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**Author Affiliation:**

Department of Environmental Microbiology, Babasaheb Bhimrao Ambedkar University, Lucknow-226025, India

**\*Corresponding author:**

Jay Shankar Singh,  
Department of Environmental Microbiology, Babasaheb Bhimrao Ambedkar University, Lucknow-226025, India; E-mail: jayshankar\_1@yahoo.co.in

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# Assessment of sugar mill effluent discharge on soil physico-chemical attributes and its correlation with soil microbial biomass in paddy fields

Rahul Kumar Nigam, Prateek Singh, Jay Shankar Singh\*

## ABSTRACT

The open discharge of untreated sugar mill effluent (SME) onto agricultural land and into water bodies is widely recognized as one of the most polluting ecological disturbances adversely affecting soil systems and microbial processes. Our findings provide novel insights into the impact of SME discharge on soil physico-chemical properties and their correlation with soil microbial biomass (SMB)-C, -N, and -P across three selected paddy fields adjacent to sugar mills in Uttar Pradesh, India. From each selected paddy field, soil samples were collected randomly in triplicate from a depth of 10-15 cm. ANOVA revealed statistically significant ( $P < 0.05$ ) differences in soil physico-chemical attributes and SMB-C, -N, and -P levels across the experimental sites. The variations in soil physico-chemical properties and SMB-C, -N, and -P levels across the selected experimental paddy sites could be due to the variations in quantity and quality of discharged SME from different sugar mill industries to the soil. The SMB-C, -N, and -P levels were found to be comparatively higher in the paddy soil of the Haidergarh site than in the Ayodhya and Burhwal sites. The SMB-C, -N, and -P exhibited positive correlation with total C and total N,  $\text{NH}_4^+\text{-N}$ ,  $\text{NO}_3^-\text{-N}$ , soil moisture, and soil porosity. In contrast, a negative correlation was found between bulk density (BD) and electrical conductivity, as well as SMB-C, -N, and -P, suggesting that a compact soil with higher BD and ions suppresses the levels of SMB. These findings indicate that SME discharge in paddy soils influences the soil physico-chemical characteristics, which in turn govern the SMB-C, -N, and -P levels, playing a crucial role in maintaining soil fertility and regulating soil nutrient availability to the paddy crop.

**Keywords:** Paddy, sugar mill effluent, soil microbial biomass, soil pollution, soil N.

## 1. INTRODUCTION

India, the world's second-largest producer of sugarcane, contributes about 18.18% of Global output (FAO, 2023). The country's sugarcane yield has increased from 33 tonnes/ hectare in 1950-51 to around 78.9 tonnes/ hectare in 2023-24 (GOI, 2024).

However, rapid industrial expansion has intensified hazardous pollution in water sources, agricultural productivity, and food security (Albrizio et al., 2017; Shomanova et al., 2025). The Sugar industry is a primary environmental polluter (Sarfranz et al., 2019) that generates a considerable amount of effluents and solid waste, including bagasse, press mud, and filter cake (Kolhe et al., 2009), and continuously struggles to manage this effluent and its associated pollution load (Verma et al., 2021).

Sugar mill effluent (SME), a major agro-industrial wastewater, is generated in huge quantities (Atila and Kazankaya, 2024). For every tonne of crushed cane, sugar mills consume 1,500 to 2,000 L of water and produce approximately 1,000 L of effluent (Saranraj and Stella, 2014). Despite regulation limits in India allowing effluent discharge of 200 L/ton of cane, infiltration of untreated SME discharge poses a serious threat to aquatic bodies, microbial population, agricultural land, and ecosystem (Mohan and Bajpai, 2018; Verma et al., 2021; Lad et al., 2022; Rehman et al., 2024; Tisha et al., 2025). The SME wastewater contains high concentrations of dissolved organic matter (Thuangchon et al., 2025). It is characterized by low pH, high temperature, turbidity, electrical conductivity (EC), alkalinity, including elevated levels of BOD, COD, total acidity, total solids, total hardness, sulphate, phosphate, chloride, calcium, magnesium, TDS, TSS, etc. (Haider et al., 2022; Hussein, 2023; Gbadeyan et al., 2024).

Prolonged use of SME for irrigation accumulates heavy metals in the soil. This increases the risk of their entry into the food chain through bio-accumulation, geo-accumulation, and bio-magnification, thereby posing significant environmental and health hazards (Muchuweti et al., 2006; Chopra et al., 2009; Khan et al., 2018). Furthermore, SME irrigation can disrupt soil fertility, plant growth, and microbial communities due to its highly poisonous chemical composition (Khan et al., 2019). The unknowingly consumption of groundwater contaminated by SME may lead to the accumulation of toxic metal ions in bones and other organs, causing health issues like diarrhoea, cancer, renal disorders, methemoglobinemia, and neurological problems (Rahim et al., 2021; Banerjee et al., 2023). Finally, when released into water bodies, the elevated BOD and COD levels of SME rapidly deplete dissolved oxygen, severely affecting aquatic flora and fauna (Nawaz et al., 2021; Rahim et al., 2021).

The physico-chemical properties of SME significantly influence soil health, notably by increasing levels of organic carbon, EC, sulphur, and potassium. Although SME can enhance soil fertility through nutrient supply, but excessive accumulation of elements like sulphur and potassium can become toxic and subsequently decline the soil quality (Daulta et al., 2014). Furthermore, SME can inhibit paddy seed germination, disrupt soil structure, and adversely affect soil microbial populations (Shivappa et al., 2007; Samuel and Muthukkaruppan, 2011). Nitrogen in wastewater primarily exists as organic nitrogen, ammonium ( $\text{NH}_4^+$ ), nitrite ( $\text{NO}_2^-$ ), and nitrate ( $\text{NO}_3^-$ ), and poses a significant ecological threat (Hurse and Connor, 1999). In Paddy soils, anaerobic conditions hinder the transformation and turnover of organic matter, thereby disrupting nutrient cycling (Zhu et al., 2017; Wei et al., 2021). In contrast, the alternating oxic and anoxic conditions found in rotation soils promote carbon stabilization (Wei et al., 2022). Given these challenges in anaerobic environments, microbial consortia present an efficient and economical approach for treating SME (Xu et al., 2010; Madariaga-Navarrete et al., 2017).

Soil microbial biomass (SMB) plays a crucial role in soil organic matter formation, nutrient cycling and availability, and the maintenance of soil fertility (Xu et al., 2020). It serves as an essential index for soil health, ecosystem productivity, and nutrient dynamics (Singh and Gupta, 2018). The SMB contributes to plant nutritional availability through mineralization of soil organic content and plays a crucial role in soil nutrient cycles. The pool sizes of SMB-C, -N, and -P in ecosystems are often large enough to significantly impact plant nutrient availability and soil productivity. In soils, microbial biomass measurements can provide the critical data necessary for ecosystem-level monitoring of disturbance and recovery. In a well-established ecosystem, the microbial biomass determines its overall functioning (Smith and Paul, 2017). It can therefore be hypothesized that following a disturbance, such as land-use change, a soil ecosystem with a higher microbial diversity and biomass will have a higher potential to sustain the microbially mediated processes. This capacity buffers soil nutrients and facilitates productivity restoration through efficient nutrient turnover (Singh, 2016). Conversely, ecological disturbance, including land use changes and pollutants, like heavy metals, can negatively impact the pool sizes of SMB-C, -N, and -P. This reduction, in turn, impairs beneficial microbial activities, ultimately leading to reduced soil fertility and productivity (Sun et al., 2023; Xu et al., 2023; Elrys et al., 2025). Previous investigations have demonstrated that SME contains organic compounds, macro-nutrients, micro-nutrients, heavy metals, and toxic components in dissolved or suspended forms, which adversely affect the physical, chemical, and physiological conditions of terrestrial and aquatic ecosystems (Lavanya et al., 2019; Arivoli et al., 2021). Studies have also demonstrated its negative impact on microbial population, enzymatic activities, seed germination, plant growth, and crop yields (Siddiqui and Waseem, 2012; Kumar and Srikantaswamy, 2015; Pandey et al., 2018; Verma et al., 2021). Nevertheless, a detailed study on the impact of SME on soil physico-chemical attributes, microbial diversity, microbial biomass, and their correlations remains unreported. Furthermore, there is a significant lack of information concerning the impact of quality and quantity of SME discharge, along with key environmental drivers and affected soil parameters, influence SMB-C, -N, and -

P levels in paddy agriculture soils. Therefore, the objectives of the present study were to explore (i) the impact of SME discharged quantity on physico-chemical properties and SMB-C, -N, and -P, and (ii) the correlation between soil physico-chemical properties and SMB-C, -N, and -P across different experimental paddy fields located in different districts of Uttar Pradesh, India.

