Article

Impact of Integrated Agronomic Practices on Soil Fertility and Respiration on the Indo-Gangetic Plain of North India

Rama Kant Dubey [1], Pradeep Kumar Dubey [1], Rajan Chaurasia [1], Ch Srinivasa Rao [2] and Purushothaman Chirakkuzhyil Abhilash [1,*]

1   Institute of Environment and Sustainable Development, Banaras Hindu University, Varanasi 221005, India; ramakant.sls@gmail.com (R.K.D.); pradeep.dubey4@bhu.ac.in (P.K.D.); rajan.chaurasiya92@gmail.com (R.C.)
2   ICAR National Academy of Agricultural Research and Management, Rajendranagar, Hyderabad 500030, India; cherukumalli2011@gmail.com
*   Correspondence: pca.iesd@bhu.ac.in; Tel.: +91-94-1564-4280

Abstract: Global agricultural production is accountable for the emission of ~30% of greenhouse gases. Therefore, the wide-scale adoptions of low-input, soil-friendly, and resource-conserving agronomic practices are imperative for the 'planet healthy food production' and also for reducing the carbon emissions from agricultural soil. In this context, the present study aimed to analyze the impacts of integrated agronomic interventions i.e., the application of arbuscular mycorrhizal fungi (AMF) + reduced tillage (RT), biochar + RT, and AMF + biochar + RT, on spatiotemporal variations in soil-quality and soil-sustainability indicators, including microbial and soil respiration, in the Indo-Gangetic Plain (IGP) of North India. For this, field experiments on the above-mentioned agronomic interventions were employed using three different staple crops (*Zea mays*, *Vigna mungo*, and *Brassica juncea*) growing in three different agro-climatic zones of IGP (Varanasi, Sultanpur, and Gorakhpur) in a randomized block design. Periodic data collection was done to analyze the changes in physiochemical, biological, and biochemical properties of the soil, and statistical analyses were done accordingly. Irrespective of the sites, the experimental results proved that the integrated application of AMF + biochar + RT in *V. mungo* resulted in the highest soil organic carbon (i.e., 135% increment over the control) and microbial biomass carbon (24%), whereas the same application (i.e., AMF + biochar + RT) in *Z. mays* had the maximum reduction in microbial (32%) and soil (44%) respiration. On the other hand, enhanced occurrence of glomalin activity (98%) was noted in *Z. mays* cropping for all the sites. Significant negative correlation between soil respiration and glomalin activity under AMF + biochar + RT (−0.85), AMF + RT (−0.82), and biochar + RT (−0.62) was an indication of glomalin's role in the reduced rate of soil respiration. The research results proved that the combined application of AMF + biochar + RT was the best practice for enhancing soil quality while reducing respiration. Therefore, the development of suitable packages of integrated agronomic practices is essential for agricultural sustainability.

Keywords: agricultural sustainability; carbon sequestration; farming practices; integrated agronomic practices; microbial respiration; soil respiration; soil sustainability indicators

1. Introduction

Improving soil fertility while reducing carbon emissions is a key component of sustainable crop production [1–3]. During the last few decades, agricultural soils have lost two-thirds of their original carbon pool due to intensive farming practices [4]. Principally, soil respiration (CO_2 efflux) is a significant process that releases soil carbon into the atmosphere at an approximated rate of 68–98 picograms $C/year^{-1}$ [5]. The global rate of soil respiration shows a continuous increasing trend, which is likely similar in Indian agriculture (net carbon emissions of 1727×10^6 mg CO_2 equivalents) as well [6,7]. Enhancing soil carbon and reducing its emission into the atmosphere is therefore a concurrent subject to ensure soil quality and agricultural sustainability [8–11].

