



## Moisture sensitivity alteration of soil organic carbon and nitrogen mineralization in Vertisol

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### ABSTRACT

Uneven distribution and intensity of rainfall in subtropical semi-arid Vertisol creates different hydro-climatic condition due to its mineralogy and expansive type of clays. Indeed, information on impact of moisture levels on soil C- and N-mineralization in presence of residue and recommended fertilization is scarce. Thus, this incubation experiment was conducted at ICAR-Indian Institute of Soil Science, Bhopal during 2018–19 with four moisture, two fertilizer, and two residue levels to evaluate the effects of fertilization and residue incorporation on moisture sensitivity of soil C- and N-mineralization. Application of fertilizer induced positive priming effect for CO<sub>2</sub>-C and increased by ~28%. Residue- and fertilizer × residue accelerated the C<sub>cum</sub> by ~108 and 125%, respectively. Residue application caused initial N-immobilization. A significant interaction was observed among fertilizer, residue and moisture content for both C- and N-mineralization. C-mineralization was amplified with increase in soil moisture content from 24 to 40% moisture levels, but decreased at 48% moisture level. Irrespective of fertilization moisture sensitivity of C and N mineralization was reduced by residue incorporation. But, fertilization led to highest moisture sensitivity of N-mineralization. Thus, residue incorporation could be recommended as obligatory practice in sub-tropical Vertisol to decrease its moisture sensitivity and to maintain positive C balance and soil health.

**Keywords:** Crop residue, C- and N- mineralization, Fertilizer, Moisture sensitivity, Rate kinetics, Vertisol

Typically, in central and Deccan region of India, the dominance of swell-shrink clay creates challenges for farming community. The expanding type smectitic clay minerals in Indian Vertisol have high specific surface area and surface negative charges, which resulted adsorption of organic molecules, nutrient fixation and reduced enzymatic activities (Sarkar *et al.* 2017, Roy *et al.* 2019, Ghosh *et al.* 2020, Sarkar *et al.* 2021a). Indeed, synergistic relation between soil temperature and heterotrophic respiration has been well established; in most cases they follow Arrhenius or Van't Hoff laws (Guntinas *et al.* 2013). However, influence of soil moisture on soil health and nutrient dynamics are not well explained and validated in large scale.

Carbon (C) and nitrogen (N) dynamics are having major significance in terrestrial ecosystems. Proper C:N ratio regulate soil functionality. Indeed, C and N are often supplanted through crop residues, manures and fertilizers for soil health and fertility restoration (Moharana *et al.* 2019, Ghosh *et al.* 2019). Though residue incorporation increases native soil organic C (SOC) and even provide good quantity of other nutrients during decomposition (Biswas *et al.* 2017, Ghosh *et al.* 2018, Moharana *et al.* 2019), but addition of fertilizer hastens the rate of decomposition (known as priming effect) (Moyano *et al.* 2013). On spatial and temporal scale, soil moisture regulates residue decomposition through alterations of solute, soil temperature and oxygen diffusion (Dijkstra and Cheng 2007, Saha *et al.* 2020, Sarkar *et al.* 2021b). Nutrient cycling is also subject to non-linear shifts in dynamics with changing hydro-climatic conditions (Biswas *et al.* 2021). Fertilization and its interaction with crop residues can incite C and N mineralization or immobilization relying upon other soil conditions, like soil moisture content (Muhammad *et al.* 2011, Saha *et al.* 2021). Better understanding on impact of different levels of soil moisture under fertilization and crop residue incorporation on C and N mineralization would be helpful to manage the soil C cycle. Therefore, our objective

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was to assess the impact of residue incorporation on soil moisture sensitivity of C and N mineralization in presence or absence of fertilizer.