## 2. MATERIALS AND METHODS

### Location, vegetation, and climatic conditions of the selected experimental area

Uttar Pradesh is the leading sugarcane producer in India, contributing 46.21% of the total area under cultivation and 43.65% of the total production (Singh and Singh, 2022). Therefore, for this study, sugar mills in selected districts of the Awadh region were selected, including Balrampur Chini Mills (Haidergarh, Barabanki), K.M. Sugar Mills Limited (Ayodhya), and UP State Sugar and Cane Development Corporation Ltd. (Burhwal, Barabanki) (Fig. 1). The coordinates of the selected sites are given in Table 1.

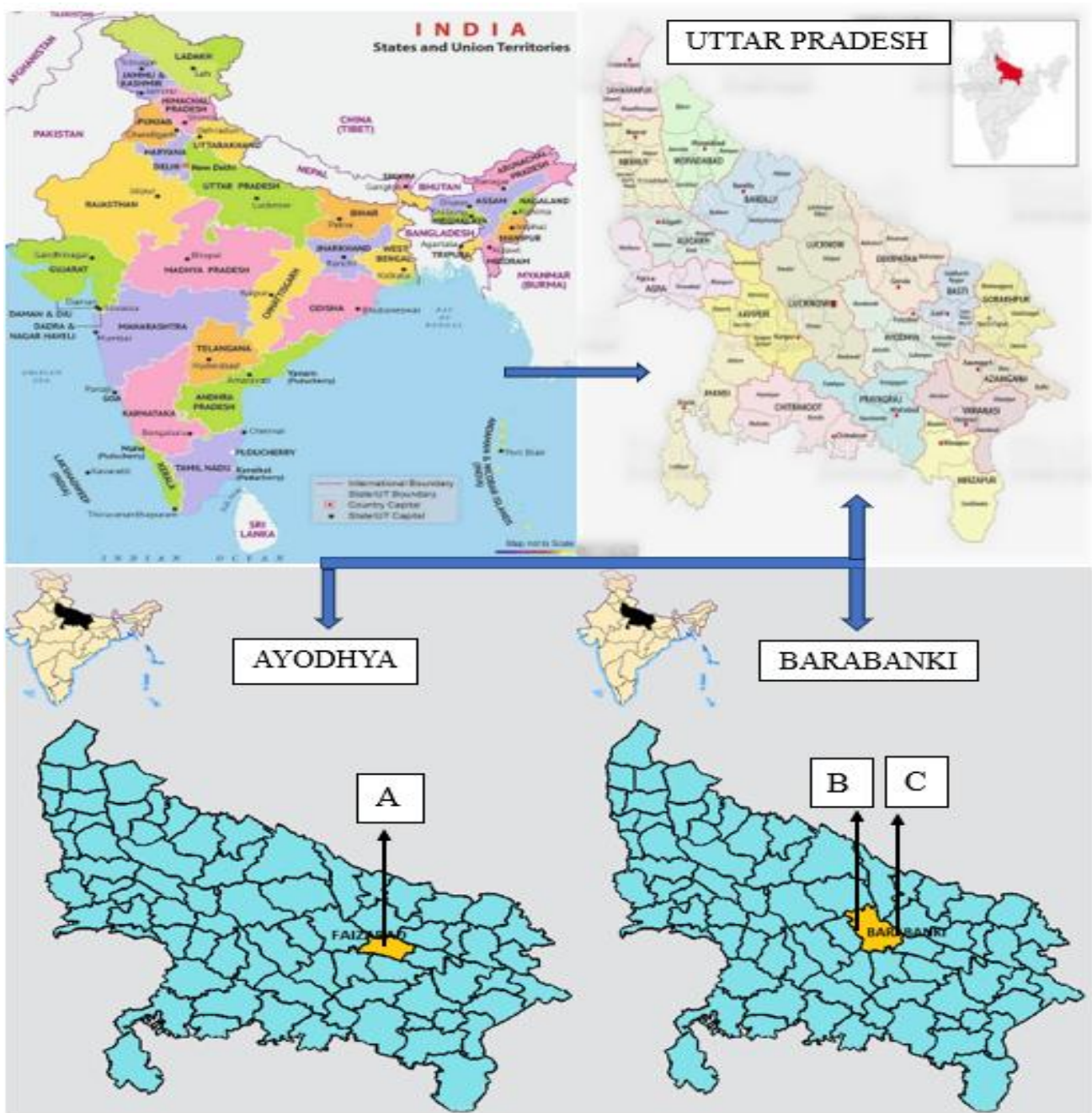


Figure 1 Location of selected experimental sites in different districts of Uttar Pradesh, India.

The Ayodhya district covers an area of approximately 2,764 km<sup>2</sup>, situated within an alluvial plain dominated by deposits from the Gomati and Ghaghara rivers. The region has a sub-tropical climate, characterized by hot, dry summers (with temperatures up to 45 °C and desiccating winds from March to June) and cold winters (down to 5 °C from November to February). High potential evaporation occurs from March to October. The average annual rainfall is about 1,070 mm, 85% of which is received during the southwest monsoon (July to October). Primary crops include *Oryza sativa* and *Triticum aestivum*. *Saccharum officinarum* and *Brassica juncea* and horticultural crops (*Mangifera indica*, *Phyllanthus emblica*, *Psidium guajava*, and *Musa* sp.) are also cultivated (Sundar and Kittur, 2012; Kumar, 2018; Kumar et al., 2022). The Barabanki district, located 29 kilometres east of Lucknow, covers an area of 4,696.8 km<sup>2</sup> within the fertile Ganga-Yamuna basin. Agriculture is the primary occupation for 72.6% of the working population. The average annual rainfall is about 1,056 mm. Major crops include *Triticum aestivum*, *Oryza sativa*, and *Saccharum officinarum*, alongside pulses, oilseeds, and *Zea mays*. Key vegetable crops cultivated are cucurbits, *Solanum tuberosum*, *Capsicum annuum*, *Lycopersicon esculentum*, *Abelmoschus esculentus*, *Solanum melongena*, *Allium cepa*, and *Allium sativum* (Kumar et al., 2015; Singh et al., 2017). These sugar mills release a considerable amount of SME into adjacent paddy agricultural lands. Farmers often use this SME as a substitute for Farm Yard Manure (FYM) or as a soil amendment in paddy fields to enhance paddy crop productivity. According to the Directorate of Economics and Statistics (DES), GOI, during the 2022-2023 season, paddy was cultivated on 99,090 hectares in Ayodhya and 188,580 hectares in Barabanki.

**Table 1** Coordinates and soil types of selected experimental sites.

Experimental sites	Name of the sugar mill industries	Soil colour	Soil texture	Coordinates
A	K.M. Sugar Mills Limited, Ayodhya	Brown	Loamy soil	(26°87'57" - 26°99'17" N & 81°02'74" - 81°60'81" E)
B	Balrampur Chini Mills, Haidergarh, Barabanki UP State Sugar and Cane	Dark brown	Loamy soil	(26°59'31" - 26°59'30" N & 81°30'41" - 81°30'63" E)
C	Development Corporation Ltd., Burhwal, Barabanki	Brownish yellow	Loamy soil	(27°08'18" - 27°08'19" N & 81°38'91" - 81°39'13" E)

### Soil sampling and analyses

For this study, soil samples were collected from three paddy fields at the crop maturity stage. Fields were selected with careful consideration to ensure homogeneity and uniformity in elevation, slope, and distance from each sugar mill. A completely randomized block design was employed for the paddy fields located in different districts of Uttar Pradesh, as described in Table 1.

At each selected site, a 10 m × 10 m plot was demarcated for soil sampling. On October 19, 2023, soil samples were collected from a depth of 10-20 cm using a 2.5 × 45 cm soil core. From each marked plot, triplicate samples were collected from three different points (Fig. 2), resulting in a total of nine samples per paddy field (three sampling points × three replicates). Consequently, a total of 27 soil samples (three sites × three sampling points × three replicates) were collected from the three selected experimental fields. The samples were wrapped in aluminum foil, placed in sterilized airtight polythene bags, and transported to the laboratory (Singh et al., 2021). They were stored at 4 °C until physico-chemical and SMB-C, -N, and -P analyses. All soil parameter analyses were performed using analytical grade chemicals and reagents (HiMedia and Sigma-Aldrich).

