

INTERACTIVE EFFECT OF SILICATE SOLUBILIZING BACTERIUM *Pseudomonas baetica* AND FLY ASH APPLICATION ON WHEAT CROP

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ABSTRACT

Fly ash, a byproduct of thermal power plants, is a rich source of essential nutrients, including silicon, which plays a key role in plant growth, stress tolerance, and overall physiology. Silicon is typically found in fly ash in the form of water-insoluble silicates, which can be made bioavailable to plants through the action of silicate-solubilizing bacteria. This study investigates the synergistic effects of silicate-solubilizing bacterial inoculation and fly ash amendment on wheat (*Triticum aestivum*) growth under controlled pot-house conditions. The results demonstrate that soil amendment with up to 3% fly ash, in conjunction with bacterial inoculation, supported the survival of the bacteria and significantly enhanced wheat growth and yield. These findings suggest that the combination of fly ash and silicate-solubilizing bacteria could offer a sustainable approach to improving crop productivity.

Keywords: Fly ash, silicate solubilizing bacteria, silicon, wheat

The demand and consumption of electricity are steadily increasing, driven by population growth and urbanization. In India, coal accounted for 53.37% of the total non-renewable sources used to produce 4,11,649 megawatts of electricity in 2023, while renewable energy sources, including hydro and nuclear, contributed only 14% of the energy mix (Ministry of Power, GoI, 2023). The combustion of coal in thermal power plants generates a fine powdered residue, known as fly ash, with particle sizes ranging from 0.5 to 300 microns (Bhatt *et al.*, 2019). This byproduct is produced in vast quantities worldwide and requires extensive ash ponds for disposal. If improperly managed, fly ash can escape from these ponds and, once released into the atmosphere and water bodies, poses a significant environmental threat, as well as a risk to human and animal health. Fly ash is commonly utilized in various industries, particularly for its pozzolanic properties, which enhance the durability of concrete, roads, bricks, and even for mine reclamation (Gorai, 2018). However, using fly ash in agriculture may represent one of the most sustainable disposal options. When applied to agricultural land, fly ash has been shown to increase soil pH, improve aeration and percolation, enhance soil conditioning, and provide a better supply of micronutrients for crops (Basu *et al.*, 2009; Kishor *et al.*, 2010; Usmani *et al.*, 2019). Additionally, coal fly ash has been used successfully in the reclamation of land rendered infertile by mining activities (Tsadilas *et al.*, 2018).

Fly ash can serve as a cost-effective carrier for

biofertilizer formulations when compared to liquid or other solid carrier materials. Utilizing fly ash as a carrier for biofertilizers, such as *Azotobacter*, not only reduces production costs but also provides an effective alternative for fly ash disposal (Parab *et al.*, 2013). Furthermore, the application of fly ash in rice cultivation has been shown to mitigate soil organic carbon loss. When combined with organic nitrogen fertilizers, such as manure or vetch, fly ash can reduce the reliance on chemical fertilizers in agriculture (Lim *et al.*, 2017). Additionally, soil amendments with up to 30% fly ash have proven effective in controlling root-knot nematodes (*Meloidogyne incognita*) in carrot crops (*Daucus carota* L.) (Haris *et al.*, 2019).

Silicates are compounds that contain silicon in forms such as silicon dioxide and aluminosilicates, which typically have low bioavailability of silicon. The bioavailable form of silica for plants is monosilicic acid, which, when absorbed by plant roots through its aqueous solution, serves as the primary source of silicon. The dissolution of silicate compounds into monosilicic acid occurs through microbial acid production and the chelating action on metals associated with silicates. The dissolution rate is significantly higher when silicate-solubilizing bacteria (SSB) are introduced into a medium containing silicates (Santi & Goenadi, 2017). The role of silicon in enhancing microbial, soil, and plant enzymatic activities has also been studied, with a positive correlation observed between silicon application and enzymatic activity levels (Pereira *et al.*, 2018; Abdelaal *et al.*, 2020; Alam *et al.*, 2021). Increased enzymatic activity is crucial for improved plant growth, development, and tolerance to heavy metal toxicity. This activity is a key factor in nutrient transport and cell

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Date of receipt: 01.08.2023, Date of acceptance: 18.07.2024

differentiation in plants. Furthermore, rock weathering and mineral solubilization by SSB contribute to the increased availability of bioavailable potassium in the soil, which can further enhance plant health and productivity (Rasouli-Sadaghiani *et al.*, 2017).