## MATERIALS AND METHODS

Present study was carried out at ICAR- Indian Institute of Soil Science (23.3072° N, 77.4050° E), Bhopal, India during 2018–19. Soil was sampled from soybean (*Glycine max* L.) -wheat (*Triticum aestivum* L.) cropping system. Soil was clayey in texture (20%, 22%, and 58% of sand, silt and clay, respectively) and non-saline (0.26 dS/m EC) with slightly alkaline (pH- 7.9) in reaction, which had 5.5 g/kg SOC, 75.1 mg/kg NH<sub>4</sub><sup>+</sup> - N, 46.2 mg/kg NO<sub>3</sub><sup>-</sup> - N, 19.7 mg/kg Olsen's P, and 264 mg/kg exchangeable K. Bulk density and water holding capacity (w/w) of soil was correspondingly 1.46 Mg/m<sup>3</sup> and 56.7%. Soil of the region belongs to Vertisol (*Typic Haplustert*) and contained smectitic clay minerals. Processed soil samples were divided, where one portion treated with crop residues at 10 tons/ha equivalent (R1) and another remain devoid of residues (R0). Then, R1 and R0 were treated with (F1; 150: 60: 60:: N: P<sub>2</sub>O<sub>5</sub>: K<sub>2</sub>O kg/ha) and without (F0) fertilizer. Carbon and N-mineralization were studied at four different moisture levels, viz. 24% (M1), 32% (M2), 40% (M3) and 48% (M4) (w/w) for 60 days at 25 °C. Total 16 treatment combinations were tested in this investigation in triplicates.

The C-mineralization study was conducted with 100 g equivalent dry soil at different moisture levels in 500 ml Erlenmeyer flask for 60 days. Evolved CO<sub>2</sub> was trapped by 20 ml 0.5 N NaOH and CO<sub>2</sub> flux was measured by using Eq. 1.

$$\text{CO}_2 - \text{C evolved (mg/kg)} = (A - B) \times N \times 6 \quad (1)$$

where, A and B are volume of HCl consumed for titrating NaOH in control (without soil) and in soils, N is normality of HCl and 6 is equivalent weight of C. On the other hand, N-mineralization was conducted separately with destructive method of sampling. Equivalent to 100 g oven dry soil were incubated in plastic vessel at respective moisture levels for 60 days at 25°C. Total inorganic N (TIN; NH<sub>4</sub><sup>+</sup>-N + NO<sub>3</sub><sup>-</sup>-N) were measured by steam distillation after 2M KCl extraction (Keeney and Nelson 1982).

The rate kinetics of C and N mineralization with time was determined by using the model developed by Stanford and Smith (1972) (Eq. 2).

$$X_t = X_0(1 - e^{-k_X t}) \quad (2)$$

where, X<sub>0</sub> represents potentially mineralizable C or N; X<sub>t</sub> is the pool of C or N mineralized at time t, with mineralization rate k<sub>X</sub>. Fertilizer and residue incorporation causes priming effect. Hence, it was not assumed that the interaction of moisture × residue, moisture × fertilization and moisture × residue × fertilization to be additive. Net C- or N-mineralized from soil due to wheat straw (Eq. 3), fertilizer (Eq. 4) and fertilizer+residue (Eq. 5) addition was calculated as;

$$X_{\text{residue amended soils}} - X_{\text{control soils}} = M_i (X_{\text{cum}}^{\text{F0R1}} - X_{\text{cum}}^{\text{F0R0}}) \quad (3)$$

$$X_{\text{fertilized soils}} - X_{\text{control soils}} = M_i (X_{\text{cum}}^{\text{F1R0}} - X_{\text{cum}}^{\text{F0R0}}) \quad (4)$$

$$\frac{X_{\text{residue+fertilized soils}} - X_{\text{control soils}}}{X_{\text{cum}}^{\text{F0R0}}} = M_i (X_{\text{cum}}^{\text{F1R1}} - X_{\text{cum}}^{\text{F0R0}}) \quad (5)$$

where, M<sub>i</sub> is i<sup>th</sup> moisture level, X<sub>cum</sub> is cumulative C- (CO<sub>2</sub> evolution) or N- mineralization. Among linear, parabolic, logarithmic, semi-logarithmic, semi-parabolic, exponential regression equations, best R<sup>2</sup> values were obtained for exponential regression equations. For each of the moisture X<sub>cum</sub> after 60 days were fitted to the following equations (Eq. 6 and 7).

$$X_{\text{cum}} = ae^{\theta_X M} \quad (6)$$

$$k_X = ae^{\theta_X M} \quad (7)$$

where, X is either C or N, M is moisture content, 'a' is a fitting parameter (referring to as the soil respiration rate at 0% soil moisture content), θ<sub>X</sub> is also a fitting parameter commonly referred to as the moisture coefficient.

