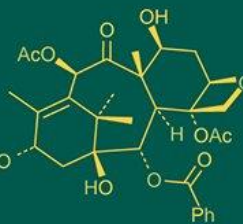
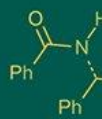
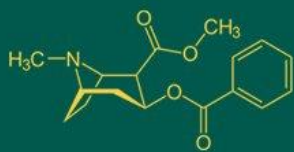


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Influence of liquified humic acid application on chemical and biological characteristics of *Vertisol* of Chhattisgarh

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Abstract

A field experiment was conducted during the *Kharif* seasons of 2023 and 2024 at the Research cum Instructional Farm, Indira Gandhi Krishi Vishwavidyalaya, Raipur (C.G), to evaluate the effect of foliar application of liquified humic acid on chemical and biological characteristics of *Vertisol* under rice cultivation. The experiment comprised of ten treatments arranged in a randomized block design with varying combinations of RDF (120:60:40 NPK kg ha⁻¹), 80% RDF, root dipping, and foliar sprays of liquified humic acid (2.5-10 ml L⁻¹). The results revealed that the combined application of liquified humic acid and inorganic fertilizers had no significant effect on soil pH, electrical conductivity (EC), organic carbon, available macronutrients (N, P, and K), and micronutrients (Fe, Mn, Cu, and Zn) during both years of study. However, the highest soil microbial biomass carbon (SMBC) (213.09 µg C g⁻¹ soil) and soil microbial biomass nitrogen (SMBN) (52.58 µg N g⁻¹ soil) were recorded under applied treatment @ RDF + liquified Humic Acid FS @ 10 ml L⁻¹ (T₆) followed by @ RDF + liquified Humic Acid FS @ 5 ml L⁻¹ (T₅), both treatments were statistically at par with the remaining treatments. Overall, the findings indicate that the integration of inorganic fertilizers with foliar-applied humic acid did not significantly influence soil chemical properties or microbial biomass in *Vertisols* under rice cultivation during the study period.

Keywords: Liquified humic acid, foliar spray, inorganic fertilizer, soil microbial biomass carbon and nitrogen, organic carbon

1. Introduction

Maintaining and enhancing soil fertility is increasingly important due to the growing complexity of nutrient-related constraints. Organic matter plays a critical role in improving soil physical, chemical, and biological properties (Stevenson, F. J. 1994) ^[1]. Reduced soil organic matter also limits the availability of secondary and micronutrients. Humic and non-humic substances reflect the quality of soil organic matter, with humic substances formed through microbial, enzymatic, and chemical decomposition of plant and animal residues (Flaig, 1975) ^[2]. Their chemically reactive functional groups directly and indirectly influence plant growth.

Humic substances, comprising humins, humic acids, and fulvic acids, represent the largest fraction of soil organic matter and play a dominant role in soil conditioning and plant development. Physically, they improve soil structure and water-holding capacity; chemically, they function as nutrient adsorption and retention complexes, keeping essential elements available to plant roots (Randhawa and Broadbent, 1965) ^[3] and biologically, they stimulate microbial activity by serving as a source of nutrients and energy.

Humic acid is recognized as one of the most significant natural organic compounds in agriculture (Ampong *et al.*, 2022) ^[4]. Derived from the decomposition of plant and animal materials, it improves soil structure, texture, microbial activity, and water-retention capacity (Nardi *et al.*, 2021; Shah *et al.*, 2018) ^[5, 6] enhances micronutrient availability through chelation, and facilitates nutrient uptake (Yang *et al.*, 2021) ^[7], while reducing the absorption of toxic heavy metals (Wu *et al.*, 2017) ^[8]. Furthermore, humic acid stimulates growth-regulating hormones such as auxins and cytokinin's, improves photosynthesis, nutrient metabolism, and stress tolerance (Canellas *et al.*, 2020; Jindo *et al.*, 2020) ^[9, 10].

