

## Original Research Article

### **Bioprospects of Tree Species in Bioremediation of Bauxite Residue**

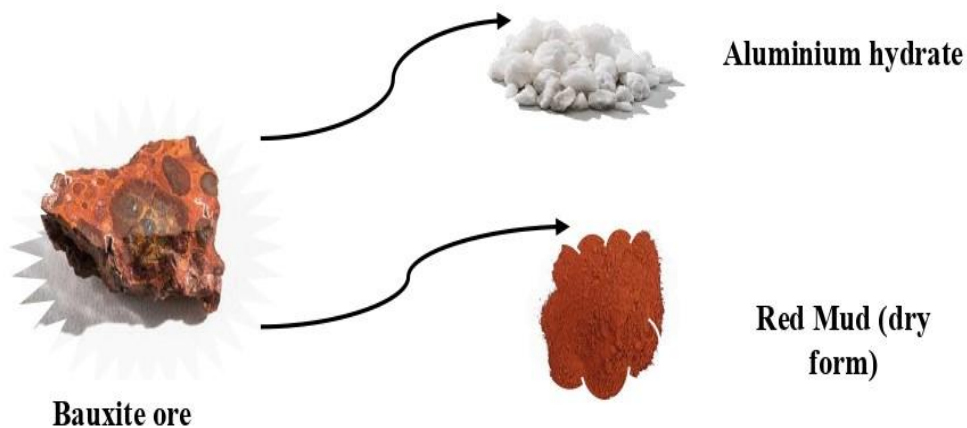
#### **Abstract**

Red mud or bauxite residue is a highly alkaline toxic by-product that is generated during the production of aluminium by Bayer's process. Around 1.5 ton of bauxite residues are produced for generating 1 ton of alumina. The toxicity of bauxite residue is a major catastrophe due to its disposal management as it limits the growth of plant species because of high alkalinity and salinity. In the present study, bioprospects carried through the cyanobacterial strains (viz. *Phormidium* and *Oscillatoria*) with planting of tree species have potentially shown successful outputs on the physical and chemical attributes of bauxite residue. The cyanobacterial strains were combined along with the microbial cultures of VAM (vesicular arbuscular mycorrhiza) and PSB (phosphate solubilising bacteria) to enhance root length, leaf area, fresh and dry weight of shoot and root, plant height, and chlorophyll content. The inoculated seeds of few plant species namely, *Siamese cassia*, *Acacia auriculiformis* A. Cunn. ex. Benth., *Dalbergia sissoo* Roxb. ex-DC., *Pithecellobium dulce* (Roxb.), and *Prosopis juliflora* var. *juliflora* were raised to study the effect of bioremediation of bauxite residue for revegetation. Further, we also found that *Siamese cassia*, *Acacia auriculiformis* A. Cunn. ex. Benth., *Dalbergia sissoo* Roxb. ex DC., *Pithecellobium dulce* (Roxb.) Benth., and *Prosopis juliflora* var. *juliflora* thrived well on the amended bauxite residue with other useful inoculums.