### Physico-chemical analyses of collected soil samples

The collected soil samples were air-dried at room temperature and gently homogenized by sieving through a 2 mm mesh to break down aggregates for physico-chemical analyses. Soil texture was determined using the standard sieving method (Indian standards, 1965), and soil colour was assessed with the Munsell soil colour chart (Munsell, 1975). EC and pH were measured using EC and pH meters in soil-water suspensions at ratios of 1:5 and 1:2.5 (w/v), respectively (Wilcox, 1950; Pansu and Gautheyrou, 2006). Soil bulk density (BD) and water holding capacity (WHC) were evaluated according to Piper (1944). Soil moisture content (%) was determined by drying soil sub-samples at 105 °C for 24 h in a hot air oven as described by Buresh (1991), while porosity was calculated using the method of Bouma (1991). Total organic carbon (TOC) was estimated by the Walkley & Black (1947) method, while inorganic nitrogen forms (NH<sub>4</sub><sup>+</sup>-N and NO<sub>3</sub><sup>-</sup>-N) and P were quantified based on the Eno (1960) method. The total soil N was analysed as described by Jackson (1958).



**Figure 2** Soil sample collection from SME affected experimental paddy fields from (A) Ayodhya, (B) Haidergarh, and (C) Burhwal sugar Mill area (D) SME discharged into the adjacent water body and the nearby paddy field.

### Soil microbial biomass -C, -N, and -P analyses

The SMB-C, -N, and -P were estimated by the chloroform ( $\text{CHCl}_3$ ) fumigation-extraction procedures (Brookes et al., 1985; Vance et al., 1987). Purified liquid chloroform was used for soil fumigation (Srivastava and Singh, 1988), followed by extraction with  $0.5 \text{ mol L}^{-1} \text{ K}_2\text{SO}_4$  (1:4 soil: extract) for about 30 min. Similar extraction procedures were also adopted for non-fumigated soil samples. The SMB-C and -N were determined from  $0.5 \text{ mol L}^{-1} \text{ K}_2\text{SO}_4$  extract by dichromate digestion (Vance et al., 1987), while SMB-P was analyzed as inorganic-P (Pi) in  $0.5 \text{ mol L}^{-1} \text{ NaHCO}_3$  extract of both fumigated and non-fumigated soil samples by ammonium molybdate-stannous chloride method (Sparling et al., 1985).

### Quality assurance and quality control

We adhered to standard protocols for sampling, transporting, and handling of soil samples from SME impacted paddy fields. Soil samples were collected using a metal soil corer and sterilized hand gloves, placed in polypropylene bags, and stored in refrigerated conditions until laboratory analyses. All reagents and calibration standards were prepared using analytical grade chemicals using Milli-Q (Millipore) water or double-distilled de-ionized water. Sterilized glassware was used to prevent microbial contamination. Experimental procedures followed thorough standard operating procedures (SOPs) and protocols, with continuous monitoring and detailed techniques to generate reliable and high-quality data.

### Statistical analyses

A one-way analyses of variance (ANOVA) was performed to compare the soil physico-chemical properties and SMB-C, -N, and -P values across the study sites. The presented values are means of three replicates  $\pm$  standard error (SE). Since ANOVA requires a regular data distribution pattern, a positive and a homogeneity of variance test were also performed before the analyses of variance. Tukey's HSD (Honestly Significant Difference) test was used to identify specific differences between group means. Pearson's correlation analyses was also performed to evaluate the interrelationships between soil physico-chemical properties and SMB-C, -N, and -P levels. All statistical analyses were conducted using SPSS version 27.0 (IBM, Armonk, NY, USA) at a 95% confidence level.

## 3. RESULTS

### Soil physico-chemical properties

The physico-chemical properties of soil samples collected from the experimental sites are presented in Table 2. The results suggest that soil texture was loamy at all sites. The soil colour was predominantly brown except at the Burhwal site, where it was brownish-yellow. The soil pH varied from 7.4 to 7.8, with significant variations observed between experimental sites ( $F = 20.885$ ;  $N = 27$ ;  $P < 0.001$ ). The mean soil moisture (SM) content (21.96 %), water holding capacity (WHC) (42.5 %), and soil porosity (SP) (46.77 %) were noted as maximum in Haidergarh soil, followed by Ayodhya and Burhwal soil samples. Variations across the experimental sites was significant for SM ( $F=18.921$ ;  $N= 27$ ;  $P < 0.001$ ), WHC ( $F=22.091$ ;  $N= 27$ ;  $P < 0.001$ ) and SP ( $F=13.584$ ;  $N= 27$ ;  $P < 0.001$ ). BD ( $1.73 \text{ gm cm}^{-3}$ ) and EC ( $4.67 \mu\text{mho ms}^{-1}$ ) were significantly higher in Burhwal soil samples and lowest in Haidergarh soil samples. Both BD and EC varied significantly ( $F = 17.692$ ;  $N = 27$ ;  $P < 0.001$ ) and ( $F = 22.527$ ;  $N = 27$ ;  $P < 0.001$ ), due to differences in study sites.

**Table 2** Description of soil physico-chemical properties across the experimental sites. For each parameter and experimental site, the value represents the mean of 9 replicates  $\pm$  SE. One-way ANOVA showed significant differences ( $P < .001$ ) in parameters across the experimental sites. The total number of soil samples (N) analyzed for each selected parameter was 27 (three experimental sites  $\times$  three sampling points  $\times$  three replicates).

Soil parameters	Experimental sites			F value (N=27)	Sig.
	(A) Ayodhya	(B) Haidergarh	(C) Burhwal		
Soil pH	7.8 $\pm$ 0.003	7.6 $\pm$ 0.007	7.4 $\pm$ 0.006	20.885	< .001
WHC (%)	34.03 $\pm$ 0.003	42.55 $\pm$ 0.007	30.39 $\pm$ 0.003	22.091	< .001
Soil porosity (%)	44.44 $\pm$ 0.006	46.77 $\pm$ 0.011	40.07 $\pm$ 0.005	13.584	< .001
Bulk density ( $\text{gm cm}^{-3}$ )	1.55 $\pm$ 0.006	1.45 $\pm$ 0.002	1.73 $\pm$ 0.004	17.692	< .001
Soil moisture (%)	20.03 $\pm$ 0.004	21.96 $\pm$ 0.002	17.76 $\pm$ 0.005	18.921	< .001
EC ( $\mu\text{mho ms}^{-1}$ )	4.52 $\pm$ 0.005	4.13 $\pm$ 0.003	4.67 $\pm$ 0.004	22.527	< .001
NH <sub>4</sub> <sup>+</sup> -N ( $\mu\text{g g}^{-1}$ dry soil)	5.66 $\pm$ 0.004	5.96 $\pm$ 0.004	5.23 $\pm$ 0.003	20.095	< .001
NO <sub>3</sub> <sup>-</sup> -N ( $\mu\text{g g}^{-1}$ dry soil)	4.67 $\pm$ 0.006	4.89 $\pm$ 0.006	4.41 $\pm$ 0.007	23.185	< .001
Total-C ( $\mu\text{g g}^{-1}$ dry soil)	15439.32 $\pm$ 1.59	16870.60 $\pm$ 12.92	14630.21 $\pm$ 2.38	8.368	< .001
Total-N ( $\mu\text{g g}^{-1}$ dry soil)	732.44 $\pm$ 0.027	783.80 $\pm$ 0.013	717.55 $\pm$ 0.025	19.544	< .001
Total-P ( $\mu\text{g g}^{-1}$ dry soil)	729.91 $\pm$ 0.011	747.00 $\pm$ 0.022	681.62 $\pm$ 0.030	17.229	< .001

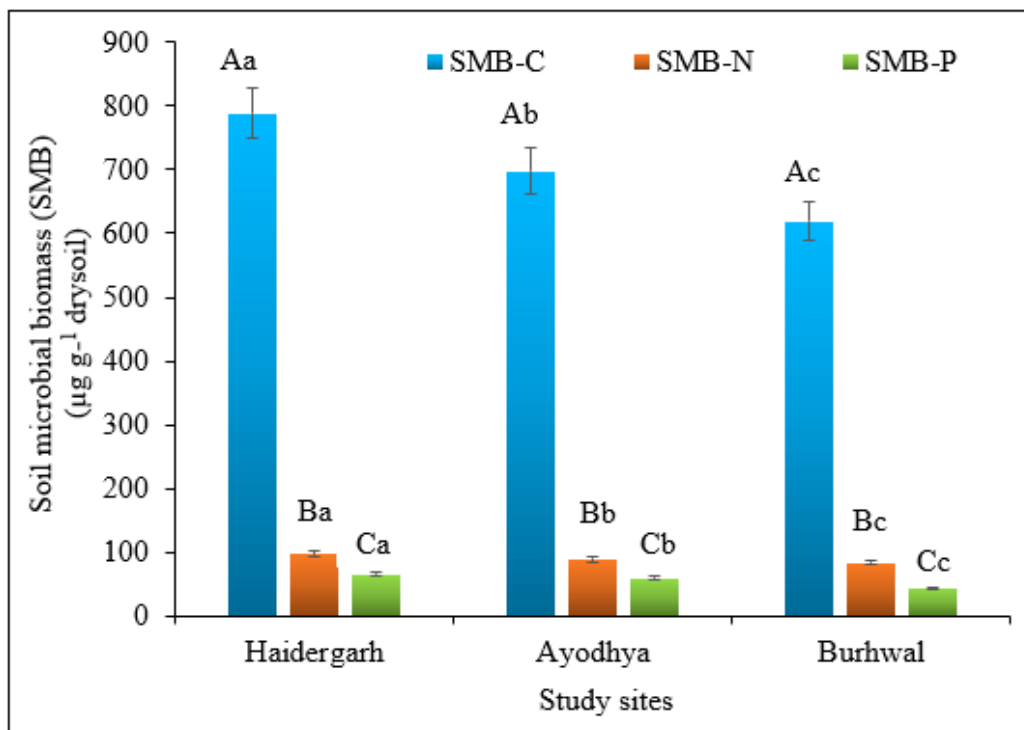
WHC: water holding capacity, EC: electrical conductivity