The concept of a circular economy is a crucial element of sustainable development, where waste products generated in one process are repurposed as substrates in other processes. The coinoculation of fly ash, a byproduct of energy production, with silicate-solubilizing bacteria (SSB) in agricultural crops offers a promising approach to improve crop growth and yield, while simultaneously providing an effective strategy for fly ash management. The present study was conducted to evaluate the impact of various concentrations of fly ash on the survival of SSB and to identify the optimal concentration of fly ash for enhancing the growth and yield characteristics of wheat.

MATERIAL AND METHODS

Isolation of silicate solubilizing bacteria and characterization of fly ash

Silicate-solubilizing bacterial isolates were obtained from the rhizospheric soil of paddy plants collected from the farms of CCS Haryana Agricultural University (CCSHAU) in Hisar. The isolates were identified based on their ability to form zones on Bunt and Rovira agar medium (Bunt and Rovira, 1955). The fly ash used in this study was collected from the Rajiv Gandhi Thermal Power Plant in Khedar (latitude: 29.34°N, longitude: 75.85°E), located in Barwala, Hisar. The fly ash was analyzed for its nitrogen (N) content (Subbiah and Asija, 1956), potassium (K) content (Pratt, 1965), and phosphorus (P) content (John, 1970).

Effect of fly ash on survival of silicate solubilizing bacterial isolate in soil

The effect of fly ash on the survival of the promising silicate-solubilizing bacterial isolate *Pseudomonas*

baetica was investigated by inoculating an active bacterial culture into sterile soil amended with 0.5–5% fly ash. Samples were withdrawn at various time intervals post-inoculation, plated on nutrient agar medium, and incubated at 30°C. Viable bacterial counts were recorded at different time points to assess bacterial survival.

Effect of fly ash and SSB application on wheat crop under pot culture conditions

Earthen pots containing five kilograms of air-dried soil were used for the pot house study. A total of 14 treatments, including the control, recommended dose of fertilizer (RDF), and 75% RDF, were applied in triplicate (Table 1). Wheat seeds of the variety WH-1105 were surface sterilized, treated with a silicate-solubilizing bacterial culture (10^8 cells/mL), and sown at a depth of 4–5 cm. After thinning, three healthy plants were maintained per pot.

Height of plants was measured on 60, 90 and at 120 days after sowing. Data on seed weight, number of seeds and plant biomass were taken as growth and yield attributes of the crop.

Statistical analysis

The data obtained from the various experiments were statistically analyzed using OPSTAT software (<http://14.139.232.166/opstat/>)

RESULTS AND DISCUSSION

Silicate-solubilizing bacterial isolates were retrieved on Bunt and Rovira agar medium by evaluating their ability to solubilize insoluble silicate (0.25% magnesium trisilicate) through zone formation, following 72 hours of incubation at 30°C. A total of forty silicate-solubilizing bacterial isolates (SSB1-SSB40) were obtained. Among these, the isolate SSB2, identified as *Pseudomonas baetica*, was found to be the most promising, exhibiting the highest silicate solubilization index (2.67) of all the

Table 1. Treatments for pot house experiment

Treatments	Description	Treatments	Description
T ₁	Soil (control)	T ₈	FA (4%)
T ₂	RDF	T ₉	FA (5%)
T ₃	75% RDF	T ₁₀	FA (1%)+SSB
T ₄	SSB	T ₁₁	FA (2%)+SSB
T ₅	FA (1%)	T ₁₂	FA (3%)+SSB
T ₆	FA (2%)	T ₁₃	FA (4%)+SSB
T ₇	FA (3%)	T ₁₄	FA (5%)+SSB

RDF: Recommended dose of fertilizers

SSB : Silicate solubilizing bacterial culture *Pseudomonas baetica*

FA: Fly ash

isolates.