**Keywords:** Bio prospects, Bauxite residue, Bioremediation, Cyanobacteria, PSB, VAM

#### **Introduction**

Aluminium is the earth's third most abundant element obtained from the ore of bauxite through the rigorous Bayer's process which remains the most economical and feasible process to date (Habashi, 1995). It is one of the most used metals on a worldwide scale. At present scenario, nearly four metric tons of dry bauxite is required to generate two metric tons of alumina that in turn yields one metric ton of prime aluminum metal (Figure 1). It was found that around 390 million metric tons of bauxite were produced globally in 2021. In order to meet high demands of the growing population, the extraction of most widely used metal has also increased parallel to which a never ending source of by-product is being generated redundantly. This by-product is characterized as a highly alkaline (Novais et. al., 2018) and toxic material known as bauxite residue. Bauxite residue is known by several other names, including ferrosilt (Spitz & Trudinger, 2019), bauxaline (Taneez & Hurel, 2019), red oxide sand bauxsol (Lyu et al., 2021), Bayer process residue (Chao et al., 2022), bauxite tailings (Santini & Fey, 2015), cajunite (Gore, 2015) and alkaline waste product. As a result, the alumina industries faces a massive difficulty for its proper disposal which might be a challenging task for the entire world. The occurrence of ferric oxide gives bauxite residue a reddish rusty colour (Liu & Li, 2015). About one part of bauxite residue is eliminated as a waste formed after the manufacture of every part of alumina, produced by this traditional method since years (Pera et. al., 1997). The substantially expanding amount of bauxite residue in storage sites focuses on the need for appropriate remediation measures to regulate the environmental consequences of metallurgical processes and contribute to the industry's long-term profitability (Santini & Fey, 2015). Bauxite residue or red sludge has just a few uses in construction materials, land reclamation, and pavement so far (Piga et. al., 1993). According to Dauvin (2010); Varnavas and Achilleopoulos (1995), using huge areas for bauxite residue storage and disposal not only increases prices but it also raises the threat to the environment, including ground water pollution and dust in the nearby areas. Hence, bauxite residue must always be treated before discharge, since it has the potential to significantly damage the area within its reach. The handling of bauxite residue seems to be a global challenge addressing the alumina industries and regulatory agencies because of the huge volumes created and the detrimental repercussions ensuing from its disposal. This red sludge has already been amassing at such an unprecedented rate across the globe. Many options for improving bauxite residue management have been proposed but so far no cost-effective techniques have been applied and implemented. Because of its potentially dangerous toxic properties, bauxite residue poses a severe and worrisome environmental issue, presenting a major challenge to researchers in developing alternative techniques for bauxite residue utilization. The paper highlights the contemporary insight into the current prospects of bauxite residue characterization, management, different neutralizing techniques, and applications throughout the world and in India as well. Bioremediation is a methodology that utilizes biological processes (especially microorganisms) to mitigate or remove environmental impacts associated with toxic substances as well as other hazardous waste build up. Hence from the perspective of various organisms' microbial communities and phenomena taking place in relatable habitats, can be utilized to investigate the technical feasibility of a microbial-driven methodology towards bauxite residue bioremediation.



**Fig1.** Bauxite ore is converted into aluminium hydrate and red mud (by-product)

## **Materials and Methods**

### ***Study site, sample collection and analysis***

Red mud/ bauxite residue was collected from random dumping sites (which were dry stacked) from Hindalco company, as this is the only functioning alumina company at Renukoot, Sonbhadra district, Uttar Pradesh. (Latitude: 24.2207° N, Longitude: 83.0323° E). The site survey with the concerned authorities was concluded. Sample was collected from four different dumping sites and they were thoroughly mixed altogether (Dubey & Dubey, 2011, 2012). Heavy metal analysis of red mud was conducted thereafter. The samples were examined under the parameters such as pH, available nitrogen and organic matter, conductivity as characterized by Piper (1944).

### ***Culturing of cyanobacteria and mass propagation***

The cyanobacterial strains used for mass propagation are *Oscillatoria* sp., *Lyngbya* sp., *Phormidium* sp. and *Microcystis* sp. collectively. The medium utilized for their propagation is BG11. The strains can later be used as an inoculum source (Rippka et al., 1979).

### ***Method of preparation of bulk propagation***

Approximately 10 kg of farm soil (gathered from the open field for 1.0 metre square area of the tank) and 100 g of superphosphate (SSP) were introduced and evenly mixed. The tank was levelled up to the height with 10 cm of water and it was maintained throughout. By adding lime to the tank, the pH level (7.0) was maintained. In order to prevent the breeding of mosquitoes and other pests in the culture tank, pesticide malathion (2.0 ml) was also introduced. After stirring the prepared mixture, it was allowed to settle into the soil. After the water started to become clear, 100 g of starting inoculums was strewn on the bottom over the period of time. To ensure that cyanobacteria could grow to their maximum potential, the temperature was regulated between 35 and 40° C (the required range for summer). Throughout the experiment, the water level was held steady at about 10 cm. The algal mat and the soil which had parted shortly after drying, had produced flakes (the area and species used for propagation influence the production). These algal flakes can be further utilized as a starting inoculum for further propagation.

