



Effect of *Crotalaria* Species on the Dynamics of Soil Biological Activity in the Northern Sudanian Zone of Burkina Faso

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Authors' contributions

This work was carried out in collaboration among all authors. All authors read and approved the final manuscript.

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Abstract

Declining soil biological activity threatens the sustainability of agricultural production in the Sudanian zone of Burkina Faso. Split-plot experiments conducted in 2023 and 2024 at Yarci, Arboillé, evaluated the effects of *Crotalaria mucronata* and *Crotalaria retusa* combined with four amendment treatments: an unamended control (T0), organic matter (OM), Burkina phosphate (BP), and OM+BP. Daily CO₂ emissions, microbial biomass carbon (MBC), microbial yield (MY), overall mineralization rate (OMR), and metabolic quotient (qCO₂) were assessed. Results showed that both *Crotalaria* species produced similar effects. Cumulative CO₂ emissions increased between 2023 and 2024, reaching maximum values of 320.90 and 298.98 mg 100 g⁻¹ soil under *C. retusa* and *C. mucronata*, respectively. For both species, OM+BP generally produced the highest CO₂ emissions and improved most soil biological indicators, followed by OM alone. MY was stable at approximately 48-51%, whereas qCO₂ and OMR remained below 2 mg CO₂-C g⁻¹ MBC day⁻¹ and 2%, respectively, suggesting relatively low microbial metabolic stress and moderate organic matter mineralization. These findings indicate that combining *Crotalaria*-based improved fallows with organic matter and Burkina phosphate can enhance soil biological functioning and represent a promising agroecological strategy for restoring degraded soils in Burkina Faso.

Keywords: Agroecology; Burkina phosphate; improved fallow; microbial activity; soil restoration.

1. Introduction

Soil degradation is a major threat to agricultural sustainability and food security, particularly in sub-Saharan Africa, where population growth, climate variability, continuous cultivation, and inadequate soil management accelerate soil organic matter and nutrient depletion (Tully *et al.*, 2015; Bayata, 2024). Approximately 65% of arable land in sub-Saharan Africa is estimated to be degraded, resulting in annual soil nutrient losses valued at nearly US\$4 billion (Kihara *et al.*, 2023). Soil erosion further exacerbates this degradation by removing nutrient-rich topsoil and reducing the productive capacity of agricultural land (Guo *et al.*, 2019; Singh *et al.*, 2020).

In the Sudanian zone of Burkina Faso, agricultural soils are generally characterized by low organic carbon contents, often below 1%, total nitrogen contents below 0.1%, limited phosphorus availability, and low exchangeable base contents (Somda *et al.*, 2022). The long-term consequences of continuous cultivation are substantial. In southwestern Burkina Faso, for example, conversion of natural vegetation to continuous cropping resulted in losses of approximately 49 t C ha⁻¹ from the upper 30 cm of soil after 29 years, corresponding to a 45% decline in soil organic carbon stocks (Hounkpatin *et al.*, 2018). Traditionally, fallow periods of up to 20 years allowed cultivated soils to recover part of their fertility. However, increasing pressure on agricultural land has considerably shortened or eliminated these periods (Dipama, 2009; Mugizi & Matsumoto, 2021). Productive alternatives to long-term natural fallows are therefore needed.

Soil microorganisms play a central role in organic matter decomposition, nutrient cycling, soil aggregation, and plant nutrient availability. Microbial biomass, soil respiration, microbial yield, overall mineralization rate, and metabolic quotient are consequently useful indicators of soil biological functioning and its response to management practices (Bardgett & van der Putten, 2014; Maron *et al.*, 2018). Practices that increase organic inputs and maintain permanent or temporary vegetation cover can stimulate these processes and contribute to the restoration of degraded soils. Recent reviews likewise emphasize that microbial functional and metabolic biomarkers can provide complementary evidence of nutrient cycling, carbon dynamics, and soil stress responses (Oyediran *et al.*, 2025).

Leguminous cover crops, particularly *Crotalaria* species, offer considerable potential for improving soil fertility. Through biological nitrogen fixation, legumes may contribute approximately 20-200 kg N ha⁻¹ year⁻¹, depending on the species, environmental conditions, and crop management (Kebede, 2021). *Crotalaria* species can also produce substantial biomass, increase organic matter inputs to the soil, improve soil structure and water infiltration, and reduce erosion and compaction (Kocira *et al.*, 2020; Hao *et al.*, 2023; Arone *et al.*, 2024). In addition, some species suppress weeds and plant-parasitic nematodes and stimulate soil microbial and enzymatic activities (Çerçioglu *et al.*, 2025). These effects may be enhanced when *Crotalaria* biomass is combined with locally available amendments, such as organic matter and Burkina phosphate, which provide complementary sources of carbon, nitrogen, and phosphorus. Evidence from Burkina Faso also shows that locally sourced rock phosphate can improve legume nodulation and crop performance where phosphorus availability is limiting (Zongo *et al.*, 2022). Recent field evidence indicates that legume cover crops can modify soil phosphorus availability and microbial community composition, linking plant cover with biologically mediated phosphorus cycling (Dong *et al.*, 2024). Complementary evidence from carbon-poor agricultural soil shows that incorporated legume biomass can enhance microbial activity and abundance alongside soil nutrient availability (Morales-Salmerón *et al.*, 2025).

