

**PHYTO-VIABILITY ON RESTORED LAND WITH COAL ASH AGGREGATES AS
BACKFILLING AMENDMENTS**

by

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ABSTRACT

One of the main concerns of people in today's world is the protection of the environment. Unnecessary constructions, inadequate waste management, and heavy transportation are the main causes of the deterioration of what is known as "green" environment. Due to increasing population, the demand for construction materials such as sand and gravel has increased over time. These basic materials are being used for concrete, filling, plumbing and for other purposes.

An alternative engineering method for conserving the environment was studied in order to restore the disturbed lands with available soils and coal ash aggregates (CAAs). Seed germination in the presence of manufactured CAAs was evaluated in order to assess the rate of growth of dissimilar plants in the restored land with the CAAs. The influence of the presence of CAAs on bacteriological and enzymatic activities in the rhizosphere was quantified.

Various experimental methods were developed. First, systems were prepared to evaluate the development of seeds (beans and pumpkins) using water from the water infiltrated through the CAAs. The growth of the plants in the systems with various factors such as backfilling ratio (soil and CAAs), layered or mixed application of the CAAs, type of water (rain or tap water), and type of seeds (bean or pumpkin), was observed. In addition, potential physical and chemical influences of the CAAs addition on the growth of such different plants were studied for beans, pumpkins, botellas and papayas. Later, a system was designed to assess the number of total heterotrophic bacteria and the activity of soil dehydrogenase in the rhizosphere in the presence and absence of the CAAs.

Experimental results indicated that the CAAs could be used as backfilling amendments for an open pit restoration to bio-viable land. There were no negative impacts on the germination and growth of the plants due to the presence of the CAAs. In some cases, the CAAs enhanced the germination and growth of the plants. Therefore, the CAAs can be utilized for restoration of disturbed lands to bio-viable ones, conserving environmental resources and achieving waste minimization.

RESUMEN

Uno de los principales propósitos de las personas hoy día en el mundo es proteger el medio ambiente. La construcción innecesaria, el manejo inadecuado de desperdicios, la transportación pesada, son las causas principales del deterioro de lo que conocemos como áreas verdes. Debido al incremento en población, la demanda por materiales de construcción, como arena y grava, ha aumentado con el tiempo. Estos materiales básicos se utilizan en el país para el concreto, relleno, tuberías y demás sistemas.

La restauración de tierras afectadas y agregados de las cenizas de carbón puede ser una alternativa de método de ingeniería para la conservación del ambiente. Los objetivos principales de esta investigación envuelven el evaluar la germinación de las semillas en presencia del agregado manufacturado (o agregado de la ceniza de carbón, CAAs en inglés), evaluar la razón de crecimiento de diferentes plantas en tierras restauradas con este agregado; y cuantificar la influencia de la presencia del agregado de las cenizas de carbón por medio de actividades bacteriológicas y enzimáticas en la rizosfera.

Para cumplir con estos objetivos, varios métodos experimentales fueron desarrollados. Primeramente se prepararon sistemas en donde se observó el desarrollo de las semillas (habichuelas y calabazas) utilizando agua proveniente de la infiltración con el agregado manufacturado. La razón de crecimiento de las plantas en sistemas con diferentes factores: modo de relleno (suelo y agregado de las cenizas de carbón), estructura de mezcla o en etapa, tipo de agua (de lluvia o grifo), y tipo de semilla (habichuela o calabaza), fueron evaluados. Además, el potencial físico y químico debido a la influencia del agregado de cenizas de carbón en el desarrollo de las plantas fue estudiado con: habichuelas, papayas, calabazas y botellas. Posteriormente, se diseñó un sistema para el conteo de bacterias heterótrofas y la actividad deshidrogenasa del suelo en la rizosfera en presencia y ausencia del agregado de las cenizas de carbón.

Los resultados experimentales indicaron que el agregado de las cenizas de carbón puede ser utilizado para relleno de canteras abiertas utilizadas para luego desarrollar vegetación. No se

mostró impacto negativo del material en la germinación y crecimiento de las plantas estudiadas. En algunos casos, el agregado aumentó la germinación y crecimiento de las plantas. Por tanto, este material puede ser utilizado para la restauración de tierras afectadas, conservando así los recursos naturales y logrando la reducción de desperdicios.

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1 INTRODUCTION

Increasing population and imposing necessary and unnecessary exploitation have compelled us to conserve our natural resources. Today, the environmental impacts due to transportation, infrastructure and construction have increased, and it is extremely remarkable. The population itself is still responsible for the degradation and negative impacts on our natural resources. Over the years, it has been shown that many materials wasted can be reusable for others. Some of those materials reflect an environmental impact but others may be a beneficial material to the people and environments.

The AES (Applied Energy Systems) power plant in Puerto Rico generates electricity by burning coal. It encourages reducing dependence on petroleum so as to strengthen the economy and protect the environment. This power plant produces byproducts: fly ash (FA), bottom ash (BA) and manufactured aggregate (MA) (AES Puerto Rico, 2006).

The main material of this project is the MA (herein, it is defined as the CAAs (coal ash aggregates)). The CAA is a mixture FA, BA and water. Because of its properties, studies showed that the CAA can be used for: a pavement base, a fill material, landfill cover, asphalt and agriculture. The CAA possesses attractive properties for use as a fill material provided it is allowed to drain reasonably well and does not become saturated for extended periods of time (Kochyil, 2004).

The utilization of CAA for restoration of disturbed lands is the main purpose of this study. The experiments conducted in this investigation were to assess bio-viability on the land after restoration with the CAA. Successful utilization of the CAA in the area of disturbed land restoration would minimize the unnecessary use of natural resources and garner benefits to the existing natural environments.

1.1 JUSTIFICATION

Human activities are causing several and unwanted impacts on the environment. The demand for construction materials such as sand and gravel has increased a lot due mainly to the development of industries and civil infrastructure. This has, subsequently, caused several environmental and economical problems around the world. For this reason, the implementation of resource recovery to minimize resource deterioration and waste production is warranted to avoid such problems aforementioned.

Coal combustion byproducts (CCPs) can be beneficially used for civil, environmental and agricultural purposes: waste stabilization, beneficial construction applications, and agricultural applications (Pando and Hwang, 2006). In the United States, over 108 million tons of CCPs were produced in 2000. The total CCP utilization was increased by 29% from 28.6 million tons to 36.9 million tons (Kalyoncu, 2001). AES is a global power company with electricity generation and distribution business in 29 countries, including a local plant in Guayama, Puerto Rico (PR). This plant produces 454 Megawatts, representing about 15% of the electricity consumed in PR (AES Puerto Rico, 2006).

A private mining company has excavated construction gravels in the heart of agricultural areas in Santa Isabel, PR since 1985. Its operation ended in 2006. This excavation activity resulted in approximately 420 ha of the open pits at the site. Some of the open pits were already restored to the agricultural areas, large portion of the open pits are, however, still need to be restored.

Therefore, a need to restore open pit quarry has to be met to sustain agricultural development and to achieve environmental conservation. In line with resource recovery and waste minimization, environmental restoration was tested with beneficial utilization of waste materials. In this regard, CAAs was evaluated as potential backfilling amendments material. Since the site is to be re-developed for agricultural purposes after restoration, phyto-viability of the restored land was evaluated.

1.2 OBJECTIVES

The overall goal of the proposed study was to achieve environmental restoration in conjunction with resource recovery and waste minimization. To this end, phyto-viability on the restored land with the CAAs as reclamation amendment materials was tested. This was done with a series of laboratory experiments which specifically aimed:

- To assess the germination of seeds in the presence and influence of the CAAs,
- To evaluate the growth rate and extent of germinated plants in the restored land under different restoration schemes with the CAAs amendment, and
- To quantify the influence of the CAAs amendments on rhizospheric biochemical properties.

2 LITERATURE REVIEW

2.1 INTRODUCTION

An adequate and thorough characterization of the CCPs is very important to be able to determine the feasibility of their use for the different possible applications (Pando and Hwang, 2006). The utilization of CCPs is well established in some countries of the world, based on long term experience and technical as well as environmental benefits. The CCPs are mainly utilized in the building material industry, in civil engineering, in road construction, for construction work in underground coal mining as well as restoration purposes in open cast mines (Feuerborn, 2005). Introducing CCPs for agricultural purpose can be an alternative way to improve soil characteristics and also plants growth.

2.2 APPLICATIONS OF COAL COMBUSTION BYPRODUCTS

As time passes, in United States and China, the burning of coal to generate electricity will continue to increase, and ash can be expected to increase too. Fly ash represents a major component (58%) of CCPs produced, followed by flue gas desulfurization (FGD) material (24%), bottom ash (15.5%), and boiler slag (2.5%). Figure 1 shows the CCP production from 1996 to 2000. Each component of CCPs is suitable for a particular application. CCPs are used in cement and concrete, mine backfill, agriculture, blasting grit, and roofing applications, also waste stabilization, road base, and wallboard production. Fly ash and FGD materials boast the highest use rate, about 32% of the amount produced. Figure 2 shows the CCP amount produced and used (Kalyoncu, 2001).

Fly ash was used in the largest quantities and found widest range of applications, with about 60% of annual consumption used in various structural applications, as in cement and concrete production comparing with 1.6% for agricultural uses of total FGD material use. However, potential FGD material use in agriculture exceeds even its use in wallboard (Kalyoncu, 2001).

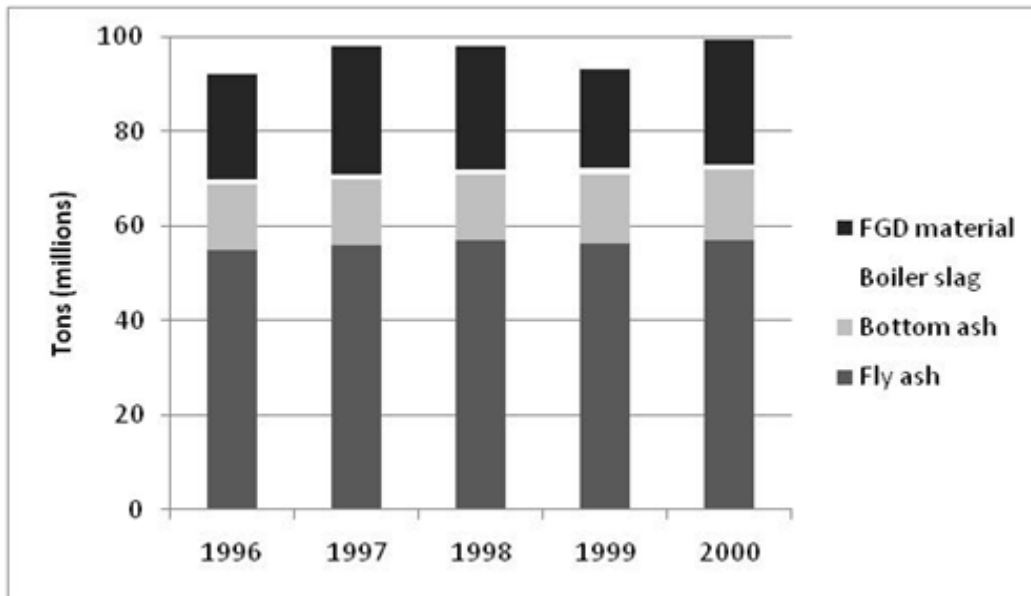


Figure 1. Historical CCP production data (1996-2000) (Source: ACAA)

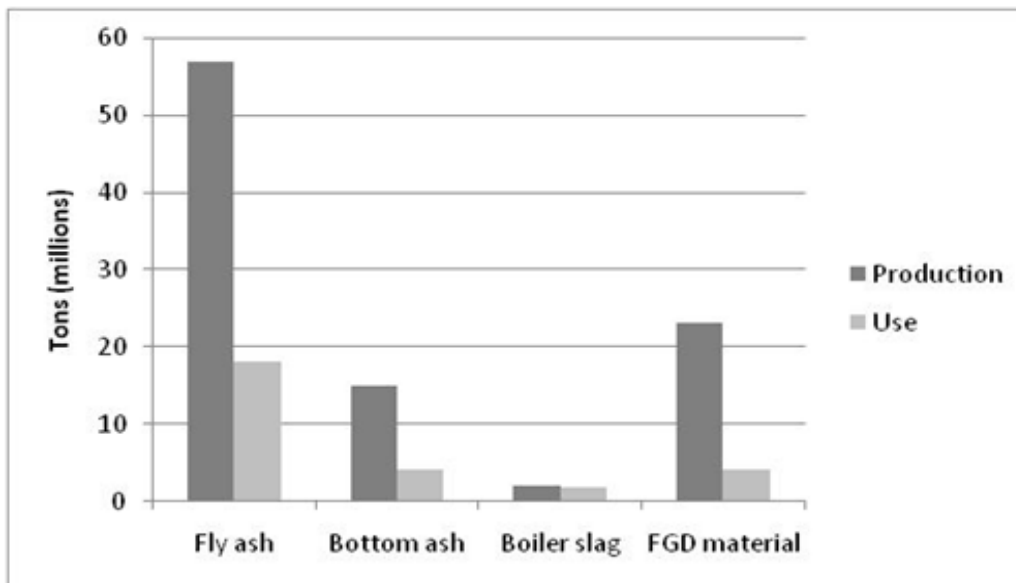


Figure 2. CCP production and use in the United States in 2000 (Source: ACAA)

A number of potential benefits applying CCPs to agricultural soils have been reviewed. Agricultural use of byproducts for improving acidic soils has potentials for both beneficial utilization and disposal cost reduction. But there are many applications of CCPs that contributed

to the resource recovery aspect. It was hypothesized that the beneficial effect of each amendment on plants could be enhanced, when more than two amendments were combined at proper proportion (Baligar et al., 1997). The application of CCPs decreased acidity effects and increased crop production on acidic soils (Baligar et al., 1997), but it also present relatively little risk to the environment when used as soil amendments (Clark et al., 2001).

Approximately 30% of CCPs are used in constructions, engineering, and manufacturing, with the remainder being disposed of in landfills and surface impoundments. A limited amount of coal ash is also used for land application purposes, such as alkaline adjustment of acidic overburden associated with coal refuse, covers for landfills, and agricultural applications (Brake et al., 2004).

2.3 CHARACTERISTICS OF FLY ASH, BOTTOM ASH AND COAL ASH AGGREGATES

The chemical and physical characteristics of fly and bottom ashes can determine the usage as agricultural purposes. It is important to be able to determine the feasibility of their use for the different possible applications (Pando and Hwang, 2006). The properties of fly ash are dependent on the composition of the parent coal conditions during the coal combustion efficiency of emission control devices and practices used during storage and handling (Gupta, 2002).

Fly ash is a material that results from the combustion of pulverized coal produced during the steam generation process in the power plant AES in Guayama, Puerto Rico (Pando and Hwang, 2006). Comprise primarily of fine sand and silt sized particles, therefore if applied at sufficient rates, it can be used to change soil texture to increase soil water holding capacity. The physical structure of FA often consists of “hollow spheres” and these particles show an increased surface area, capillary, aeration, and nutrient holding capacity compared with sands (Gupta, 2002). It is used as a pozzolan, reacting in the presence of water with calcium hydroxide at ordinary temperatures to produce cementitious compounds.

Fly ash is composed of silt sized, spherical, amorphous ferroaluminosilicate minerals (Garau, 2007). The American Society for Testing and Materials (ASTM), the FA can be classified in Class C and F. The former is an FA with high calcium content, more than 20% by weight, and the latter a low calcium FA, as less than 10% by weight. For that reason, the principal factors that influence on the classification of it are silica (SiO_2), alumina (Al_2O_3), and ferric oxide (Fe_2O_3). Table 1 shows the chemical composition of FA provided by AES, which shows that the content of sulfur dioxide of 12.57%. Based on the results provided, it exceeds the 5% maximum percentage of sulfur dioxide specified in ASTM Standard C 618, concluding that it complies with the most of the chemical requirements for a Class C fly ash (Pando and Hwang, 2006).

Table 1. Chemical composition of FA (Data provided by AES, PR)

Chemical Analysis of Fly Ash (% by weight)	Results
Silica, SiO_2	39.41
Alumina, Al_2O_3	12.59
Ferric Oxide, Fe_2O_3	4.35
$\text{SiO}_2 + \text{Al}_2\text{O}_3 + \text{Fe}_2\text{O}_3$	56.35
Titania, TiO_2	0.51
Lime, CaO	27.02
Magnesia, MgO	1.27
Potassium Oxide, K_2O	1.17
Sodium Oxide, Na_2O	0.44
Sulfur Trioxide, SO_3	12.57
Phosphorus Pentoxide, P_2O_5	0.28
Strontium Oxide, SrO	0.14
Barium Oxide, BaO	0.23
Manganese Oxide, Mn_3O_4	0.02
Undetermined	0
Alkalis as Na_2O , Dry Coal Basis	1.12
Base to Acid Ratio	0.65

The smaller portion of the total ash produced during the coal combustion process is the bottom ash. It is a CCP consisting of coarse grained particles that fall to the bottom of the furnace as a result of the coal combustion procedures. According to analysis, the particles have a greater tendency to fuse together; displays less pozzolanic properties than fly ash. Table 2 summarized the chemical analysis provided by AES, Puerto Rico for bottom ash. This type of ash is grey to black in color and has a large particle size, and is commonly used as replacement for aggregate because it is well graded in sizes which avoid the need for blending with other aggregates to meet construction gradation requirements (Pando and Hwang, 2006).

Table 2. Chemical composition of BA (Data provided by AES, PR)

Chemical Analysis of Bottom Ash (% by weight)	Results
Silica, SiO ₂	30.83
Alumina, Al ₂ O ₃	12.2
Ferric Oxide, Fe ₂ O ₃	3.95
SiO ₂ +Al ₂ O ₃ +Fe ₂ O ₃	46.98
Titania, TiO ₂	0.57
Lime, CaO	36.02
Magnesia, MgO	1.58
Potassium Oxide, K ₂ O	0.66
Sodium Oxide, Na ₂ O	0.55
Sulfur Trioxide, SO ₃	12.82
Phosphorus Pentoxide, P ₂ O ₅	0.37
Strontium Oxide, SrO	0.15
Barium Oxide, BaO	0.27
Manganese Oxide, Mn ₃ O ₄	0.03
Undetermined	0
Alkalis as Na ₂ O, Dry Coal Basis	0.95
Base to Acid Ratio	0.98

The main coal ash material used of this project is the CAAs. It is an agglomerate of fly and bottom ash particles. The material is 60 to 70% of the weight of traditional, natural soils, the optimum moisture content for a compaction is 50%, and the maximum density is 70 pounds per cubic feet (Kochyil, 2004). A chemical analysis was also provided by AES and it is shown in Table 3.