### ***Effect of cyanobacterial strains on amended bauxite residue with various treatments***

Examining the impact of cyanobacteria (blue green algae), including *Lyngbya*, *Phormidium*, *Oscillatoria*, and *Microcystis* species, on bauxite residue amendments with varied treatments under nursery circumstances were performed using trays in order to select potential *cyanobacteria* for further studies. Different bio-amendments were investigated for their impacts on bauxite residue. Study was conducted on cyanobacterial growth development on bauxite residue with various amendments. The interventions were Control 1 (bauxite residue), Control 2 (bauxite residue: normal soil (1:1) amended with 10 g of bone meal). Bone meal is an organic source of phosphorous.

#### ***Effect of cyanobacterial strains on selected plant species (Growth performance)***

The investigation was carried to study how well blue green algae (BGA)/ bio-inoculants when combined together, promote the growth of various plant species. In order to study the effects of integrating BGA with bio-amendments on the growth performance of selected plant species, which include *Siamese cassia*, *Acacia auriculiformis* A. Cunn. ex. Benth., *Dalbergia sissoo* Roxb. ex DC., *Pithecellobium dulce* (Roxb.) Benth., and *Prosopis juliflora* var. *juliflora* on bauxite residue, an experiment in nursery pots had been established (Dubey & Dubey, 2011; Dubey, 2012). The Sonbhadra district in Uttar Pradesh, India, where the Hindalco company is located, is home to these selected tree species.

Combination of potential BGA species i.e. *Phormidium* and *Oscillatoria* was used for cyanobacterial inoculation in 1:1 proportion. Treatments carried out here were Control 1 (bauxite residue: Normal soil), Control 2 (normal soil: normal sand: farm yard manure (FYM) without bauxite residue), Treatment 1 (bauxite residue: normal soil amended with 10 g of each bone meal, FYM and *cyanobacteria*), Treatment 2 (bauxite residue: normal soil amended with 10 g bone meal, FYM and *cyanobacteria* and PSB), Treatment 3 (bauxite residue: normal soil amended with 10 g bone meal, FYM, *cyanobacteria* and VAM). Control 2 was a positive control for the trial. *Siamese cassia*, *Acacia auriculiformis* A. Cunn. ex. Benth., *Dalbergia sissoo* Roxb. ex DC., *Pithecellobium dulce* (Roxb.) Benth., and *Prosopis juliflora* var. *juliflora* were the tree species whose seeds were sown, and their growth was periodically monitored for six months.

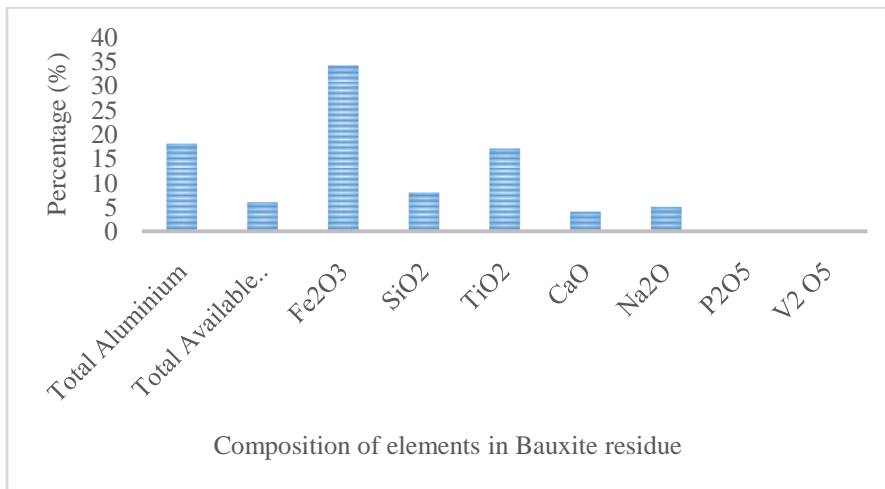
#### ***Comparative study for selecting efficient plant species on amended treatments***

A comparative data was analysed on the basis of growth pattern i.e., height parameter of the selected plant species with various bio treatments. As far as the growth was concerned, the effect of both cyanobacterial and bio amendments had shown positive effects on all the selected plant species.