The soil NH<sub>4</sub><sup>+</sup>-N and NO<sub>3</sub><sup>-</sup>-N across the experimental sites were highest (5.96 and 4.89  $\mu\text{g g}^{-1}$  dry soil, respectively) in Haidergarh samples and lowest (5.23 and 4.41  $\mu\text{g g}^{-1}$  dry soil, respectively) in Burhwal soil samples and differences in NH<sub>4</sub><sup>+</sup>-N and NO<sub>3</sub><sup>-</sup>-N

concentrations in the soil across selected sites were significant ( $F=20.095$ ;  $N= 27$ ;  $P < 0.001$ ; and  $F=23.185$ ;  $N= 27$ ;  $P < 0.001$ ). The mean soil total-C ( $16870.6 \mu\text{g g}^{-1}$  dry soil), total-N ( $783.8 \mu\text{g g}^{-1}$  dry soil), and total-P ( $747.0 \mu\text{g g}^{-1}$  dry soil) were highest in Haidergarh soil samples compared to other study sites and across experimental sites variations in soil total-C, total-N, and P ( $F=8.368$ ;  $N= 27$ ;  $P < 0.001$ ;  $F=19.544$ ;  $N= 27$ ;  $P < 0.001$ ; and  $F=17.229$ ;  $N= 27$ ;  $P < 0.001$ , respectively) were noted to be significant.

#### Soil microbial biomass -C, -N, and -P

ANOVA revealed statistically significant variations ( $P < 0.001$ ) in SMB-C, -N, and -P (Fig. 3) values across the experimental sites. The mean SMB-C across the experimental sites was significantly highest ( $789.09 \mu\text{g g}^{-1}$  dry soil) in Haidergarh samples and lowest ( $618.89 \mu\text{g g}^{-1}$  dry soil) in Burhwal samples, with an overall significant difference across selected sites ( $F=22.313$ ;  $N= 27$ ;  $P < 0.001$ ). Similarly, the highest mean SMB-N ( $97.21 \mu\text{g g}^{-1}$  dry soil) and SMB-P ( $65.76 \mu\text{g g}^{-1}$  dry soil) were also recorded in Haidergarh soil. Variations in soil SMB-N and SMB-P ( $F= 8.756$ ;  $N= 27$ ;  $P < 0.001$ ;  $F=9.944$ ;  $N= 27$ ;  $P < 0.001$ ) were noted to be significant. The trend of increasing order for SMB-C, -N, and -P was Haidergarh < Ayodhya < Burhwal (Fig. 3).



**Figure 3** Variations in soil microbial biomass (SMB) -C, -N, and -P content across the experimental sites. Values are means of nine replicates  $\pm$ SE. On each bar, different letters indicate a significant difference between study sites for SMB-C, -N, and -P ( $P < 0.001$ ).

#### Relationship between soil physico-chemical properties and SMB-C, -N, and -P

The SMB -C, -N, and -P exhibited significant positive correlations with pH, WHC, soil porosity, soil moisture,  $\text{NH}_4^+$ -N,  $\text{NO}_3^-$ -N, total-C, -N, and -P contents. In contrast, a strong inverse correlation was observed between BD and EC with SMB -C, -N, and -P across all experimental sites (Table 3).

## 4. DISCUSSION

#### Impact of SME on soil physico-chemical characteristics

Most essential plant nutrients remain soluble and available for plant uptake within a pH range of 6.5 to 6.8. The soil pH in this study ranged from 7.4 to 7.8, consistent with previous research, which reported that the pH ranged from 7.6 to 7.9 (Nagaraju et al., 2009; Balkhande et al., 2013; Daulta et al., 2014). The mean pH of the sugar mill effluent (SME) itself was  $7.32 \pm 0.21$ , which falls within the permissible range of 5.5 to 9.0. The low pH value can be attributed to the use of phosphoric acid and sulfur dioxide during cleaning processes of sugarcane juice (Bhutiani et al., 2015). As pH deviates from the optimal range can cause nutrients to chemically bind, reducing their availability for plant uptake (Daulta et al., 2014). These findings suggest that the SME application influences nutrient

dynamics through pH alteration. Bulk density is a critical factor in assessing soil porosity and water retention capacity. The application of organic amendments, such as SME, can significantly alter the soil's bulk density by reducing its compactness. The BD and WHC were 1.41 gm cm<sup>-3</sup> and 41.86% respectively, on application of 100% SME concentration (Baskaran et al., 2009; Kumar, 2014). Increase in WHC and decrease in BD at the Haidergarh site suggest a more porous soil structure and improved hydrothermal conditions compared to the other two sites. Furthermore, application of SME may enhance cation exchange capacity (CEC) and improve WHC of the soil (Daulta et al., 2014), supporting the observed improvements in soil physical properties.

The higher soil moisture at the Haidergarh site may be attributed to more organic matter content and improved water aeration than Ayodhya and Burhwal sites. The sugar mill wastewater contains dissolved organic and inorganic compounds, which elevates electrolyte content and consequently, electrical conductivity increases at the Burhwal site. Samuel and Muthukkaruppan, (2011) also reported a greater EC of 0.43  $\mu\text{mho ms}^{-1}$  in effluent irrigated soil compared to non-treated soils (Arivoli et al., 2021).

Nitrogen present in untreated wastewater is mainly as ammonia and organic nitrogen, in both soluble and particulate forms (Sahu and Chaudhari, 2015). Mean values of NH<sub>4</sub><sup>+</sup>-N and NO<sub>3</sub><sup>-</sup>-N in the present study were recorded as higher in the Haidergarh site, whereas, Burhwal site observed comparatively lower values (Table 3). The increase in NH<sub>4</sub><sup>+</sup>-N in Haidergarh soils may result from higher net ammonification rates and subsequent accumulation in the topsoil. Additionally, lower immobilization by soil microbial biomass and reduced plant uptake contributed to the accumulation of inorganic nitrogen (NH<sub>4</sub><sup>+</sup>-N and NO<sub>3</sub><sup>-</sup>-N) in these soils. The reason for higher N-mineralization rates may result from higher soil organic-C and -N content in the soil (Dhangar and Lohar, 2013). Furthermore, the variations in inorganic nutrient levels across sites receiving different quantities of SME may often be linked to the use of varied quantities and quality of phosphoric acid during sugar processing, cleaning, and other operations (Daulta et al., 2014).

**Table 3** Pearson's correlation (2-tailed) between physico-chemical properties and soil microbial biomass of SME. Total soil samples analyzed were N=27 (three sites  $\times$  nine replicates). Numbers denote respective physico-chemical parameters.

