1 Substrates

The combination of 50% wood chips and 50% sawdust consistently resulted in the most growth during the growth and resiliency trials of the 2012 study. One substrate that was not examined in the study was spent mushroom substrate (SMS), or substrate that has already been used in commercial growing applications that is typically composted or discarded. In commercial growing, mushrooms can be grown on the same substrate for a number of harvests, but the growing efficiency can decrease with increased numbers of harvests. Once growing efficiency decreases, the substrate becomes SMS and is often discarded. Due to the growing amount of

SMS waste, studies have examined potential uses of SMS [17]. Since SMS contains viable mycelium, enzymes produced by the mycelial networks within SMS can be used for biodegradation of contaminants. Studies have investigated the potential uses of SMS to treat polycyclic aromatic hydrocarbons (PAHs), biocides, petroleum, heavy metals, and other contaminants [17]. Since SMS is readily available and can be acquired at little to no cost, $S^3$ included SMS as a substrate to test in the bench-scale study.

### 5.1.2 Filter Preparation

Based on the results of the resiliency tests from 2012, $S^3$ developed a design for a bench-scale test that can be conducted at DIA. Three batches will be prepared, with each batch consisting of the eight filters shown in Table 5. Although *Pleurotus sp.* did not exhibit high levels of resiliency, the species grow locally and have been tested in some stormwater applications, and will therefore be considered for this bench scale study. Since the species *Irpex* isn't typically commercially grown, only *Stropharia Rugoso-annulata* and *Pleurotus sp.* will be tested with SMS.

*Table 5: Proposed fungal species and substrate combinations for the bench-scale study*

| Filter Number | Fungal Species | Substrate |
|---|---|---|
| 1 | *Stropharia Rugoso-annulata* | 50% Alder Chips; 50% Sawdust |
| 2 | *Irpex sp.* | 50% Alder Chips; 50% Sawdust |
| 3 | *Pleurotus sp.* | 50% Alder Chips; 50% Sawdust |
| 4 | *Stropharia Rugoso-annulata* | Spent Mushroom Substrate |
| 5 | *Pleurotus sp.* | Spent Mushroom Substrate |
| 6 | None (control) | 50% Alder Chips; 50% Sawdust |
| 7 | None (control) | 50% Alder Chips; 50% Sawdust |
| 8 | None (control) | 50% Alder Chips; 50% Sawdust |

The testing procedure will be adjusted so that DIA can easily conduct the resiliency test at the airport. The freshly inoculated filters (filters 1-3 in Table 5) and the SMS filters (filters 4-5) can be sent to DIA from a commercial growing company. After receiving the filters, the study can be conducted in an empty garage or storage room. The filters will be transferred to five gallon buckets, and subjected to trickle-flow of synthetic stormwater. Two different hydraulic loading rates will be tested in order to determine treatment potential for a moderate one-year storm and a 100-year storm of 60 minute duration: 0.5 ml/min and 2.2 ml/min, respectively. Two different types of water will be used for the trickle flow: synthetic water created by mixing tap water from DIA with concrete sludge, and water collected from the stormwater conveyance systems. The pH of the water will be measured and adjusted using chemical addition to a pH of 9.5. The experiment will be run for six weeks. Visual checks will be performed throughout the treatment phase, and growth of the mycelial networks and degradation of the filter will be recorded. The mycelial networks are easy to identify with the naked eye as shown in Figure 12.

*Figure 12: Picture of visible mycelium within the mycofilter bucket. [34]*

The levels of success will be measured by three criteria: resiliency, treatment capability, and observed lifespan. The possible outcomes of the study are shown in Table 6. The first outcome would be considered a "failure" for that criterion, the second outcome would indicate that further investigation may be necessary along with potential adjustment of testing parameters, and the third outcome would be considered a "success" for that criterion. The criterion with the most importance would be treatment capability, followed by resiliency, and the least important criterion would be observed lifespan. If the results indicate a failure in treatment capability, mycoremediation would not move on towards the pilot test phase. However, if resiliency or observed lifespan indicates failure, further study may be warranted to determine whether or not mycoremediation can move on to the pilot test phase.