It also enhances membrane permeability, mineral transport, protein synthesis, enzymatic activity, and beneficial microbial populations (Khaled *et al.*, 2011) ^[11]. Given these multifaceted benefits, humic acid represents a promising tool for improving soil fertility, nutrient-use efficiency, and crop productivity. Thus, evaluating the influence of liquified humic acid on rice performance under *Vertisol* conditions of Chhattisgarh is essential for developing sustainable nutrient management strategies.

2. Materials and Methods

The experimental site was located at the Research cum Instructional Farm, Indira Gandhi Krishi Vishwavidyalaya, Raipur (C.G.), situated in the central agro-climatic zone of Chhattisgarh. The field experiment was carried out during the *Kharif* seasons of 2023 and 2024 on rice crop to study the effect of foliar application of liquified humic acid on the chemical and biological properties of *Vertisol* under rice cultivation (cv. Rajeshwari). The field experiment was laid out in a Randomized Block Design (RBD) with ten treatments replicated three times: T₁-Control (N₀:P₀: K₀), T₂-RDF (120:60:40), T₃-RDF + Root dipping with liquified Humic Acid (1ml L⁻¹), T₄-RDF + liquified Humic Acid FS @ 2.5 ml L⁻¹, T₅-RDF + liquified Humic Acid FS @ 5 ml L⁻¹, T₆-RDF + liquified Humic Acid FS @ 10 ml L⁻¹, T₇-80% RDF + Root dipping with liquified Humic Acid (1ml L⁻¹), T₈-80% RDF + liquified Humic Acid FS @ 2.5 ml L⁻¹, T₉-80% RDF + liquified Humic Acid FS @ 5 ml L⁻¹, T₁₀-80% RDF + liquified Humic Acid FS @ 10 ml L⁻¹. The experimental soil was a *Vertisol* (locally known as Kanhar), classified as fine montmorillonitic, hyperthermic Ustic Chromustert, deep and clayey in texture, and neutral to alkaline in reaction. Rice variety Rajeshwari was used as the test crop. The Soil pH was determined in 2.5:1 water-soil suspension (Piper, 1966) ^[12] soil suspension after pH determination was stored overnight and EC of the supernatant liquid was determined by Solu-bridge as described by Black ^[13]. The organic carbon was determined by Walkley and Black's rapid titration method. Available nitrogen was estimated by alkaline potassium permanganate method, available phosphorus by Olsen's method (1954) ^[14] method, and available potassium was determined by neutral normal ammonium acetate extractant and detected by Flame photometer as described by Hanway and Heidal ^[15]. Available micronutrients (Fe, Zn, Mn, Cu) were extracted using DTPA (Lindsay and Norvell, 1978) ^[16]. Soil microbial biomass carbon (SMBC) and nitrogen (SMBN) were determined by the chloroform fumigation-extraction method (Vance *et al.*, 1987; Brookes *et al.*, 1985) ^[17, 18]. The calculation for microbial biomass of soil:

1. Microbial biomass carbon in soil was calculated as

SMBC =
$$\frac{(CF-CUF) \mu g \text{ g}^{-1} \text{ dry soil}}{0.45}$$

Where

CF = Total weight of extractable C in a fumigated sample (μg C ml⁻¹)

CUF = Total weight of extractable C in Non-fumigated sample (μg C ml⁻¹)