Substantial research identifies agronomic interventions such as organic amendments, microbial inoculum and reduced tillage as methods to increase soil carbon stock [4,12–15]. Enhanced soil carbon is noted as a key responsive factor to increase both soil fertility and crop performances [16–19]. Among various studied organic amendments, biochar is one known for improving soil quality by stabilizing the occluded particulates and mineral-bound organic matter [20]. Meta-analysis on biochar indicates a significant increase in microbial biomass carbon (MBC; 26%) and microbial biomass nitrogen (MBN; 21%). However, maize (*Zea mays* L.) stover (7.5 t ha^{-1}) and its extracted biochar application (2.63 t ha^{-1}) were found to increase both soil organic carbon (SOC) and MBC, while a simultaneous rise in soil CO_2 efflux was also observed under both amendments (stover, 129%; biochar, 24%) [21]. In addition to this, biochar application also tends to show soil physicochemical properties varying with soil structure. For example, in the clay over sandy loam soil profile, biochar addition increased the relative moisture retention, aggregation, and nitrogen-use efficiency; and reduced qCO_2 (CO_2 efflux per unit MBC) [22]. These results suggest a need to further investigate the soil sustainability (quality and CO_2 efflux) aspects of biochar-based practices using varied soil types. Likewise, the microbial inoculum, viz. arbuscular mycorrhizal fungi (AMF), which enhances plant nutrient availability through its extraradical hyphae/long hyphal network, also potentially regulates the soil carbon pool (microbial biomass, 25%; soil respiration, ~15%) [23]. Zhang et al. [23] highlighted a negative relationship between the soil respiration and AMF association, i.e., reduced AMF colonization increased soil microbial respiration. Besides, reduced tillage (RT) is one of the widely applicable practices for improving soil carbon stock [24]. Reduced tillage, for example in maize cropping, increases crop residues, which in turn improves aggregate stability, organic matter, dehydrogenase, urease, phosphatase, and glomalin enzyme activity in the soil [25]. Similarly, in a maize–cowpea–rice system, RT improved soil organic carbon (water-soluble, readily mineralizable, and microbial biomass), dehydrogenase (13%), β-glucosidase (15%), and fluorescein diacetate (27%) enzymatic activity [14].

It is noteworthy that the above-mentioned agronomic interventions (organic amendments, microbial inoculum, and reduced tillage) are not sustainable when applied alone, because they either enhance soil respiration or release carbon into the atmosphere, or mainly suited for limited crops/cropping patterns, viz. a rice–wheat system [26–30]. Specifically, the high soil respiration rate reduces soil carbon (a key soil-fertility factor) stock and thereby reducing agroecosystem sustainability. Thus, integration of agronomic interventions is essential to improve soil carbon and agricultural sustainability. However, there is limited investigation of simultaneously improving the SOC while reducing carbon loss (soil respiration) by integrated agronomic practices. Notably, there is a need to discover the integrated impacts of different crop species, soil type, and agronomic interventions on soil quality; and microbial and soil respiration with their spatiotemporal variations [27–31]. Therefore, we hypothesize that integrated sustainable agronomic interventions (SAIs) could reduce the rate of soil and microbial respiration, which in turn could stabilize the soil carbon pool. In the present study, we validated the integrated effect of distinct types of crop species (cereal, legume, and oilseeds), soil types (covering three locations on the Indo-Gangetic Plain (IGP) of India) and SAIs (reduced tillage (RT), AMF, and rice-husk biochar) on soil- fertility parameters (SOC, MBC, N, P cycling-related enzymes, etc.) and microbial and soil respiration.

2. Materials and Methods

2.1. Experimental Sites and Cropping System

The three selected experimental sites (ES) fall under three different agro-climatic subzones on the Indo-Gangetic Plain (IGP) of North India: ES1-Varanasi (V) (25.2820° N, 82.9563° E), on the Eastern Gangetic Plain; ES2-Sultanpur (S) (26.2500° N, 82.0000° E), on the Central Plain of Agro-climatic zone V; and ES3-Gorakhpur (G) (26.7588° N, 83.3697° E), on the North-Eastern Plain of Agro-climatic zone IV (Figure 1). The agricultural background of the selected field plots at site ES1 (research plots inside BHU campus) and