*Table 6: Bench-Scale Study Outcomes*

| Criteria | Description of Rating |
|---|---|
| **Resiliency** | 1. Did not achieve significant growth after resiliency testing<br>2. Resiliency testing affected growth, but mycofilter remained viable<br>3. Resiliency testing had little to no effect on growth |
| **Treatment Capability** | 1. Had little to no effect on pH of the effluent relative to influent, or resulted in elevated pH levels above that of the influent<br>2. The effluent pH was lowered compared to influent pH, but the results were insignificant.<br>3. Effluent pH was lowered to below a pH of 8.5, and results were significant. |
| **Observed lifespan** | 1. Filter showed significant degradation or fungal species showed a decrease in growth at the end of the testing period<br>2. Filter showed some degradation or fungal species had some growth limitation by the end of the testing period<br>3. Filter showed little to no degradation, and fungal species maintained growth by the end of the testing period |

## 5.2 Pilot-Scale Test

If the bench-scale treatability study indicates that mycoremediation may be a viable solution for treating elevated pH in stormwater, $S^3$ recommends further investigation through a pilot test investigation. The pilot test will have a growth and inoculation phase, followed by the treatment phase. The growth and inoculation phase will involve the preparation of the mycofilters. For the pilot-scale, mycofilters will consist of burlap sacks filled with inoculated substrate. Mycofilters can be produced and shipped to DIA from a commercial company. Specific substrate combinations and appropriate fungal species will be determined from the results of the bench-scale study. The number of filters needed will also be determined from the results of the bench-scale study; the treatment capability of one filter can be determined, and the number of filters necessary can be calculated given the influent pH, desired effluent pH, and buffering potential of the filter.

Determining the placement of the filters within Pond 927 depends on the observed water levels within the pond from previous years. In high flow events, certain areas of Pond 927 can become inundated, or otherwise run dry during other parts of the year. DIA has indicated that the wing walls of Pond 927 may be the best location to avoid elevated water levels during high flow events. Alternatively, the mycofilters could be anchored into the ground, and wood blocks or other floatation devices can be attached to the mycofilters so that the mycofilters float in the water during high flow events.

## 5.3 Monitoring

After placement of the mycofilters, the monitoring system will be set up. This monitoring system will work to determine if the mycoremediation design is effectively lowering the pH of the stormwater to acceptable levels. The monitoring system will be comprised of two pH meters: one pH meter will be placed at a location upstream of the mycofilters, and a second pH meter will be placed a few meters downstream of the mycofilters. These pH meters will be Milwaukee Instruments- Standard pH Mini Bench Meter that continuously monitor pH. The placement of these meters will allow for the continuous measurement of pH data for the stormwater prior to and after treatment from the mycofilters. Treatment from the mycofilters will be considered a success if the downstream pH values remain below 8.5. The extent of treatment can be evaluated when the upstream pH values exceed 9.0 by comparing the upstream and downstream pH values. The documentation of extent of treatment can inform treatment plans for a full-scale implementation at DIA or other airports.

## 5.4 Maintenance

Different studies indicate that mycofilters can remain viable for at least one year [18], but visual monitoring checks should be performed once a month in order to monitor any degradation or growth limitation of the mycofilters. The inner contents of the mycofilters should be checked, and the growth of the mycelial network should be monitored. If a decrease in the mycelial network is observed during a visual monitoring check, fresh substrate should be added to the mycofilter. If the addition of fresh substrate does not increase the growth of the mycelial network within two to three weeks, the filter should be replaced. $S^3$ does not expect the occurrence of extended periods where Pond 927 runs completely dry, but if such conditions are to occur, the moisture of the mycofilters should be monitored, and occasional manual watering of the mycofilters may be necessary.

Once the pilot-scale test is complete, the monitored pH data will inform the success or failure of the test. A success is indicated if pH levels downstream of the mycofilters remained below 8.5 throughout the test. The pilot test will also inform needs for maintenance and visual monitoring frequency. If the mycofilters remain viable for the whole test, visual monitoring frequency during a full scale implementation can be scaled down.

# 6.0 Airport Interactions

From the inception of the project to the final design, $S^3$ has worked closely with contacts at DIA. $S^3$ reached out to DIA in the winter of 2015 to see if a potential project could be recruited, and was fortunate to receive prompt communication from DIA's Director of Environmental Services, Scott Morrissey. Morrissey introduced our team to Kim Ohlson, an Environmental Public Health Analyst and member of the Environmental Services team, who became our main point of contact. Ohlson arranged for two site visits to DIA, the first one to investigate issues on the land side area of the airport, and the second to investigate issues on the airside. Throughout the project process, $S^3$ has communicated with DIA via email and biweekly conference calls. Keith Pass and Craig Schillinger on the Environmental Services team also provided information and valuable input for the project. Any necessary data or reports including water quality monitoring data and airport regulations were provided promptly by DIA upon request. After each phase of the project, reports and presentation slides were submitted to DIA for review in order to check that the project was proceeding according to DIA's satisfaction. For the alternatives analysis phase, DIA was consulted to develop and finalize the MCDA so that the evaluation of alternatives reflected DIA's main priorities such as environmental safety and cost.