2. Microbial biomass nitrogen in soil was calculated as

SMBN =
$$\frac{Nf-Nuf}{KEC}$$

Nf = Concentration of N in fumigated sample

Nuf = Concentration of N in unfumigated sample

KEC = Efficiency of extraction of MBN & the value is 0.68

3. Results and Discussion

3.1 Soil chemical properties

Application of inorganic fertilizer either alone or in combination with liquified humic acid were not significantly influenced soil pH, EC, and organic carbon (OC) (Table-1) during *Kharif* 2023 and 2024. The stability in pH may be attributed to the buffering capacity of humic substances (Tahir *et al.*, 2011) ^[19]. Slightly higher EC values were observed under 100% RDF (120:60:40 NPK) treatments compared to the control, the differences were non-significant and may be due dissolution of fertilizers in the soil solution, releasing higher concentrations of soluble salts such as nitrates, phosphates, potassium, and sulfates. The results were in close conformity to those obtained by Sathiya Bama *et al.* (2003) ^[20], while higher OC was recorded under @ RDF + liquified humic acid FS @ 10 ml L⁻¹ (T₆) and the lowest in the control. Pooled data across both years confirmed the non-significant influence of liquified humic acid and inorganic fertilizer combinations on soil available nitrogen, phosphorus and potassium. This may be attributed due to short-term experimentation, adequate initial soil fertility, rapid plant uptake, and the indirect role of humic acid in improving nutrient use efficiency rather than directly contributing nutrients. No variation in the Soil N was also reported by Layek *et al.* (2018) ^[21] and Banjare *et al.* (2022) ^[22]. Application of inorganic fertilizer either alone or in combination with liquified humic acid were not significantly influenced available Fe, Zn, Cu, and Mn may be due to the foliar application of humic acid, which has minimal interaction with soil, and the strong buffering and fixation of micronutrients in soil, limiting changes in their availability. These findings are supported by earlier studies conducted by Clapp *et al.* (1999) ^[23], (Stumm and Morgan, 1970) ^[24] and Sharif *et al.* (2002) ^[25].

Table 1: Effect of liquified humic acid on soil pH, EC (dSm⁻¹) and organic carbon (%) after harvest of crop during 2023 and 2024.

Treatment Details	Soil pH (1:2.5)			Electrical conductivity (dSm ⁻¹)			Organic carbon (%)		
	2023	2024	Pooled	2023	2024	Pooled	2023	2024	Pooled
T ₁	7.43	7.41	7.42	0.20	0.21	0.20	0.45	0.47	0.46
T ₂	7.45	7.46	7.46	0.25	0.33	0.28	0.52	0.54	0.53
T ₃	7.50	7.49	7.49	0.29	0.32	0.27	0.55	0.53	0.54
T ₄	7.47	7.48	7.48	0.27	0.31	0.32	0.54	0.53	0.54
T ₅	7.45	7.44	7.44	0.30	0.29	0.27	0.54	0.51	0.53
T ₆	7.47	7.46	7.47	0.32	0.30	0.32	0.55	0.53	0.54
T ₇	7.43	7.41	7.42	0.24	0.28	0.31	0.50	0.52	0.51
T ₈	7.41	7.51	7.46	0.28	0.22	0.25	0.48	0.45	0.46
T ₉	7.40	7.38	7.39	0.29	0.28	0.29	0.48	0.48	0.48
T ₁₀	7.42	7.41	7.42	0.31	0.26	0.27	0.49	0.51	0.50
SEm (±)	0.04	0.06	0.04	0.03	0.05	0.02	0.02	0.03	0.02
C.D. (P = 0.05)	NS	NS	NS	NS	NS	NS	NS	NS	NS

Table 2: Effect of liquified humic acid on soil available macronutrients after harvest of crop during 2023 and 2024.