Parameters ↓	1	2	3	4	5	6	7	8	9	10	11	12	13	14
1. Soil pH	1													
2. WHC (%)	0.259	1												
3. Soil porosity (%)	0.614**	0.920**	1											
4. BD (gm cm <sup>-3</sup> )	-0.618**	-0.912**	-0.994**	1										
5. Soil moisture (%)	0.508**	0.963**	0.992**	-0.985**	1									
6. EC ( $\mu\text{mho ms}^{-1}$ )	-0.241	-0.998**	-0.910**	0.904**	-0.956**	1								
7. NH <sub>4</sub> <sup>+</sup> -N ( $\mu\text{g g}^{-1}$ dry soil)	0.561**	0.944**	0.997**	-0.992**	0.997**	-0.936**	1							
8. NO <sub>3</sub> <sup>-</sup> -N ( $\mu\text{g g}^{-1}$ dry soil)	0.507**	0.958**	0.987**	-0.977**	0.995**	-0.951**	0.993**	1						
9. Total-C ( $\mu\text{g g}^{-1}$ dry soil)	0.324	0.997**	0.945**	-0.936**	0.979**	-0.994**	0.964**	0.974**	1					
10. Total-N ( $\mu\text{g g}^{-1}$ dry soil)	0.181	0.997**	0.886**	-0.877**	0.938**	-0.997**	0.914**	0.933**	0.989**	1				
11. Total-P ( $\mu\text{g g}^{-1}$ dry soil)	0.685**	0.879**	0.996**	-0.990**	0.975**	-0.868**	0.986**	0.971**	0.910**	0.838**	1			
12. SMB-C ( $\mu\text{g g}^{-1}$ dry soil)	0.434*	0.982**	0.977**	-0.970**	0.996**	-0.977**	0.989**	0.992**	0.993**	0.964**	0.953**	1		
13. SMB-N ( $\mu\text{g g}^{-1}$ dry soil)	0.393*	0.990**	0.966**	-0.959**	0.991**	-0.985**	0.981**	0.986**	0.997**	0.975**	0.938**	0.999**	1	
14. SMB-P ( $\mu\text{g g}^{-1}$ dry soil)	0.655**	0.898**	0.999**	-0.993**	0.983**	-0.887**	0.992**	0.979**	0.926**	0.860**	0.999**	0.965**	0.951**	1

\* Significant at  $P < 0.05$ ; \*\* Significant at  $P < 0.01$  level.

The high concentrations of total-C (16,870.60 $\pm$ 12.92  $\mu\text{g g}^{-1}$  dry soil), total-N (783.8 $\pm$ 0.013  $\mu\text{g g}^{-1}$  dry soil), and total-P (747.0 $\pm$ 0.022  $\mu\text{g g}^{-1}$  dry soil) observed at the Haidergarh site in SME irrigated soil are likely due to elevated organic-C, -N, and -P nutrient load in the SME wastewater used for irrigation. These findings are consistent with those of Tabriz et al., (2011), who observed 2.14% of organic carbon in the effluent-affected soil, while Chopra and Pathak, (2013) reported 1.37% of organic carbon in SME amended soil. Similarly, Singh et al., (2010a), who also demonstrated that the application of farmyard manure amendment rich in organic contents significantly increases the organic-C, -N, and -P levels in saline paddy fields. Singh, (2018) and Singh et al., (2021) also reported that crop residues-

based biochar amendment showed significant variation in total-N ( $P < 0.01$ ) and organic-C ( $P < 0.001$ ) in the soil of the paddy field. The significant variations ( $P < 0.001$ ) in total-C, -N, and -P levels across different study soils due to high influx of organic litter and greater microbial-mediated decay and decomposition processes (Boudh et al., 2024). Furthermore, Tiwari et al., (2019) reported land use changes and soil depths ( $P < 0.001$ ) exhibited significant differences in total-C, -N, and -P levels across agricultural land, mixed forest, savanna, and natural forest. In this investigation, the variations in total-C -N and -P composition across the selected paddy agriculture fields reflect high qualitative and quantitative heterogeneity in the SME discharge that might likely be due to variations in tailing disposal time, microbial activity, and other soil and environmental drivers, as suggested by Liu et al., (2014).

### Impact of SME on SMB -C, -N, and -P

A significant variation ( $P < 0.001$ ) in SMB -C, -N and -P compositions, being maximum at Haidergarh site (789.09  $\mu\text{g g}^{-1}$  dry soil, 97.21  $\mu\text{g g}^{-1}$  dry soil and 65.76  $\mu\text{g g}^{-1}$  dry soil, respectively) in the soil of selected paddy experimental sites, affected by SME discharge are presented in Fig. 3. In present study the variations in SMB-C, -N and -P across different paddy soils can be attributed to the changes in the quantity and quality of SME discharge, as suggested by Singh and Gupta, (2018) regarding variations in SMB pools due to variations in organic-C and -N inputs plants residues for different forest ecosystems. Numerous studies show that the abundance of soil microbes and microbial biomass is directly proportional to precipitation and soil water content (Chen et al., 2015; Ma et al., 2015; Shah et al., 2024). Furthermore, the MB-N dynamics are influenced by environmental factors like moisture and temperature, as well as biological processes such as litter input and microbial-feeding fauna activity, which affect microbial growth (Chen et al., 2003).

The results of this research work strongly to our hypothesis that SME application influences the SMB -C, -N, and -P in the upper soil layer (depth 0-20 cm) of paddy fields. The SMB -C, -N, and -P levels were observed to be highest in the Haidergarh site might be due to the elevated concentration of dissolved organic, inorganic compounds, macronutrients, and micronutrients in SME (Kumar and Srikantaswamy, 2015; Lavanya et al., 2019; Malik, 2019; Arivoli et al., 2021). An elevated microbial activity in SME affected paddy soils may likely enhance the decay and decomposition processes. This, in turn, improves soil physico-chemical properties and promotes the accumulation of soil microbial biomass, as evidenced by the increase in SMB -C, -N, and -P levels.

The significant variations in SMB-C, -N, and -P values in this study indicate considerable variations in organic matter quantity and quality across the effluent-affected paddy field, diverse vegetation compositions, and soil characteristics across the SME affected paddy fields. These variations may also be due to the greater microbial activities, such as organic matter mineralization, which may have resulted in greater organic-C inputs and subsequently increased soil microbial biomass (Tiwari et al., 2018). The SME discharge can influence the soil microbial-mediated chemical transformations and nutrient cycling, potentially influencing the soil microbial biomass and composition (Tripathi and Tripathi, 2014; Pandey et al., 2018). Across the experimental paddy fields, soil organic matter and SMB may both vary due to variations in crop residue inputs due to differences in crop density variations, and paddy cultivars diversification (Cerny, 2008; Maharjan et al., 2017). Additionally, variations in TDS concentration of SME discharge may directly impact the soil microbial communities and their biomass (SMB-C, -N, and -P) (Kaur et al., 2010; Verma et al., 2021). This is supported by studies reporting significantly higher ( $P < 0.001$ ) microbial population in SME irrigated soil, including a two-fold increase in bacterial and fungal population (Reddy et al., 2013). Srivastava et al., (2017) further observed that a maximum population of bacteria, rhizobia, and actinomycetes occurred at 50% SME concentration, whereas yeast and fungal populations peaked at 75% SME concentration. Therefore, in this study, an increase in SMB-C, -N, and -P may be associated with organic matters present in SME, which provides available more nutrients to microbial communities and therefore, builds up more soil microbial biomass (Ahmad et al., 2008; Mahmood et al., 2017; Xu et al., 2018). Additionally, the variations in soil physico-chemical properties, environmental drivers, and geography of selected paddy agriculture fields may be instrumental in SMB-C, -N, and P variations (Singh et al., 2010b; Singh, 2015).

The SMB -C, -N, and -P exhibited significant positive correlations with pH, WHC, soil porosity, soil moisture,  $\text{NH}_4^+\text{-N}$ ,  $\text{NO}_3^-\text{-N}$ , total-C, -N, and -P contents. Conversely, they were negatively correlated with BD and EC across all the experimental sites (Table 3). The positive correlation of SMB with inorganic-N nutrients, organic nutrients, and soil moisture in this investigation may have resulted because a greater soil microbial biomass and microbial communities may enhance the decay and decomposition of complex organic contents of paddy crop plant residues into simpler soil inorganic and organic contents, which in turn favours the soil to hold more WHC and soil moisture. Contrary to the present result, the study of Singh et al., (2009) from dry tropical soils demonstrated that microbial biomass-N was negatively correlated with soil moisture and such situations raised because of the enhanced SM conditions in rainy season was in demand for a greater microbial mediated soil organic matter mineralization to satisfy inorganic-N availability to the vigorously growing plants during active growth period (monsoon season). The low levels of SMB-C, -N, and -P in this study, along with their negative correlation with BD and EC across sites, could be due to unfavourable soil conditions. The quality of SME discharge

may disturb the soil microbial activities, growth, and multiplication, thereby lowering SMB. This inhibitory effect is reflected in the reciprocal relationship between the magnitude of SMB-C, -N, and -P and both soil BD and EC (Table 3).