ES2 and ES3 (both farmer field plots) includes seasonal vegetable cultivation for more than one decade. Two-year field experimental validations at aforesaid sites for selected crops were completed over consecutive years. The IGP is a rice–wheat producing region (150% cropping intensity) with frequent usage of straw for feeding livestock. However, since rice-husks largely remain left after rice milling, rice-husk-derived biochar were used for the present study. Maize (*Zea mays* var. K-65), black gram (*Vigna mungo* var. Shekher−3-599305), and mustard (*Brasicca juncea* var. Kala Sona) crops were selected in particular for this study, owing to dietary diversification and the taste/market/palate preferences that prevail within North Indian communities.
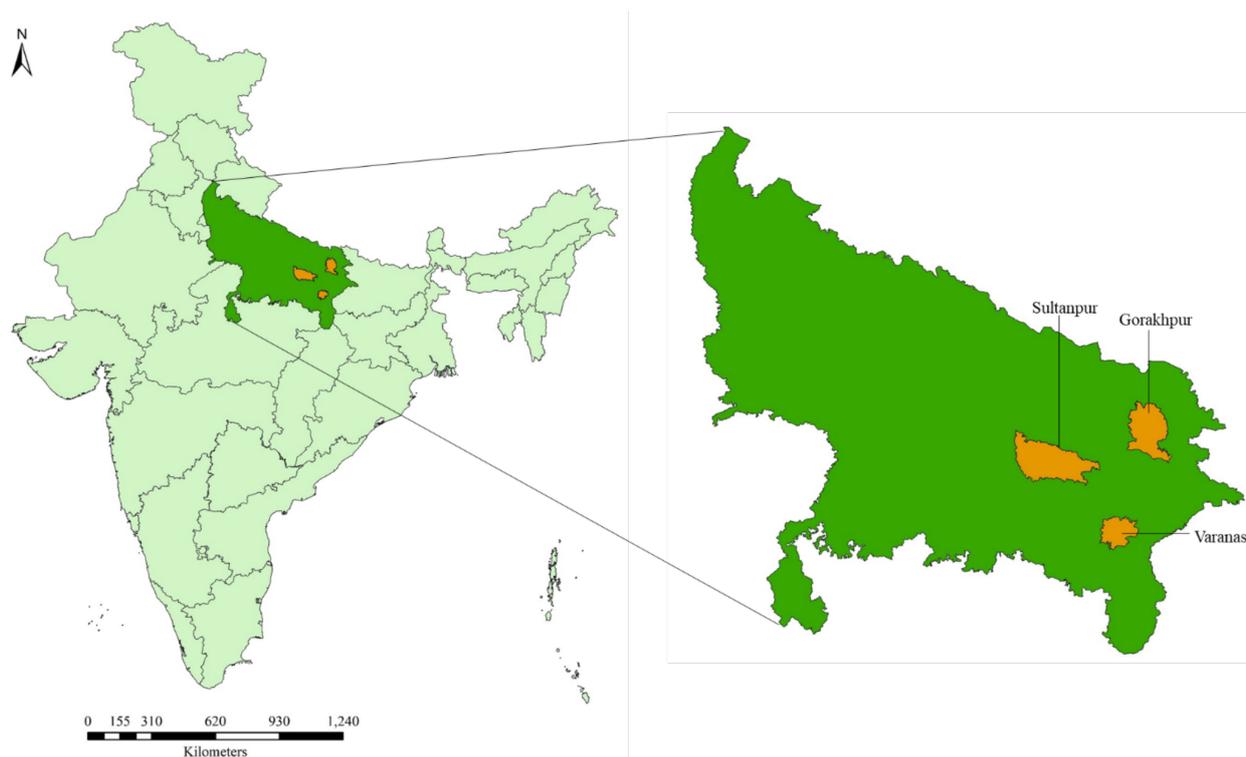

Figure 1. Study area: ES1-Varanasi (V), ES2-Sultanpur (S), and ES3-Gorakhpur (G), which fall under different agro-ecological subzones on the Indo-Gangetic Plain (IGP) of India.