The fly ash used in this study was grey in colour, with an alkaline pH of 8.38, and contained 0.05%, 0.13%, and 0.06% nitrogen (N), potassium (K), and phosphorus (P), respectively. The colour of fly ash can vary from light tan to dark grey, and its pH typically ranges from 2 to 12. The colour of fly ash is influenced by the iron and lime content, while the amount of sulfur in the parent coal determines the pH of the residual fly ash after combustion (Bhatt *et al.*, 2019). Fly ash is composed of approximately 15-60% SiO₂, 5-35% oxides of aluminum and iron, and contains oxides of calcium, magnesium, and other elements in concentrations of 2-10% by weight.

Effect of fly ash on survival of silicate solubilizing bacterial culture

The survival of microorganisms in an environment is influenced by various factors, including the availability of bioavailable nutrients, temperature, pH, and both abiotic and biotic stresses. Under adverse conditions, when microbial existence is threatened, adaptation and strain variations are key strategies for survival. The amendment of fly ash to soil alters its structural, nutrient, and mineral composition, which can either support or hinder the survival of the microbial population.

The survival of the silicate-solubilizing bacterial culture *Pseudomonas baetica* at different fly ash concentrations was assessed in terms of colony-forming units (cfu) at various incubation times on nutrient agar medium. The survival of *P. baetica* was supported by fly ash concentrations up to 3% for 72 hours, after which it was inhibited (Fig. 1). The maximum Log cfu mL⁻¹ (7.82) was observed in soil amended with 3% fly ash after 72 hours. The presence of silicate-solubilizing bacteria (SSB) in diverse environments has been reported by several researchers. Hu *et al.* (2018) isolated SSB from the gut of earthworms (*Pheretima guillelmi*) and soil samples, noting low cfu of SSB retrieved from soil

samples. Singh *et al.* (2016) suggested that fly ash can have a beneficial effect on microbial populations and enzymatic activity. Their investigation reported the highest microbial biomass, organic carbon, and bacterial populations in soil amended with 20 t/ha of fly ash used for cultivating *Vigna radiata* L. Fly ash significantly enhanced microbial populations when organic materials such as kitchen waste or jaggery were added to the culture medium (Mandpe *et al.*, 2019). Pichtel and Hayes (1990) studied the effect of fly ash on soil-inhabiting microorganisms, stating that the concentration of fly ash in soil is a determining factor for microbial survival and enzymatic activity. They observed a decrease in viable counts of bacteria, fungi, and actinomycetes when 20% fly ash was added to the soil.

Effect of fly ash on growth and yield parameters of wheat crop

The height of wheat plants at 60, 90, and 120 days after sowing was measured, and the results are presented in Table 2. Application of fly ash, both independently and in combination with silicate-solubilizing bacterial culture, resulted in increased plant height at 2% and 3% amendment rates. The maximum plant height at 120 days was recorded in treatment 12 (3% fly ash + SSB), followed by treatment 11 (2% fly ash + SSB 2). Plants in treatment 6 (2% fly ash) exhibited the tallest growth among treatments that contained only fly ash, without SSB. A significant increase in plant height was observed in treatment 12 (3% fly ash + SSB) after 120 days of sowing, compared to the control treatment. Pani *et al.* (2015) studied the effect of fly ash concentration on the plant height of sunflower, *Helianthus annuus* L. at different time intervals. In their study, the plant height in the treatment with 25% fly ash was 25.36 cm after 30 days of sowing, compared to 20.2 cm in the control treatment. Detrimental effects of fly ash were observed at higher concentrations (50%, 75%, and 100%). A similar trend was observed at 60 and 90 days after sowing. The findings of Shakeel *et al.* (2019) also supported the use of fly ash amendment (up to 30%) for promoting better growth, as evidenced by increased dry shoot and root weight in Indian mustard, *Brassica juncea*.