# 7.0 Immediate Impacts

## 7.1 Financial Analysis

The costs provided included the total amount for a bench scale-study as well as a two-year pilot test. The design and cost may be brought to a full-scale time period of 20 years if this treatment is proven to work effectively for this pH problem. In general, mycoremediation is a low-cost, low maintenance solution compared to other remediation solutions [19].

*Table 7: Cost of Mycoremediation*

| Project Phase | Description | No. of Units | Cost | Total Construction & Capital Costs (Based on O&P and Contingency) | Annual Costs (Cost per year for a two-year life cycle) |
|---|---|---|---|---|---|
| **Bench-Scale Study** | pH meters | 16 | $1,600 | $2755 | $1502.68 |
| | Buckets | 24 | $5/bucket= $120 | $206.63 | $112.70 |
| | Inoculated substrate (spawn) | 9 | $20/spawn= $180 | $309.94 | $169.05 |
| **Total** | | | | $3,271.56 | $1,784.43 |
| **Pilot Test** | pH meters | 2 | $400 | $688.75 | $375.67 |
| | Inoculated Mycofilters | 5-10 | $200/filter (assuming $20/burlap, $20/spawn, for 5 filled bags)= $1,000 | $1,721.87 | $939.17 |
| **Total** | | | | $2,410.62 | $1,314.84 |

The following information is an example calculation used in order to calculate the total and annual costs for the pH meters in the mycoremediation bench-scale study based on the information provided in Table 7:

$$Capital\ cost\ for\ pH\ meters\ (16) = \$100 \times 16 = \$1{,}600$$

$$Installation\ of\ pH\ meters = \$1{,}600 \times 0.3 = \$480$$

$$General\ site\ work = \$1{,}600 \times 0.15 = \$240$$

$$Subtotal = \$1{,}600 + \$480 + \$240 = \$2{,}320$$

$$Overhead\ \&\ Profit = \$2{,}320 \times 0.15 = \$348$$

$$Contingency = \$348 \times 0.25 = \$87$$

$$Total\ cost\ for\ 16\ pH\ meters = \$2{,}320 + \$348 + \$87 = \$2{,}755$$

$$Amortized\ cost = \frac{P}{\left[\frac{(1+i)^n - 1}{i(1+i)^n}\right]} = \$1{,}502.68\ per\ year\ for\ two\ years$$

Where $P$ is the total cost for 16 pH meters, $i$ is the selected interest rate of 6% (0.06), and $n$ is the number of years of investment which is 2 years for this two-year long bench-scale study.

These calculations were produced using a multiplier method in order to determine estimated installation, general site work, overhead and profit, and contingency costs. The equations were used to cost all pieces of equipment within the study and then totaled to find the cost of bench-scale and pilot-test studies.

# 8.0 Future Impacts

$S^3$ has decided to look into other various applications of mycoremediation outside of the pH treatment scope. Mycoremediation has been used as a form of remediation to clean up contaminants such as petroleum hydrocarbons, PAHs, polychlorinated biphenyls (PCBs), polymers, heavy metals and other pollutants. Due to the versatility of mycoremediation, airports could consider investigating mycoremediation applications for the cleanup of spilled airplane fuel and deicing fluid that may occur on site.

## 8.1 Commercial Potential

In order for mycoremediation to become a more standard treatment option at airports, a larger number of bench-scale and pilot-scale studies must be conducted to investigate treatment potential for different contaminants that are commonly present at airports. Once treatment potential is determined for a particular contaminant, mycoremediation is a low-cost, environmentally friendly option for treatment, and has the possibility of replacing traditional treatment options. Aside from mycofilters, mycoremediation can take appearance in other forms such as fungal bioreactors or as broken down inoculated substrate to mix into soils. Exploring these different options can lead to innovations not only in stormwater applications but other remediation areas in airports.