## 5. CONCLUSION

It may be concluded that the quality and quantity of SME discharged in paddy agro-ecosystems, located in the adjoining area of sugar mills, is one of the major causes for significant variations in soil physico-chemical properties and microbial biomass. The release of SME into paddy cropland has resulted in an increase in soil inorganic and organic nutrients, soil moisture, and SMB-N, -C, and P pools. These findings and analyses indicate that the SME substantially enhances the fertility and productivity of paddy cropland soils by playing an important role in enhancing microbial biomass and regulating carbon, nitrogen, and phosphorus status. The improved soil physico-chemical properties, as well as nutrient sources due to SME discharge, may positively support to the growth and multiplication of beneficial agriculturally important soil microbial communities and soil microbial biomass, which may act as a soil fertility index. Therefore, there is further need for better SME management, detailed research investigations, and policy/plans for its efficient use and sustainable benefits in paddy agriculture farming located in the adjoining area of sugar mill industries.

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### Conflict of interest

The authors declare that there are no conflicts of interests.

### Ethical approval

Not applicable.

### Informed consent

Not applicable.

### Data availability

All data associated with this work are present in the paper.

## REFERENCES

- Ahmad R, Arshad M, Khalid A, Zahir ZA. Effectiveness of organic-/bio-fertilizer supplemented with chemical fertilizers for improving soil water retention, aggregate stability, growth and nutrient uptake of maize (*Zea mays* L.). *J Sustain Agric* 2008; 31(4):57-77. doi: 10.1300/J064v31n04\_05
- Albrizio S, Kozluk T, Zipperer V. Environmental policies and productivity growth: Evidence across industries and firms. *J Environ Econ Manag* 2017; 81:209-226. doi: 10.1016/j.jeem.2016.06.002
- Arivoli A, Naveen XAJ, Thangavelu S. Impact of Sugar Mill Effluent on the Quality of Groundwater in Cuddalore District, Tamil Nadu, India. In: Marimuthu PD, Sundaram R, Jeyaseelan A, Kaliannan T (eds) *Bioremediation and Green Technologies: Sustainable Approaches to Mitigate Environmental Impacts*. Springer, Switzerland, 2021, 41-52.
- Atila F, Kazankaya A. Evaluation of the yield and heavy metal bioaccumulation in the fruit body of *Pleurotus ostreatus* grown on sugar mill wastewaters. *Biomass Convers Biorefin* 2024; 14(16):19177-19186. doi: 10.1007/s13399-023-03913-7
- Balkhande JV, Jalander V, Balkhande SV. The influence of Hingoli and Nanded districts sugarcane industry effluent on physicochemical properties and mycoflora of soil. *Res Biotechnol* 2013; 4(3):1-6.

6. Banerjee P, Garai P, Saha NC, Saha S, Sharma P, Maiti AK. A critical review on the effect of nitrate pollution in aquatic invertebrates and fish. *Water, Air, & Soil Pollution* 2023; 234(6):333. doi: 10.1007/s11270-023-06260-5
7. Baskaran L, Sankar GK, Chidambaram ALA, Sundaramoorthy P. Amelioration of sugar mill effluent polluted soil and its effect of green gram (*Vigna radiata* L.). *Bot Res Int* 2009; 2(2):131–135.
8. Bhutiani R, Khanna DR, Kumar A. Physico-chemical characterization of sugar mill effluent. *Ind J Sci Res Technol* 2015; 3(2):13-16.
9. Boudh S, Tiwari S, Singh C, Singh JS. Lindane degradation potential of methanotrophs and soil microbial biomass from HCH contaminated sites. *Environ Adv* 2024; 17:100581. doi: 10.1016/j.envadv.2024.100581
10. Bouma J. Influence of soil macroporosity on environmental quality. *Adv Agron* 1991; 46:1-37. doi: 10.1016/S0065-2113(08)60577-5
11. Brookes PC, Landman A, Pruden G, Jenkinson DS. Chloroform fumigation and release of soil N: a rapid direct extraction method to measure microbial biomass N in soil. *Soil Biol Biochem* 1985; 17(6):837-842. doi: 10.1016/0038-0717(85)90144-0
12. Buresh RJ. Extraction of ammonium, nitrate and nitrite from soil. IFDC/IRRI Collaborative Project 1991; 1-3.
13. Černý J, Balík J, Kulháněk M, Nedvěď V. The changes in microbial biomass C and N in long-term field experiments. *Plant, Soil and Environ* 2008; 54(5):212-218. doi: 10.17221/393-PSE
14. Chen CR, Xu ZH, Blumfield TJ, Hughes JM. Soil microbial biomass during the early establishment of hoop pine plantation: seasonal variation and impacts of site preparation. *Forest Ecol Manage* 2003; 186(1-3):213-225. doi: 10.1016/S0378-1127(03)00275-5
15. Chen D, Mi J, Chu P, Cheng J, Zhang L, Pan Q, Xie Y, Bai Y. Patterns and drivers of soil microbial communities along a precipitation gradient on the Mongolian Plateau. *Landsc Ecol* 2015; 30(9):1669-1682. doi: 10.1007/s10980-014-9996-z
16. Chopra AK, Pathak C, Prasad G. Scenario of heavy metal contamination in agricultural soil and its management. *J Appl Nat Sci* 2009; 1(1):99-108. doi: 10.31018/jans.v1i1.46
17. Chopra AK, Pathak C. Enrichment and translocation of heavy metals in soil and *Spinacea oleracea* grown in sugar mill Effluent irrigated soil. *Sugar Tech* 2013; 15(1):77-83. doi: 10.1007/s12355-012-0194-5
18. Daulta R, Rani J, Yadav A. Effect of sugar mill effluent on physico-chemical properties of soil at Panipat city, India. *Int Arch App Sci Tech* 2014; 5(2):6-12. doi: 10.5555/20153245770
19. Dhangar SD, Lohar PS. Effect of sugar factory effluent on physico-chemical properties and cellulase activity of soil-A case study. *Int Multidiscip Res J* 2013; 3(1):18-22.
20. Directorate of Economics and Statistics, Department of agriculture and farmers welfare, Ministry of agriculture and farmers welfare, Govt. of India. crops apy reports web.
21. Elrys AS, Wen Y, Feng D, El-Mekawy RM, Kong M, Qin X, Lu Q, Dan X, Zhu Q, Tang S, Wu Y. Cadmium inhibits carbon and nitrogen cycling through soil microbial biomass and reduces soil nitrogen availability. *J Hazard Mater* 2025; 489:137524. doi: 10.1016/j.jhazmat.2025.137524
22. Eno CF. Nitrate production in the field by incubating the soil in polyethylene bags. *Soil Science Society of America Journal* 1960; 24(4):277-279. doi: 10.2136/sssaj1960.03615995002400040019x
23. FAO. FAOSTAT database of the food and agriculture organization of the United Nations; 2023.
24. Gbadeyan OJ, Sibiya L, Linganiso LZ, Deenadayalu N. Waste-to-energy: the recycling and reuse of sugar industry waste for different value-added products such as bioenergy in selected countries—a critical review. *Biofpr* 2024; 18(5):1639-1657. doi: 10.1002/bbb.2579
25. GOI. Price Policy for Sugarcane: Commission for Agricultural Cost and Prices. Ministry of Agriculture, New Delhi; 2024.
26. Haider MI, Mustaqeem M, Ullah F, Baloch MK, Fatima F, Luqman A, Usmani MY, Ahmad F. Impact of Sugar Cane Industry Effluents on The Environment of its Surroundings. *SciTech* 2022; 3(3):44-56.
27. Hurse JT, Connor AM. Nitrogen removal from wastewater treatment lagoons. *Water Sci Technol* 1999; 39(6):191-19. doi: 10.1016/S0273-1223(99)00139-0
28. Hussein AMSA. Sugar Industry Wastewater Treatment (Case Study: Kom Ombo Factory). Doctoral thesis, Faculty of Engineering Sanitary Engineering Department, Alexandria University, 2023.
29. Indian Standards. Indian standard methods of test for soils, part 4: grain size analysis. Bureau of Indian Standards, New Delhi, 1965; 1-43.
30. Jackson ML. Soil chemical analysis. Prentice-Hall, Englewood Cliffs, NJ, 1958; 498:183-204.
31. Kaur A, Vats S, Rekhi S, Bhardwaj A, Goel J, Tanwar RS. Physico-chemical analysis of the industrial effluents and their impact on the soil microflora. *Procedia Environ Sci* 2010; 2:595-599. doi: 10.1016/j.proenv.2010.10.065
32. Khan ANMAI, Habib MAB, Hossain MS. Chemical characteristics of different concentrations of sugarcane industry wastes for algal culture. *Int J Fish Aquat Stud* 2018; 3(2):21-24.