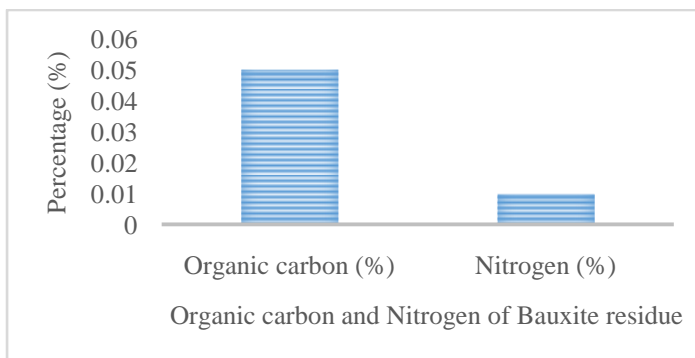
### **Results and Discussion**

The mineralogical composition of bauxite residue was provided by the Hindalco Company as illustrated in figure 2. Following that, the bauxite residue sample was evaluated for pH, EC, organic carbon, and nitrogen, as shown in figures 3 and 4, respectively. Further in the experiment cyanobacterial strain growth was observed after 45 days, based on this, the physico-chemical characteristics of the cyanobacteria strains in bauxite residue amended medium were observed. The two prominent species (*Phormidium* and *Oscillatoria* species) that exhibited effective results were selected for bioremediation. According to the aforementioned experiment, *Phormidium* and *Oscillatoria* species outperformed *Lyngbya* and *Microcystis* species on bauxite residue-amended medium on various parameters such as pH, EC, organic carbon and nitrogen as depicted in figure 5. According to El-Sheekh & Mahmoud (2017) and Kaur et al. (2019), *Phormidium* and *Oscillatoria* species can thrive within alkaline conditions and can also acclimatize to the alkalinity of bauxite residue. These two cyanobacterial species can therefore be utilized to bioremediate bauxite residue (Dubey, 2012; Dubey & Dubey, 2011). Additionally, the effect of selected cyanobacterial species (*Phormidium* and *Oscillatoria*) collectively was employed to observe the growth performance of selected plant species. The species selected for the trial viz., *Siamese cassia*, *Acacia auriculiformis* A. Cunn. ex. Benth., *Dalbergia sissoo* Roxb. ex DC., *Pithecellobium dulce* (Roxb.) Benth., and *Prosopis juliflora* var. *juliflora* were inoculated with *Phormidium* and *Oscillatoria* species, PSB, and VAM along with amended bauxite residue to re-vegetated these plant species. The observation on growth (height in meter) after six months is represented in figure 11. As far as the growth was concerned the effect of cyanobacterial strains and bioamendments was found to have positive effect in comparison to control 1 and it was significantly at par with the control 2 in case of *Prosopis juliflora* var. *juliflora*, *Pithecellobium dulce* (Roxb.) Benth., *Siamese cassia*, *Acacia auriculiformis* A. Cunn. ex. Benth. Control 2 was considered as positive control without red mud. Hence in case of *Dalbergia sissoo* Roxb. ex DC the growth with treatments was significantly less in comparison to control 2. As the comparative study is concerned the growth performance was noted in the selected plant species, in decreasing order: *Prosopis juliflora* var. *juliflora* > *Pithecellobium dulce* (Roxb.) Benth > *Siamese cassia* > *Acacia auriculiformis* A. Cunn. ex. Benth > *Dalbergia sissoo* Roxb. ex DC. Treatments with the amendments of Cyanobacteria *Phormidium* & *Oscillatoria* with PSB & VAM were at par and performed well over other treatments. The above mentioned findings contribute strong support to the idea that *cyanobacteria* may play a crucial role in the detoxification of bauxite residue by fixing heavy metals, in the formation of a resistant layer on bauxite residue dumping sites, in

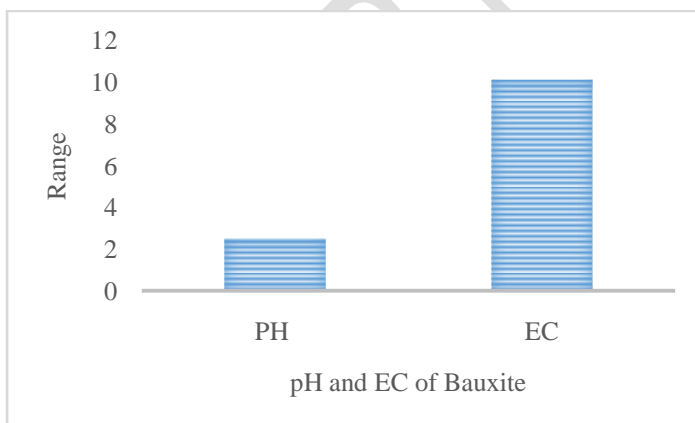
limiting the ecological threat posed by water and wind erosion, and in enhancing the nutrient status of the mud in a manner that is ecologically sound.