Crotalaria mucronata and *Crotalaria retusa* are adapted to the climatic conditions of the northern Sudanian zone of Burkina Faso, characterized by annual rainfall of approximately 600-900 mm and marked seasonal variations in temperature. Previous trials at Yarci showed that combining these species with organic matter and Burkina phosphate enhanced biomass production. Under the combined OM+BP treatment, *C. mucronata* and *C. retusa* produced sixfold and 3.3-fold increases in biomass, respectively, in 2024 compared with 2023 (Coulibaly *et al.*, 2025). Although these findings demonstrate their biomass-production potential, their effects on the temporal dynamics of soil respiration and other microbial indicators remain insufficiently documented under the relatively dry and nutrient-poor conditions of this agroecological zone.

This study therefore assessed the effects of *C. mucronata* and *C. retusa*, combined with organic matter and Burkina phosphate, on soil biological functioning at Yarci, Burkina Faso. Specifically, it evaluated daily and cumulative CO₂ emissions, microbial biomass carbon, microbial yield, overall mineralization rate, and metabolic quotient over two consecutive years. The study hypothesized that combining *Crotalaria*-based improved fallows with organic matter and Burkina phosphate would stimulate soil microbial activity more effectively than either amendment applied alone.

2. Material and Methods

2.1 Study Site Description

The experiment was conducted during the 2023 and 2024 cropping seasons at Yarci, a village located approximately 80 km northwest of Ouagadougou in the commune of Arbolle, Passoré Province, within the northern Sudanian zone of Burkina Faso. The study area is characterized by a northern Sudanian climate, with an average annual rainfall of approximately 700 mm, generally ranging from 600 to 900 mm. The relatively flat landscape comprises two main topographic positions along the toposequence, the middle and lower glacia, and is dominated by tropical ferruginous soils with low nitrogen and phosphorus contents (Coulibaly *et al.*, 2025).

2.2 Plant Material Used

The plant material consisted of seeds of two *Crotalaria* species, *Crotalaria retusa* (L.) and *Crotalaria mucronata* (Desv.), as described by Ouédraogo *et al.* (2024). The seeds were collected from Noumousso and Arbolle, the study sites of Project No. 0087 FONRID/AAP8/NCP/PC/202, entitled “Use of Herbaceous Papilionaceae in the Restoration of Degraded Soil Fertility.”

2.3 Fertilizers and Soil Amendments Used

Two types of fertilizers were used in this study: (i) organic matter (OM), derived from decomposed crop residues and animal manure, and (ii) Burkina phosphate (BP). BP is a naturally occurring phosphate rock found in abundance in eastern Burkina Faso, particularly at Kodjari, Arly National Park, and W National Park. It is a rich source of phosphorus, although its short-term solubility is low. Research conducted at INERA has shown that the solubility of BP increases when it is applied in combination with organic matter (Traoré *et al.*, 2023). These two fertilizers were selected because of their local availability and affordability for farmers in Burkina Faso (Sanon *et al.*, 2021). The chemical compositions of OM and BP before the experiment are presented in Table 1.

Table 1. Chemical composition of Burkina Phosphate (BP) and Organic Mater (OM)

Designation	BP (%)	OM (g kg ⁻¹)
Total phosphorus (Pt)	12.35	1.95
Phosphorus pentoxide (P ₂ O ₅)	28.3	
Water P	0.19	
Citrate P	30	
Ferrous oxide (Iron II) (FeO)	1.7	
Calcium oxide (CaO)	38.3	
Silicon dioxide (SiO ₂)	23.33	
Aluminium oxide (Al ₂ O ₃)	2.35	
Potassium oxide (K ₂ O)		2.35
Nitrogen (N)		5.99
Carbon (C)		4.30

BP: Bikienga (2011); OM: current study

2.4 Experimental Design and Treatments Applied

The experiment was established as a two-factor split-plot design with four replicates. Each replicate constituted a block containing two main plots, to which the two *Crotalaria* species, *C. mucronata* and *C. retusa*, were randomly assigned. The main plots measured 38 m x 16 m (608 m²).