Table 3. Chemical composition of CAA (Data provided by AES, PR)

Chemical Analysis of CAA (% by weight)	Results
Silica, SiO ₂	34.79
Alumina, Al ₂ O ₃	11.97
Ferric Oxide, Fe ₂ O ₃	4.19
SiO ₂ +Al ₂ O ₃ +Fe ₂ O ₃	50.95
Titania, TiO ₂	0.51
Lime, CaO	29.67
Magnesia, MgO	1.11
Potassium Oxide, K ₂ O	0.76
Sodium Oxide, Na ₂ O	1.52
Sulfur Trioxide, SO ₃	14.66
Phosphorus Pentoxide, P ₂ O ₅	0.32
Strontium Oxide, SrO	0.23
Barium Oxide, BaO	0.24
Manganese Oxide, Mn ₃ O ₄	0.03
Undermined	0
Alkalis as Na ₂ O, Dry Coal Basis	1.76
Base to Acid Ratio	0.79

2.4 INFLUENCE OF CCPS IN PLANTS AND SOILS

Plants grow in different and specific ways. They obtain resources from the environment to survive. Some environments contain abundant resources and offer few physical or chemical constraints on growth (Fitter and Hay, 1987). Studies show the extraction or behavior of the plants with heavy metals from CCPs. Phytoextraction of heavy metals from FA contaminated soil was evaluated using *Phaselous vulgaris* and showed that the accumulation of Fe, Mn, Ni, Cu and Co was found more in the roots while Zn, Pb and Cd were more in the aerial parts (Gupta, 2002). FA contains most of the essential elements for the growth and development of the plants although they are in the environment is deficient in N and unavailable P. Moreover, FA can improve the physic-chemical properties of the soil, such as pH, texture (Gupta, 2002) and the addition of BA, FA and/or flue gas desulfurization materials to agricultural soils may also increase water holding capacity which could decrease irrigation frequency, thereby increasing water savings (Lombard et al., 2006).

In a study by Gupta (2002), the results of physic-chemical analysis for the plant revealed an increase in pH with an increase of FA amendment ratio (Table 4). Fe, Mn, Zn, Pb and Ni were high in the amendments of FA than the control soil except for Cu, which did not show significance changes. It was also found that accumulation of metals such as Zn, Pb and Cd were measured from roots to shoots in the plants grown in FA amended soil like other studies (Gupta, 2002). The accumulations in the plants of different metals depend on the type of soil, and also plant species.

Table 4. Accumulation of metals (mg/kg, in dry weight) in the plants grown in the systems amended with FA (Gupta, 2002)

Amendments	Plants Parts	Metals Accumulation (mg/kg dw)							
		Fe	Mn	Zn	Cu	Ni	Co	Pb	Cd
C	Shoot	115.6±23.6	11.0±3.6	57.2±2.3	3.4±3.1	3.4±0.8	1.7±0.4	2.2±0.1	0.9±0.4
	Root	162.8±15.1	16.3±3.8	20.9±2.5	4.8±0.3	4.5±0.9	2.5±0.6	1.2±0.3	1.0±0.4
10% FA	Shoot	191.8±37.9	16.6±1.3	51.5±7.8	4.8±0.5	7.8±2.9	8.2±0.4	18.1±0.3	5.0±1.5
	Root	254.2±36.2	20.6±1.6	32.0±4.8	5.5±1.7	9.8±3.1	10.3±0.8	16.8±0.8	3.1±0.3
25% FA	Shoot	199.1±19.7	20.9±1.1	69.0±4.1	7.2±0.8	8.9±0.4	15.8±1.1	22.3±1.2	6.1±0.4
	Root	321.7±30.2	25.8±4.6	43.9±4.1	11.6±0.9	13.3±0.8	24.0±0.6	19.3±0.2	4.4±0.8

On the other hand, the use of FA to agriculture was not always beneficial; however, utilization of lower concentrations of it as soil amendment was suitable for better management of few crops (Singh, 2008). The application of FA caused significant reductions in growth, biomass and yield responses of *B. vulgaris* plants at different ages of observations. The concentrations of all heavy metals increased with increasing concentrations of FA. It showed higher levels of heavy metals such as Ni, Cd, and Pb compared to the cases with the control soil.

Reductions in growth parameters such as the number of leaves, leaf area, root and shoot, and total plant lengths were observed (Singh, 2008); also leaf area, leaf fresh weight, and leaf dry weight were reduced by the coal bottom ash media (Coffindaffer, 2000). On the other hand, a study with strawberries was made in response to coal bottom ash root media. It exhibited higher germination percentages and it did not present negative impacts on plants (Coffindafer, 2000).

Moreover, the use of CCPs at moderate rates with phosphate rock and dolomitic lime appeared to be the best combination in increasing crop yields on infertile acidic soils. The low nutrient availability and acidity related toxicity in these soils made normal plant growth difficult. Application of limiting materials, fertilizers, and organic manures were generally essential for

reduction of acidity-related constraints and to improve the crop production potential of these soils (Baligar et al., 1997).

Chemical and physical improvements included ameliorating nutrient status for plant growth, altering soil pH, and enhancing root penetration and water holding capacity. The BA had the least influence on electrical conductivity and pH among the CCPs. Leaf copper content also improved with BA cultivation. Soil pH increased above the control in FA treated soil. The addition of FA increased the plant tissue micronutrients Zn, Fe, Mn and Cu over the control (Lombard et al., 2006).

Amendment addition increased significantly plant biomass production by a maximum of 26% using 1120 mg ha⁻¹ FA and 10 mg ha⁻¹ poultry biosolid (PB). Application of the highest rate of FA significantly increased the plant tissue concentrations of Mg, Ar, Se and B (Adriano et al., 2002). However, CCPs can act as a supplementary source of Ca, S, B, Mo, Se, and other trace elements when soil contents are deficient for adequate plant growth. Selenium is not an essential element for higher plant growth, although it has been shown to be a required element for some lower species plant species. A similar result was also reported that CCPs could be used in agriculture to supplement crops and soils low in Se, Mo, Cu, Zn, or B (Korcak, 1998). Another study also reflected a significantly increasing of B in the plants with the BA amendments (Coffindaffer, 2000).

Soil pH was initially increased from 4.6 to 6.1 by CCPs amendments. Concentrations of plant essential trace elements (B, Cu and Zn) that were marginally deficient in the unamended eroded soil were increased to those in typical soil concentrations due to the amendment with FA and PB (Adriano et al., 2002). However, FA has a number of inherent qualities that under certain circumstances may limit its usefulness for soil stabilization, and which may even result in increased erosion and soil loss (Fine et al., 2006). The co-application of both FA and PB successfully promoted the revegetation of the eroded burrow area with no apparent adverse environmental side effects (Adriano et al., 2002)

Organics have received little attention in studies on agricultural use of CCPs. The primary potential hazards for agricultural use of CCPs were excessive trace element loadings, which increased food chain metals; high soluble salt loadings, which may reduce initial plant growth; high Na loadings, which reduce water infiltration; sulfite damage to crops; and leaching of toxic substances into the groundwater (Korcak, 1998).

According to a study of effects of FA compost and yard waste on the tomato leaves (Bryan et al., 2002), concentrations of Cd, Pb and Ni in tomato fruits were below detection limits. Fruits, a consumable part of plants, normally accumulate lower amounts of the trace metals than other plant organs. The treatments with the coal ash mixture did not significantly affect the concentrations of Zn, Cu, Fe, Cd, Pb, and Ni in tomato leaves, while others have reported increased concentrations of iron and decreased concentrations of Zn in tissue samples of rice and wheat. After tomato harvest in this study, cover crop (sorghum) was planted. The height and total dry biomass of the sorghum were significantly greater than that of control. Although the concentrations of N, P, and K in sorghum root and shoot were not significantly higher than that of control, the total N and K retained by the cover crop biomass were significantly greater than that of control. The concentration of the metals in sorghum root was much higher than that in sorghum shoots.

2.5 RHIZOSPHERE ACTIVITY

The rhizosphere is a place of intense biological activity. Soil ecosystems contain remarkable numbers of bacteria, fungi, protozoa and small invertebrates. The most striking feature of the rhizosphere is the stimulation of bacterial numbers and activity (Fitter and Hay, 1987). This is a concept that was first coined by Hiltner (1904) to describe the volume of soil that was influenced by root activity by living plants. It is a unique hot spot in the soil at the viewpoint of microbial ecology as soil microorganisms are considerably stimulated in the vicinity of the roots, as a consequence of the release by roots of a range of C- compounds (Hinsinger et al., 2006).

According to Fitter and Hay (1987), the microbes in the rhizosphere were stimulated by increased concentrations of various chemicals that acted as energy sources, and these were deposited there from a number of sources: 1) by the sloughing of root cap cells as roots grow through soil, 2) by the production of mucigel by the root cap, which facilitates its passage through the soil, 3) by exudation or secretion of compounds from intact cells, and 4) by the death of root cells.

Soil microbial activities play an important role in organic matter turnover, element cycling and plant growth. Knowing this role can be helpful in order to evaluate the efficiency of a remediation treatment and its influence on soil functional recovery (Garau et al., 2007). Moreover, the measurement of soil enzyme activities has proved to be suitable and valuable tool for detecting natural and anthropogenic disturbance (Garau et al., 2007).

The exudates from roots act as messengers that stimulate biological and physical interactions between roots and soil organisms. They modified the biochemical and physical properties of the rhizosphere and contribute to root growth and plant survival. The rhizosphere environment had a lower pH, lower oxygen and higher carbon dioxide concentrations but exudates could make the soil more acid or alkaline (Lines-Kelly, 2005).

2.6 RESTORATION ASPECT USING CCPs

Scientific progress in the restoration of derelict lands requires an understanding of successional sequence of colonization, species' autoecology, and the dynamics of recruitment, and it is based on the population variability for metal tolerance or similar adaptive genetic criteria; therefore, both community and ecosystem level interactions among species must be investigated. Land restoration is therefore an acidic test of our ecological understanding of adaptive processes at all three levels (communities, species, populations) in an integrative applied science (Bradshaw, 1992).

There are certain levels of strategies that must be considered in restoration projects. A variety of factors must be considered in a successful mined-land revegetation program. Soil properties, other site characteristics, the time of seeding, the species seeded, and soil amendment application rates will affect revegetation success. The most effective way to match between soil conditions, species, and postmining land use is to select and place surface soil materials so as create a soil that is favorable to vegetation compatible with the postmining land use declared in a mining permit. Lime, fertilizer, and organic additions can be used to remedy problems of low soil fertility and or moderate acidity (Skousen and Zipper, 1996).

Organic matter content and others chemical properties did not show any relationships with the stability of aggregates in the experimental area. After 13 years of revegetation practices, a slight recovery of the stability was observed, although this was still lower than stability in soils of similar edaphic conditions in the original topsoil of experimental area (Alves et al., 2009).

The term reclamation is used to describe efforts that aim to improve the quality of the land by restoring some pre-disturbance functions. The potential of reclamation efforts to impact conservation of both species and ecosystems services grows. Most legislative mandates for land reclamation require evaluating the success of such efforts after a relatively short time period. The effect of initial vegetation composition on succession, particularly in old fields in the eastern USA, has long been discussed and many studies have highlighted the importance of vegetation composition at the time of abandonment on the successional trajectory. Many mine reclamation efforts focus on establishing rapid growing non-native species that control erosion but may complete with later successive, native species. In 1992 and 1993, the vegetation was surveyed on mined sites reclaimed 2-30 years prior and on reference sites in unmined and periodically logged forests. Results of this study suggest that a large number of native species colonize coal surface mined sites after 10-15 years (Holl, 2002).

3 PROCEDURE AND EXPERIMENT METHODS

3.1 INTRODUCTION

The general methodology undertaken for this project involved a series of procedures including choice of materials used and an experimental approach. The major materials used were: the coal ash aggregates, plants (beans, pumpkins, papayas, coco-plumoso and botellas), and an organic rich soil from Coamo Lake used as top soil. An experimental approach included: the growing process of seeds, extent and rate growth, physical hindrance and chemical enhancement, and also rhizosphere environment.

The area of interest of this project is located in the municipality of Santa Isabel, Puerto Rico. It is located in the south of the country. Figure 3 shows an aerial image of the place. Previously, this place was used by a private mining company since 1985, which ceased operations in 2006. It is currently an area surrounded by vegetation and plantation.

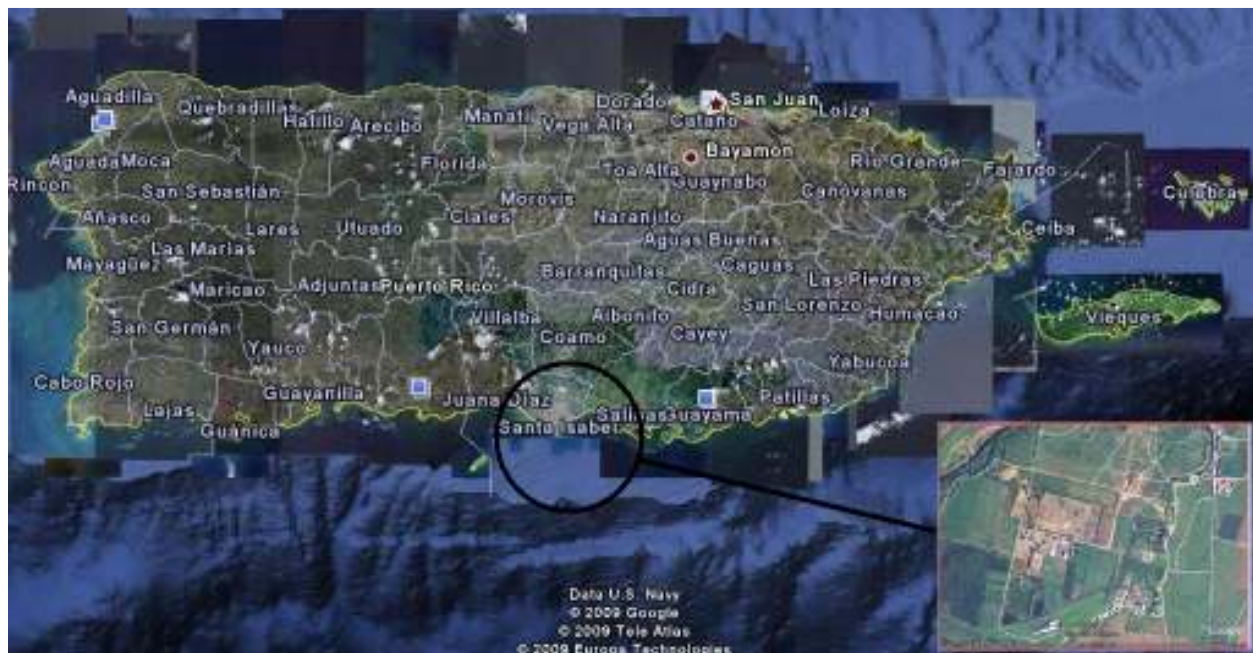


Figure 3. Aerial image of the study area (Source: Google Earth)

After the extraction of construction aggregates, the open pits represent an environmental hazard. Factors such as security risks because of the depth, inappropriate accumulation of waste, among others, are the worries to people near the area.

All analyses and experiments were made with equipment and instruments in the Environmental Engineering Laboratory located in Civil Engineering Building, the University of Puerto Rico at Mayagüez.

3.2 MATERIALS

3.2.1 SOILS

Different types of soil were used according to the structural scenario for the restoration of the open pits in the area. The open pits in the area were already filled with soils from the Bay of Guayama (BS) at a depth of approximately 0.3 meters. For future vegetation, soils from the Coamo Lake (TS) will be used as a top soil at a depth of 1 meter. Figure 4 shows the schematic of the backfilling.

Top soil was used for the current experiment after being transported to the laboratory area. The TS samples passed a sieved size 3 / 8 " were used for the experimentation. Then, it was put in an oven at a temperature of 105 ° C and dried for 24 hours.

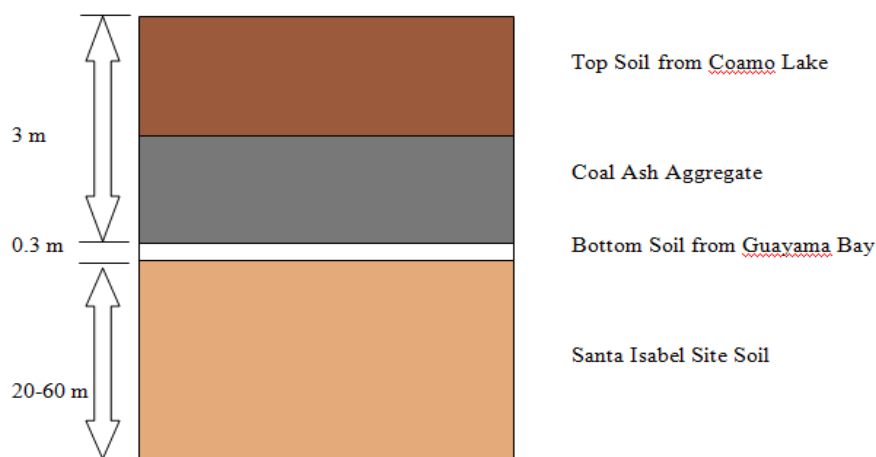


Figure 4. Backfilling of the site (Santa Isabel Area)

There were several characterizations of the soil samples used in the project. At the end of each experiment, a digestion process was developed for the soil samples according to the Digestion Digesdahl HACH method. The following parameters were determined (Table 5): pH, nitrogen and phosphorus. The concentration of lead was determined by the Lead Track method (HACH), and the amount of organic matter analyzed by the Loss-on-Ignition method (Standard Methods, 2002). A duplicate for each sample was made.

Table 5. Characteristics of the soils

Samples	pH	Total Nitrogen (as N) (mg/g)	Total Phosphorous (as P) (mg/g)	Lead (mg/g)	Soil Organic Matter (%)
TS	6.50±0.099	0.202±0.009	0.044±0.002	0.00±0.000	5.69±0.172
BS	7.94±0.057	0.000±0.000	0.002±0.001	0.00±0.000	1.92±0.086

Particle size distributions of the soils were evaluated with a sieve analysis (Figure 5). The principal component of the soils analyzed was sand.