The present study was carried out using three seasonal crops: mustard (plant-to-plant and row-to-row spacing of 10 and 30 cm, respectively) as winter or Rabi crops; and black gram and maize with 20×30 cm (plant $\times$ row) spacing as monsoon or Kharif crops, for consecutive years 2013–2014. Three seeds were sown at a 10 cm soil depth and aforesaid spacing. Initial soil samplings were done during field bed preparation using a hand-driven auger. Six soil subsamples were collected and mixed to make a composite soil sample per plot, and collected soil samples were analyzed for initial soil conditions of the study sites as described in Section 2.3. Similarly, soil samples were collected and analyzed prior to amendments to see the impact of years on soil quality. Periodic rainfall and relative humidity data were taken from regional meteorological stations and analyzed for the mean. Agro-metrological data are represented in Table 1. Each separated crop plot (6×6 m^2 plot size) was maintained in its own randomized block design (RBD) with four different treatments (i.e., sustainable agronomic interventions) at each selected location in triplicate. The crops sown in winter (October–November) and monsoon (June–July) were harvested during March–April and September–October, respectively. Crop-specific basal fertilization was given at 75% of the recommended dose in each field plot, followed by periodic irrigation and manual weeding as per the requirement. Basal doses of fertilizers were 120 kg N + 40 kg P + 20 kg S ha^{-1} for mustard; 25:50 and 25:25 kg of NPK and ZnSO$_4$ ha^{-1} for black gram; and 150, 75, and 37.5 kg N, P$_2$O$_5$, and K$_2$O ha^{-1}, respectively,

for maize. Nitrogen was applied in the form of urea (46% N), phosphate as single super phosphate (16% P_2O_5), and potassium as a muriate of potash (60% K_2O). Soil sampling, processing, and analysis were done as illustrated in our previous publications [32].

Table 1. Agrometeorological and soil quality of the study areas. For soil-quality values, means with different letters ($^{a-c}$) within a row indicate significant difference at $p \leq 0.05$.

Variables	ES1-Varanasi (V)	ES2-Sultanpur (S)	ES3-Gorakhpur (G)
(Agro-climatic zones) (Plain)	Agro-climatic zone IV Eastern Gangetic Plain	Agro-climatic zone V Central Plain	Agro-climatic zone IV North-Eastern Plain
Mean monsoon soil temperature (MMT in °C)	26.1 [b]	25.8 [c]	28.2 [a]
Mean winter soil temperature (MWT in °C)	18.3 [c]	17.5 [b]	20.6 [a]
Annual mean rainfall (mm)	1084	950	1607
Mean relative humidity (%)	76	58	80
Altitude amsl (m)	83.0	105	82
Soil types	Sandy fine loam	Sandy loam	Fine silty
Sand	48.9%	61.3%	55.1%
Silt	33.7%	22.7%	25.6%
Clay	17.4%	16.0%	19.3%
pH (1/4: soil/H_2O)	7.63 ± 0.07 [b]	8.57 ± 0.03 [a]	7.24 ± 0.05 [c]
Electrical conductivity (dS m^{-1})	0.167 ± 0.02 [b]	0.163 ± 0.04 [b]	0.178 ± 0.01 [a]
Moisture content (%)	5.01 ± 0.37 [b]	4.02 ± 0.19 [c]	6.13 ± 0.72 [a]
Bulk density (Mg m^{-3})	1.28 ± 0.05 [b]	1.45 ± 0.04 [a]	1.19 ± 0.01 [c]
Total organic carbon (g kg^{-1})	3.93 ± 0.09 [a]	3.11 ± 0.08 [b]	4.03 ± 0.11 [a]
Microbial biomass carbon (µg g^{-1})	141.67 ± 0.64 [b]	138.90 ± 6.51 [c]	147.66 ± 6.73 [a]