Within each main plot, four amendment treatments were randomly assigned to the subplots: an unamended control (T0), organic matter alone (OM), Burkina phosphate alone (BP), and the combined application of organic matter and Burkina phosphate (OM+BP). Each elementary subplot measured 5 m x 2 m (10 m²) and constituted the experimental unit for amendment application and soil sampling. Organic matter and Burkina phosphate were applied at rates of 5 t ha⁻¹ and 500 kg ha⁻¹, respectively.

2.5 Crop Management Practices

In 2023 and 2024, shallow tillage was performed under moist soil conditions across the entire experimental area using a Manga hoe. This operation facilitated soil penetration by the implement and improved soil aggregate fragmentation. Before plot demarcation, the soil was manually levelled to a depth of approximately 5-10 cm, corresponding to the shallow rooting zone and the soil layer into which the amendments were incorporated. After plot establishment, the organic and mineral amendments were applied at rates of 5 t ha⁻¹ for OM and 500 kg ha⁻¹ for BP, and sowing was carried out on the same day. Three hundred seeds of each *Crotalaria*

species were evenly distributed in the corresponding subplots and incorporated into the upper 5-10 cm of soil. No phytosanitary treatment was applied during the experiment.

At the end of the first cropping season, the aboveground biomass produced in each plot was harvested, weighed, and incorporated into the soil of the corresponding plot. The determination of soil carbon content in the second year accounted for the biomass incorporated after the first cropping season, which was considered a residual organic input contributing to the soil carbon stock.

2.6 Soil Sampling and Analysis

Before the experiment was established in 2023, five composite soil samples were randomly collected across the entire experimental area from the 0-30 cm soil layer to characterize the initial soil conditions. At the end of the 2023 and 2024 cropping seasons, soil samples were collected from each subplot corresponding to the different treatments. Within each subplot, five soil cores were collected from the 0-30 cm layer and thoroughly mixed to obtain one composite sample. As each treatment was replicated four times, four composite samples were obtained per treatment and cropping season. After collection, the samples were air-dried, passed through a 2-mm sieve, and analysed at the Soil-Water-Plant Laboratory of Farako-Bâ in Bobo-Dioulasso.

2.7 Microbial Biomass Assessment

Soil microbial biomass (MB) was determined using the fumigation-incubation method described by Jenkinson and Powlson (1976) and calculated using the following equation:

$$MB \text{ (mg 100 g}^{-1}\text{)} = \frac{F(0-7)+F(7-14)}{K_c} \quad (1)$$

where K_c is the proportionality coefficient ($K_c = 0.41$ according to Anderson and Domsch (2010)), and $F(0-7)$ and $F(7-14)$ represent the amounts of carbon dioxide (CO_2) released during the 0-7-day and 7-14-day incubation periods, respectively.

2.8 Measurement of Daily Soil Carbon Dioxide (CO_2) Emissions

Daily soil carbon dioxide (CO_2) emissions were measured using the respirometric method described by Dommergues (1960). CO_2 emissions were determined daily during the first seven days of incubation and every two days from day 9 to day 21, resulting in a total of 14 measurements. Soil respiration rate ($\mu g \text{ } CO_2 \text{ g}^{-1} \text{ soil h}^{-1}$) was calculated from the measured CO_2 concentrations using the following equation:

$$CO_2 \text{ (mg)} = (V_{blank} - V_{sample}) \times 2,2 \quad (2)$$

where V_{blank} is the volume of 0.1 N HCl (mL) used to titrate the control jars, and V_{sample} is the volume of 0.1 N HCl (mL) used to titrate the jars containing the soil samples. One millilitre of 0.1 N HCl corresponds to 2.2 mg of CO_2 (Somé *et al.*, 2007).

2.9 Calculation of Microbial Yield (MY), Overall Mineralization Rate (OMR), and Metabolic Quotient (qCO_2)

The collected data were also used to calculate microbial yield (MY), the overall mineralization rate (OMR), and the metabolic quotient (qCO_2).

Microbial yield corresponds to the ratio of microbial biomass carbon (MBC) to total organic carbon (TOC) and was calculated as follows:

$$MY \text{ (\%)} = \frac{MBC}{TOC} \times 100 \quad (3)$$

The metabolic quotient (qCO_2) represents the amount of carbon mineralized per gram of microbial biomass carbon per day. It was calculated according to Fotio *et al.* (2013) as follows:

$$qCO_2 = \frac{C-CO_2}{MBC \times t} \quad (4)$$

where qCO_2 is the metabolic quotient, $C-CO_2$ is the amount of carbon mineralized during the 21-day incubation period, MBC is the microbial biomass carbon expressed as $mg \text{ } 100 \text{ g}^{-1} \text{ soil}$, and t is the incubation time expressed in days.