### 8.1.1 Airplane Fuel

DIA and other airports run into problems dealing with fuel spills. Whether the spill is due to an overloading at the fueling stations or because a storage tank has leaked, mycoremediation could be an easy and effective way to remove the spill and stains left behind. Mycoremediation has been seen through several studies to work effectively in eliminating petroleum hydrocarbon contaminants within soil and other mediums. A study performed by Adenipekun and Lawal in 2011 analyzed the effect of mycoremediation on crude oil. The results of this test showed that the Total Petroleum Hydrocarbons (TPH) decreased at a percentage loss of 40.8% at 1% crude oil concentration and a 9.28% reduction for 40% crude oil contaminated soil [20]. The ability of fungi to breakdown this type of contaminant is a result of the released enzymes which occurs in order to maintain its own metabolism. These enzymes are able to breakdown a wide range of toxic hydrocarbons. Other studies have focused on the mycoremediation of PAH contaminated mediums. A study within the Department of Agriculture and Environmental Engineering at Rivers State University of Science and Technology in Nigeria used spent white-rot fungi substrate to biologically treat oil-based drill cuttings containing PAHs. Results showed that after some time there were significant decreases in the concentration of residual PAHs within the cuttings [21].

### 8.1.2 Airplane Deicing Fluid

Throughout the year, airports must store, collect, recycle, and properly handle deicing fluids on site. When deicing a plane, spills are a constant threat, potentially leading to the spread of deicing fluid into the surrounding environment. Propylene glycol is toxic to animals in high concentrations and due to its degradation rate, may remain the environment for long periods of time. It has been observed that some airports use various forms of bioremediation in order to clean and treat the deicing fluids, such as glycol, from contaminated areas. The Office of Aviation Research in Washington D.C. began some research in 1998 to determine the effectiveness of bioremediation techniques for reducing the biochemical oxygen demand (BOD) of aircraft deicing fluid contamination and runoff [22]. The tested bioremediation systems involved combinations of bacteria, nutrients, and enzymes. Results demonstrated that bioremediation was capable of reducing the effectiveness of the propylene glycol deicing fluid. Based on these results, mycoremediation may be a useful form of treatment for any deicing fluid contamination.

### 8.1.3 Assisting Revegetation

Another issue that occurs at DIA and that airports can experience is erosion along slopes adjacent to runways and roadways. Erosion and slope stability can become an issue when sediments are carried by precipitation into stormwater drainage channels. The sediments not only raise the TSS levels in stormwater, but can also lead to undesired vegetative growth within the channels. Utilizing revegetation to stabilize the slopes with root structures can be one solution to address erosion issues, and is a fairly industry standard practice [23]. Fungal species are often known to have mycorrhizal associations with plants, or mutually beneficial relationships with vegetation, providing nutrients to the plants in exchange for carbohydrates [24]. The mycorrhizal associations of fungi can help assist with the revegetation process. In slopes affected by erosion, the necessary mycorrhizal fungi can be absent, calling for the external addition of the fungi to improve growth. During the hydroseeding step of revegetation, mycorrhizal fungi can be used as an additive to the slurry of seed, water, compost, etc. [23].

## 8.2 Social/Environmental Impacts

Mycoremediation has a relatively low impact on the environment. No harsh chemicals are involved, and no large equipment is needed to implement a mycoremediation solution. If SMS proves viable for stormwater applications of mycoremediation, waste can be diverted from commercial mushroom growing operations and used for remediation purposes, a solution that is both economical and environmentally friendly. Mycoremediation also provides an opportunity for public outreach, and a heightened general awareness of environmental issues and potential treatment options. In the Dungeness River Targeted Watershed Initiative, public outreach included open workshops and discussions about contaminants and the treatment systems, prompting increased interest in the local community [14]. Although public workshops may be outside of the scope of an airport's agenda, mycoremediation can still act as a positive impact on the community through news pieces on airport websites so that travelers can be informed about the environmental innovations at airports.

## 9.0 Conclusion

DIA must address the problem of an elevated pH measured in their stormwater discharge to ensure no future violations of their stormwater permits occur. $S^3$ proposed three options for solutions to help DIA reduce pH levels in their stormwater discharge at Pond 927, evaluating each alternative through research, preliminary design considerations, development of an MCDA score, cost. The mycoremediation solution was recommended as the best solution based on a high MCDA score and low cost. $S^3$ decided to move forward into the design phase for mycoremediation, consisting of a bench study and pilot scale test. $S^3$ believes that the low cost of mycoremediation and the ease of implementation will best fit DIA's needs in neutralizing pH in their stormwater.