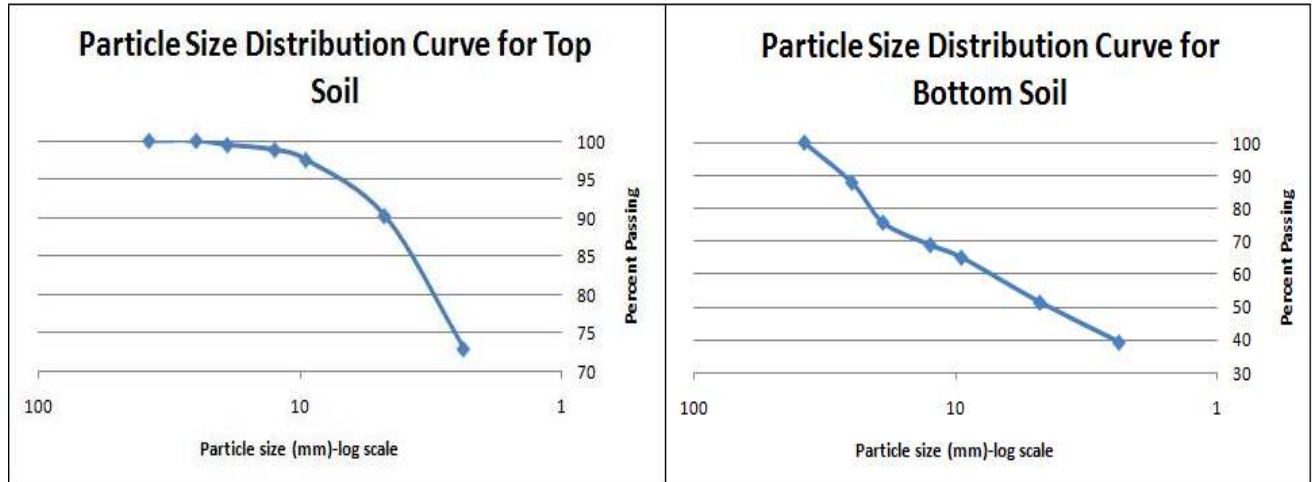


Figure 5. Particle-size distribution curves

Also, a hydrometer analysis was conducted to classify soil texture. As shown in Table 6 and Figure 6 both soils had a texture of loamy sand.

Table 6. Soil classification (Hydrometer analysis)

Sample	% Sand	% Clay	% Silt	Soil texture
TS	75.13±1.95	11.62±2.10	13.25±0.16	Loamy sand
BS	74.47±0.00	16.19±0.00	9.34±0.00	Loamy sand

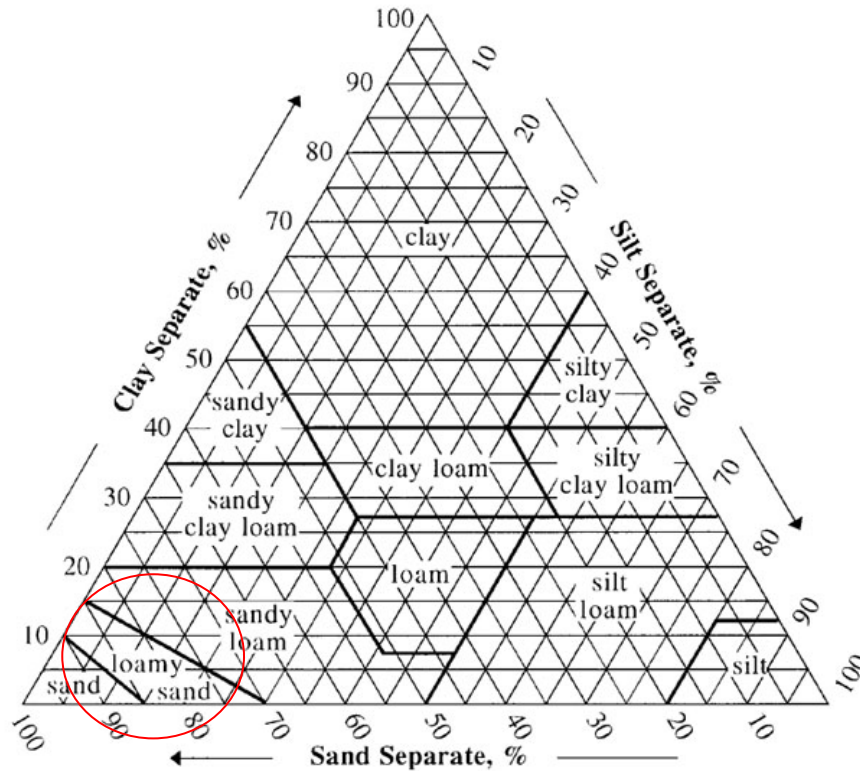


Figure 6. Soils texture classification (Source: USDA, 2009)

3.2.2 COAL ASH AGGREGATE

CAAs (Figure 7) was collected from a coal burning plant, AES in Guayama, Puerto Rico. This material is a solidified mixture of FA and BA with water and the main chemical components, by weight, are: 51% ($\text{SiO}_2 + \text{Al}_2\text{O}_3 + \text{Fe}_2\text{O}_3$), 30% lime (CaO), and 15% SO_3 . More detailed characteristics of the CAA can be found in Pando and Hwang (2006) (Table 3). They were oven dried at 105°C for 24 hours, crushed with a mechanical mixer located in the Material's Laboratory in Civil Engineering Department, and then sieved to avoid unwanted particles and to collect sizes of 2.36 to 9.53 mm for the experiments.



Figure 7. CAAs from AES

Kochyl and Little (2004) made a gradation tests for CAAs and found that it had a similar gradation as natural gravel (Pando and Hwang, 2006). Table 7 shows the results from the gradation experiment and Figure 8 presents the particle size distribution curve.

Table 7. Gradation results (Source: Pando and Hwang, 2006)

Sieve Identification	Sieve size (mm)	Total % passing
2 inches	50.8	100
1.5 inches	37.5	93.27
1 inches	25.4	84.55
0.75 inches	19	77.12
0.5 inches	12.5	66.99
0.375 inches	9.5	59.7
#4	4.75	44.84
#8	2.36	34.16
#16	1.18	25.72
#30	0.6	19.65
#50	0.3	14.66
#100	0.15	5.25
#200	0.075	1.92
P-200	0	0.00

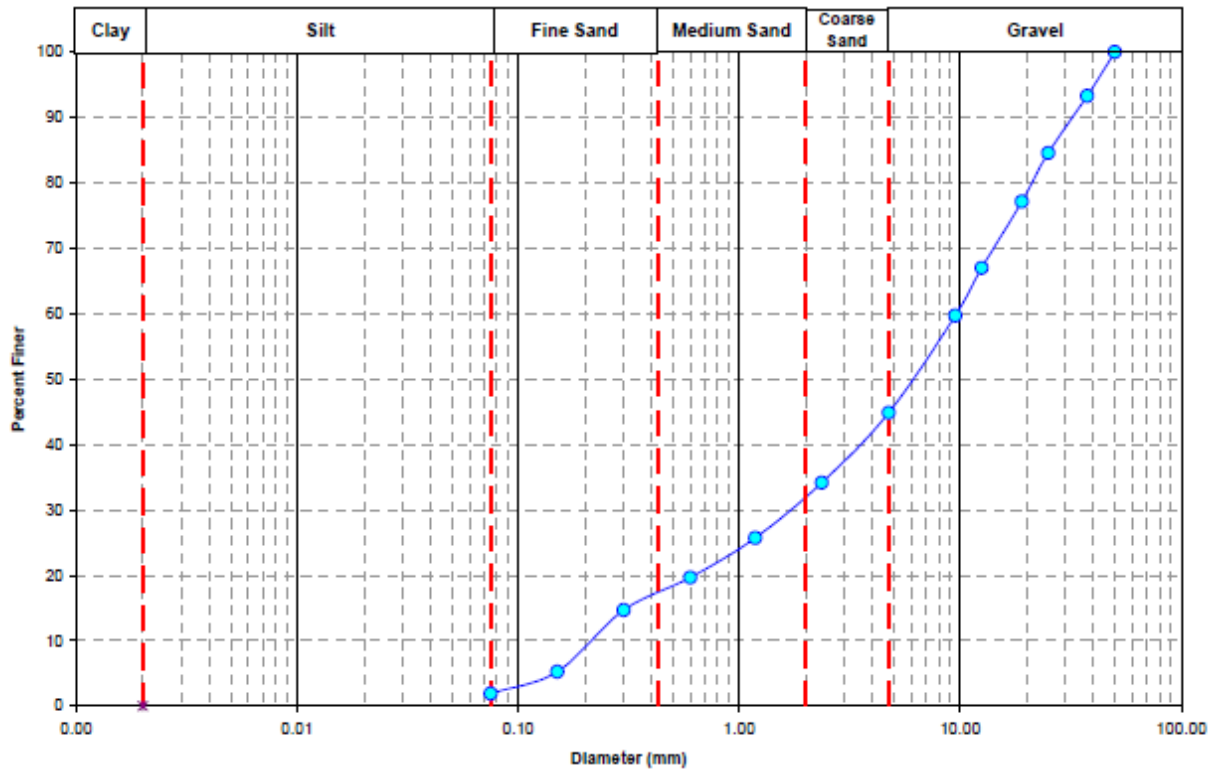


Figure 8. Gradation curve for AES (Source: Pando and Hwang, 2006)

3.2.3 PLANTS

For the proposed laboratory study, plants such as beans, pumpkins, papayas and botellas were used. Scientific names for each are: *Phaselous vulgaris* (United States Department of Agriculture (USDA), 2009), *Cucurbita maxima* (Department of Horticulture and Crop Science, 2009), *Carica papaya* (United States Department of Agriculture (USDA), 2009) and *Hyophorbe lagenicaulis* (Euroresidentes, 2000), respectively. Bean and pumpkins seeds were provided by a local nursery farm, and botellas and papayas were obtained from nursery farm on site. However, beans were studied as the main target plant because of their fast growing characteristics and convenient sizes for the laboratory study. Figure 9 shows the different plants used for project purposes.



Figure 9. Plants used for the experimental purposes: (a) botellas, (b) beans, (c) pumpkins, (d) papayas

3.3 EXPERIMENTAL METHODS

Phyto-viability tests on the restored land with the CAAs were conducted with respect to germination and growth of the plants. Also, physicochemical influence of the CAAs was evaluated for their potential physical hindrance to the roots and for their fertilizing effects. Lastly, rhizospheric biochemical changes due to the application of the CAAs amendment to restoration were evaluated.

3.3.1 GERMINATION OF SEEDS

It was necessary to determine the possibility of germination of selected plant seeds in direct contact with water from the CAAs. For this, a ceramic reactor was prepared (150 mm diameter, 250 mm height), which included 835 grams of gravel at the bottom, 1,500 grams of sand and 1,080 grams of CAAs. After having this structure (Figure 10) 1,000 mL de-ionized water was added for 5 days. The infiltrated water was collected and was used for the seed germination process. Cadmium and lead were analyzed from water collected, both using an AAnalyst 400 Atomic Absorption Spectrometer (Perkin Elmer).

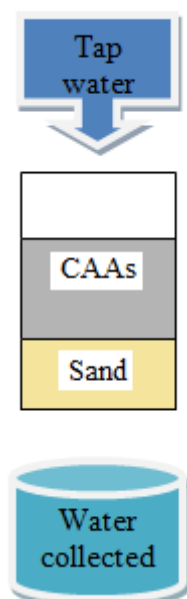


Figure 10. Diagram of CAAs reactor to collect infiltrated water for the germination experiment

For the seed germination process, 5 plastic reactors were prepared with a height of about 11 inches and a diameter of 4 inches (Table 8). To support the soils, 2.5 inches of gravel were added on the bottom. The TS from Coamo Lake (6.5 inches) was added on the top of the gravels. Prior to the use, the gravel was washed with tap water and then placed in oven at 105°C for 24 hours. The top soil passed a sieve size 3/8” and then placed in the oven at 105° C for 24 hours.

The reactors were placed in an environmental chamber (35 in L, 25 in W, 24 in H). Temperature was controlled using a chiller (Thermo NESLAB RTE 10 Digital One) at a temperature of 25°C. It was also equipped with a light (GRO-LUX, Sylvania, 20 W), which remained on for 10 hours a day (Figure 11). A timer was used to control the duration of illumination (Global Heavy Duty Indoor / Outdoor Light Timer Kit, # 82,428,101, Globe Electric).

Table 8. Configuration of reactors for the germination experiment

Reactors	Gravel (g)	Top Soil (g)	Type of seed
CR	197	1253	N/A
1	201	1262	Beans
2	202	1264	Beans
3	200	1270	Pumpkin
4	196	1262	Pumpkin



Figure 11. Temperature controller and environmental chamber used for the germination experiment

Data from (National Oceanic and Atmospheric Administration (NOAA), 2009) was used for maximum and minimum precipitation. Using an average recurrence interval of 5 years, a maximum of 6.04 inches and minimum of 0.83 inches of precipitation frequency was obtained for the municipality of Santa Isabel. These data were used to establish the maximum and

minimum volume of water added to the reactors according to superficial area. Water from the CAAs reactor was used to the 5 reactors and the germination of seeds was monitored. The amount used was 105 mL by calculations of surface area and maximum precipitation. Minimum amount was not used because low germination rate. Cadmium and lead were analyzed both using an AAnalyst 400 Atomic Absorption Spectrometer (Perkin Elmer).

3.3.2 EXTENT AND RATE OF GROWTH

Reactors were designed to evaluate the germination rate of the seeds in the CAAs amended systems. Multiple factors were assessed on their effects on the germination rate and growth. The parameter monitored was the product of the germination rate and shoot growth. Different factors were evaluated: 1) backfilling mode with a mixed or layered application of the TS and CAAs; 2) the type of seeds, bean or pumpkin; 3) the ratio of the TS to the CAAs; and 4) the type of water sprayed to the systems, rain water and tap water.

In order to accommodate many factors governing the germination of the seed, this experiment was conducted and analyzed with a statistical design. Table 9 shows a design matrix of the experiment. Sixteen reactors (and 4 control reactors) were run for those factors aforementioned and tested in an environmental chamber, of which temperature will be regulated at 30°C (Thermo NESLAB RTE 10 Digital One) to provide a better germination environment in the laboratory and also the chamber was equipped with a lighting system (GRO-LUX, Sylvania-20 W). Those plastic reactors had an inside diameter of 2.5 inches and a height of 6 inches (Figure 12). Five seeds were placed to each reactor at 1.5 inches below the surface.



Figure 12. Sixteen reactors prepared with multiple factors

Table 9. Matrix of 4-factor, 2- level design

Reactors	Mixed/Layered	Type of seed	Distribution	Type of water	Top Soil (g)	CAAs (g)
CR1	N/A	beans	6" top soil	RW	664.9	/
CR2	N/A	beans	6" top soil	TW	674	/
CR3	N/A	pumpkin	6" top soil	RW	657.6	/
CR4	N/A	pumpkin	6" top soil	TW	677.3	/
R1	Layered	beans	4" top soil+2" aggregate	RW	440.1	134.3
R2	Layered	beans	2" top soil+4" aggregate	TW	225.1	254.3
R3	Mixed	beans	66.7% top soil+ 33.3% aggregate	RW	445.2	127.7
R4	Mixed	beans	33.3% top soil+ 66.7% aggregate	TW	222.7	258.6
R5	Layered	beans	4" top soil+2" aggregate	TW	439.6	134.5
R6	Layered	beans	2" top soil+4" aggregate	RW	227.5	254.5
R7	Mixed	beans	66.7% top soil+ 33.3% aggregate	TW	444.5	129.5
R8	Mixed	beans	33.3% top soil+ 66.7% aggregate	RW	222.5	259.5
R9	Layered	pumpkin	4" top soil+2" aggregate	RW	439.4	134.5
R10	Layered	pumpkin	2" top soil+4" aggregate	TW	227.5	254.5
R11	Mixed	pumpkin	66.7% top soil+ 33.3% aggregate	RW	444.4	129.5
R12	Mixed	pumpkin	33.3% top soil+ 66.7% aggregate	TW	222.3	256.5
R13	Layered	pumpkin	4" top soil+2" aggregate	TW	439.6	134.5
R14	Layered	pumpkin	2" top soil+4" aggregate	RW	227.5	254.5
R15	Mixed	pumpkin	66.7% top soil+ 33.3% aggregate	TW	447.2	129.5
R16	Mixed	pumpkin	33.3% top soil+ 66.7% aggregate	RW	222.9	262.5

To determine the amount of water to be added, the maximum and minimum volumes of water were calculated according to the precipitation data for Santa Isabel, Puerto Rico and the superficial area of each reactor. Because the minimum volume rainfall intensity was too small for plant experiments, the experiment was only conducted adding the maximum volume of 40 mL during 3 times per week (Mondays, Wednesday and Fridays). Reactors were also in the chamber at 30°C with a light on during 9 hours per day.

3.3.3 PHYSICAL HINDRANCE/CHEMICAL ENHANCEMENT

The previous experiment aforementioned suggested potential of physical hindrance and/or chemical enhancement due to the presence of the CAAs contacting to the roots of the plants. To check this potential, another experiment was conducted.

In order to accommodate more seeds, 4 rectangular reactors (Figure 13) were constructed of acrylic plates with effective volume of 800 in³ (13 W, 8 L, 8 D). Control reactors were also prepared without the CAAs. The reactors had different distributions of gravel as a filter, CAAs in the middle and top soil on the top (Table 10). Reactors set-ups were done in accordance to the results which were obtained from the previous experiment. The extent and rate of growth was evaluated with respect to the height of the plants, the number of leaves and cotyledon development.

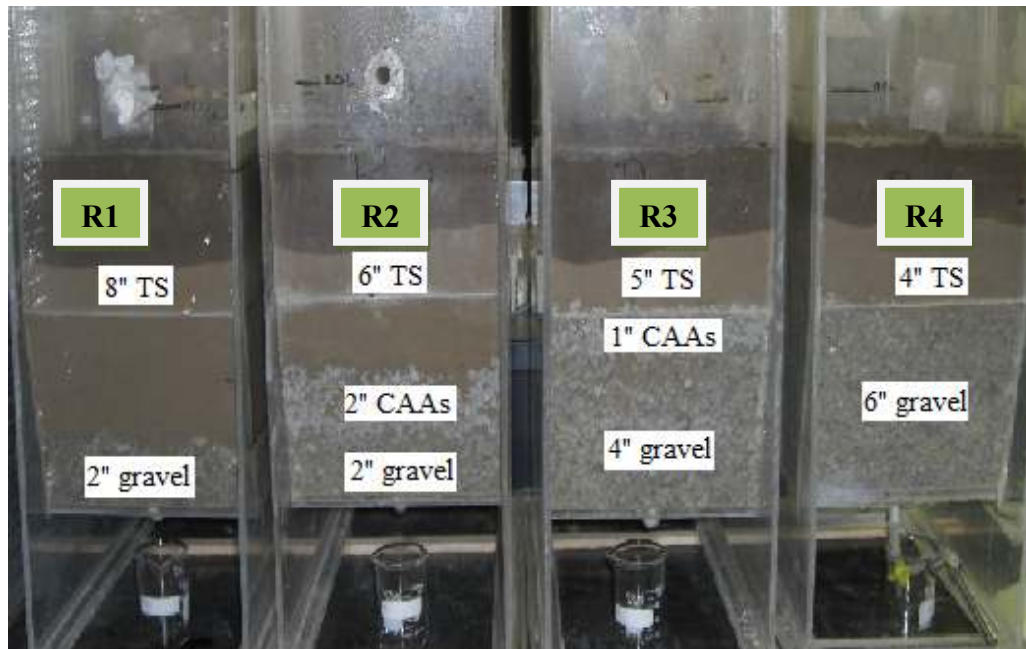


Figure 13. Reactors for extent and rate of growth experiment

Table 10. Distribution of the reactors

Reactors	TS (g)	Gravel (g)	CAAs (g)	Volume (m ³)	Density (kg/m ³)
1	20540	4830	/	0.01	1536
2	15960	4160	2730	0.01	1541
3	10470	11580	1550	0.01	1521
4	10610	13900	/	0.01	1542

Each reactor (Figure 14) was of 13 inches long and 8 inches wide (top view). Six seeds were placed 4x4.3 inches between each other at a 1.5 inches depth in each reactor. For each seed are, 140 mL of tap water were added according to volume calculated using the precipitation data from Santa Isabel, on every Mondays, Wednesdays, and Fridays.