The overall mineralization rate is the ratio of cumulative $C-CO_2$ released to the total organic carbon content of the corresponding treatment. It expresses the rate of soil organic matter mineralization (Somé *et al.*, 2007) and can be used to assess temporal changes in organic substrates. It was calculated as follows:

$$OMR \text{ (\%)} = \frac{\text{cumulative } C-CO_2}{TOC} \times 100 \quad (5)$$

where cumulative $C-CO_2$ is the total amount of carbon released as carbon dioxide during incubation, and TOC is the total organic carbon content of the soil.

2.10 Statistical Analysis

Data were entered and organized using Microsoft Excel 2016, which was also used to generate the graphs. Statistical analyses were performed using GenStat version 12.1.

Data were entered and organized using Microsoft Excel 2016, which was also used to generate the graphs. Statistical analyses were performed using GenStat version 12.1. Data from 2023 and 2024 were jointly analysed using a linear mixed model that accounted for the split-plot design and the repeated measurements across years. Year, *Crotalaria* species, amendment treatment, and all their interactions were included as fixed effects. Blocks and the block x species interaction, representing the main-plot error term, were included as random effects. Amendment treatment and its interactions were tested against the subplot residual error. Because the same experimental plots were assessed in both years, year was treated as a repeated factor, with plot specified as the experimental subject. When significant effects were detected, means were compared using Tukey's honestly significant difference (HSD) test at the 5% significance level ($P < 0.05$).

3. Results

3.1 Effect of *Crotalaria* Species on Daily CO₂ Emissions in 2023 and 2024

Daily soil CO₂ emissions followed similar temporal patterns under *C. mucronata* and *C. retusa* during the 21-day incubation period (Figs. 1A-B and 2A-B). Emissions were initially high, declined progressively until day 7, increased temporarily on day 9, and subsequently decreased to their lowest levels by day 21. Amendment treatments significantly affected emissions ($P < 0.001$). In both years and under both species, OM+BP consistently produced the highest emissions, followed by OM, whereas T0 and the initial soil produced the lowest values. Emissions were significantly higher in 2024 than in 2023 ($P < 0.001$); under OM+BP, day⁻¹ values increased from approximately 19-21 mg CO₂ 100 g⁻¹ soil in 2023 to 34-35 mg CO₂ 100 g⁻¹ soil in 2024. Differences among treatments were also more pronounced in 2024, particularly between treatments containing organic matter and those receiving BP alone or no amendment.

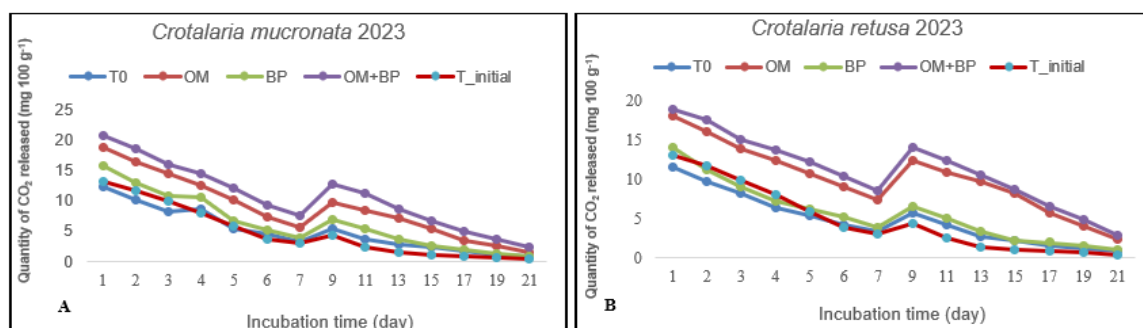


Fig. 1. Evolution of daily CO₂ released by *Crotalaria retusa* 2023 (A) and *Crotalaria mucronata* 2023 (B)

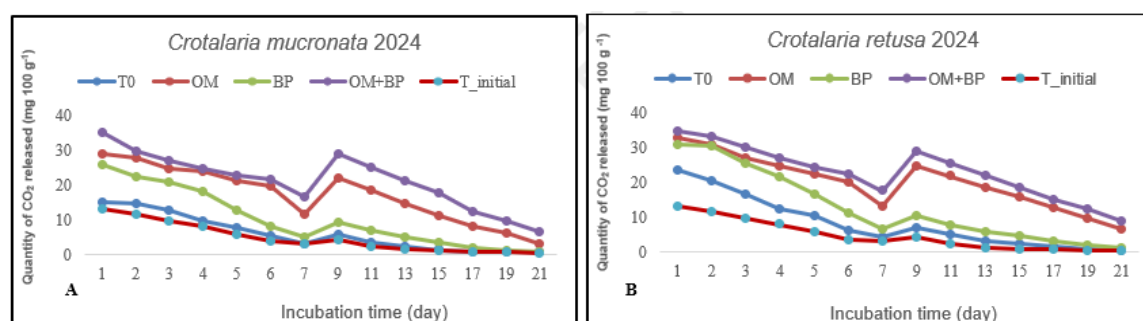


Fig. 2. Evolution of daily CO₂ released by *Crotalaria retusa* 2024 (A) and *Crotalaria mucronata* 2024 (B)