Mycoremediation is a new frontier in the field of treating stormwater, especially in regards to pH. Even though mycoremediation has not been directly used in the lowering of pH of stormwater, research of the biological process and other remediation tests suggest that this technology has strong potential to perform the necessary remediation. The success of the designed pilot scale test in lowering pH will further the innovation of mycoremediation as well as increase the already growing potential of using this technology for remediation at other airports for issues such as fuel spills, de-icing spills, and revegetation. The investigative potential, commercial potential, economic potential, and environmental/social potential of mycoremediation has yet to be fully realized. Our team believes that further evaluation of mycoremediation through our designed bench study and pilot scale test will lead to an innovative, sustainable, low-cost, and effective solution in stormwater management for DIA and other airports worldwide.

# Appendix A: Team Contact Information

## Team Members

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# Appendix B: Description of University

The University of Colorado Boulder (UCB) was founded in April 1876, consisting of just one building with a total enrollment of just 44 students. Today, the university has over 30,000 undergraduate and graduate students attending the 11 colleges that make up UCB. The campus is located near the foothills of the Rocky Mountains in beautiful Boulder, Colorado [25]. UCB has excellent faculty teaching in 53 departments. Five Nobel prizes have been awarded to its faculty and more than 50 alumni of prestigious academies teach at the university. UCB is a Carnegie Research I University, containing 11 individual research institutes and 90 research centers with more than 900 faculty, students, and supporting staff involved [26]. The mission and vision of UCB is to serve the nation and world through leadership, public service, and advancing research and knowledge.

UCB is a leading university in sustainability research and practices in the nation. The school's official statement on sustainability is as follows: "For more than half a century, UCB has been a leader in climate and energy, research, interdisciplinary environmental studies programs and engaging in sustainability and 'green' practices both on campus and in the larger world" [27]. The university's student body established a student-led Environmental Center that has helped provide the school with sustainable programs such as an NCAA Division 1 zero-waste athletics program and the CU Green Labs Program. UCB is ranked very high in sustainability throughout the nation and was the first school in the nation to receive a Gold rating in 2010 by the Sustainability, Tracking, Assessment, and Rating System (STARS) [27].

The high prowess and focus in advancement of research and education in sustainability is reflected in UCB's Environmental Engineering (EVEN) department, which is ranked 7$^{th}$ among public undergraduate programs [28]. The EVEN program branched off from the Civil, Chemical, and Mechanical departments in 1998, and is a relatively new program compared to the other engineering disciplines. It became ABET accredited in 2003 with renewal in 2006. The EVEN program gives the students opportunity to tailor a track of focus within the program. Currently, there are seven option tracks in the EVEN program which include: energy, water resources and treatment, environmental remediation, chemical processing, applied ecology, air quality, and engineering for developing countries [29].  UCB's College of Engineering and Applied Science has had a tradition of excellence in engineering education dating back to 1893, and continually updates and improve their programs to reflect the highest standards in teaching and learning, discovery and innovation.

# Appendix C: Description of Non-University Partners

## DIA

Our main contact at DIA was Ms. Kim Ohlson. She helped to arrange communication between our team and the Environmental team at DIA, and also provided information and data such as water quality monitoring data, site maps, permit regulations, hydrology reports, and wildlife information. She also arranged two field visits for our team. Additionally, Scott Morrissey, Craig Schillinger, and Keith Pass provided feedback and input through emails and conference calls throughout the project process.

## Fungi Perfecti

Our team contacted Fungi Perfecti, a commercial company that conducted the mycoremediation study in 2012 funded by the EPA. We were in correspondence with Alex Taylor at Fungi Perfecti, who provided more detailed information on the study, answered questions about mycoremediation, and provided pH data to our team.

## Marc Beutel

Our team contacted Professor Marc Beutel via email, and arranged for a call to discuss details of the 2012 mycoremediation study conducted by Fungi Perfecti and Washington State University. We discussed potential feasibility of modeling bench-scale studies investigating pH buffering after the 2012 study, and clarified some details of the study procedure.

Sustainable Stormwater Solutions

# Appendix D: Sign-Off Forms

## Airport Cooperative Research Program
## University Design Competition for Addressing Airport Needs
## Design Submission Form (Appendix D)

*Note*: This form should be included as Appendix D in the submitted PDF of the design package. The original with signatures must be sent along with the required print copy of the design.