33. Khan MM, Yang Y, Din I. Impacts of sugar mill's effluent on soil and plant's seed germination, Punjab, Pakistan. *Earth Sci EVS* 2019; 4(3):623-637. doi: 10.25177/JESES.4.3.RA.507
34. Kolhe AS, Sarode AG, Ingale SR. Study of effluent from sugar cane industry. *Sodh Samiksha Mulyankan*, 2009; 2:303-306.
35. Kumar A, Singh D, Nainwal RC, Verma RK, Thomas T. Assessment of various soil and their properties of different districts of Uttar Pradesh. *Progress Res- Int J* 2015; 10:2757-2759.
36. Kumar DS, Srikantaswamy S. Evaluation of effluent quality of a sugar industry by using physico-chemical parameters. *Int J Adv Res Eng App Sci* 2015; 4(1):16-25.
37. Kumar V. Sugar mill effluent utilization in the cultivation of maize (*Zea mays* L.) in two seasons. *J Waste Manag* 2014; 1:408509. doi: 10.1155/2014/408509
38. Kumar S. Cultural landscape and heritage of Ayodhya-Faizabad: A geographical analysis. Ph.D. Thesis submitted to Department of Geography, Banaras Hindu University, Varanasi, Uttar Pradesh, 2018.
39. Kumar A, Kumar S, Prakash V. Effects of varied cropping systems on vertical distribution of soil properties under an alluvial Soil in Ayodhya, Uttar Pradesh. *J Indian Soc Soil Sci* 2022; 70(3):313-318. doi: 10.5958/0974-0228.2022.00033.0
40. Lad PD, Patil ST, Kadam US, Ingle PM. Effect of sugar factory effluent on physico-chemical characteristics of groundwater in surrounding area. *Pharma Innov* 2022; 11(11):2183-2189.
41. Lavanya Y, Reddy KKK, Chandana P. Industrial Waste: Impact on soil health and agriculture and its management. In: Naresh RK (eds) *Advances in agriculture sciences*. Akinik Publications, 2019; 9-34.
42. Liu J, Hua ZS, Chen LX, Kuang JL, Li SJ, Shu WS, Huang LN. Correlating microbial diversity patterns with geochemistry in an extreme and heterogeneous environment of mine tailings. *Appl Environ Microbiol* 2014; 80(12):3677-3686. doi: 10.1128/AEM.00294-14
43. Ma L, Guo C, Lü X, Yuan S, Wang R. Soil moisture and land use are major determinants of soil microbial community composition and biomass at a regional scale in northeastern China. *Biogeosciences* 2015; 12(8):2585-2596. doi: 10.5194/bg-12-2585-2015
44. Madariaga-Navarrete A, Rodríguez-Pastrana BR, Villagómez-Ibarra JR, Acevedo-Sandoval OA, Perry G, Islas-Pelcastre M. Bioremediation model for atrazine contaminated agricultural soils using phytoremediation (using *Phaseolus vulgaris* L.) and a locally adapted microbial consortium. Part B: *J Environ Sci Health* 2017; 52(6):367-375. doi: 10.1080/03601234.2017.1292092
45. Maharjan M, Sanaullah S, Razavi BS, Kuzyakov Y. Effect of land use and management practices on microbial biomass and enzyme activities in subtropical top and sub-soils. *Appl Soil Ecol* 2017; 113:22-28. doi: 10.1016/j.apsoil.2017.01.008
46. Mahmood F, Khan I, Ashraf U, Shahzad T, Hussain S, Shahid M, Abid M, Ullah S. Effects of organic and inorganic manures on maize and their residual impact on soil physico-chemical properties. *J Soil Sci Plant Nutr* 2017; 17(1):22-32. doi: 10.4067/S0718-95162017005000002
47. Malik MS. Waste-Water of Sugar Industries-A Serious Threat to the Natural Environment. *Int J Sci Technol Res* 2019; 6(3):345-353. doi: 10.32628/IJSRST1966180
48. Mohan N, Bajpai A. New technique for effluent treatment in sulphitation sugar factories. *Proc Annu Congr S Afr Sugar Technol Assoc* 2018; 91:341-349.
49. Muchuweti M, Birkett JW, Chinyanga E, Zvauya R, Scrimshaw MD, Lester JN. Heavy metal content of vegetables irrigated with mixtures of wastewater and sewage sludge in Zimbabwe: implications for human health. *Agric Ecosyst Environ* 2006; 112:41-48. doi: 10.1016/j.agee.2005.04.028
50. Munsell AH. Munsell soil color charts. Macbeth division of Kollmorgen corporation. Baltimore (Maryland), 1975.
51. Nagaraju M, Narasimha G, Rangaswamy V. Impact of sugar industry effluents on soil cellulase activity. *Internat Biodeterior Biodegrad* 2009; 63(8):1088-1092. doi: 10.1016/j.ibiod.2009.09.006
52. Nawaz MZ, Bilal M, Tariq A, Iqbal HM, Alghamdi HA, Cheng H. Bio-purification of sugar industry wastewater and production of high-value industrial products with a zero-waste concept. *Crit Rev Food Sci Nutr* 2021; 61(21):3537-3554. doi: 10.1080/10408398.2020.1802696
53. Pandey SN, Abid M, Khan MMAA. Diversity, functions, and stress responses of soil microorganisms. In: Egamberdieva D, Ahmad P (eds) *Plant Microbiome: Stress Response*. Springer, Singapore, 2018; 1-19.
54. Pansu M, Gautheyrou J. pH measurement. *Handbook of Soil Analysis: Mineralogical, Organic and Inorganic Methods*. Springer-Verlag, New York, 2006; 551-579.
55. Piper CS. Soil and plant analysis, a laboratory manual of methods for the examination of soils and the determination of the inorganic constituents of plants. *Inter Science Publication Inc, Adelaide*, 1944; 1-368.
56. Rahim MA, Mostafa MG. Impact of sugar mills effluent on environment around mills area. *AIMS Environ Sci* 2021; 8(1):86-99. doi: 10.3934/environsci.2021006
57. Reddy KV, Vijayalakshmi T, Hemalatha M, Kumar YD, Raju MN. Changes in soil enzyme activities under the influence of sugar industry effluents. *Int J Fut Sci Eng Tech* 2013; 1(1):6-10.
58. Rehman TU, Waseem H, Babar A, Haleem A, Abid R, Ahmed S, Gilbride KA, Ali M. Mitigation of Sugar Industry Wastewater Pollution: Efficiency of Lab-Scale Horizontal