Legend: T0: control; OM: organic matter; BP: Burkina phosphate

3.2 Effects of *Crotalaria* Species and Organo-mineral Amendments on Soil Microbial Biomass Carbon

Soil microbial biomass carbon (MBC) was significantly affected by species, amendment treatment, and year (Table 2). Mean MBC increased significantly between 2023 and 2024, from 100.4 ± 6.1 to 185.6 ± 16.4 mg 100 g⁻¹ soil under *C. mucronata* and from 102.2 ± 9.0 to 207.8 ± 16.5 mg 100 g⁻¹ soil under *C. retusa* ($P < 0.001$).

Treatment responses varied between species and years. Under *C. mucronata* in 2023, OM+BP and OM produced the highest and statistically comparable MBC values, whereas BP and T0 formed the lower statistical group. In 2024, MBC differed significantly

among all treatments and followed the order OM+BP > OM > BP > T0, ranging from 81.3 ± 4.4 mg 100 g⁻¹ soil under T0 to 279.9 ± 5.4 mg 100 g⁻¹ soil under OM+BP.

A different response was observed under *C. retusa* in 2023. T0 and OM produced the highest and statistically comparable MBC values, whereas BP and OM+BP produced significantly lower values. In 2024, the treatment ranking was again OM+BP > OM > BP > T0, with values ranging from 93.0 ± 2.2 to 294.3 ± 6.2 mg 100 g⁻¹ soil. The year $\times$ species interaction was significant ($P < 0.05$), while the year $\times$ treatment and species $\times$ treatment interactions were highly significant ($P < 0.001$). The year $\times$ species $\times$ treatment interaction was not significant ($P > 0.05$).

The initial soil MBC was 57.8 mg 100 g⁻¹ soil and was used as a descriptive baseline. Relative to this value, MBC ranged from approximately 1.1-fold under BP with *C. retusa* in 2023 to 5.1-fold under OM+BP with *C. retusa* in 2024.

Table 2. Variation of soil microbial biomass (MB) as affected by *Crotalaria* species and organo-mineral fertilizer

Species	Treatments	MBC 2023 (mg 100 g ⁻¹)	MBC 2024 (mg 100 g ⁻¹)
<i>Crotalaria mucronata</i>	BP	82.4 ± 3.9^b	148 ± 13.1^c
	OM	114.1 ± 6.2^a	233 ± 8.0^b
	OM+BP	135.5 ± 7.6^a	279.9 ± 5.4^a
	T0	69.6 ± 6.3^b	81.3 ± 4.4^d
	Pr_Treatments (T)	***	***
	Mean/Years (Y)	100.4 ± 6.12	185.6 ± 16.4
	Pr	***	
	T_initial	57.8	
<i>Crotalaria retusa</i>	BP	66.0 ± 2.5^b	181.5 ± 7.7^c
	OM	128.4 ± 16.9^a	262.2 ± 6.2^b
	OM+BP	73.0 ± 3.9^b	294.3 ± 6.2^a
	T0	141.7 ± 17.1^a	93 ± 2.2^d
	Pr_Treatments (T)	***	
	Mean/Years	102.15 ± 9.02^b	207.8 ± 16.5^a
	Pr	***	***
	T_initial	57.8	
	Pr_species (Sp)	***	
	Y $\times$ Sp	*	
	Y $\times$ T	***	
	Sp $\times$ T	***	
	Y $\times$ Sp $\times$ T	NS	

Legend: *** highly significant ($P < 0.001$); * : significant ($P = 0.019$); NS ($P > 0.05$); no significant; T0: control; OM: organic matter; BP: Burkina phosphate; within each column, means followed by the same letter are not significantly different according to Tukey's HSD test at the 5% significance level

3.3 Effect of *Crotalaria* Species and Organo-mineral Fertilizer on Overall Mineralization Rate (OMR) and Soil Microbial Yield (MY)

The overall mineralization rate (OMR) varied significantly among amendment treatments and between years (Table 3). Under *C. mucronata*, treatment effects were highly significant in both years ($P < 0.001$) with OM+BP and OM consistently producing the highest and statistically comparable values. Mean OMR increased significantly from 0.34% in 2023 to 0.44% in 2024 ($P < 0.001$).