All data were analysed by using Analysis of Variance (ANOVA) for a factorial completely randomized design. Tukey's Honestly Significant Difference test was used as a post hoc mean separation test (P<0.05) using SAS 9.1 (SAS Institute, Cary, North Carolina, USA). A simple t-test was performed to evaluate the effect of fertilization and residue incorporation.

## RESULTS AND DISCUSSION

*Effects on C- and N-mineralization:* Irrespective of moisture content, C<sub>cum</sub> from residue (F0R1) and fertilizer+residue (F1R1) treatments were higher than the fertilizer treatments (F1R0) (Fig 1), which could be due to abundant quantity of substrates supplied through residue incorporation and higher microbial activities. The F1R1 had even higher C<sub>cum</sub> than F0R1, which is simple indication of 'priming effect' (Moyano *et al.* 2013). The F1R0, F0R1, and F1R1 correspondingly caused ~28, 108 and 125% higher C<sub>cum</sub> than the control (F0R0) soils. Surprisingly, F1R0 showed a plateau after 25 days after incubation (Fig 1), which might be due to diminishing of labile C. However, F0R1 led to net immobilization of N initially, while F1R0 significantly increased (~46%) N<sub>cum</sub>. The F0R1 and F1R1 also enhance the N<sub>cum</sub> by ~6 and 41%, respectively. It can be assumed that fertilizer addition with or without residue helps to maintain soil C-N, C-P, C-S stoichiometry, which provides a better niche for microbial interaction and provides sufficient nutrients for plant growth (Gutiñías *et al.* 2013, Ghosh *et al.* 2018). Overall, higher quantity of C<sub>cum</sub> and N<sub>cum</sub> was recorded at M1 and M2, which might be due to closeness of these moisture levels to the field capacity of the soil.

*Effects on rate constants of C- and N-mineralization:* The k<sub>C</sub> and k<sub>N</sub> were regularly higher up to 25 days of incubation (Table 1), which is an indicator for consumption of labile C and nutrients at earliest. The k<sub>C</sub> was decreased by ~27, 14, 17 and 17% under F0R0, F1R0, F0R1 and F1R1, respectively. The k<sub>N</sub> was hastened by ~35 and 81%