Plant biomass, number of seeds, and seed weight were measured as yield attributes of the crop. The dry biomass of plants ranged from 1.77 to 5.00 g per pot. Inoculation with silicate-solubilizing bacteria (SSB) and the amendment of soil with 3% fly ash resulted in significantly higher plant biomass production, which was 2.82 times greater than the control. Higher concentrations of fly ash had a negative impact on plant biomass production (Table 3). Similarly, Pani *et al.* (2015) reported that in treatments with a soil-to-fly

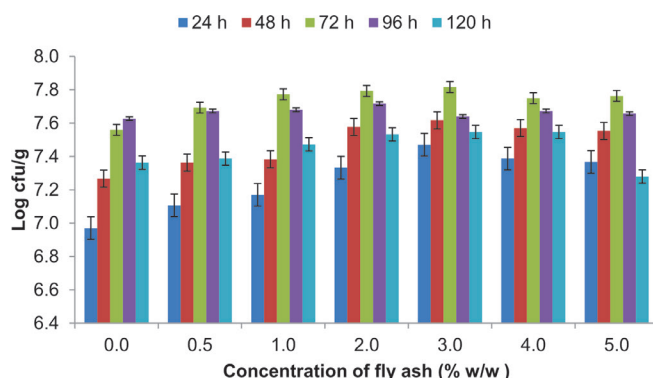


Fig. 1. Survival of silicate solubilizing bacterial culture in soil amended with different concentrations of fly ash

Table 2. Effect of fly ash application along with silicate solubilizing bacterial culture on plant height of wheat

Treatments	Plant height (cm) after		
	60 Days	90 Days	120 Days
T ₁ : Soil (control)	21.91	38.44	40.74
T ₂ : RDF	40.03	51.22	52.14
T ₃ : 75% RDF	36.50	46.56	47.86
T ₄ : SSB2	26.30	42.78	44.10
T ₅ : FA (1%)	23.31	39.67	43.10
T ₆ : FA (2%)	24.17	40.67	48.00
T ₇ : FA (3%)	24.44	35.89	46.99
T ₈ : FA (4%)	32.77	42.78	45.91
T ₉ : FA (5%)	24.48	38.33	42.68
T ₁₀ : FA (1%) + SSB	32.53	46.11	48.07
T ₁₁ : FA (2%) + SSB	37.19	48.33	49.37
T ₁₂ : FA (3%) + SSB	35.59	49.53	53.11
T ₁₃ : FA (4%) + SSB	32.74	47.56	48.62
T ₁₄ : FA (5%) + SSB	33.32	45.22	47.71
CD ($p \leq 0.05$)	4.57	3.81	2.52

RDF: Recommended dose of fertilizers

SSB : Silicate solubilizing bacterial culture *Pseudomonas baetica*

FA: Fly ash

ash ratio of 3:1, the dry root and shoot weights were 1.2 and 1.11 times greater, respectively, than those of the control. However, higher concentrations of fly ash exhibited an inhibitory effect. In line with this, Singh *et al.* (2015) found that when soil was amended with 50% fly ash, the dry root and shoot weights were reduced by 71% and 54%, respectively, compared to the control.

The number of seeds per pot varied between 37.67 and 146.37, while seed weight ranged from 0.63 to 3.50 g per pot. The highest number of seeds was recorded in treatment 12 (3% fly ash + SSB). A continuous increase in the number of seeds was observed in treatments until the fly ash concentration reached 3%. The amendment of 2% fly ash was found to be more promising in terms

Table 3. Effect of fly ash alongwith silicate solubilizing bacterial culture on growth and yield parameters of wheat