Under *C. retusa*, treatment effects were significant in 2023 ($P = 0.02$) and highly significant in 2024 ($P < 0.001$). In 2023, OM+BP produced the highest numerical OMR but did not differ significantly from OM or T0. In 2024, OMR followed the order OM+BP > OM > BP > T0, with a maximum of 0.61% under OM+BP. Mean OMR increased from 0.28% in 2023 to 0.44% in 2024. The species effect was not significant, whereas the year $\times$ treatment interaction was highly significant ($P < 0.001$).

Table 3. Variation of overall mineralization rate (OMR) and soil microbial yield (MY) as affected by *Crotalaria* species and organo-mineral fertilizer

Species	Treatments	OMR (%)		MY (%)	
		2023	2024	2023	2024
<i>Crotalaria mucronata</i>	BP	0.26 ± 0.02^b	0.41 ± 0.04^b	48.74 ± 0.20	51.13 ± 0.22
	OM	0.37 ± 0.03^a	0.54 ± 0.05^a	48.05 ± 0.33	49.26 ± 0.25
	OM+BP	0.45 ± 0.03^a	0.58 ± 0.02^a	47.70 ± 0.38	48.37 ± 0.13
	T0	0.26 ± 0.01^b	0.25 ± 0.02^c	49.12 ± 1.7	49.52 ± 1.54
	Pr_Treatments (T)	***	***	NS	NS
	Means/years	0.34 ± 0.02^b	0.44 ± 0.03^a	48.41 ± 0.43	49.57 ± 0.42
	Pr	***		NS	
	T_initial	0.15		53.50	
<i>Crotalaria retusa</i>	BP	0.19 ± 0.02^b	0.36 ± 0.01^c	49.50 ± 0.69	50.51 ± 0.22^a
	OM	0.34 ± 0.05^{ab}	0.53 ± 0.01^b	47.60 ± 0.40	48.21 ± 0.13^b

Species	Treatments	OMR (%)		MY (%)	
		2023	2024	2023	2024
	OM+BP	0.36 ± 0.06 ^a	0.61 ± 0.01 ^a	48.10 ± 0.76	47.84 ± 0.08 ^b
	T0	0.25 ± 0.01 ^{ab}	0.27 ± 0.01 ^d	47.50 ± 0.33	44.62 ± 1.01 ^c
	Pr_treatments	*	***	NS	***
	Means/years	0.28 ± 0.02 ^b	0.44 ± 0.03 ^a	48.19 ± 0.31	47.79 ± 0.50
	Pr	***		NS	
	T_initial	0.15		53.50	
	Pr_ species (Sp)	NS		**	
	Y x Sp	NS		*	
	Y x T	***		***	
	Sp x T	NS		NS	
	Y x Sp x T	NS		*	

Legend: *** and ** highly significant ($P < 0.001$ and $P \leq 0.001$); * : significant ($P = 0.02$ et $P = 0.03$); NS ($P > 0.05$): no significant; T0: control; OM: organic matter; BP: Burkina phosphate; within each column, means followed by the same letter are not significantly different according to Tukey's HSD test at the 5% significance level.

Microbial yield (MY) responded differently from OMR. Under *C. mucronata*, amendment treatments had no significant effect on MY in either year, and annual means remained statistically comparable at approximately 48–50%. Under *C. retusa*, treatment effects were not significant in 2023 but were highly significant in 2024 ($P < 0.001$). In 2024, BP produced the highest MY (50.51%), whereas T0 produced the lowest (44.62%). Mean MY did not differ significantly between years. However, MY was significantly affected by species and by its interactions with year and treatment.

The initial soil, used only as a descriptive baseline, had a lower OMR (0.15%) but a higher MY (53.5%) than the treatment means.

3.4 Effects of *Crotalaria* Species and Amendment Treatments on the Metabolic Quotient (qCO₂) and Total Organic Carbon (TOC)

Amendment treatments had no significant effect on the metabolic quotient (qCO₂) under either *Crotalaria* species in 2023 or under *C. mucronata* in 2024 (Table 4). Under *C. retusa*, however, treatment effects were highly significant in 2024 ($P < 0.001$). T0 produced the highest qCO₂ (1.25 mg C-CO₂ g⁻¹ MBC day⁻¹), whereas BP produced the lowest value (0.98 mg C-CO₂ g⁻¹ MBC day⁻¹). Mean qCO₂ did not differ significantly between years under either species.