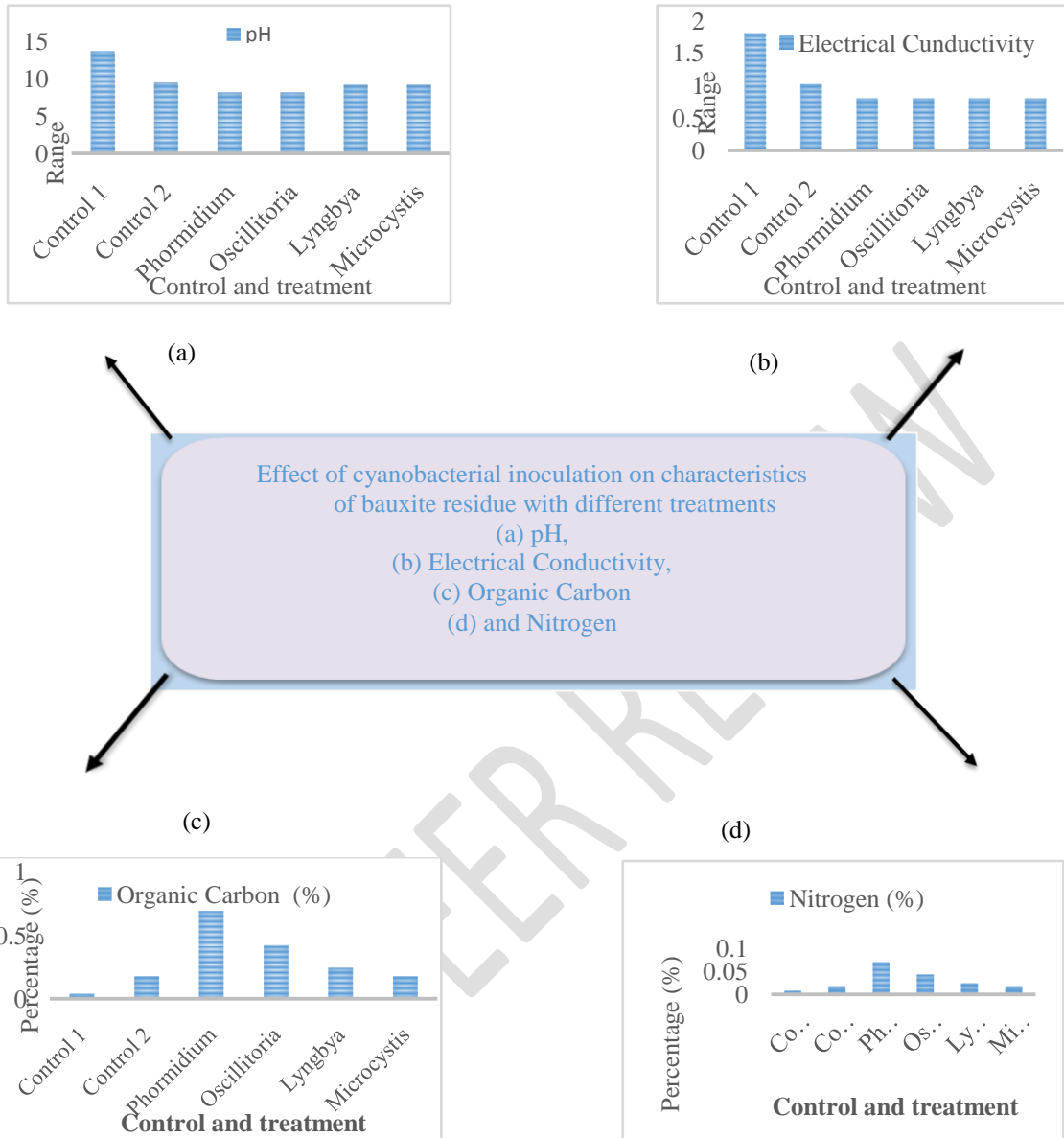
**Fig2.** Mineralogical composition of bauxite residue