- Subsurface Flow Wetlands. *Processes* 2024; 12(7):1400. doi: 10.3390/pr12071400
59. Sahu OP, Chaudhari PK. The characteristics, effects, and treatment of wastewater in sugarcane industry. *Water Qual Expo Health* 2015; 7(3):435-444. doi: 10.1007/s12403-015-0158-6
60. Samuel S, Muthukkaruppan SM. Physico-chemical analysis of sugar mill effluent, contaminated soil and its effect on seed germination of paddy (*Oryza sativa* L.). *Intern J Pharm Biolog Arch* 2011; 2(5):1469-1472.
61. Saranraj P, Stella D. Impact of sugar mill effluent to environment and bioremediation: a review. *World Appl Sci J* 2014; 30(3): 299-316. doi: 10.5829/idosi.wasj.2014.30.03.1656
62. Sarfraz S, Siddique A, Qamar F, Majeed A, Raza S. A perspective review on methodologies for treating sugar industry effluents. *Int J Environ Sci Nat Res* 2019; 21(4):117-122. doi: 10.19080/IJESNR.2019.21.556067
63. Shah A, Huang J, Han T, Khan MN, Tadesse KA, Daba NA, Khan S, Ullah S, Sardar MF, Fahad S, Zhang H. Impact of soil moisture regimes on greenhouse gas emissions, soil microbial biomass, and enzymatic activity in long-term fertilized paddy soil. *Environ Sci Eur* 2024; 36(1):120. doi: 10.1186/s12302-024-00943-4
64. Shivappa D, Puttaiah ET, Kiran BR. Physico-chemical characteristics of sugar mill effluents-current scenario in Bhadravathi Taluk Karnataka, India. *J Ind Pollut Control* 2007; 23(2):217-221.
65. Siddiqui W, Waseem M. A comparative study of sugar mill treated and untreated effluent-a case study. *Orient J Chem* 2012; 28(4):1899-1904. doi: 10.13005/ojc/280451
66. Singh JS, Singh DP, Kashyap AK. A comparative account of the microbial biomass-N and N-mineralization of soils under natural forest, grassland and crop field from dry tropical region, India. *Plant Soil Environ* 2009; 55(6):223-230. doi: 10.17221/1021-PSE
67. Singh JS, Pandey VC, Singh DP, Singh RP. Influence of pyrite and farmyard manure on population dynamics of soil methanotroph and rice yield in saline rain-fed paddy field. *Agric Ecosyst Environ* 2010a; 139(1-2):74-79. doi: 10.1016/j.agee.2010.07.003
68. Singh CP, Tiwari S, Rai PK, Singh JS. Sustainable management of paddy crop residues: effects on methanotrophs diversity and value for soil health restoration. *Land Degrad Dev* 2021; 32(14):4121-4131. doi: 10.1002/ldr.3982
69. Singh JS, Gupta VK. Soil microbial biomass: A key soil driver in management of ecosystem functioning. *Sci Total Environ* 2018; 634:497-500. doi: 10.1016/j.scitotenv.2018.03.373
70. Singh JS. Microbes: the chief ecological engineers in reinstating equilibrium in degraded ecosystems. *Agric Ecosyst Environ* 2015; 203:80-82. doi: 10.1016/j.agee.2015.01.026
71. Singh JS, Singh DP, Kashyap AK. Microbial biomass C, N and P in disturbed dry tropical forest soils, India. *Pedosphere* 2010b; 20(6):780-788. doi: 10.1016/S1002-0160(10)60068-9.
72. Singh JS. Capping methane emissions. *Sci Rep* 2016; 47: 29-30.
73. Singh JS. Crop residues management in agro-environmental sustainability. *Clim Change* 2018; 4:653-660.
74. Singh PK, Pandey N, Pratap SG. Soil Carbon Sequestration as Climate Change Mitigation: A Case Study of Change in Soil Organic Carbon Stock and Carbon Sequestration During Last Three Decades in Dewa Block of District Barabanki (UP) India. *Sristi JEEE* 2017; 1-16.
75. Singh S, Singh AK. The recent developments in sugarcane mechanization in India. *AMA-Agric Mechanization Asia Afr Latin Am* 2022; 53(3):24-30.
76. Smith JL, Paul EA. The significance of soil microbial biomass estimations. In: Bollag J-M, Stotzky G (eds) *Soil biochemistry*. Routledge, New York, 2017; 357-398.
77. Shomanova Z, Nossenko Y, Yerkibayeva M, Yessimova D, Kuspanova A, Aldasheva A, Kaimuldinova K, Safarov R. Environmental risk assessment for sustainable industrial urban development: The case of northern industrial zone of Pavlodar, Kazakhstan. *PloS one* 2025; 20(4):1-32. doi: 10.1371/journal.pone.0320835
78. Sparling GP, Whale KN, Ramsay AJ. Quantifying the contribution from the soil microbial biomass to the extractable P levels of fresh and air-dried soils. *Soil Res* 1985; 23(4):613-621. doi: 10.1071/SR9850613
79. Srivastava SC, Singh JS. Carbon and phosphorus in the soil biomass of some tropical soils of India. *Soil Biol Biochem* 1988; 20(5):743-747. doi: 10.1016/0038-0717(88)90161-7
80. Srivastava S, Chopra AK, Sharma P, Kumar V. Amendment of sugar mill wastewater irrigation on soil bihydrological properties and yield of *Vigna umguiculata* L. Walp in two seasons. *Commun. Soil Sci Plant Anal* 2017; 48(5):511-523. doi: 10.1080/00103624.2016.1254788
81. Sundar KSG, S Kittur. Methodological, temporal and spatial factors affecting modeled occupancy of resident birds in the perennially cultivated landscape of Uttar Pradesh, India. *Landsc Ecol* 2012; 27(1):59-71. doi: 10.1007/s10980-011-9666-3
82. Sun X, Sun M, Chao Y, Shang X, Wang H, Pan H, Yang Q, Lou Y, Zhuge Y. Effects of lead pollution on soil microbial community diversity and biomass and on invertase activity. *Soil Ecol Lett* 2023; 5(1):118-127. doi: 10.1007/s42832-022-0134-6
83. Tabriz SS, Mojid MA, Wyseure GCL. Irrigation suitability of north Bengal sugar mill's effluent and its impact on soil

- properties. *J Bangladesh Agri Univ* 2011; 9(2):283-290. doi: 10.22004/ag.econ.208676
84. Thuangchon S, Somboot P, Sungthong B, Punyapalakul P, Siripattanakul-Ratpukdi S, Phungsai P, Ratpukdi T. Dissolved organic matter removal from sugar mill effluent by activated carbon-catalytic ozonation: Performance, organic carbon change, system characterization, and catalyst reusability. *J Water Proc Eng* 2025; 69:106808. doi: 10.1016/j.jwpe.2024.106808
85. Tisha FN, Rahman MA, Rahman MM. Assessing the Impact of Sugar Mills Wastewater on the Surface Water Quality in Natore District. In: Miah MK, Islam MN, Alam MA, Taiyub MA, Karim MR, Choudhury BI, Hossain MM, Anwar MS, Hasan MZ, Mia MS (eds) 1st International Conference on Recent Innovation in Civil Engineering and Architecture for Sustainable Development. Atlantis Press, Netherland, 2025; 40-50.
86. Tiwari S, Singh C, Singh JS. Land use changes: a key ecological driver regulating methanotrophs abundance in upland soils. *Energy Ecol Environ* 2018; 3(6):355-371. doi: 10.1007/s40974-018-0103-1
87. Tiwari S, Singh C, Boudh S, Rai PK, Gupta VK, Singh JS. Land use change: A key ecological disturbance declines soil microbial biomass in dry tropical uplands. *J environ manag* 2019; 242:1-10. doi: 10.1016/j.jenvman.2019.04.052
88. Tripathi DM, Tripathi S. Assessment of distillery effluent irrigation on soil microbes and its bioremediation. *Octa J Environ Res* 2014; 2:303-309.
89. Vance ED, Brookes PC, Jenkinson DS. An extraction method for measuring soil microbial biomass C. *Soil Soil Biochem* 1987; 19(6):159-164. doi: 10.1016/0038-0717(87)90052-6
90. Verma DK, Mandal P, Singh P, Chand D. Quality and Treatment of Sugar Industry Effluent-A Study. *J Indian Assoc Environ Manag* 2021; 41(1):01-07.
91. Walkley A, Black IA. Critical examination of rapid method for determining organic carbon in soils, effect of variance in digestion conditions and of inorganic soil constituents. *Soil sci* 1947; 63:251-264. doi: 10.1097/00010694-194704000-00001
92. Wei L, Ge T, Zhu Z, Luo Y, Yang Y, Xiao M, Yan Z, Li Y, Wu J, Kuzyakov Y. Comparing carbon and nitrogen stocks in paddy and upland soils: Accumulation, stabilization mechanisms, and environmental drivers. *Geoderma* 2021; 398:115121. doi: 10.1016/j.geoderma.2021.115121
93. Wei L, Ge T, Zhu Z, Ye R, Penuelas J, Li Y, Lynn TM, Jones DL, Wu J, Kuzyakov Y. Paddy soils have a much higher microbial biomass content than upland soils: A review of the origin, mechanisms, and drivers. *Agric Ecosyst Environ* 2022; 326:107798. doi: 10.1016/j.agee.2021.107798
94. Wilcox LV. Electrical conductivity. *J.-Am. Water Works Assoc* 1950; 42(8):775-776.
95. Xu M, Wu WM, Wu L, He Z, Van Nostrand JD, Deng Y, Luo J, Carley J, Ginder-Vogel M, Gentry TJ, Gu B. Responses of microbial community functional structures to pilot-scale uranium in situ bioremediation. *ISME J* 2010; 4(8):1060-1070. doi: 10.1038/ismej.2010.31
96. Xu S, Geng W, Sayer EJ, Zhou G, Zhou P, Liu C. Soil microbial biomass and community responses to experimental precipitation change: A meta-analysis. *Soil Ecol Lett* 2020; 2(2):93-103. doi: 10.1007/s42832-020-0033-7
97. Xu Y, Tang H, Xiao X, Li W, Li C, Sun G, Cheng K. Effects of long-term fertilization management practices on soil microbial carbon and microbial biomass in paddy soil at various stages of rice growth. *Rev Bras Ciênc Solo* 2018; 42: e0170111. doi: 10.1590/18069657rbcsc20170111
98. Xu Z, Zhang Z, Peng S, Yuan Y, Wang X. Influences of lithium on soil microbial biomass, bacterial community structure, diversity, and function potential. *Environ Tech Innov* 2023; 32:103361. doi: 10.1016/j.eti.2023.103361
99. Zhu Z, Ge T, Hu Y, Zhou P, Wang T, Shibistova O, Guggenberger G, Su Y, Wu J. Fate of rice shoot and root residues, rhizodeposits, and microbial assimilated carbon in paddy soil-part 2: turnover and microbial utilization. *Plant Soil* 2017; 416(1):243-257. doi: 10.1007/s11104-017-3210-4