Table 4. Variation of respiration quotient and carbon content as affected by *Crotalaria* species and organo-mineral fertilizer

Species	Treatments	qCO ₂ (mg g ⁻¹ jour ⁻¹)		TOC (mg 100 g ⁻¹)	
		2023	2024	2023	2024
<i>Crotalaria mucronata</i>	BP	1.05 ± 0.01	0.96 ± 0.01	650.80 ± 28.95	715.70 ± 21.35 ^b
	MO	1.08 ± 0.01	1.03 ± 0.01	656.30 ± 48.47	892.20 ± 45.07 ^a
	MO+BP	1.10 ± 0.02	1.07 ± 0.01	626 ± 16.35	1007.20 ± 14.67 ^a
	T0	1.05 ± 0.07	1.03 ± 0.06	537.80 ± 62.64	675.60 ± 30.54 ^b
	Pr_treatments	NS	NS	NS	***
	Means/years	1.07 ± 0.02	1.02 ± 0.02	617.70 ± 22.30	822.70 ± 31.30
	Pr	NS		***	
<i>Crotalaria retusa</i>	T_initial	0.90		730	
	BP	1.02 ± 0.03	0.98 ± 0.01 ^c	807 ± 44.52 ^a	1001.70 ± 14.89 ^a
	MO	1.10 ± 0.02	1.08 ± 0.01 ^{bc}	802.70 ± 34.77 ^a	1023.40 ± 21.94 ^a
	MO+BP	1.08 ± 0.04	1.09 ± 0.00 ^b	843.20 ± 45.26 ^a	1000.80 ± 10.56 ^a
	T0	1.1 ± 0.01	1.25 ± 0.05 ^a	537.80 ± 25.23 ^b	774.90 ± 13 ^b
	Pr_treatments	NS	***	***	***
	Means/years	1.08 ± 0.01	1.10 ± 0.02	747.70 ± 31.10 ^b	950.20 ± 22.4 ^a
	Pr	NS		***	
	T_initial	0.9		730	
	Pr_ species (Sp)	**		***	
	Y x Sp	*		NS	
	Y x T	***		*	
	Sp x T	NS		**	
	Y x Sp x T	*		*	

Legend: *** highly significant ($P < 0.001$); NS ($P > 0.05$): no significant; T0: control; OM: organic matter; BP: Burkina phosphate; within each column, means followed by the same letter are not significantly different according to Tukey's HSD test at the 5% significance level.

Total organic carbon (TOC) was not significantly affected by amendment treatments under *C. mucronata* in 2023, but treatment effects were highly significant in 2024 ($P < 0.001$). In 2024, OM+BP and OM produced the highest and statistically comparable TOC contents, while BP and T0 formed the lower statistical group. Mean TOC increased significantly from 617.7 mg 100 g⁻¹ soil in 2023 to 822.7 mg 100 g⁻¹ soil in 2024.

Under *C. retusa*, treatment effects on TOC were highly significant in both years ($P < 0.001$). BP, OM, and OM+BP produced statistically comparable TOC contents and significantly higher values than T0. Mean TOC increased significantly from 747.7 mg 100 g⁻¹ soil in 2023 to 950.2 mg 100 g⁻¹ soil in 2024.

qCO₂ and TOC were significantly affected by species and by species interactions with year and treatment.

The initial soil, used as a descriptive baseline, had a qCO₂ of 0.90 mg C-CO₂ g⁻¹ MBC day⁻¹ and a TOC content of 730 mg 100 g⁻¹ soil.

4. Discussion

4.1 Effect of *Crotalaria* Species on Daily CO₂ Emissions in 2023 and 2024

Daily CO₂ emissions under *C. mucronata* and *C. retusa* followed comparable temporal patterns, characterized by high initial values, a significant decline until day 7, a temporary increase on day 9, and a subsequent decrease until day 21. This pattern is characteristic of organic substrate mineralization during soil incubation. The initial peak probably reflected the rapid microbial use of readily decomposable carbon compounds, whereas the subsequent decline may indicate the progressive depletion of these labile substrates and a shift towards more resistant organic fractions. Similar patterns have been reported by Marhan *et al.* (2008), who observed high CO₂ production during the early stage of incubation followed by a progressive decline, and by Datta *et al.* (2019), who demonstrated that residue characteristics and placement influence the rate and dynamics of soil carbon mineralization.

The temporary increase observed on day 9 may reflect renewed microbial utilization of organic compounds released during the decomposition process. However, because microbial community composition and the biochemical fractions of the residues were not measured, the mechanism responsible for this rebound cannot be established from the present data.

The consistently higher emissions under OM+BP and OM indicate that treatments containing organic matter provided more mineralizable carbon than BP alone or the unamended control. The strong response under OM+BP may also suggest that the combined supply of carbon and phosphorus reduced nutritional constraints on microbial activity. Nevertheless, the direct contribution of Burkina phosphate to microbial respiration cannot be separated from that of organic matter in the present study.