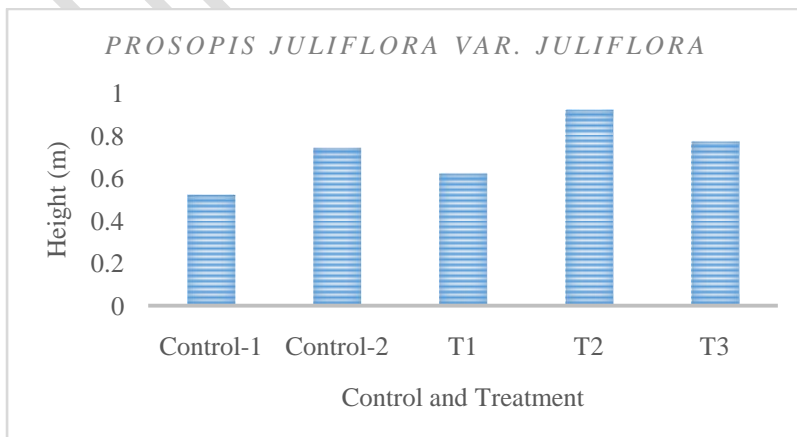
**Fig3.** Organic carbon and Nitrogen present in the bauxite residue sample



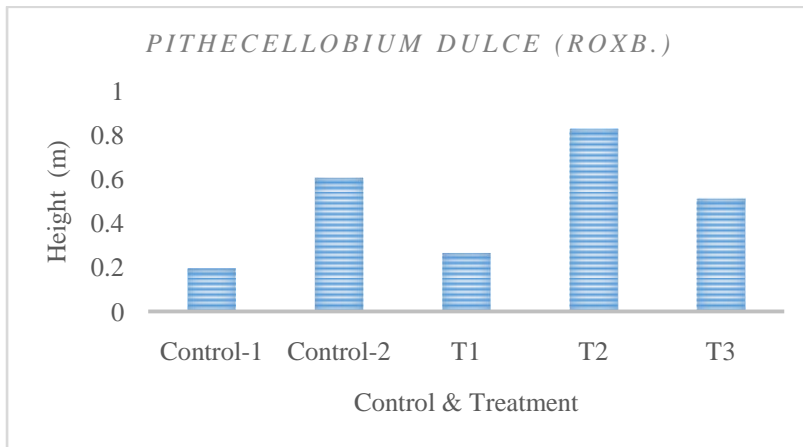
**Fig 4.** pH and EC of bauxite residue sample



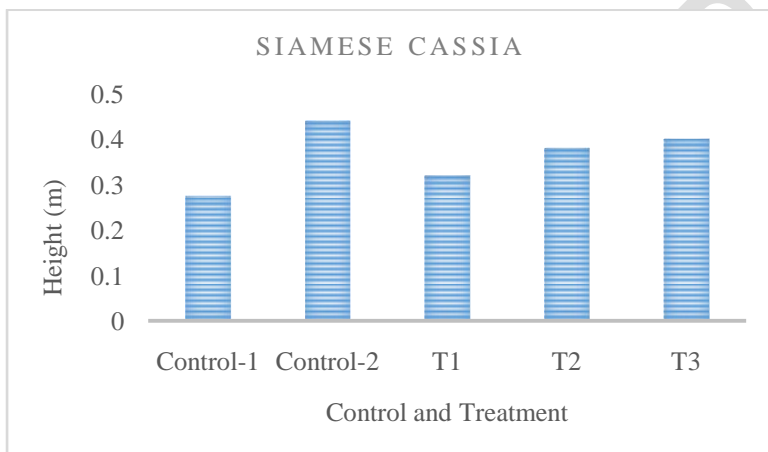
**Figure 5:** Effect of cyanobacterial inoculation on characteristics of bauxite residue with different treatments on parameters such as (a) pH, (b) Electrical Conductivity (EC), (c) Nitrogen, (d) Organic Carbon