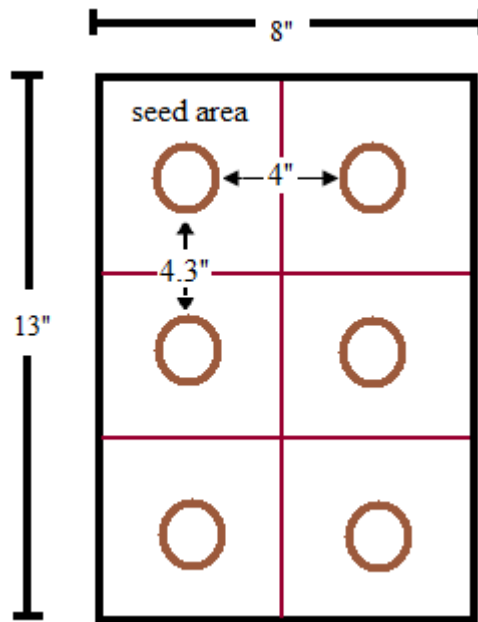


Figure 14. Top view of reactor

During the 5 weeks of experiment, measurements were taken such as heights of shoots, the number of leaves, and first cotyledon/flowering. Heights of the shoots and the number of leaves were recorded 2 times per week. At the end of the experiment, beans were sacrificed

(Figure 15) taking them from the reactors, washing off any loose soil, counting the numbers of roots, and weighing the root and shoot structure wet and dried in an analytical balance (ACCULAB, L-Series) in the laboratory. They were dried in an oven at 100 °C overnight and stored in a Ziploc bag for further analysis. Root was separated from the plants and measurements of root-shoot ratio were calculated in a dry weight ratio.



Figure 15. Root shoot ratio procedure: (a) sacrificed beans, (b) washed structure, (c) after dried

3.3.4 EXPANSION OF PHYSICAL HINDRANCE/CHEMICAL ENHANCEMENT WITH VARIOUS PLANTS

This experiment was developed to further evaluate the behavior of the plants with respect to the growth patterns of plants, according to different distance of the CAAs from the surface which might chemically and physically affect plant growth and healthiness. It was expanded for influence plant types such as botellas, papayas and pumpkins. Table 11 shows the distribution of those reactors.

Table 11. Distribution of expansion of physical hindrance with various plants

Reactors	R1	R2	R3	R4
Plant	Botella	Botella	Beans, papayas, pumpkins*	Beans, papayas, pumpkins*
TS (in)	7	7	5	5
CAAs (in)	0	2	2	0
Gravel (in)	4	2	4	6

*pumpkins were seeded after completion with beans

Beans were sacrificed after sacks development. Seeds in the sacks were digested using Digesdahl Apparatus (Figure 16) and lead was measured by Lead Track method (HACH). Pumpkins were added after the completion with beans. The numbers of leaves, first flowering, and heights were recorded. Like the Physical Hindrance experiment, 140 mL of tap water was sprayed to each seed area two times per week, and measurements were taken on Mondays and Fridays.



Figure 16. Digesdahl Apparatus (HACH) for digestion purposes

3.3.5 RHIZOSPHERE ENVIRONMENT

To test phyto-viability on the restored land with the same backfilling scenario and to evaluate consequent rhizospheric biochemical changes, an experiment was designed to conduct. Microbial activity in rhizosphere, growth rate and extent, and shape/conditions of the roots were analyzed.

Two acrylic reactors with a volume of 9,340 cm³ (19" H, 12" L, 2" W) were constructed with different application scenarios (Figure 17). Thimbles were prepared with gravel (6.35 mm-9.525 mm) for control reactor and CAAs (2.36 mm-9.53 mm) for R2. According to the surface area of the reactors and the precipitation data, total 300 mL of tap water were added to the thimbles, with 60 mL being added to each thimble using a plastic syringe. Tap water was added Mondays and Thursdays for 2 months of experiment. Heights of shoots, the numbers of leaves, and chlorophyll using Chlorophyll Meter SPAD-502, were recorded.

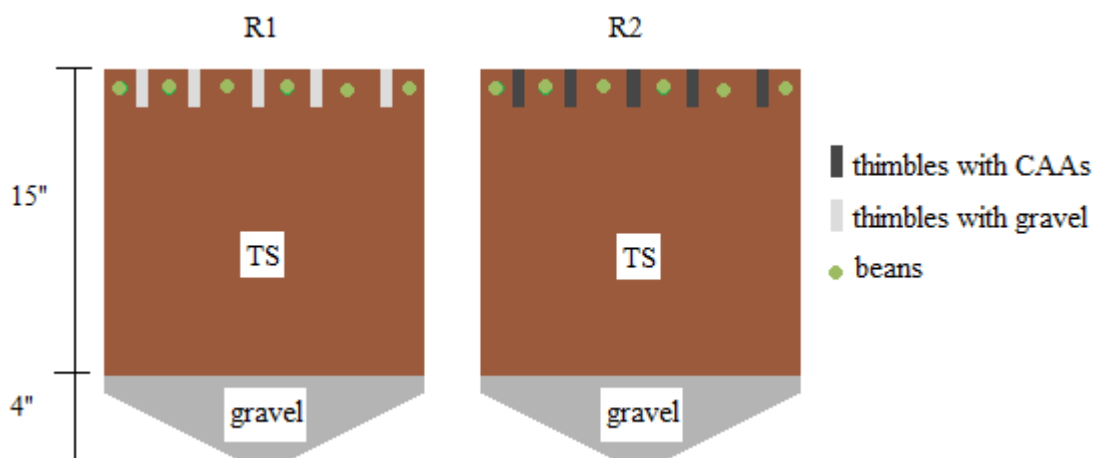


Figure 17. Reactor design for rhizosphere experiment.

The method used for determination of dehydrogenase activity was developed by Lenhard (1956). It involves colorimetric determination of 2, 3, 5-Triphenyltetra Zolium Formazan (TPF) produced by reduction of 2, 3, 5 Triphenyltetrazolium Chloride 95% (TTC) by soil microorganism. After following the Lenhard's method, samples were measured using a spectrophotometer (Portable Datalogging Spectrophotometer, HACH DR/2010).

The microbial activity in terms of total heterotrophic bacteria (THB) was monitored. THB were analyzed via a membrane filtration method (HACH) after 48 hours of incubation at 35°C. Ten grams of wet soil were dissolved in sodium chloride (NaCl) and a series of dilutions were made.

The membrane filtration method was used to estimate bacterial population in water that is low in turbidity (HACH Method, 2007) and is a simple way to estimate bacterial populations in water. The heterotrophic plate count medium (m-HPC) broth was used to enumerate heterotrophs in the samples. To retain the bacteria an appropriate sample volume (100 mL) was passed through a membrane filter with a pore size of 0.45 µm. The filter was placed on an absorbent pad saturated with a culture medium that was selective for heterotrophic bacteria growth. The numbers of colonies were observed through colony counter (Leica Quebec Darkfield) (Figure 18).

Materials used were sterilized in laboratory oven at 105°C (Erlenmayer flasks and cylinders). Sodium chloride solution and filtration funnels were sterilized with an autoclave (Tuttnauer Brinkmann 2340M) (Figure 18).

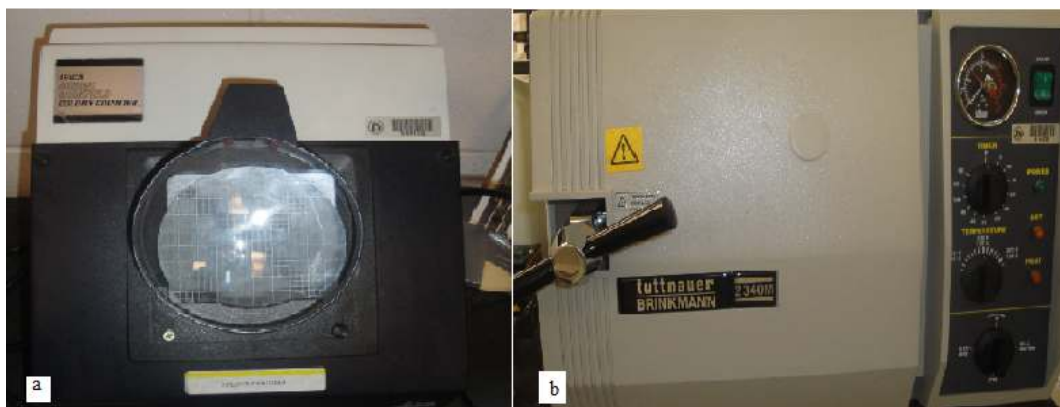


Figure 18. Equipment for THB analysis: a) Colony Counter and b) Autoclave

3.4 GENERAL ANALYSIS

Various physical, chemical, and biological analyses were conducted throughout the current experiment. The following Table 12 provides synopsis of those analyses:

Table 12. Synopsis of the analyses performed throughout the experiment

TYPE OF ANALYSIS	PARAMETERS	METHOD
Physical	Shoot height	Measurement by a ruler
	The number of leaves	Visual monitoring
	The number of roots	Visual monitoring
	The first cotyledon	Visual monitoring
	Sieve analysis	Methods of Soil Analysis
	Particle size distribution	Hydrometer analysis
Chemical	Lead (Pb)	LeadTrak (HACH)
	Cadmium (Cd)	Ion selective electrode
	Organic matter	Standard Methods
	Nitrogen (N)	HACH method
	Phosphorus (P)	HACH method
Biological	Total heterotrophic bacteria	Membrane filtration
	Chlorophyll measurement	Chlorophyll meter SPAD-502
	Soil dehydrogenase	Methods of Soil Analysis (Lenhard's Method)

4 RESULTS AND DISCUSSIONS

4.1 INTRODUCTION

This chapter provides the discussions of results obtained in the study. The results presented are: analysis of factors involved in plant growth studies, results of Cd and Pb as the main heavy metals, root and shoot ratio, and bacteriological and enzyme activity results in the rhizosphere.

4.2 GERMINATION OF SEEDS

A worst case scenario was developed to determine the germination of seeds in a direct contact of the seeds with the water infiltrated from the CAAs. The water collected from a column filled with CAAs was sprayed to the seeds as a worst case scenario and their germination was monitored. Both beans and pumpkins germinated well and grew in a good shape as shown Figure 19. Pb and Cd were the target heavy metals. After two weeks, roots, leaves, and stems were analyzed and both heavy metals were not detected. Similar results were obtained from a study by Li (2002), where FA did not significantly affect Cd and Pb concentrations in tomatoes (leaves, roots, stems, and fruit). However, beans plants had great capacity to accumulate heavy metals (Sharma et al., 2007).

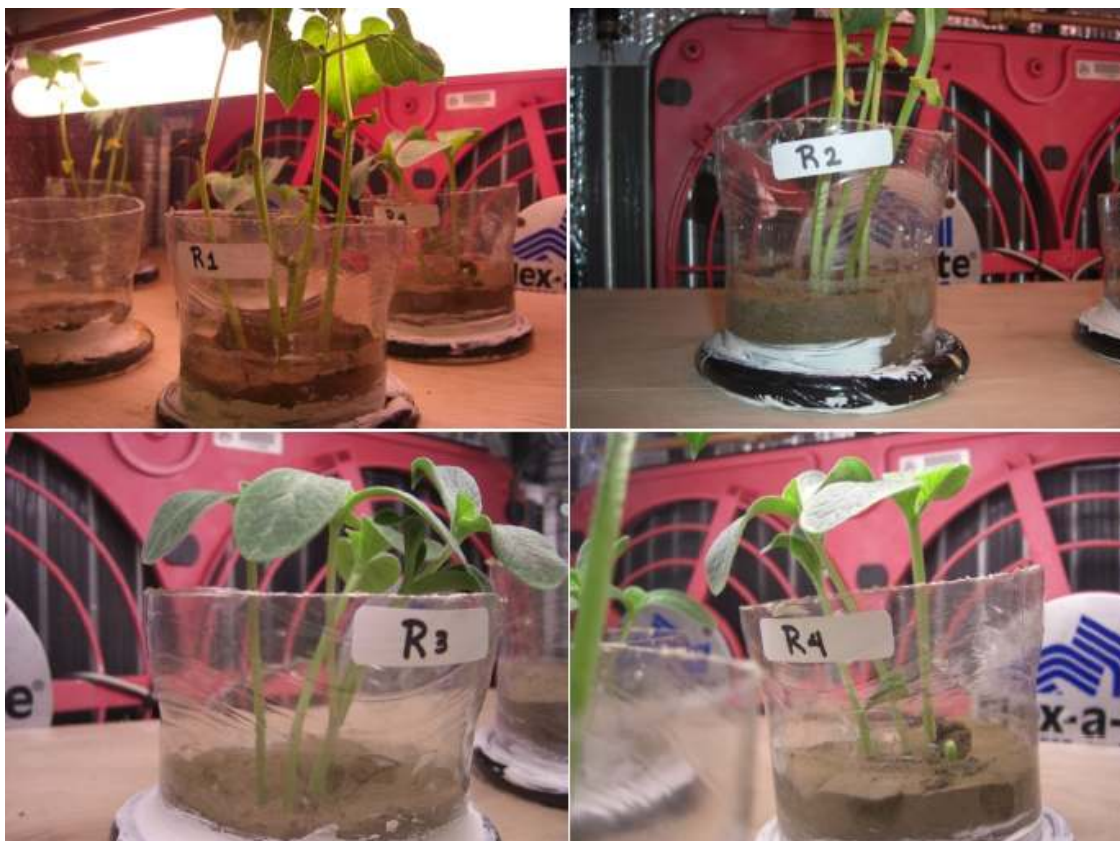


Figure 19. Germination results of beans and pumpkin seeds (top and bottom, respectively)

4.3 EXTENT AND RATE OF GROWTH

Soils vary in their ability to supply nutrients, and the factors which give rise to this variation are those which determine the development of soils: parent material, climate, topography, age, and vegetation (Fitter & Hay, 1987). To determine which factor influence more than others for the growth of the beans, we took into account the lengths of the roots to be harvested. Figure 20 shows at a glance the average of the lengths of the roots of plants in 16 different growth environments.

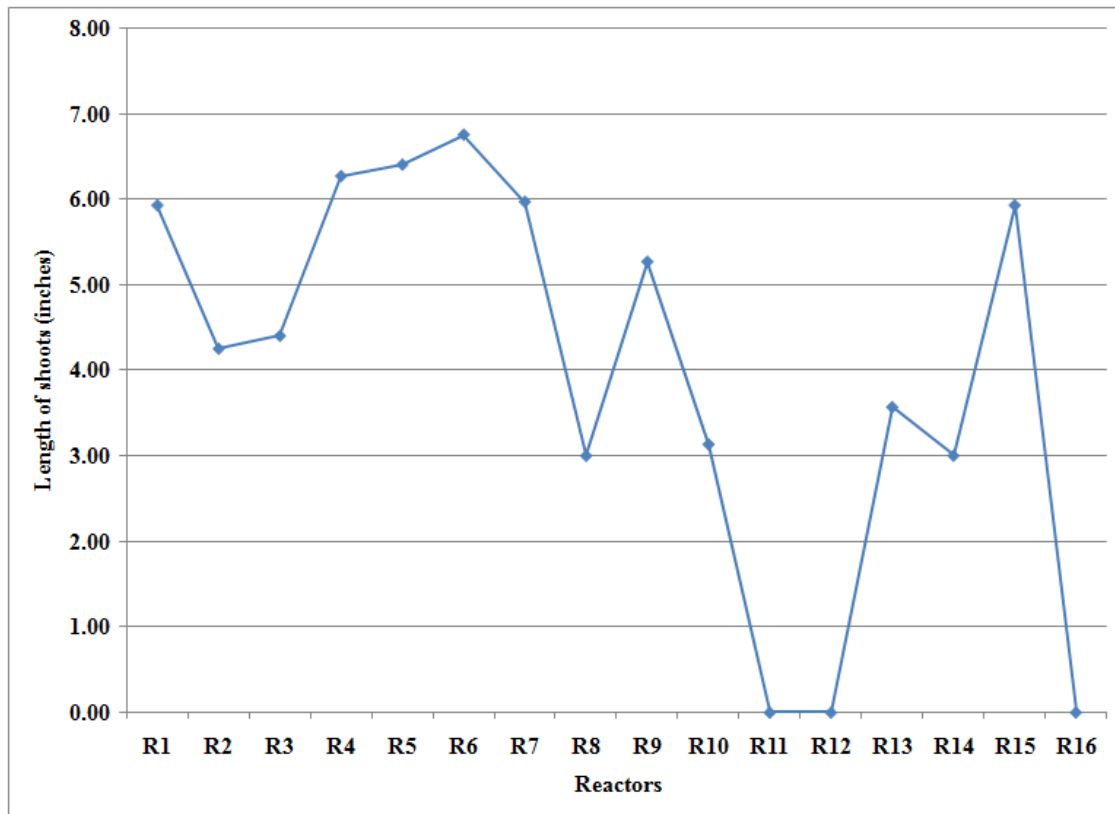


Figure 20. Average length of shoot in different environment

As shown in Figure 20, better length development was notable for reactors in a layered backfilling mode, compared to mixed mode (Reactors: 3, 4, 7, 11, 12, 15 and 16). Bean seeds were better at germination compared to pumpkin seeds (8 out of 8 beans, 5 out of 8 pumpkins). Type of water did not significantly affect the germinations of any seeds. Retarded growth of beans was observed in an experiment (Singh et al., 2008). Also, the slowed growth of *B. vulgaris*, grown in FA, reflected a decrease in root and shoot lengths (Singh et al., 2008; Mishra et al., 1986).