CO₂ emissions were significantly higher in 2024 than in 2023, with day-1 values under OM+BP increasing from approximately 19-21 to 34-35 mg CO₂ 100 g⁻¹ soil. This increase may partly reflect the residual effects of the amendments and the incorporation of *Crotalaria* biomass produced during the first year. The higher rainfall recorded in 2024 (915 mm, compared with 821 mm in 2023) may also have indirectly promoted plant growth and organic inputs before soil sampling. Overall, the two species showed similar emission dynamics, while the magnitude of the treatment differences was greater in 2024. The higher emissions observed under treatments containing organic matter indicate increased carbon mineralization and microbial respiration.

4.2 Effect of *Crotalaria* Species and Organo-Mineral Amendment on Soil Microbial Biomass (MB), Microbial Yield (MY), Overall Mineralization Rate (OMR) and Metabolic Quotient (qCO₂)

Microbial indicators responded differently to *Crotalaria* species, amendment treatments, and year. The significant increase in microbial biomass carbon (MBC) from 2023 to 2024, particularly under OM+BP and OM, suggests that organic inputs stimulated microbial growth by increasing the availability of decomposable substrates. This response is consistent with Hori *et al.* (2011), who reported rapid increases in microbial biomass and respiration following the incorporation of *Crotalaria* leaves and stems. However, the response was not uniform: under *C. retusa* in 2023, T0 and OM produced higher MBC than BP and OM+BP, confirming that amendment effects depended on species and year.

The overall mineralization rate (OMR) was generally highest under OM+BP and OM and increased significantly in 2024. This pattern indicates greater mineralization of organic substrates in treatments receiving organic matter. The significant year × treatment interaction further shows that amendment effects became more pronounced during the second year, possibly because of residual organic inputs and the incorporation of first-year *Crotalaria* biomass. Residue quality and placement can strongly influence carbon mineralization dynamics, as reported by Datta *et al.* (2019).

In contrast, microbial yield (MY) remained relatively stable across most treatments, generally ranging from approximately 48% to 50%. This suggests that the increase in respiration and mineralization was not consistently accompanied by proportional changes in the ratio of microbial biomass to total organic carbon. Under *C. retusa* in 2024, BP produced the highest MY, whereas T0 produced the lowest, demonstrating that OM+BP did not systematically maximize this indicator.

The metabolic quotient (qCO₂) was relatively stable under *C. mucronata* but differed among treatments under *C. retusa* in 2024. The higher qCO₂ recorded under T0 (1.25 mg C-CO₂ g⁻¹ MBC day⁻¹) than under the amended treatments indicates greater respiration per unit of microbial biomass. This may reflect lower microbial metabolic efficiency or greater physiological stress under the unamended treatment. Nevertheless, qCO₂ should be interpreted as an index of biomass-specific respiration rather than as a direct measurement of microbial carbon-use efficiency. It was found that the combined OM+BP treatment generally enhanced MBC, OMR, and TOC, particularly in 2024, whereas its effects on MY and qCO₂ were less consistent. These results suggest that combining *Crotalaria*-based fallows with organic matter and Burkina phosphate can stimulate soil microbial functioning.

5. Conclusion

This study showed that *Crotalaria mucronata* and *Crotalaria retusa*, combined with organic matter and Burkina phosphate, improved soil biological functioning in the northern Sudanian zone of Burkina Faso. Both species showed similar CO₂ emission patterns, with high initial values followed by an overall decline during incubation. OM+BP generally produced the highest CO₂ emissions, reaching approximately 35 mg CO₂ 100 g⁻¹ soil in 2024, and the highest microbial biomass, reaching 279.9 and 294.3 mg 100 g⁻¹ soil under *C. mucronata* and *C. retusa*, respectively. The overall mineralization rate remained below 0.61%, while microbial yield was generally stable at approximately 48-50%.

These findings indicate that *Crotalaria*-based improved fallows combined with organic matter and Burkina phosphate represent a promising strategy for enhancing soil biological functioning.

6. Limitations

The study was conducted at a single site over two cropping seasons using four replicates, which may limit the spatial and temporal generalisability of the findings. Soil biological responses were assessed mainly through incubation-based respiration, microbial biomass, microbial yield, mineralisation rate, metabolic quotient, and total organic carbon, without characterising microbial community composition or residue biochemical properties. Differences in annual rainfall and residual effects of incorporated biomass may also have influenced year-to-year responses. Future multi-site, longer-term studies incorporating microbial community and residue-quality analyses would strengthen the interpretation and applicability of the findings.

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Declaration of AI Use

This manuscript was prepared through the intellectual contributions of the author(s) to the study design, data analysis, interpretation of results, and scholarly content. Grammarly and ChatGPT were used solely to assist with grammatical refinement and reference formatting during manuscript revision. The author(s) accept full responsibility for the accuracy, integrity, and final content of the manuscript.

Competing Interests

Authors have declared that they have no known competing financial interests OR non-financial interests OR personal relationships that could have appeared to influence the work reported in this paper.

Competing Interests

Authors have declared that no competing interests exist.

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