Treatment	Weight of seeds/pot (g)	No. of seeds/ pot	Dry plant biomass/pot(g)
T ₁ : Soil (control)	0.63	37.67	1.77
T ₂ : RDF	3.77	99.33	3.87
T ₃ : 75% RDF	4.33	119.00	3.27
T ₄ : SSB2	2.07	89.67	3.07
T ₅ : FA (1%)	2.17	56.33	3.13
T ₆ : FA (2%)	2.23	70.00	3.53
T ₇ : FA (3%)	1.70	73.67	2.47
T ₈ : FA (4%)	2.13	86.67	2.30
T ₉ : FA (5%)	2.63	70.00	2.27
T ₁₀ : FA (1%) + SSB	3.77	125.67	3.63
T ₁₁ : FA (2%) + SSB	4.07	134.00	3.73
T ₁₂ : FA (3%) + SSB	4.27	146.67	5.03
T ₁₃ : FA (4%) + SSB	3.50	106.00	4.87
T ₁₄ : FA (5%) + SSB	3.30	94.00	3.79
CD ($p \leq 0.05$)	0.36	7.80	0.56

RDF: Recommended dose of fertilizers

SSB : Silicate solubilizing bacterial culture *Pseudomonas baetica*

FA: Fly ash

of seed number and seed weight compared to the 3% concentration. The control treatment (T₀) showed a decrease in seed weight (3.64 g), number of seeds (109), and plant dry biomass (3.26 g) per pot, compared to treatment 12 (3% fly ash + SSB). The presence of SSB around the plant roots increases the bioavailability of silicon, which enhances plant growth. Increased silicon absorption has been shown to promote better growth in crops like maize (Hu *et al.*, 2018). Similarly, Dhindsa *et al.* (2016) reported that the application of 10% fly ash in clay soil resulted in a wheat grain yield of 35.25 q/ha, compared to 31.00 q/ha in the control treatment. The addition of fly ash improved the soil's capacity to support plant growth, enhancing water holding capacity and aeration. Shakeel *et al.* (2019) found that fly ash amendment in soil led to increased pods per plant, seeds per pod, and seed weight in mustard plants. In their study, a 30% fly ash incorporation resulted in 46.4 pods per plant, 12.4 seeds per pod, and a 100-seed weight of 0.931 g, compared to 30 pods, 11.2 seeds per pod, and 0.481 g in the control treatment. Furthermore, Ou *et al.* (2020) reported that the use of fly ash-based soil conditioners enhanced the yield of wheat and corn by 15% and 10%, respectively. Additionally, plant height increased by 8.73%, and the number of grains increased by 24% in treatments containing fly ash compared to those with conventional fertilizers.

The patterns of plant height, seed weight, plant biomass, and seed number observed in the present study suggest that higher concentrations of fly ash can be applied to agricultural soil in the presence of silicate-solubilizing bacteria (SSB) without inhibiting crop growth and development, leading to enhanced yield. Different crops exhibit varying levels of tolerance to fly ash, with no adverse effects on growth and yield at certain concentrations. For example, soil amendment with fly ash (up to 15%) has been shown to provide yield and physiological benefits for potato crops, while up to 20% fly ash amendment can benefit tomato crops (Tomar & Khan, 2015; Dhindsa *et al.*, 2016). Similarly, fly ash amendment (up to 15%) has been reported to support increased yield and growth attributes of beetroot (Shakeel *et al.*, 2020).

CONCLUSION

In conclusion, the disposal of the ever-increasing amount of fly ash generated from electricity production remains a significant environmental challenge. However, this study demonstrates that fly ash can be effectively utilized in agricultural soil, either alone or in combination with reduced amounts of conventional chemical fertilizers or silicate solubilizing bacteria. The application of 3% fly ash, along with the silicate solubilizing bacterial culture *Pseudomonas baetica*, was found to be particularly beneficial for the growth

and yield of wheat crops. The findings highlight the potential of fly ash as a valuable resource in agriculture, supporting both enhanced crop productivity and sustainable development. Thus, the use of fly ash in agricultural practices could play a pivotal role in addressing waste disposal issues while contributing to more sustainable farming practices.

Authors' contribution

Conceptualization of research work and designing of experiments (LW); Execution of experiments (GC and AKS); Analysis of data and interpretation (LW and MR); Preparation of manuscript (GC and LW)

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