2.2. Sustainable Agrobiotechnological Interventions (SAIs)

Reduced tillage conditions were maintained in all the amendments, including the control field. The rice-husk biochar was prepared by using a conventional drum method, and biofertilizer in the form of plant-growth-promoting fungi, i.e., arbuscular mycorrhizal fungus (AMF) containing a consortia of *Glomus* sp., a market-based product of Indore Biotech Pvt. Ltd., were used. The biochar (8 ton ha^{-1}) and AMF (12 kg ha^{-1}) were applied into the soil each year during seed-bed preparation, prior to sowing of individual crops. Seed beds were prepared by conventional methods using a hand-driven spade and amendments were given in rows at standard spacing as mentioned in Section 2.1. A total of four treatments, i.e., sustainable agrobiotechnological interventions (SAIs), were given and details are as follows in Table 2.

Table 2. Sustainable Agrobiotechnological Interventions Employed in the Present Study.

S. No.	Treatments	Details of the Treatments
1.	Control (RT)	Control with reduced tillage having no biochar or arbuscular mycorrhizal fungus amendment (Control + RT)
2.	AMF (RT)	Arbuscular mycorrhizal fungi amendment with reduced tillage (AMF + RT)
3.	Biochar (RT)	Biochar amendment with reduced tillage (Biochar + RT)
4.	AMF + biochar (RT)	Arbuscular mycorrhizal fungus amended with biochar under reduced tillage (AMF + biochar + RT)

The above mentioned SAIs were applied for the crops during both the years of study. The basic properties of the rice-husk biochar were: pH (9.5), total carbon (410 g kg^{-1}), total

nitrogen (2 g kg^{-1}), total P (0.25 g kg^{-1}), calcium (1.08 g kg^{-1}), Mg (0.46 g kg^{-1}), and K (2.1 g kg^{-1}). Prior to field application, the biochar was sieved (2 mm sieve) to achieve a uniform particle size and oven-dried at 105 °C.

2.3. Measurement of Soil Physicochemical and Biological Properties

A triplicate of composite soil samples (six random soil subsamples) per plot were collected at 10 cm depth during the early flowering stage of crops. Soil samples were examined for pH and electrical conductivity (EC) using 1:4 soil:H$_2$O solution (pH and EC meter: Cyber Scan-500) with instrumental protocols [33], moisture content (MC) was determined gravimetrically, bulk density (BD) was determined according to Blake and Harte [34], and total organic carbon (TOC) was measured [35]. Soil particle size was analyzed via a hydrometer method. Soil temperature at 15 cm depth per day (at 12 randomly selected locations per treatment combination) was recorded after reaching stabilization using an inserted sensor attached to a mercury thermometer. Estimation of total nitrogen (TN) was done using the acid-digestion-mediated Kjeldahl method, [36] and total phosphorus (TP) by using the standard protocol [37]. The carbon-to-nitrogen (C:N) ratio was known from estimated TOC and TN. Microbial biomass carbon (MBC) and microbial biomass nitrogen (MBN) were determined using a modified fumigation–extraction method [38].

2.4. Monitoring Microbial and Soil Respiration

Microbial respiration was monitored under the mesocosm setup through the soil incubation and alkali absorption method in the laboratory [39]. Composite soil samples were collected from each experimental field plot followed by sieving (2 mm mesh) to evade the root and macrofauna-mediated respiration to specifically account for the microbe respiration. Sieved soils were incubated in butyl rubber-stopper-based plastic jars with a glass beaker containing 0.5 N NaOH at room temperature for 24 h. Further, in all incubations, microbial respiration was monitored weekly during the entire crop cycle. Similarly, every week, soil respiration was monitored in the experimental field with the same alkali trap method using 0.5 N NaOH, and evolved CO$_2$ was calculated as per the standard methodology of Dubey et al. [32]. A total of 18 replications of microbial and soil respiration data were recorded during each crop cycle.