Poorter (1996) and Obert (2005) used a statistical modeling program (MINITAB ®) to show their plant responses results. MINITAB ® software was also used to statistically determine the dominant factor in the growth and seed development in the current research. Establishing a 2^4 factorial design in the program, the following factors and levels were analyzed as show in Table 13:

Table 13. Factors and levels in a 2^4 factorial design

FACTORS	LEVELS
Backfilling mode	Mixed or layered
Type of seed	Beans or pumpkins
Ratio	More CAAs or less CAAs
Type of water	Tap or rain

As shown in Figures 21 and 22, the main factor did not produce any statistical difference regardless of their levels, nor did the compounding factors.

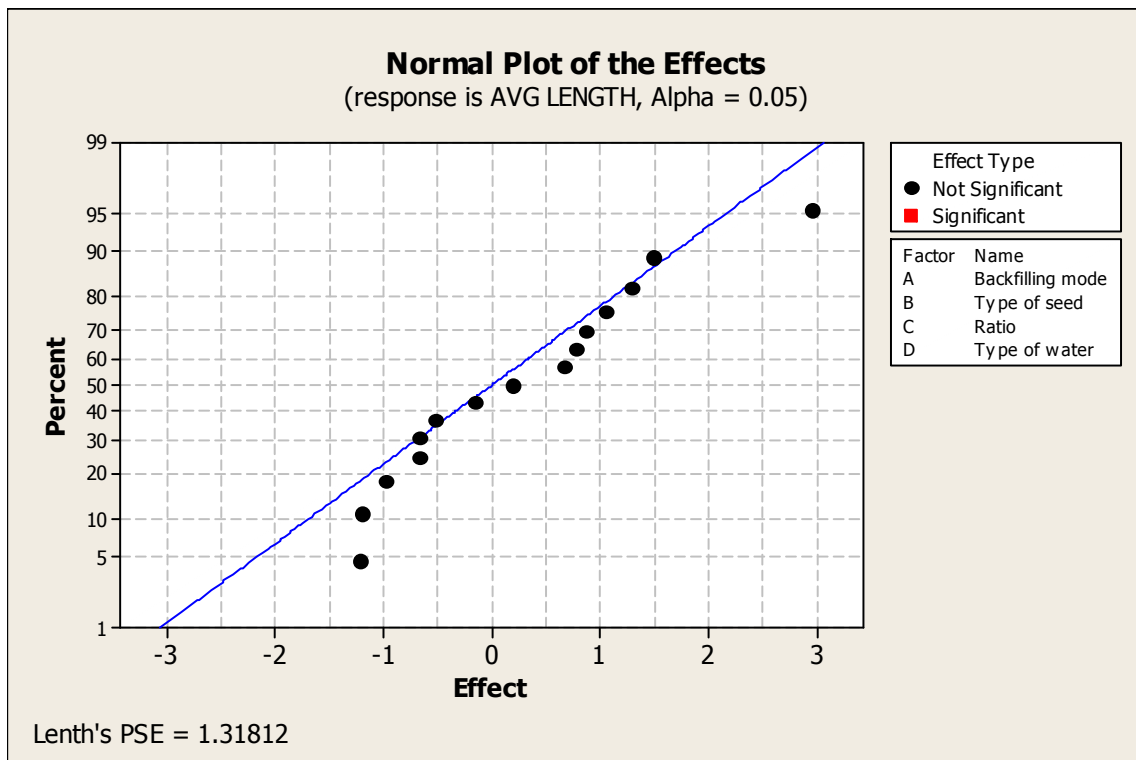


Figure 21. Normal plot of the effects (Minitab ®)

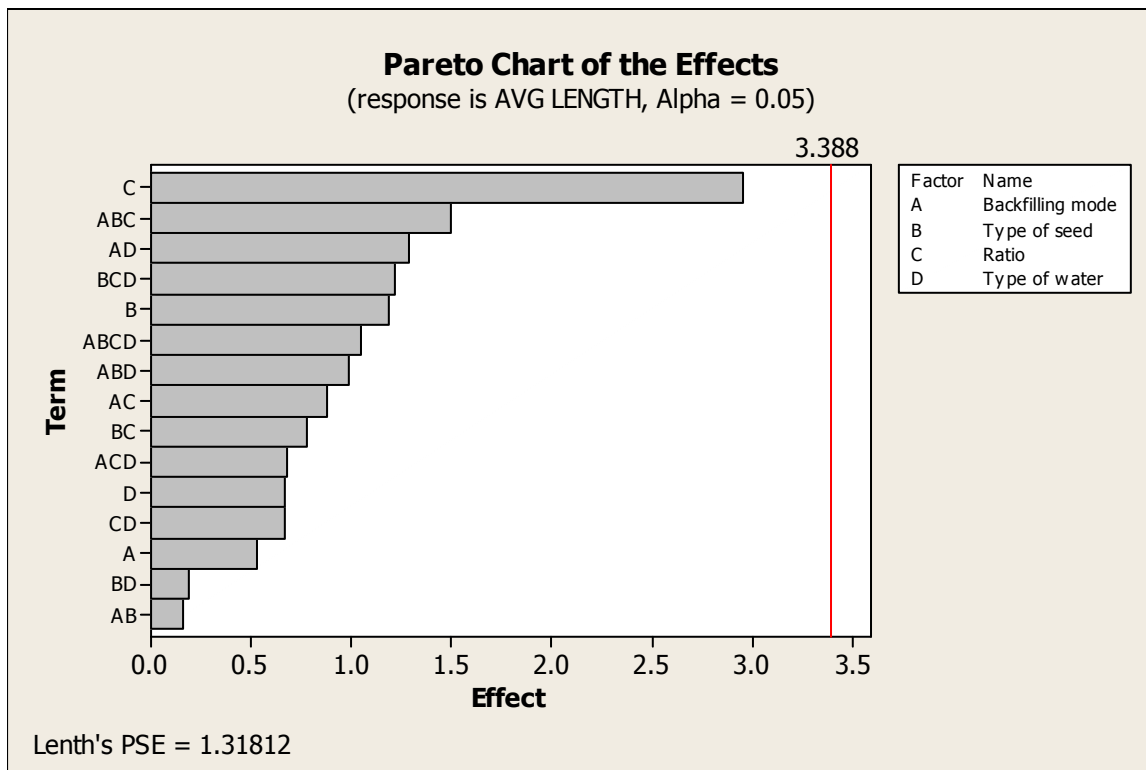


Figure 22. Pareto chart of the effects (Minitab ®)

However, based on the statistical analysis using the MINITAB ®, it can be said that both the main and compounding effects were in a normal distribution (Figure 21). In addition, among the main effects, the ratio of top soil to the CAAs produced the dominant effect on the length of shoots, followed by the seed type, the type of water, and lastly the backfilling mode (Figure 22).

An effort was made to phenomenologically explain the results obtained from the multi-factor germination and growth experiment. Accordingly, the product of shoot length and germination rate was assessed, knowing that not all seeds were germinated during the experiment.

As shown in Figure 23, better germination and growth were observed for the beans when they were seeded in the reactors with more TS in a layered application and, at the same time, when they were watered with tap water. The germination of seeds might be according to their respective characteristics. For example, the type of soil, water amount needed to survive, or

preferred environmental conditions. These findings led to additional experiments to be able to make answers to the following questions:

- Why did tap water produce better germination and growth than rain water did?
- Can the solid CAAs be a physical barrier to the roots so as to interface the growth?
- Can the CAAs possibly provide nutritional metal compounds to the plants so as to enhance their germination and growth?

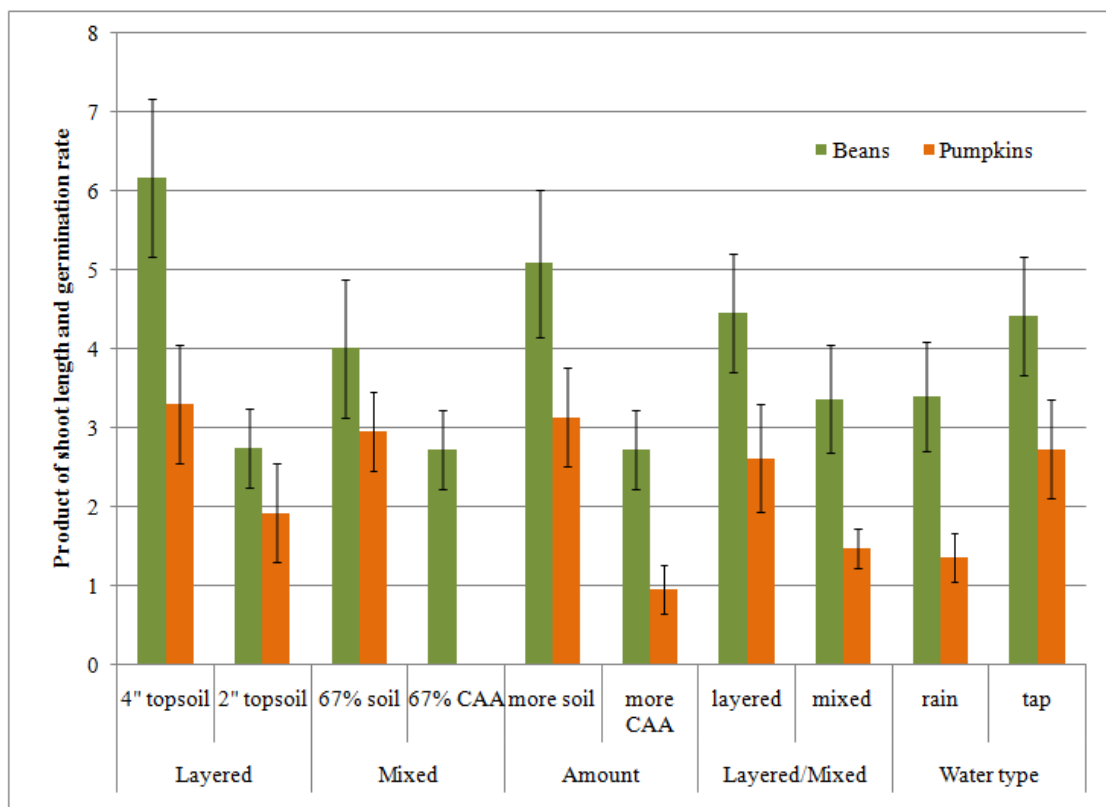


Figure 23. Product of shoot length and germination rate

4.4 TYPE OF WATER

Better germination and growth were observed with tap water compared to rain water. Water quality determined was specific conductivity, pH, and hardness of both waters (Table 14). A major difference between two type of water was found in the concentration of hardness. Tap water had 10th time greater hardness concentration than rain water.

Table 14. Results of analysis on pH, specific conductivity and hardness of tap and rain water

Water type	pH	Specific Conductivity ($\mu\text{S/cm}$)	Hardness (mg/L as CaCO_3)
Rain	7.5 $\pm$ 0.1	37.5 $\pm$ 28.1	6.3 $\pm$ 0.6
Tap	7.9 $\pm$ 0.1	42.6 $\pm$ 0.2	64.4 $\pm$ 4.0

A sub-experiment was conducted to elucidate the effect of hardness concentration on the germination and growth of the plants.

Two seeds were placed at a depth of 1.5 inches below surface in each plastic reactor (2.5 inches diameter and 6 inches long.) and were filled with TS at a depth of 5 inches. Table 15 describes the distribution of the reactors prepared. Corresponding to the actual maximum average precipitation in Santa Isabel, 40 mL of hardness water at a hardness concentration from 0 to 80 mg/L as CaCO_3 was sprayed on every other days for a month.

Table 15. Design of experiment to assess the effect of hardness on germination and growth

Reactor	A	B	C	D	E
Hardness in water sprayed (mg/L as CaCO_3)	0	4	20	40	80

Reactor that received the water with highest hardness made 100% germination as shown in Figures 24 and 25. The highest growth of the bean seeds was achieved in Reactor D which had been sprayed with the higher hardness concentration (80 mg/L as CaCO_3). In general, the number of leaves was not significantly different among the reactors (Figure 26).



Figure 24. View of the experiment to assess the effect of hardness on germination and growth

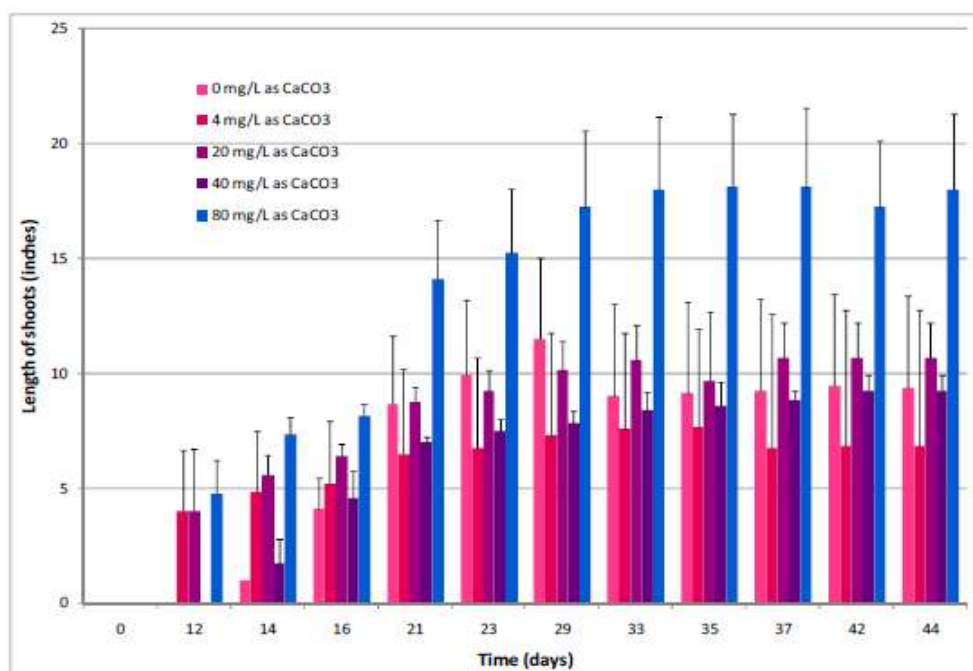


Figure 25. Length of bean shoots when receiving water at different hardness concentrations

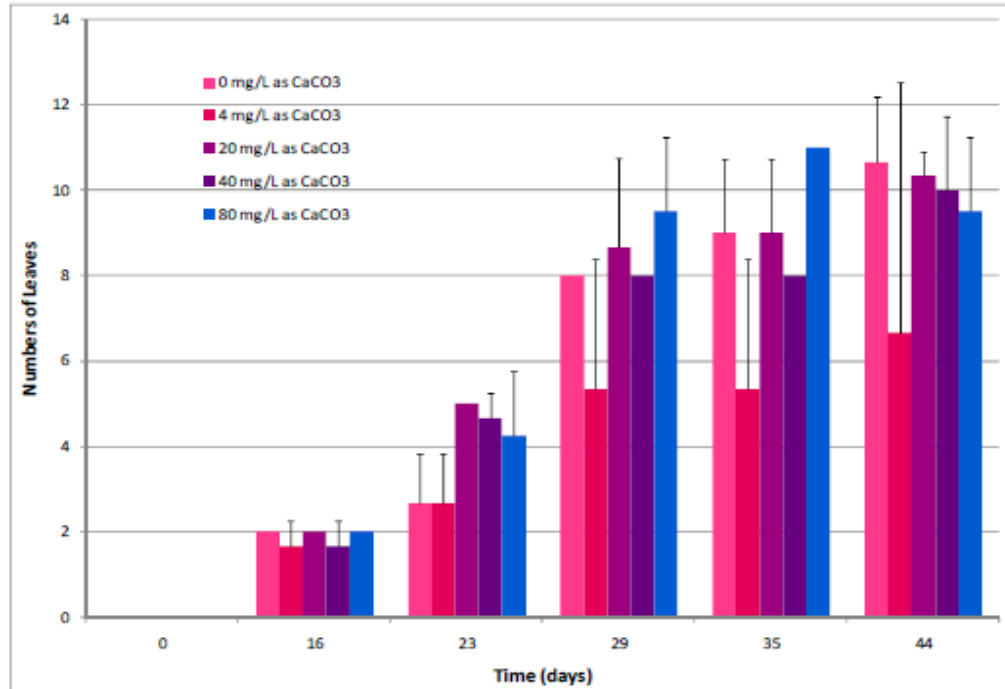


Figure 26. Number of leaves when receiving water at different hardness concentrations

Overall, water hardness showed important effects on the germination and growth of the plants. Tap water used in the previous experiment showed a better germination and development of beans or pumpkins seeds. Table 14 showed that tap water has higher hardness concentration (65 mg/L as CaCO₃ approximately) comparing rain water hardness concentration (6 mg/L as CaCO₃). Therefore, the additional hardness experiment proved that a better germination of seeds was with the higher hardness concentration.

4.4 PHYSICAL HINDRANCE/CHEMICAL ENHANCEMENT

All six bean seeds in each reactor were germinated (Figure 27). However, after a month of growth, 3 shoots died from the reactors 1 and 4 and 1 shoot died from reactor 2. The reactor 3 resulted in 100% of survivability of plants. The reactors 2 and 3 had the CAAs layers (Reactor 2: 2" CAAs and 6" TS, and Reactor 3: 1" CAAs and 5" TS) and demonstrated better shoot growth (Figures 28-31).