Treatment Details	Soil available nitrogen (Kg ha ⁻¹)			Soil available phosphorus (Kg ha ⁻¹)			Soil available potassium (Kg ha ⁻¹)		
	2023	2024	Pooled	2023	2024	Pooled	2023	2024	Pooled
T ₁	209.67	207.82	208.75	15.19	15.02	15.11	483.03	481.70	482.36
T ₂	214.42	212.73	213.57	18.24	18.19	18.21	489.05	486.82	487.94
T ₃	214.28	212.69	213.49	18.20	18.13	18.16	487.97	486.57	487.27
T ₄	214.06	212.34	213.20	17.83	17.63	17.73	487.57	485.43	486.50
T ₅	213.69	211.82	212.76	17.53	17.42	17.48	486.49	485.17	485.83
T ₆	213.32	210.53	211.93	17.44	17.31	17.38	486.19	484.71	485.45
T ₇	212.84	210.48	211.66	16.90	16.85	16.87	485.42	484.22	484.82
T ₈	212.47	210.31	211.39	16.81	16.79	16.80	485.28	483.94	484.61
T ₉	212.59	209.94	211.26	16.79	16.66	16.73	484.16	483.09	483.63
T ₁₀	211.65	209.13	210.39	16.66	16.59	16.62	483.67	482.88	483.28
Sem (±)	3.51	3.15	3.18	0.86	0.96	0.62	7.86	9.24	7.08
C.D. (P = 0.05)	NS	NS	NS	NS	NS	NS	NS	NS	NS

Table 3: Effect of liquified humic acid on soil available micronutrients after harvest of crop during 2023 and 2024.

Treatment Details	Soil available iron mg kg ⁻¹			Soil available manganese (mg kg ⁻¹)			Soil available copper (mg kg ⁻¹)			Soil available zinc (mg kg ⁻¹)		
	2023	2024	Pooled	2023	2024	Pooled	2023	2024	Pooled	2023	2024	Pooled
T ₁	17.15	17.04	17.10	9.38	9.29	9.34	0.64	0.62	0.63	1.07	1.03	1.05
T ₂	18.94	18.45	18.70	10.38	10.21	10.30	0.72	0.69	0.70	1.27	1.25	1.26
T ₃	18.88	18.25	18.56	10.31	10.18	10.25	0.70	0.69	0.70	1.25	1.23	1.24
T ₄	18.85	18.19	18.52	10.12	10.08	10.10	0.69	0.68	0.69	1.25	1.23	1.24
T ₅	18.60	18.17	18.38	10.09	9.93	10.01	0.69	0.68	0.68	1.24	1.21	1.23
T ₆	18.49	18.12	18.30	10.00	9.86	9.93	0.68	0.67	0.67	1.22	1.19	1.21
T ₇	17.92	17.84	17.88	9.99	9.96	9.98	0.67	0.66	0.66	1.20	1.18	1.19
T ₈	17.69	17.53	17.61	9.86	9.81	9.84	0.67	0.65	0.66	1.18	1.14	1.16
T ₉	17.56	17.30	17.43	9.75	9.69	9.72	0.66	0.64	0.65	1.16	1.13	1.15
T ₁₀	17.44	17.14	17.29	9.67	9.61	9.64	0.65	0.64	0.64	1.15	1.11	1.13
SEm (±)	1.07	0.89	0.93	0.46	0.45	0.44	0.12	0.14	0.11	0.12	0.14	0.11
C.D. (P = 0.05)	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS

3.2 Soil biological properties

3.2.1 Soil microbial biomass carbon

The combined application of inorganic fertilizer and foliar-applied liquified humic acid had a non-significant effect on soil microbial biomass carbon (SMBC) and nitrogen (SMBN) during both of the years as presented in Table-4. The pooled value of SMBC ranged from 180.68 to 213.09 $\mu\text{g C g}^{-1}$ soil, with the highest recorded under @ RDF + liquified Humic Acid FS @ 10 ml L⁻¹ (T₆), followed by @ RDF + liquified Humic Acid FS @ 5 ml L⁻¹ (T₅), and these both treatments were statistically at par with the remaining treatments. The increase in SMBC may be attributed to the positive effect of foliar-applied humic acid on root growth and root exudation, which provide readily available carbon substrates for soil microorganisms, thereby enhancing microbial growth and activity. The lowest (180.68 $\mu\text{g C g}^{-1}$ soil) soil microbial biomass carbon (SMBC) was noticed under unfertilized plot (T₁). Similar findings have been documented by Sellamuthu and Govindaswamy (2003) [26], Li *et al.* (2019) [27], and Sarangthem *et al.* (2023) [28].