2.5. Soil Enzyme Analysis

The soil β-glucosidase activity and urease activity were assayed according to Dubey et al. [40] using *p*-nitrophenyl- β-D-glucopyranoside as a substrate and tris buffer instead of NaOH, and Kandeler and Gerber [41] protocols through estimating the NH$_4$$^+$ released from 5 g soil after incubation at 37 °C for 2 h with tris hydroxyl methyl amino methane (THAM) buffer (pH 9.0), 0.2% of urea solution. Total glomalin content was estimated by the method given by Wright and Upadhyaya [42]. Briefly, the procedure was to mix 1 g of air-dried soil in 8 mL of 20 mM citrate (pH 7.0) and then autoclave the mixture for 30 min to remove the easily extractable glomalin. The mixture was further centrifuged (10,000× *g*) and the supernatant was removed. A further 8 mL of 50 mM citrate solution (pH 8.0) was added to the remaining soil and autoclaved for 60 min to extract the total glomalin (TG). Additional extractions with 50 mM citrate were done until the supernatant became straw-colored.

2.6. Statistical Analysis

Data of periodically collected site specific parameters were analyzed through an analysis of variance (ANOVA) followed by Tukey's honest significance difference post hoc tests. Means are represented in Table 1. All estimated soil parameters were analyzed in each of the samples collected per site and adopted practices in the *V. mungo*, *B. juncea* and *Z. mays* crops. Multifactorial ANOVA (MANOVA) was applied to decide whether the experimental site, crop species, and/or the sustainable agrobiotechnological practices (for moisture content, total organic carbon, nitrogen, C:N ratio, and microbial and soil

respiration) had a significant ($p \leq 0.05$) effect on the examined soil parameters. MANOVA was performed for multiple comparisons of examined means with a Duncan post hoc test at a 95% confidence interval. Before the MANOVA, obtained data were tested for multivariate normality and homogeneity of variance–covariance via the Shapiro–Wilk test of normality and Box's M test of equality of covariance, respectively. Soil-respiration data were log-transformed. All statistical analysis was done using SPSSv16, Chicago, USA. A linear correlation analysis was also performed to find whether there were significant ($p \leq 0.05$) relationships between the soil respiration, soil moisture, and temperature.

3. Results

3.1. Site-Specific Factors

Agrometeorological and soil characteristics of experimental site are provided in Table 1. ANOVA results indicated a significantly ($p \leq 0.05$) higher mean monsoon temperature (MMT) and mean winter temperature (MWT) of soil on the North-Eastern Plain of agro-climatic zone IV (ES3 site; MMT and MWT, 28.2 and 20.6, respectively) in comparison to the sites located on the Eastern Gangetic Plain (ES1) and Central Plain of agro-climatic zone V (ES2). The lowest bulk density was recorded at ES3, followed by ES1 and ES2 (Table 3); under the lower bulk density and higher temperature regime at ES3, there was a higher rate of soil and microbial respiration.

Table 3. Bulk density and total organic carbon changes in *V. mungo, B. juncea,* and *Z. mays* grown at three different sites. Data are the mean values of different practices. Data followed by different letters were significantly different ($p = 0.05$) CT: control field with conventional agronomic practices and tillage; T1: AMF (RT); T2: Biochar (RT); T3: AMF + Biochar (RT). AMF = Arbuscular mycorrhizal fungi; RT = Reduced tillage.