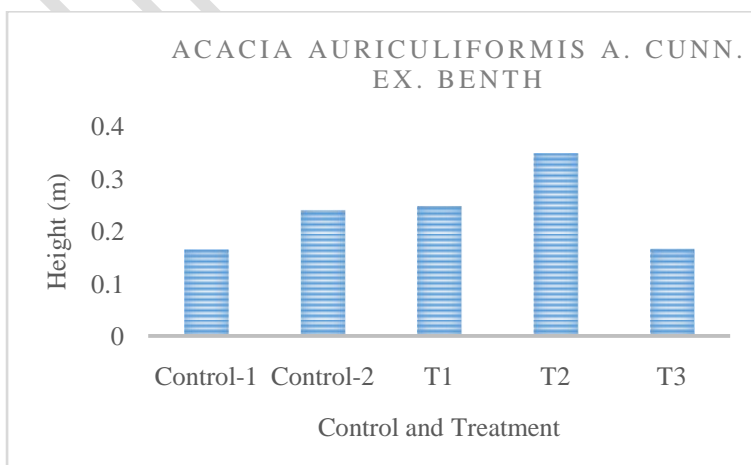
**Figure 6:** Growth effect on *Prosopis juliflora* var. *juliflora* after treating (T1- bauxite residue: normal soil amended with 10 g of each bone meal, FYM and *cyanobacteria*, T2- bauxite residue: normal soil amended with 10 g bone meal, FYM and *cyanobacteria* and PSB , T3-bauxite residue: normal soil amended with 10 g bone meal, FYM, *cyanobacteria* and VAM) with amended bauxite residue.



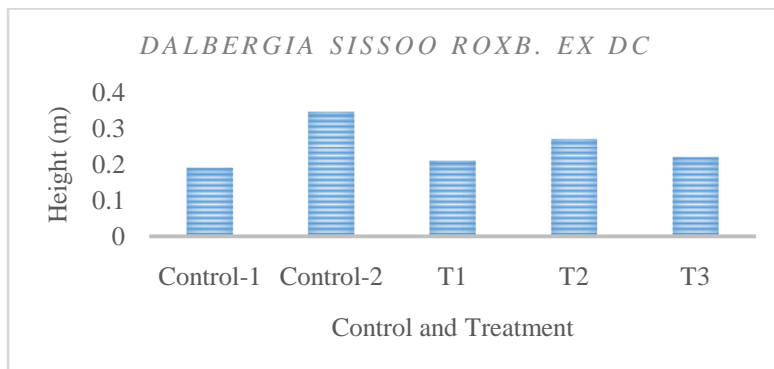
**Figure 7:** Growth effect on *Pithecellobium dulce* (Roxb.) after treating (T1- bauxite residue: normal soil amended with 10 g of each bone meal, FYM and *cyanobacteria*, T2- bauxite residue: normal soil amended with 10 g bone meal, FYM and *cyanobacteria* and PSB , T3-bauxite residue: normal soil amended with 10 g bone meal, FYM, *cyanobacteria* and VAM) with amended bauxite residue.