3.2.1 Soil microbial biomass nitrogen (SMBN)

The pooled SMBN values ranged from 35.62 to 52.58 $\mu\text{g N g}^{-1}$ soil, with the maximum under @ RDF + liquified Humic Acid FS @ 10 ml L⁻¹ (T₆), followed by @ RDF + liquified Humic Acid FS @ 5 ml L⁻¹ (T₅) and found at par with rest of the treatments. The foliar application of humic acid along with RDF enhanced SMBN by improving root growth, nutrient uptake, and root exudation, which enriched the rhizosphere environment and supported microbial proliferation. The lowest SMBN (35.62 $\mu\text{g N g}^{-1}$ soil) was observed under the unfertilized plot (T₁). Similar findings have been documented by Sellamuthu and Govindaswamy (2003) [26], Li *et al.* (2019) [27], and Sarangthem *et al.* (2023) [28] and El-Ghamry *et al.* (2009) [31]. The lowest (35.62 $\mu\text{g N g}^{-1}$ soil) soil microbial biomass nitrogen (SMBN) was noticed under unfertilized plot (T₁). These results indicate that while humic acid application slightly enhanced microbial biomass, the effects were not statistically significant.

Table 4: Effect of liquified humic acid on soil biological characteristics of rice crop at 60 DAT during 2023 and 2024.

Treatment Details		Soil microbial biomass carbon ($\mu\text{g C/g soil}$)			Soil microbial biomass nitrogen ($\mu\text{g N/g soil}$)		
		2023	2024	Pooled	2023	2024	Pooled
T ₁	Control	179.62	181.75	180.68	34.40	36.84	35.62
T ₂	RDF (120:60:40)	207.40	209.54	208.47	48.23	51.73	49.98
T ₃	RDF + RD with liquified Humic Acid (1 ml L ⁻¹)	207.65	209.78	208.72	48.83	51.13	49.98
T ₄	RDF + liquified Humic Acid FS @ 2.5 ml L ⁻¹	208.55	210.68	209.61	49.40	53.77	51.58
T ₅	RDF + liquified Humic Acid FS @ 5 ml L ⁻¹	209.47	211.60	210.53	50.67	52.90	51.78
T ₆	RDF + liquified Humic Acid FS @ 10 ml L ⁻¹	212.03	214.16	213.09	51.53	53.63	52.58
T ₇	80% RDF + RD with liquified Humic Acid (1 ml L ⁻¹)	194.69	196.82	195.76	44.37	47.03	45.70
T ₈	80% RDF + liquified Humic Acid FS @ 2.5 ml L ⁻¹	195.26	198.39	196.82	44.82	47.23	46.03
T ₉	80% RDF + liquified Humic Acid FS @ 5 ml L ⁻¹	198.24	200.37	199.30	45.97	48.20	47.08
T ₁₀	80% RDF + liquified Humic Acid FS @ 10 ml L ⁻¹	199.47	201.61	200.54	46.93	48.50	47.72
	SEm ($\pm$)	6.36	6.42	6.39	3.75	3.71	3.20
	C.D. (P = 0.05)	NS	NS	NS	NS	NS	NS

4. Conclusion

The study concluded that the application of inorganic fertilizers either alone or in combination with foliar-applied liquified humic acid did not significantly influence soil chemical and biological properties during Kharif 2023 and 2024. Soil pH, EC, organic carbon, available nutrients, micronutrients, soil microbial biomass carbon (SMBC), and soil microbial biomass nitrogen (SMBN) remained statistically non-significant across treatments. However, comparatively higher values of SMBC and SMBN were observed under @ RDF + liquified Humic Acid FS @ 10 ml L⁻¹ (T₆). The non-significant response may be attributed to the short-term nature of the experiment, adequate initial soil fertility, and the indirect role of humic acid in improving nutrient use efficiency.

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