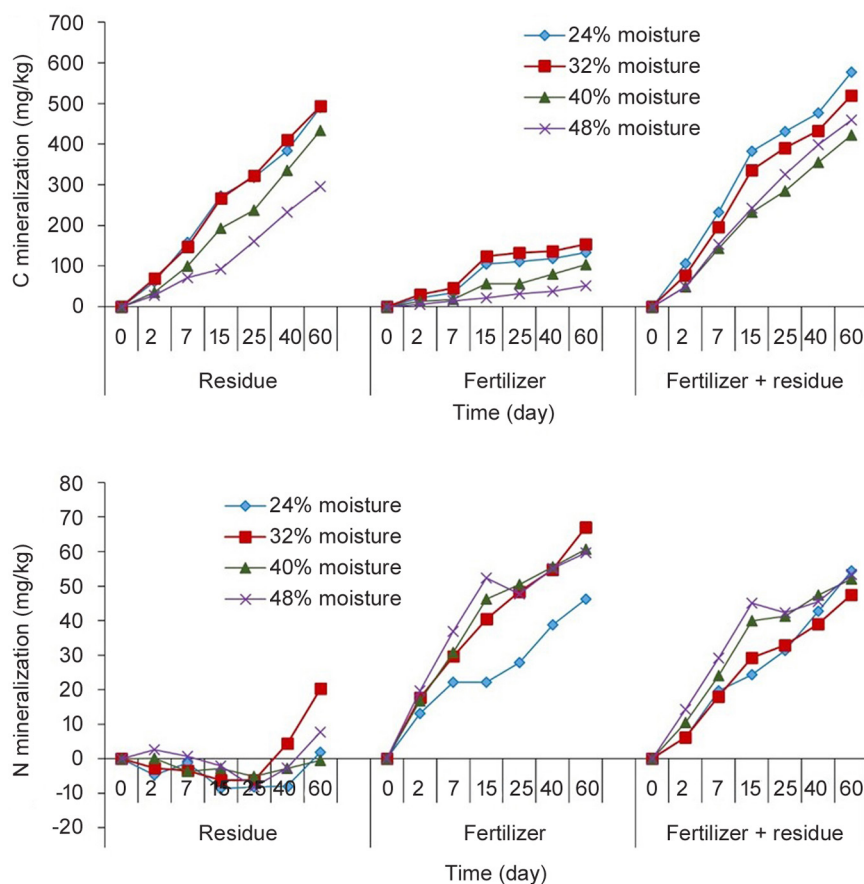


Fig 1 Impact of residue (F0R1), fertilizer (F1R0) and fertilizer+residue (F1R1) application on C- and N-mineralization over control (F0R0) in Vertisol.

due to fertilization under R0 and R1, respectively (Table 1), while residue addition decreased  $k_N$  by ~19%. The contradictory results of  $k_C$  and  $k_N$  was due to microbial assimilation of N. In the presence of available C, higher N availability stimulates the growth of r-strategists (Blagodatskaya *et al.* 2007) microbial population. The N assimilation by newly flourished microbial cells resulted in lower values of  $k_N$ , whereas  $k_C$  was higher. Upon increasing soil moisture up to 40%, water entered the interlayers of expanding clay and causes swelling of clay. During the entry of water in the interlayers, residue C, labile C, soluble C, nutrients and desorbed enzymes also enter into the interlayers; thus, their availability to microbes fall and lower the  $k_C$  value (Kolman *et al.* 2014, Biswas *et al.* 2021).

*Effects on moisture sensitivity:*

The t-test statistics revealed that exponential regression equations with  $k_C$  and  $k_N$  as dependent variables were significant in all cases (Table 2). Hence,  $\theta_C$  and  $\theta_N$  obtained from Eq. 6 and Eq. 7 was used to estimate the moisture coefficient of the reaction in each case.

Table 1 Impact of moisture, residue and fertilizer on rate kinetics of C- ( $k_C$ ) and N-mineralization ( $k_N$ ) after 25 days ( $k_{C25}$  and  $k_{N25}$ ) and after 60 days ( $k_{C60}$  and  $k_{N60}$ ) in a sub-tropical Vertisol

	$k_{C25}$ (per day)				$k_{C60}$ (per day)			
	R0		R1		R0		R1	
	F0	F1	F0	F1	F0	F1	F0	F1
24% moisture	0.101a	0.084b	0.093a	0.106a	0.074a	0.072a	0.059a	0.075a
32% moisture	0.094b	0.082b	0.086b	0.091b	0.054d	0.062b	0.049b	0.062b
40% moisture	0.084c	0.081b	0.073c	0.083c	0.060c	0.054c	0.042c	0.055c
48% moisture	0.103a	0.098a	0.083b	0.083c	0.068b	0.063b	0.044bc	0.057c
Mean F <sup>#</sup>	0.095A	0.086B	0.084B	0.091A	0.064A	0.063A	0.049B	0.062A
Mean R <sup>§</sup>	0.091A		0.087B		0.064A		0.056B	
	$k_{N25}$ (per day)				$k_{N60}$ (per day)			
	R0		R1		R0		R1	
	F0	F1	F0	F1	F0	F1	F0	F1
24% moisture	0.068c	0.104b	0.067c	0.081b	0.039b	0.043c	0.028b	0.038c
32% moisture	0.083b	0.107b	0.076b	0.088b	0.043a	0.052b	0.021c	0.047b
40% moisture	0.093a	0.114a	0.091a	0.104a	0.042a	0.061a	0.035a	0.056a
48% moisture	0.055d	0.117a	0.085a	0.099a	0.035b	0.061a	0.025b	0.055a
Mean F <sup>#</sup>	0.075B	0.111A	0.080B	0.093A	0.040B	0.054A	0.027B	0.049A
Mean R <sup>§</sup>	0.093A		0.087B		0.047A		0.038B	

<sup>#</sup>Mean for fertilization (F); <sup>§</sup> mean for residue (R); Means with similar lower-case letters within a column are not significantly different at  $P < 0.05$  according to Tukey's HSD test. Significant ( $P < 0.05$ ) effects of fertilization and residue incorporation for C- and N-mineralization rates (mean of all treatments) are denoted by different upper-case letters in the last two rows, respectively.