Figure 27. Scene of the reactors to assess physical hindrance of the CAAs

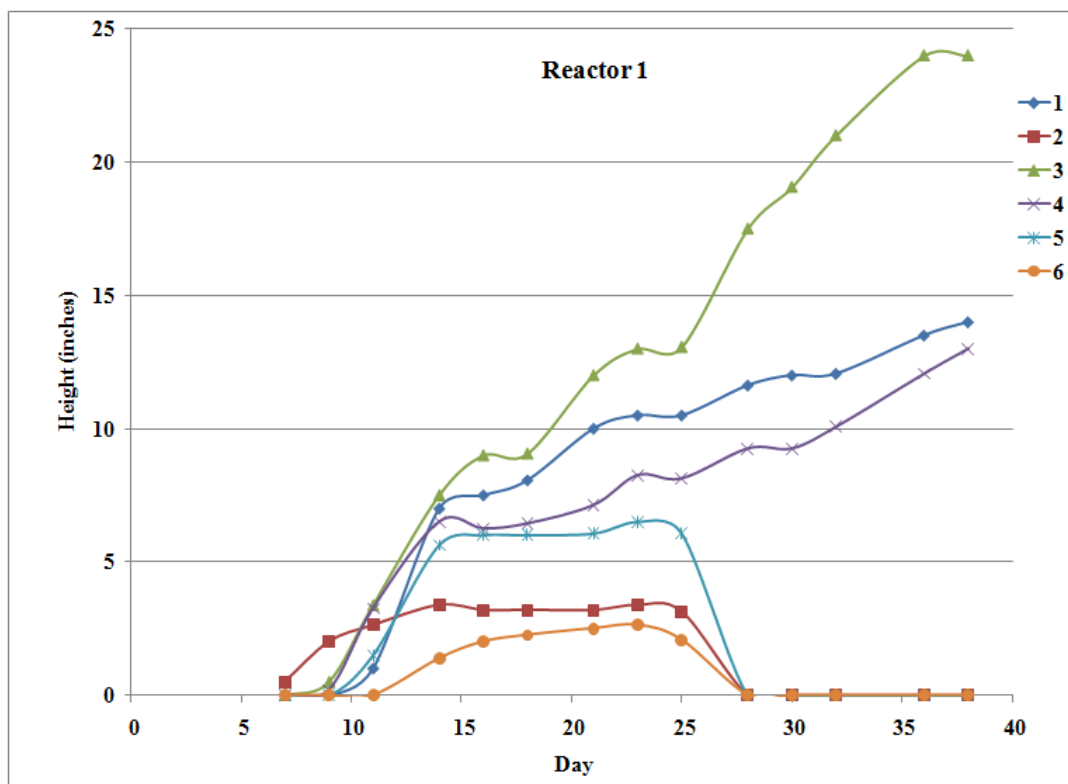


Figure 28. Height of shoots grown in reactor 1

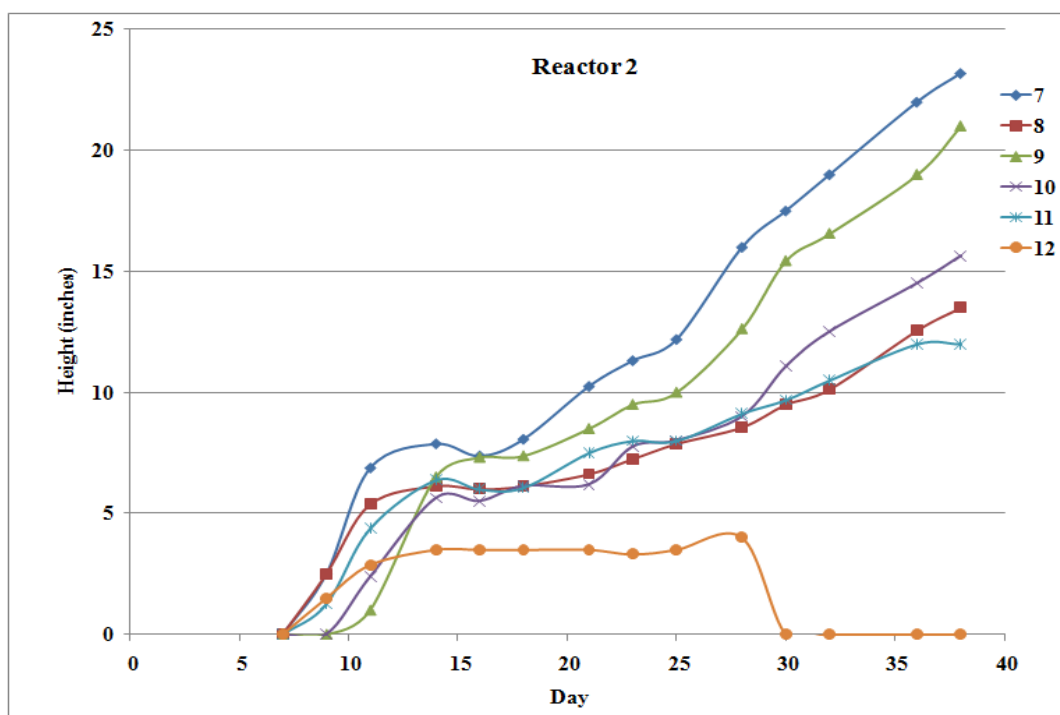


Figure 29. Height of shoots grown in reactor 2

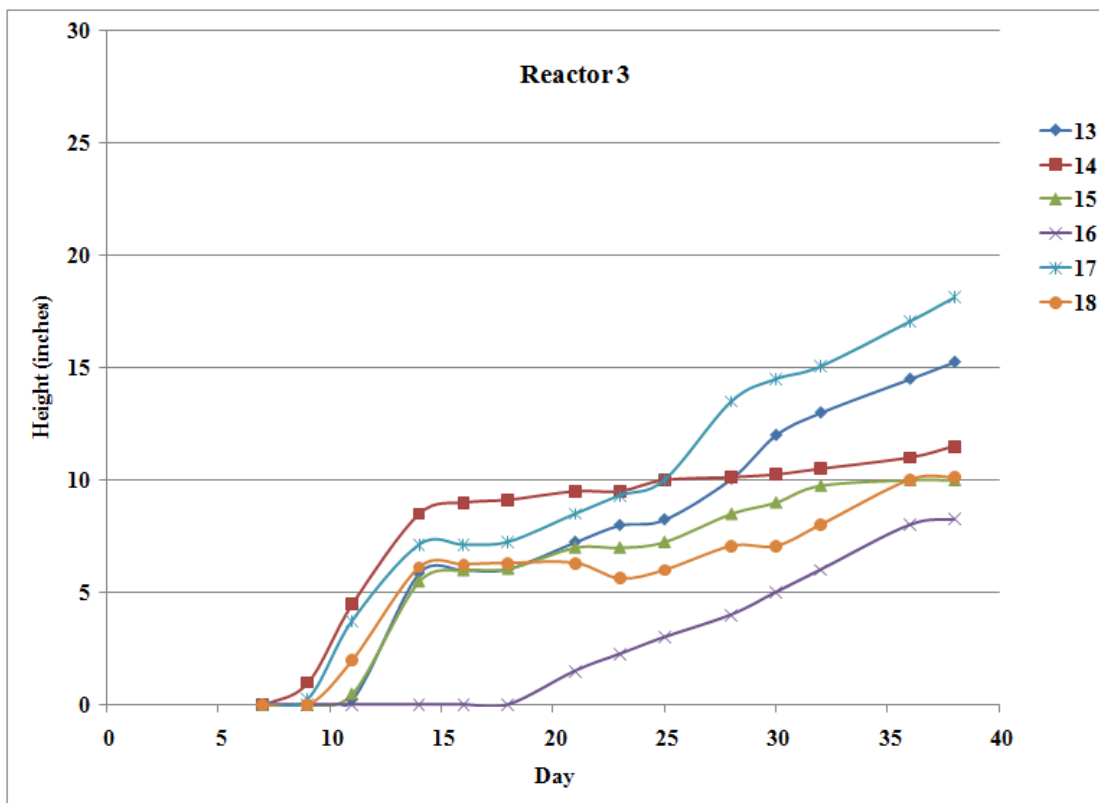


Figure 30. Height of shoots grown in the reactor 3

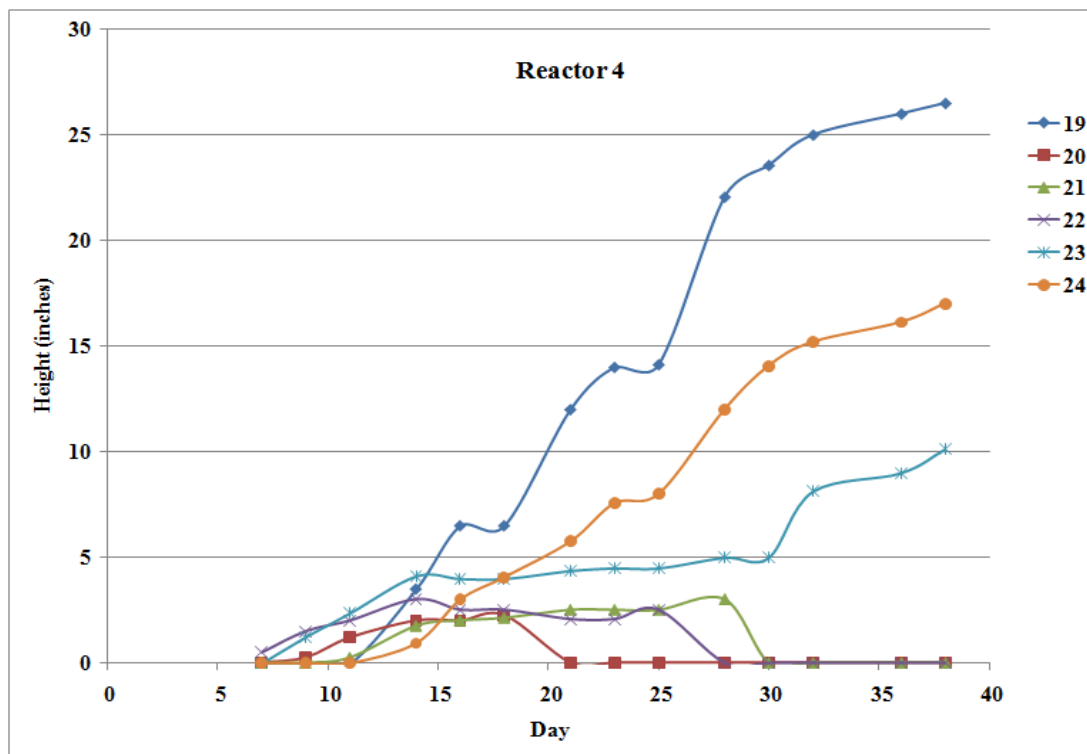


Figure 31. Height of shoots grown in reactor 4

The reactor 2 had the best shoot growth as showed in Figures (28 to 31), followed by reactor 3. Both reactors had CAAs and TS. Reactor 2 had 2 inches CAAs layer below 6 inches TS, and the reactor 3 had 1 inch CAAs layer below 5 inches TS. The shoots in the reactor 1 which had 8 inches of TS grew a similar manner that those in the reactors 2 and 3 up to 3 weeks of growth. However, its growth was limited.

Different results from those in the current study were reported in a study by Singh (2008), in which FA amended soil showed significant reductions in growth parameters such as number of leaves, leaf area, root and shoot and total plant height of *B. vulgaris* as compared to the non-FA amended soil. However, a similar result was reported that FA addition generally increases plant growth and nutrient uptake (Furr et al., 1978) and also improved the physical and chemical characteristics of the soil (Jala and Goyal, 2006).

Figure 32 shows roots and shoot weight of beans plants. It was measured at the end of the experiment to investigate the rate of growth and physical hindrance on the plant growth. The root and shoot of reactors 2 and 3 were higher than reactors 1 and 4. Inhibition of root growth in FA system may be due to compaction of FA particles which probably served as physical barrier to root degradation (Singh et al., 2008). Moreover, studies demonstrated that the FA contains high levels of heavy metals which induced inhibition of root growth by affecting root elongation due to reduced cell division (Gunse et al., 2000; Vazquez et al., 1999)

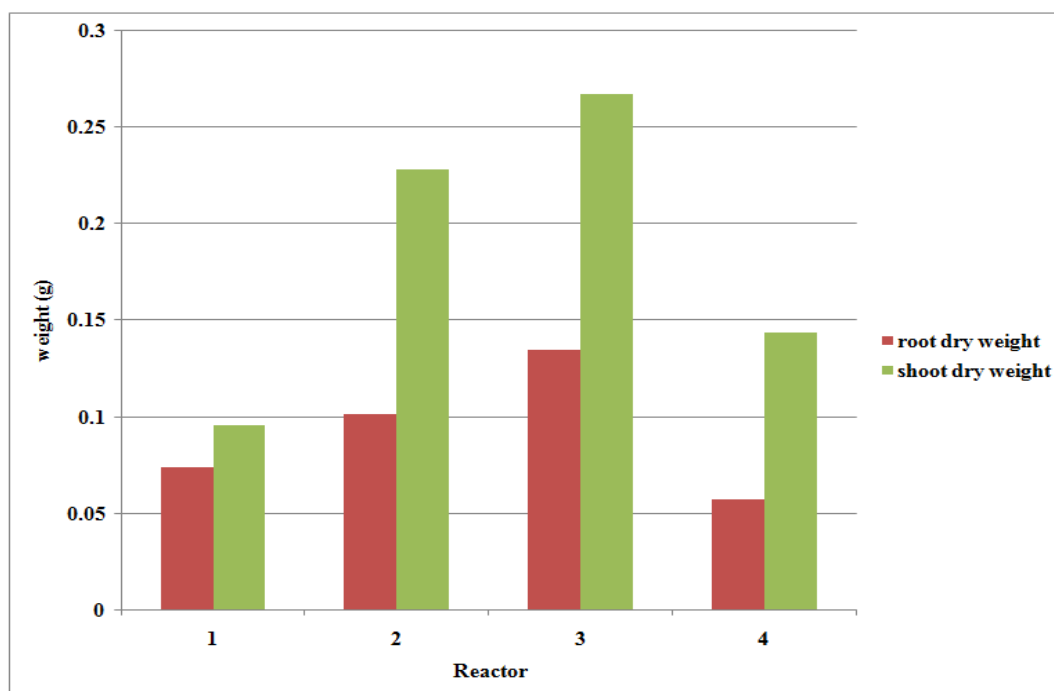


Figure 32. Root and shoot weight

4.5 EXPANSION OF PHYSICAL HINDRANCE/CHEMICAL ENHANCEMENT WITH VARIOUS PLANTS

As shown in Figure 33, botellas, papayas, beans and later pumpkins were tested with respect to physical hindrance that the CAA layer might exert to their roots and consequently their roots. Botellas at 8" and papayas 5" in height were planted directly to the reactors. Beans and pumpkins were seeded to them too.



Figure 33. Botellas, beans, papayas and pumpkins tested for potential physical hindrance

Two botellas were added to the reactors 1 and 2. No specific characterization of their growth was measured because of their physical appearance. However, it was observed that both botellas grew very well even after 4 months (Figure 34). Botellas might be a plant which does not require specific conditions to survive or grow.

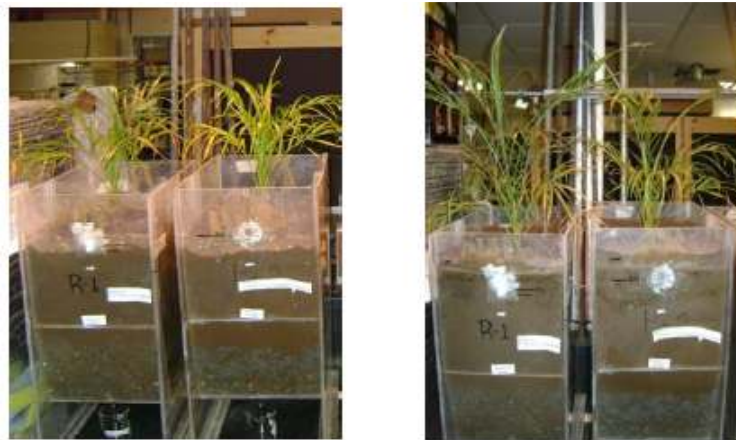


Figure 34. Comparison of the growth of botellas between the initial day (left) and after 4 months (right)

Initially, one papaya from a nursery farm was planted to the reactors 3 and 4. However, those two baby papayas died after one month due to parasites developed on the leaves. Four new baby papayas were planted again to one reactor. This time, a commercial pesticide (VEL 4283) was diluted 130 times as instructed and leaves were swabbed with it. Results are shown in Figure 35.

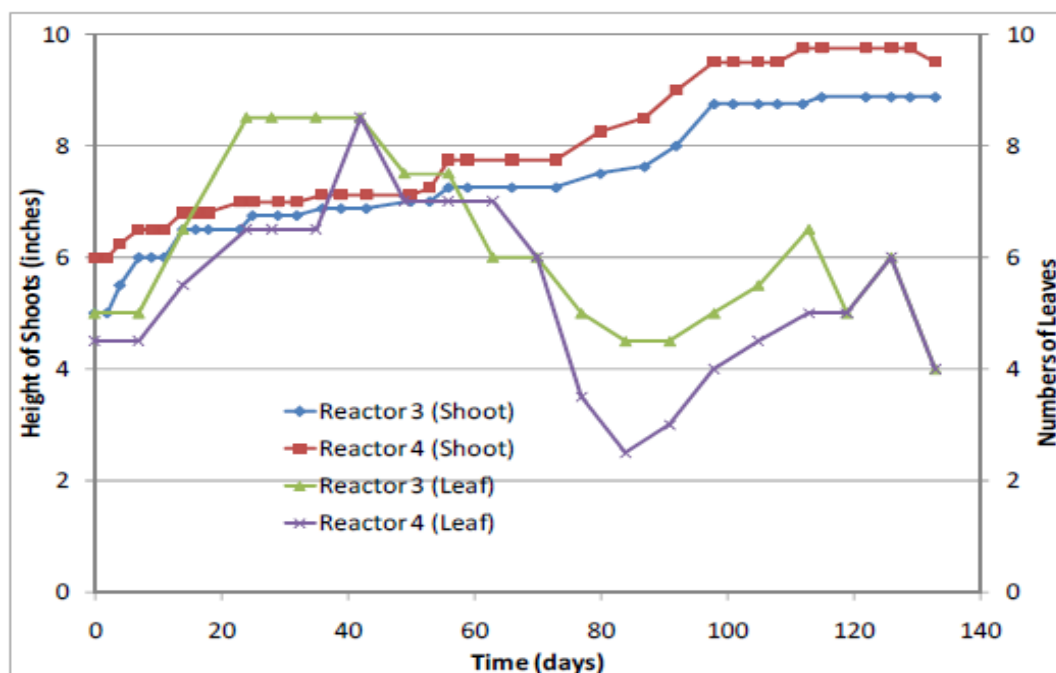


Figure 35. Height of shoot and number of leaves of papayas

As shown, shorter shoots but more leaves were found from papayas planted in reactor 3 which had the CAAs layer. The initial conditions might have influenced on this results. That is, changes in soil might be a determinant factor for their development.

A chlorophyll meter (SPAD 502, Konica Minolta) was acquired in the middle of the study and the chlorophyll intensity was monitored on leaves of papayas. Results showed a healthier growth of papayas in reactor 3 which had the CAAs layer than in reactor 4 which had a gravel layer (Figure 36). Singh et al. (2008) found that the chlorophyll intensity on the bean leaves decreased significantly with an increasing concentration of FA. The accumulation of heavy metals also led to inhibition of chlorophyll formation (Krupa and Baszynski, 1995).