University: University of Colorado Boulder

List other partnering universities if appropriate: ______

Design Developed by: ☐ Individual Student ☑ Student Team

***If individual student:***

Name ______

Permanent Mailing Address ______

Permanent Phone Number ______ Email ______

***If student team:***

Student Team Lead: Erica Wiener

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Permanent Phone Number 303-618-8882 Email erwi2783@colorado.edu

***Competition Design Challenge Addressed:***

Airport Environmental Interactions: Innovative methods for storm water management at airports

I certify that I served as the Faculty Advisor for the work presented in this Design submission and that the work was done by the student participant(s).

Signed [signature] Date 4/24/16

Name Christopher Corwin

University/College University of Colorado, Boulder

Department(s) Environmental Engineering

Street Address 1111 Engineering Drive, ECOT 447, 428 UCB

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Telephone 303-492-7651 Fax ______

# Appendix E: Evaluation of Educational Experience

## Students

**1. Did the Airport Cooperative Research Program (ACRP) University Design Competition for Addressing Airports Needs provide a meaningful learning experience for you? Why or why not?**

Being able to investigate a challenging issue at DIA was an incredibly meaningful learning experience for the whole team. The issues at DIA were such that all of our team members had the opportunity to investigate technologies that were unknown to them. We were able to utilize resources available to us in order to come up with an innovative solution for the competition, and communicated with professors, companies, and DIA in order to make our designs more robust.

**2. What challenges did you and/or your team encounter in undertaking the competition? How did you overcome them?**

Our main challenge was dealing with the many unknowns in our project. At every point, our team was either missing data or lacking information that we felt was needed to move forward with the design. These challenges forced us to become more resourceful, reaching out to a variety of individuals and doing thorough research of literature in order to address every data or information gap.

**3. Describe the process you or your team used for developing your hypothesis.**

Our team first did a study of literature surrounding the issue to try and gauge industry standards. To come up with a more innovative solution, we expanded our research to investigate topics that were similar but not quite the same as the issues at DIA. Some team members had ideas initially from previous knowledge of different treatment techniques. By using information gleaned from this process, we prepared our proposal and reviewed our ideas with DIA and our Project Principal.

**4. Was participation by industry in the project appropriate, meaningful and useful? Why or why not?**

Participation by industry in the project was appropriate, meaningful and useful. Our team needed information from knowledgeable sources in industry in order to collect information needed for our design. Even in the alternatives analysis phase, we contacted people in industry to gain deeper knowledge of what different alternatives may entail, and to determine feasibility of developing alternatives. Additionally, we reached out to researchers to gain further information on alternatives that weren't the industry standard in order to show proof of concept.

**5. What did you learn? Did this project help you with skills and knowledge you need to be successful for entry in the workforce or to pursue further study? Why or why not?**

Our team learned many things throughout this process: we learned effective ways to navigate literature, improved technical writing skills, and learned key aspects of the engineering design process. We learned to adapt to and address issues and challenges that arose during the design process. All of our experiences on this project will help us for entry in the workforce, as the challenges we faced are challenges that people face in the workforce. Learning how to address these challenges now have prepared us to address them with a greater ease in the workforce.

## Faculty

**1. Describe the value of the educational experience for your student(s) participating in this Competition submission.**

The students used this competition as a vehicle to get an authentic engineering experience in working with an actual client (DIA) on a relevant, current problem. The students develop the project with the client resulting in a proposal, then investigate several alternative solutions to the problem, and finally design the best alternative. The competition provides the opportunity for the students to combine all their undergraduate courses into this "capstone" project while improving their skills in written and oral communication.

**2. Was the learning experience appropriate to the course level or context in which the competition was undertaken?**

Yes, very much so.

**3. What challenges did the students face and overcome?**

Recruiting a participating airport, developing a project scope, and then executing the scope within the confines of a single semester.

**4. Would you use this Competition as an educational vehicle in the future? Why or why not?**

Yes. The competition provides a vehicle to motivate the students to perform their best and provides an outlet for their hard work.

**5. Are there changes to the Competition that you would suggest for future years?**

More assistance in recruiting participating airports. If there were a webpage dedicated to airports that have expressed interest in participating and a brief synopsis of the problem(s) they are facing.

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