Table 2 Effect of fertilization and residue incorporation on moisture sensitivity of C-, and N- mineralisation

	Moisture sensitivity of C ( $\times 10^{-3}$ )		Moisture sensitivity of N ( $\times 10^{-3}$ )	
	$\theta_c$ from $C_{cum}$	$\theta_c$ from $k_C$	$\theta_c$ from $N_{cum}$	$\theta_c$ from $k_N$
F0R0	12a	-2a	-2 a	- 5 b
F1R0	2 b	- 7a	1 a	15.2 a
F0R1	- 4 c	-13b	- 2 a	2 b
F1R1	- 1c	-12b	-1 a	15.9 a

Means with similar lower-case letters within a column are not significantly different at  $P < 0.05$  according to Tukey's HSD test.

The  $\theta_c$  value of  $C_{cum}$  was correspondingly  $\sim 6.5$  and  $1.9$  times lower under F0R1 than F0R0 and F1R0. Similarly,  $\theta_c$  value in F1R1 was  $6$  and  $1.7$  times lower than F0R0 and F1R0, respectively. However, F1R0 led to  $\sim 250\%$  lower  $\theta_c$  value over control (F0R0). Hence, residue incorporation without fertilization had lesser impact on  $\theta_c$  than fertilization without residue incorporation. Contrastingly with  $C_{cum}$ , fertilization led to highest moisture sensitivity of  $N_{cum}$ . The negative sign of moisture coefficients indicates that increasing soil moisture steadily up to saturation would lead to decline in sensitivity of respiration to moisture. Our results were also aligned with the findings of Xu *et al.* (2004) and Biswas *et al.* (2021). However, greater moisture sensitivity of C mineralization was observed under F0R0, which suggested the essentiality of soil water availability to SOC mineralization in Vertisol. On the other hand, residue amendment provides easily available, labile C sources to soil microbes and decrease its sole dependency on moisture for substrate availability.

*Interactive effects of residue, fertilizer and moisture:* Three-way ANOVA analysis revealed that across the soil moisture regimes, the residue  $\times$  fertilizer interaction was not significant in determining  $C_{cum}$  and  $N_{cum}$ . But the interaction effect of residue  $\times$  moisture and fertilizer  $\times$  moisture on  $C_{cum}$  and  $N_{cum}$  were significant ( $P < 0.05$ ). Most interestingly, residue  $\times$  fertilizer  $\times$  moisture interaction significantly influenced  $C_{cum}$  and  $N_{cum}$ . These findings could be justified by the concept of crop residue composition and their interactions with other experimental variables. Wheat residue generally contains higher quantity of cellulose than the lignin and fertilization helped in supply of nutrients and may provide abundant C:N ratio; while moisture modified the soil physical environment in favour of microbes; these three-way positive effects were responsible for such types of results (Dijkstra and Cheng 2007, Sarkar *et al.* 2017, 2021b).

It is important for the reliable prediction of C and N dynamics under climate change to understand the sensitivity of C and N mineralization to soil moisture. We found that impact of increasing soil moisture on C mineralization was adjusted by fertilization and residue incorporation. The  $C_{cum}$  and  $N_{cum}$  were higher in residue and fertilizer amended soils than non-amended soils,  $k_C$  was always

less in amended samples at all soil moisture levels. Our attempt to estimate moisture sensitivity by using  $k_C$  and  $k_N$  provided a better insight rather than using  $C_{cum}$  and  $N_{cum}$ . Residue addition reduced moisture sensitivity of  $C_{cum}$  and  $N_{cum}$ , thus it could be recommended in tropical and sub-tropical Vertisol for maintains positive C balance. The study highlighted that fertilization and residue incorporation interact synergistically at higher soil moisture content, but the interaction is antagonistic by nature at low levels of soil moisture in Vertisol. So, on a broader scale, the results of this study might be utilised to adjust irrigation frequency in soils amended with crop residue and fertilizer.

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