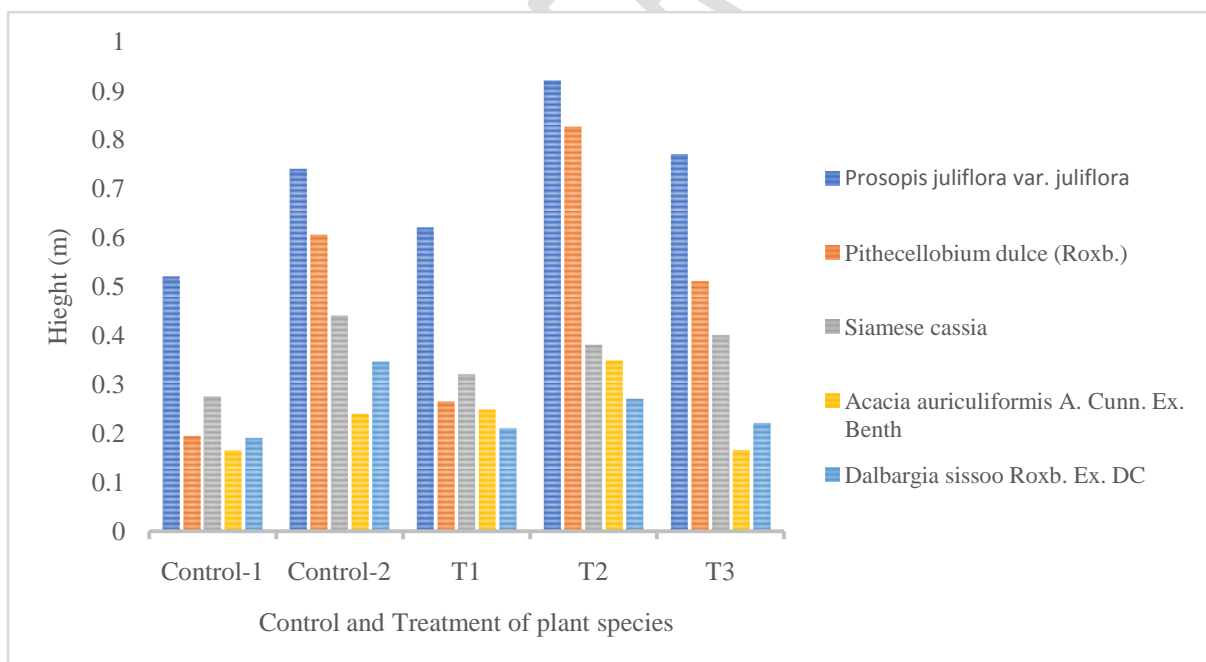
**Figure 8:** Growth effect on *Siamese cassia* after treating (T1- bauxite residue: normal soil amended with 10 g of each bone meal, FYM and *cyanobacteria*, T2- bauxite residue: normal soil amended with 10 g bone meal, FYM and *cyanobacteria* and PSB , T3-bauxite residue: normal soil amended with 10 g bone meal, FYM, *cyanobacteria* and VAM) with amended bauxite residue.



**Figure 9:** Growth effect on *Acacia auriculiformis* A. Cunn. ex. Benth after treating (T1- bauxite residue: normal soil amended with 10 g of each bone meal, FYM and cyanobacteria, T2- bauxite residue: normal soil amended with 10 g bone meal, FYM and cyanobacteria and PSB , T3-bauxite residue: normal soil amended with 10 g bone meal, FYM, cyanobacteria and VAM) with amended bauxite residue.



**Figure 10:** Growth effect on *Dalbergia sissoo* Roxb. ex DC after treating (T1- bauxite residue: normal soil amended with 10 g of each bone meal, FYM and cyanobacteria, T2- bauxite residue: normal soil amended with 10 g bone meal, FYM and cyanobacteria and PSB , T3-bauxite residue: normal soil amended with 10 g bone meal, FYM, cyanobacteria and VAM) with amended bauxite residue.



**Figure 11:** Comparative growth performance (height in meter) of the selected plant species *Prosopis juliflora* var. *juliflora*, *Acacia ariculiformis*, *Dalbergia sissoo* Roxb. ex DC, *Siamese cassia* and *Pithecellobium dulce* (Roxb.) with different bio treatments

### Conclusion

The performance of selected tree species viz. *Dalbergia sissoo* Roxb ex DC, *Prosopis juliflora* var. *juliflora*, *Acacia auriculiformis*, *Pithecellobium dulce* (Roxb.) and *Siamese cassia* inoculated with cyanobacteria with phosphor-bacteria and VAM inoculation was studied and it was observed that *Prosopis juliflora* var. *juliflora* and *Pithecellobium dulce* (Roxb.) performed well over other remaining species. *Acacia auriculiformis* A. Cunn.

*ex. Benth* and *Siamese cassia* also performed well. Therefore, from the above study, it may be concluded that these four species with *Cyanobacteria* viz. *Phormidium* and *Oscillatoria* species in combinations with PSB and VAM inoculation may be used for re-vegetating the red mud dumping site. Inoculated seedlings of *Prosopis juliflora* var. *juliflora*, *Pithecellobium dulce* (Roxb.), *Acacia auriculiformis* A. Cunn. *ex. Benth* and *Siamese cassia* performed well on red mud. These plant species with above discussed cyanobacterial strains may be used for re-vegetating the red mud site. It was expected that the findings of the proposed study will provide suitable, eco-friendly and cost-effective bioremediation methods for bauxite residue.

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