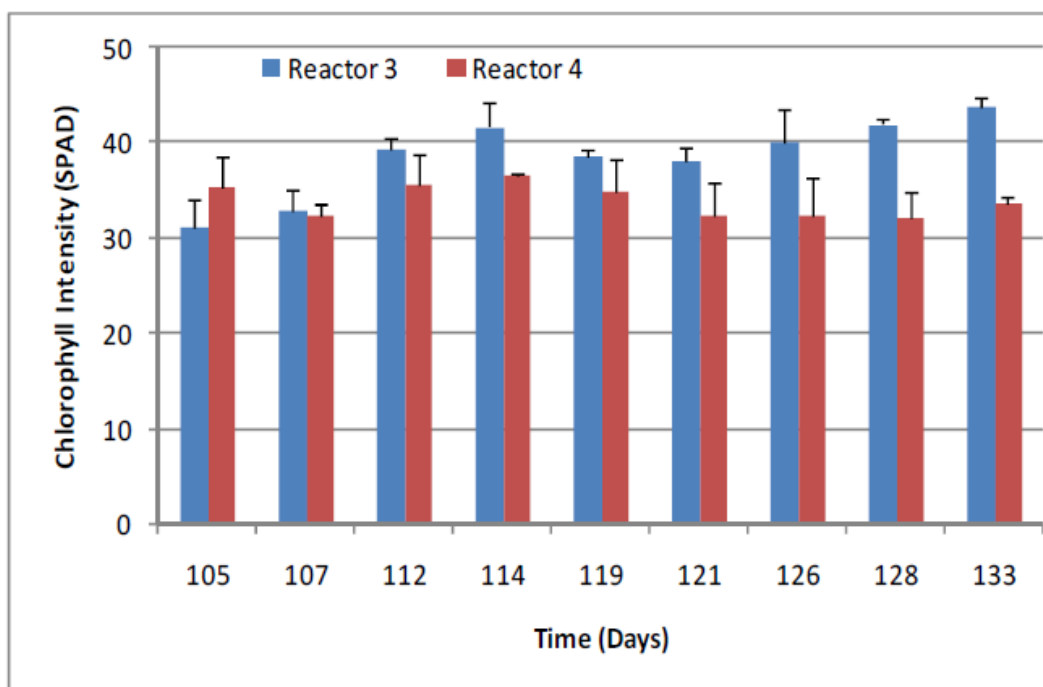


Figure 36. Chlorophyll intensity in the papaya leaves

Figure 37 shows the heights of beans. It shows that the bean seeds were germinated at the almost same time. First cotyledon was observed after 8 to 10 days approximately. In general, they grew well. Likely, they started blossoming 29 to 31 days after seeding.

The heights of shoots of the beans grown in the reactor 4 were very dissimilar between two bean plants. The number of bean leaves was found very similar except for a bean grown in reactor 4 (Figure 38).

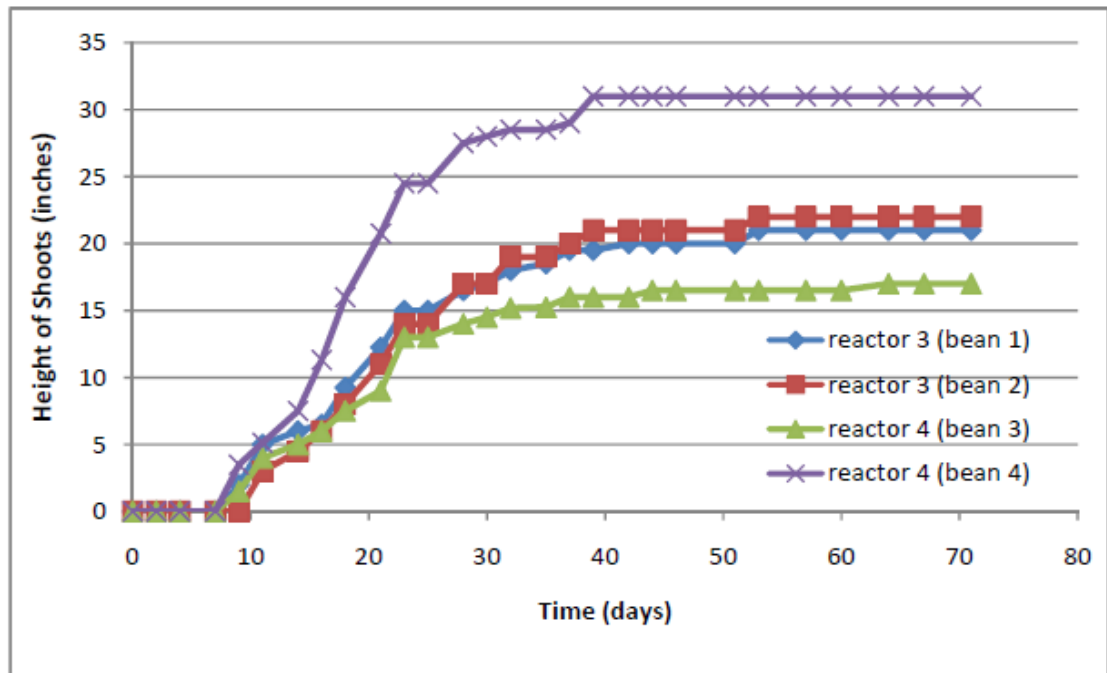


Figure 37. Height of shoots of the beans

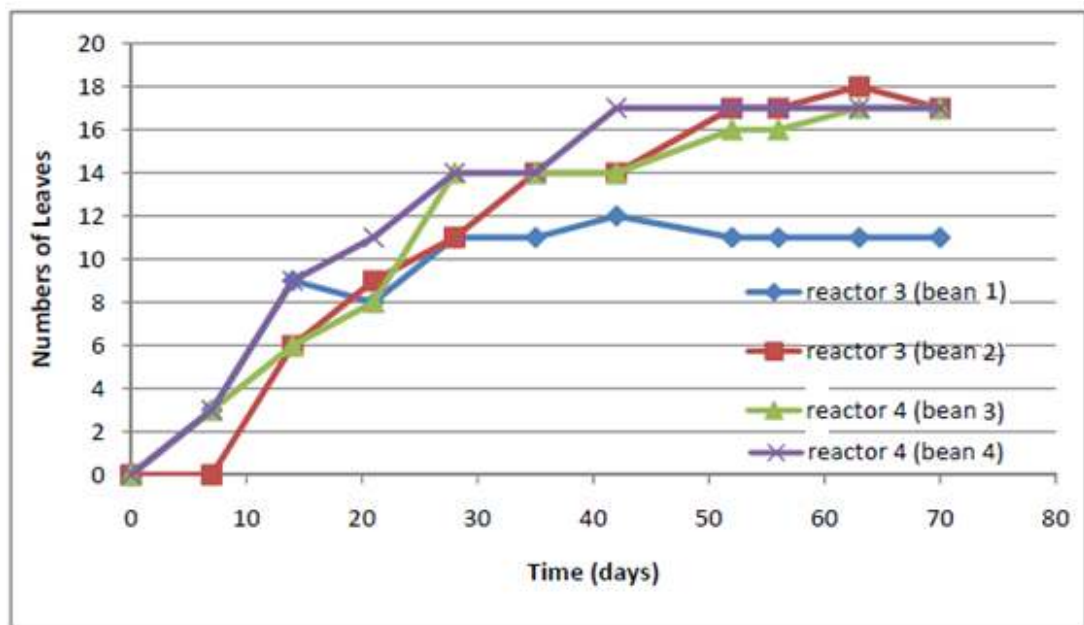


Figure 38. Number of leaves of the beans

After 40 days approximately, bean sacks were developed and their numbers and lengths were monitored (Figures 39-40). Incorporation of FA into medium textured soil influenced soil properties and crop productivity (Kalra et al., 1998) and might be helpful for bean sacks

development. Bean seeds in the sacks were harvested at the end of experiment and were extracted for Pb analysis by a HACH Digestion method. Measurements of Pb were made using an ion selective electrode and the results showed no Pb in the extractant.

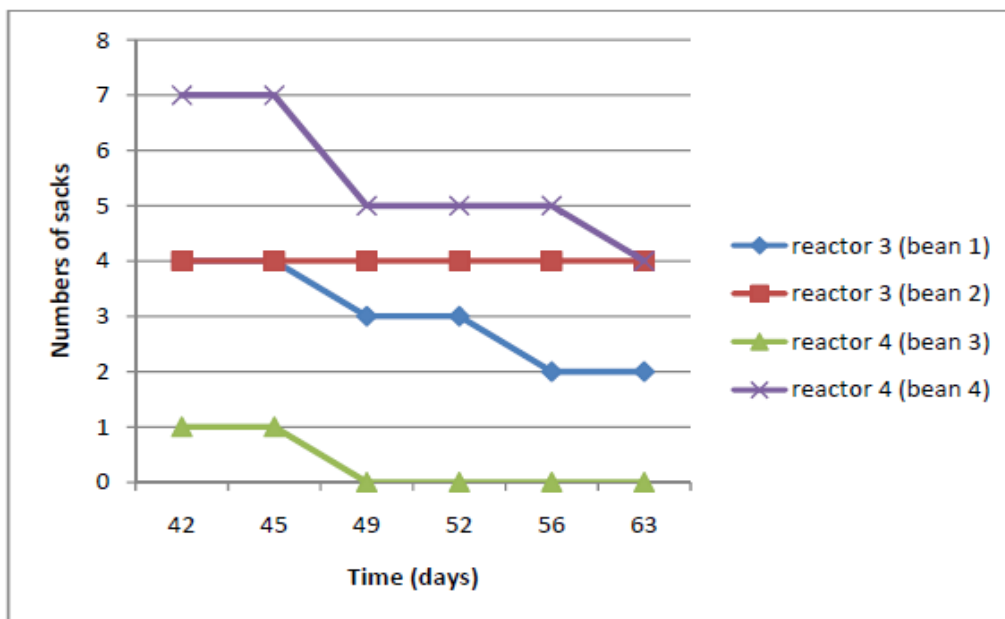


Figure 39. Number of bean sacks

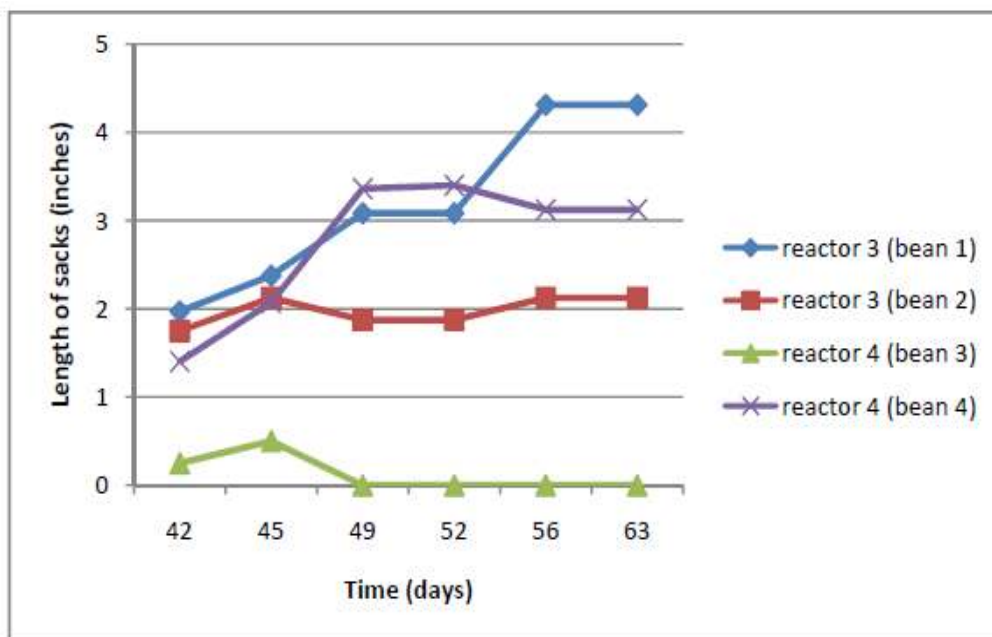


Figure 40. Length of bean sacks

Bean stalks were cut close to the roots after completion of the experiment. Then, two pumpkin seeds were planted in the reactors 3 and 4. In the reactor 4 which had a gravel layer as a physical barrier 5 inches below the TS, one seed did not germinate at all and the other died after a month of growth. However, pumpkins germinated in the reactor 3 have grown well (Figure 41).

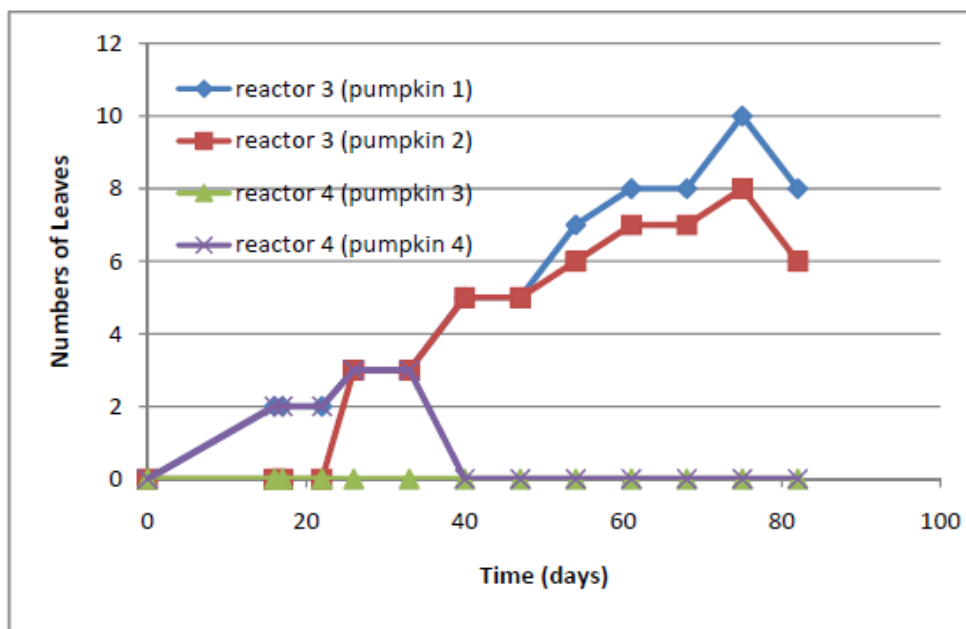


Figure 41. Number of pumpkin leaves

4.6 RHIZOSPHERE ENVIRONMENT

Figure 42 shows a trend of bean growth in terms of their height. Three bean seeds and 3 pumpkin seeds were planted in the pots. However, seeds did not germinate such that 3 additional beans were planted. The B4 bean in the control reactor was spoiled due to unknown reasons. A new bean was planted later time in the B4 spot. The B1, B3 and B6 beans in the CAAs reactor were damaged during the transport of the system to another location due to local electricity shut-down. This resulted in losses of the heights and leaves of the beans B1 and B3 as shown in Figure 43. No further efforts were provided to correct the damaged beans B1 and B3, but a new bean seed was planted to replace the dead B6 bean.

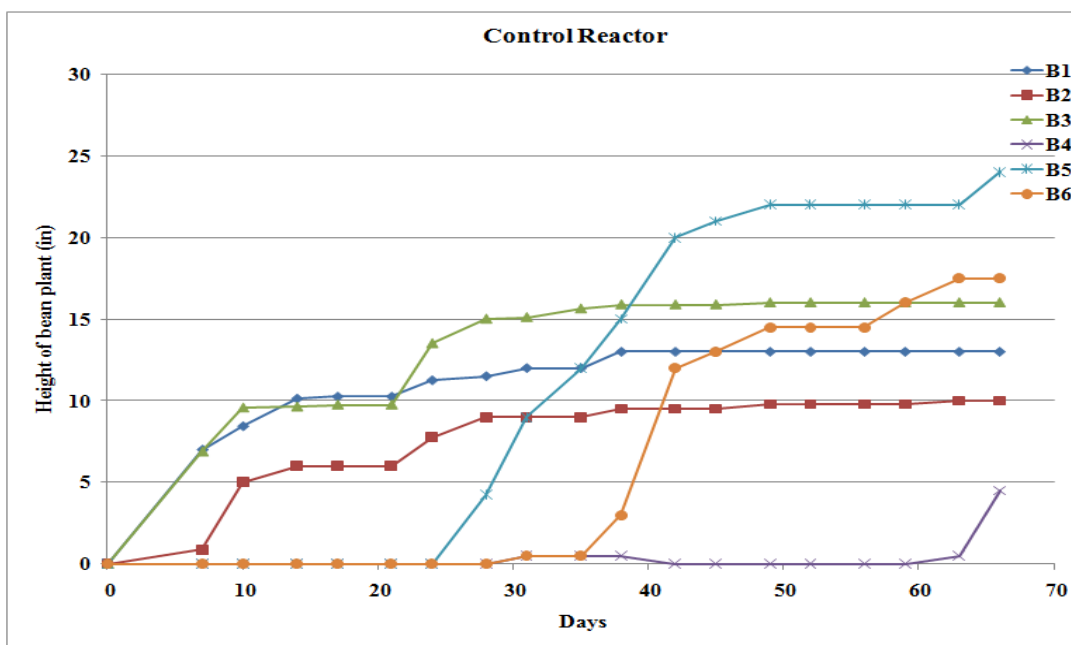


Figure 42. Height of bean from a study on plant growth and soil microbiology results
(Control reactor)

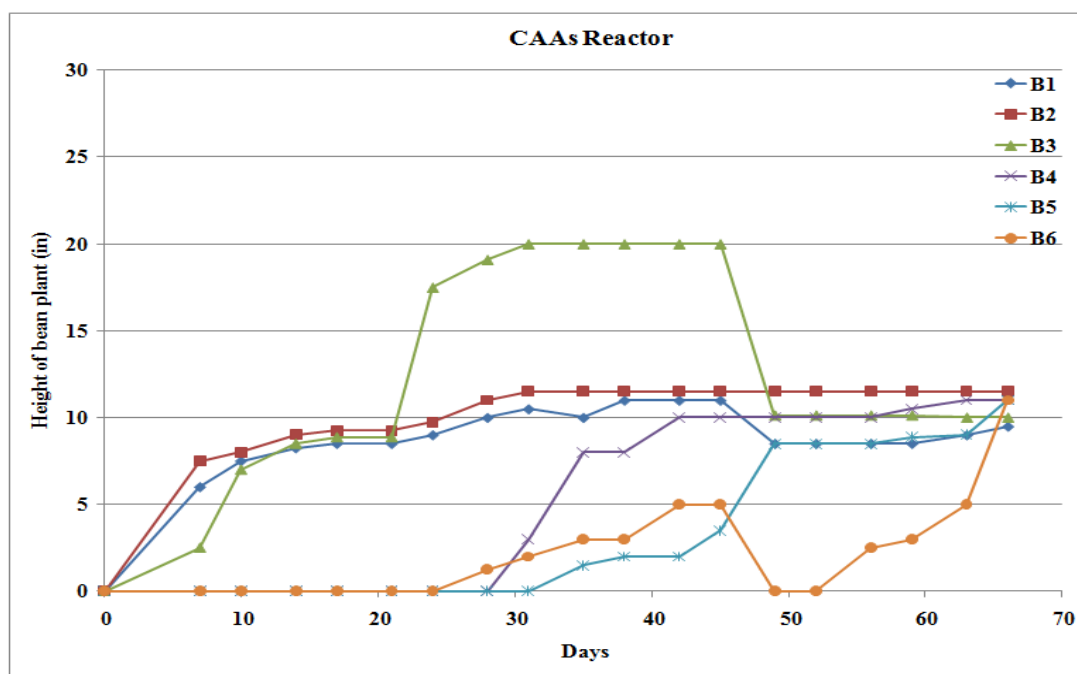


Figure 43. Height of bean from a study on plant growth and soil microbiology results
(CAAs reactor)

The number of leaves was also monitored. For the same reason that happened to the heights, leaves were also affected. Taking into consideration of the numbers of bean leaves only during the first stage of the experiment, the results of physical hindrance from the CAAs reactor were not much different from those from control reactor (Figures 44 and 45).

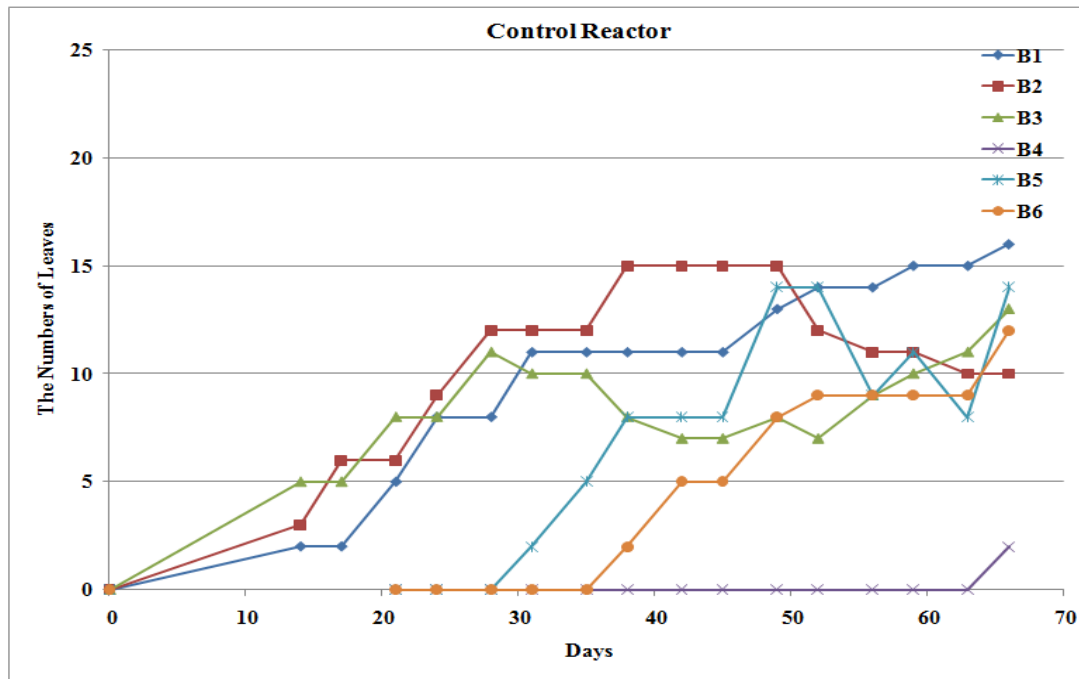


Figure 44. Number of bean leaves from a study on plant growth and soil microbiology (Control reactor)

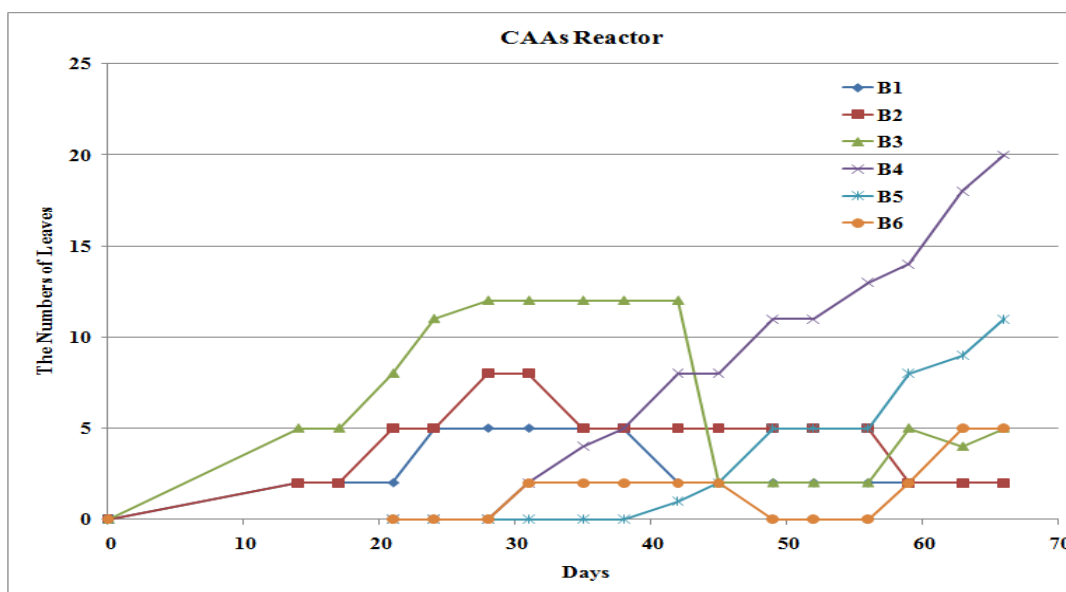


Figure 45. Number of beans leaves from a study on plant growth and soil microbiology (CAAs reactor)

Results from the chlorophyll intensity measurement (Figure 46-47) showed very similar trends between the control and CAAs reactor. The data point shown in Figure 45 was an average of chlorophyll intensity measure on the leaves. The value of zero chlorophyll intensity means no leaves available for the analysis at the respective time. An analysis showed a significant effect on age and FA on chlorophyll content. A decrease in chlorophyll contents was due to elevated levels of Cd, Cu, Zn in *B. vulgaris* plants treated with application of FA (Singh et al., 2008).

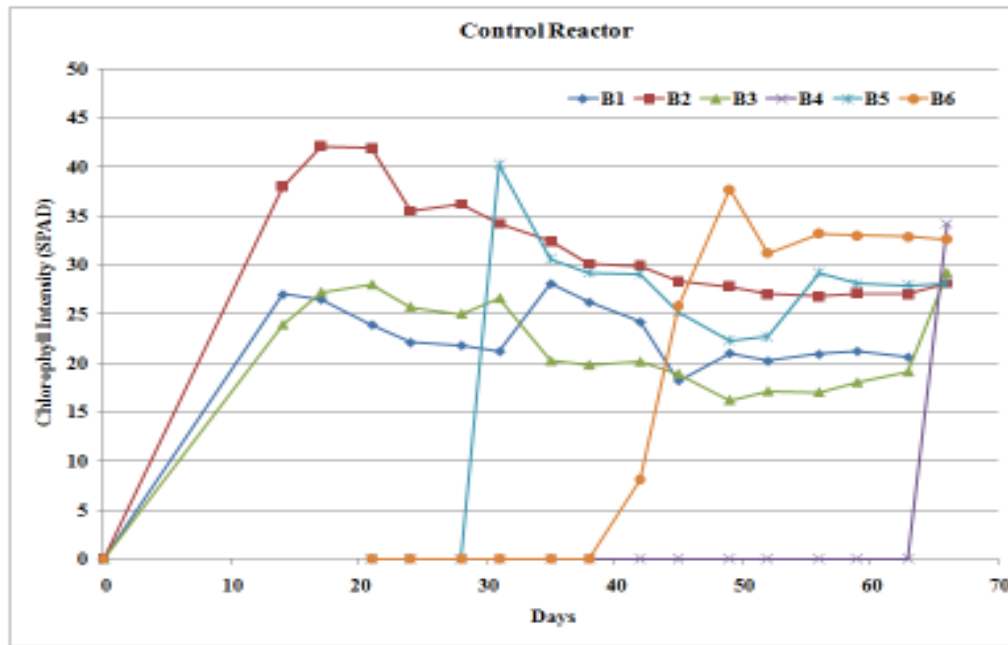


Figure 46. Chlorophyll intensity from a study on plant growth and soil microbiology (Control reactor)

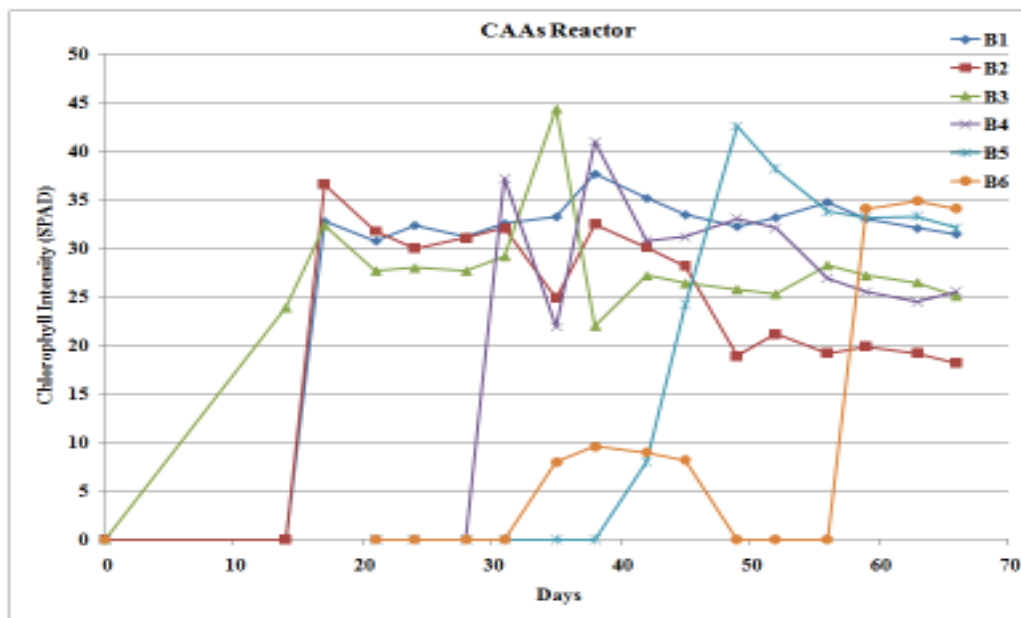


Figure 47. Chlorophyll intensity from a study on plant growth and soil microbiology (CAAs reactor)

Enzyme activities can be used to describe changes in soil microbial quality due to land use management and for understanding soil ecosystem functioning (Acosta et al., 2007). In this

case, the dehydrogenase activity was monitored to determine the enzyme activity in the rhizosphere. Figures 48 and 49 show the results of soil dehydrogenase analysis at 3 interval during the experiment. After 2 months of experimentation, less dehydrogenase activity was measured in CAAs reactor than in the control reactor, although the differences were not significant.

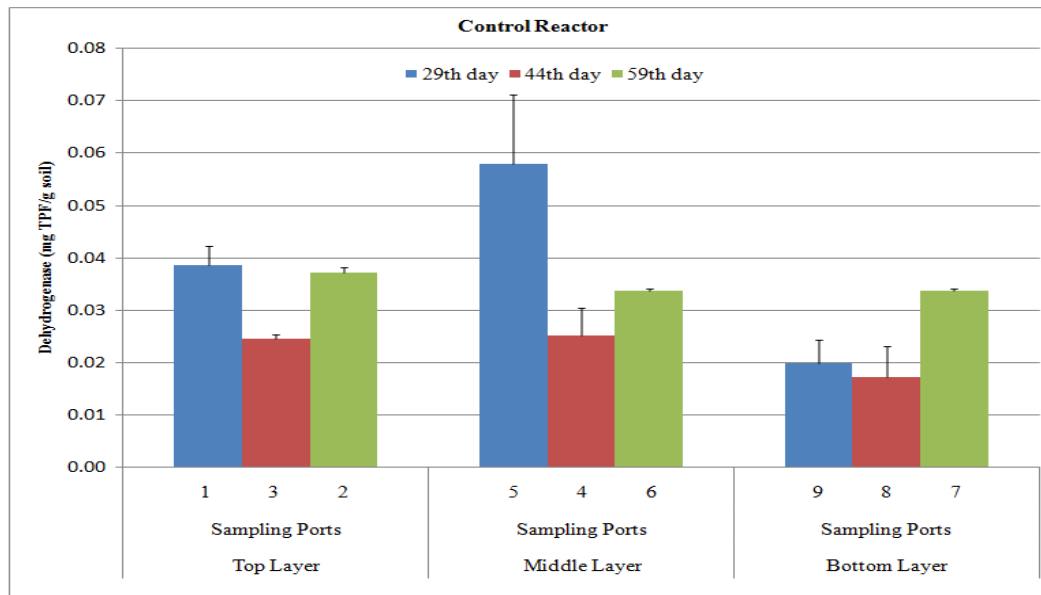


Figure 48. Dehydrogenase activity in control reactor

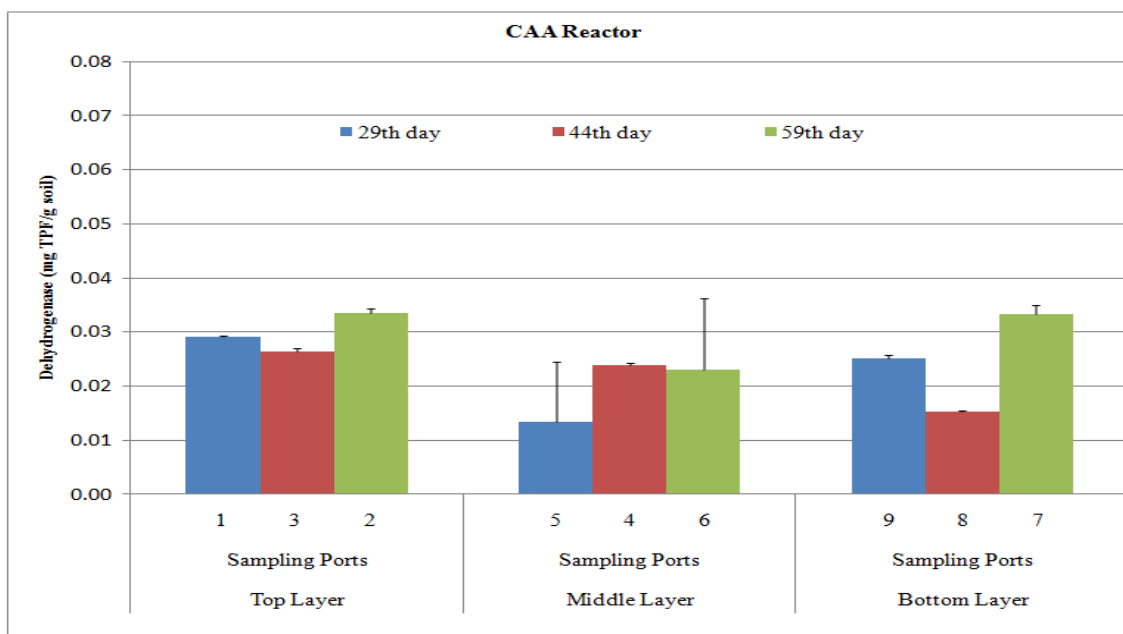


Figure 49. Dehydrogenase activity in CAAs reactor

Every two weeks, the total numbers of heterotrophic bacteria were counted in soils. The results are given in Table 16. It was shown that bacterial counts was higher in the control reactor than the CAAs reactor, although not much difference was found . This was in line with the soil dehydrogenase activity aforementioned. The results from a study by Garau (2007) reported that the only addition of red mudd was able to improve the number of heterotrophic bacteria in soil.

Table 16. Results of THB analysis

Layer	Sampling Pot	Day	CFU/g wet soil	
			Control Reactor	CCA Reactor
Top	1	29 th	TMTC	TMTC
	3	44 th	TMTC	TMTC
	2	59 th	3.08E+6	2.00E+06
Middle	5	29 th	TMTC	1.18E+07
	4	44 th	8.10E+06	5.10E+06
	6	59 th	3.20E+06	4.00E+06
Bottom	9	29 th	TMTC	TMTC
	8	44 th	TMTC	4.80E+06
	7	59 th	5.40E+06	3.20E+06

5 CONCLUSIONS AND RECCOMENDATIONS

In this project, an environmental restoration was made in conjunction with resource recovery and waste minimization. Industrial byproducts CAAs, which otherwise are regarded as wastes, were utilized as a backfilling amendment for an open pit restoration to bio-viable land. Accordingly, the physiochemical influence of the CAAs was assessed with respect to the seed germination and growth. Also, potential of the CAAs to enhance rhizospheric activities were evaluated. Based on the various feasibility experiments studied in this research, the following conclusions are made:

- Beans and pumpkins were germinated and grew when they were sprayed with the water directly infiltrated from the CAAs. The best results were observed when they seeded in the reactors with more top soil than the CAAs (2:1 ratio) in a layered configuration by tap water spraying.
- The hardness concentration in water positively affected the germination and growth of plants. It is construed that the CAAs would contribute the rhizosphere with elevated hardness concentration, which will enhance the growth of the plants.
- The layer of the CAAs underneath the top soil did not negatively impact the growth of the plants. Rather, it seemed provide a better growth environment for the plants, providing necessary chemical components to the roots which, in turn, used for the growth of the plants (e.g., the numbers of roots and leaves, the height of shoots, and chlorophyll intensity on the leaves).
- Rhizosphere activities with respect to soil bacteria counts and dehydrogenase concentrations were not significantly enhanced with the addition of the CAAs. However, the beans grew better in the system with the CAAs than in the control system without the CAAs.

Overall, the experimental results indicated that the CAAs could be utilized as a backfilling amendment for an open pit restoration to bio-viable land. They showed an enhanced germination and growth of the plants tested in this study (beans, pumpkins, papayas, and botellas). No negative impacts of the CAAs to the plant germination and growth were observed in the current study.

However, there were inherent limitations of the current lab-scale feasibility study. For example, the plants grew in an artificial environment indoors and small plants were tested. For better understanding of the disturbed land restoration to bio-viable land with the CAA amendment, the followings are recommended to conduct:

- Plant experiment should be set up in a natural environment subject to dynamic and varying weather conditions of rainfall, sunlight intensity, evaporation, etc.
- Additional research with different soils and plants should be conducted. Biological and physiochemical interactions of the CAAs and surrounding soil environment will produce dissimilar results on the phyto-viability. Deep-root plant should be tested in a larger scale experiment to accommodate the practical needs of open pit restoration to bio-viable land.

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