	Varanasi (ES1)				Sultanpur (ES2)				Gorakhpur (ES3)			
	2013		2014		2013		2014		2013		2014	
BD (g cm^{-3})												
Vigna mungo												
CT	1.25 ± 0.06 c	4.11 ± 0.21 d	1.26 ± 0.02 bc	4.15 ± 0.06 d	1.41 ± 0.05 c	4.12 ± 0.06 c	1.41 ± 0.03 c	4.13 ± 1.13 c	1.19 ± 0.05 b	4.14 ± 0.15 d	1.19 ± 0.02 c	4.21 ± 0.06 d
T1	1.26 ± 0.06 bc	5.18 ± 0.17 c	1.27 ± 0.00 b	5.26 ± 0.21 c	1.42 ± 0.02 b	5.11 ± 0.21 b	1.43 ± 0.03 b	5.14 ± 0.11 b	1.20 ± 0.08 ab	5.21 ± 0.82 c	1.21 ± 0.04 b	5.25 ± 0.03 b
T2	1.26 ± 0.04 bc	6.89 ± 0.17 ab	1.25 ± 0.06 c	6.94 ± 0.33 a	1.43 ± 0.09 ab	6.81 ± 0.33 a	1.44 ± 0.01 b	6.84 ± 0.19 ab	1.20 ± 0.09 ab	6.90 ± 0.28 ab	1.20 ± 0.03 b	6.91 ± 0.09 ab
T3	1.26 ± 0.07 bc	6.94 ± 0.63 a	1.28 ± 0.01 ab	6.98 ± 0.31 a	1.44 ± 0.07 a	6.85 ± 0.31 a	1.45 ± 0.04 ab	6.88 ± 0.09 a	1.20 ± 0.03 ab	6.96 ± 0.15 a	1.21 ± 0.03 ab	6.99 ± 0.05 a
Brassica juncea												
CT	1.26 ± 0.04 bc	4.09 ± 0.09 d	1.26 ± 0.03 bc	4.14 ± 0.68 d	1.41 ± 0.07 b	4.12 ± 0.02 c	1.41 ± 0.02 c	4.13 ± 0.43 c	1.19 ± 0.09 b	4.11 ± 0.07 d	1.19 ± 0.02 c	4.19 ± 0.02 d
T1	1.27 ± 0.09 b	5.02 ± 0.09 c	1.28 ± 0.03 ab	5.04 ± 0.39 cd	1.43 ± 0.02 ab	5.31 ± 0.14 b	1.44 ± 0.04 b	5.14 ± 0.61 b	1.20 ± 0.08 ab	5.09 ± 0.66 c	1.21 ± 0.04 ab	5.22 ± 0.04 c
T2	1.27 ± 0.03 b	6.32 ± 0.46 ab	1.28 ± 0.07 ab	6.38 ± 0.26 b	1.43 ± 0.03 ab	6.79 ± 0.43 ab	1.44 ± 0.03 b	6.82 ± 0.09 ab	1.21 ± 0.06 a	6.37 ± 0.11 b	1.21 ± 0.04 ab	6.39 ± 0.04 bc
T3	1.27 ± 0.09 b	6.91 ± 0.17 a	1.28 ± 0.07 ab	6.93 ± 2.18 a	1.44 ± 0.01 a	6.82 ± 0.41 a	1.45 ± 0.03 ab	6.85 ± 0.14 a	1.21 ± 0.08 a	6.94 ± 0.09 a	1.22 ± 0.6 a	6.54 ± 0.6 b
Zea mays												
CT	1.27 ± 0.00 b	4.02 ± 0.03 d	1.27 ± 0.02 b	4.13 ± 0.22 d	1.41 ± 0.02 c	4.11 ± 0.07 c	1.41 ± 0.07 c	4.14 ± 1.19 c	1.19 ± 0.07 b	4.18 ± 0.03 d	1.19 ± 0.02 c	4.19 ± 0.01 d
T1	1.27 ± 0.08 b	5.00 ± 0.05 c	1.28 ± 0.00 ab	5.02 ± 0.03 cd	1.43 ± 0.07 ab	5.12 ± 0.00 b	1.44 ± 0.05 b	5.13 ± 1.10 b	1.20 ± 0.01 ab	5.06 ± 0.11 c	1.21 ± 0.00 ab	5.19 ± 0.05 c
T2	1.27 ± 0.07 b	6.01 ± 0.06 b	1.28 ± 0.06 ab	6.24 ± 0.96 b	1.44 ± 0.03 a	6.77 ± 0.01 ab	1.45 ± 0.07 ab	6.80 ± 1.21 ab	1.21 ± 0.07 a	6.10 ± 0.13 bc	1.21 ± 0.06 ab	6.22 ± 0.01 bc
T3	1.28 ± 0.05 a	6.82 ± 0.49 ab	1.29 ± 0.01 a	6.87 ± 0.53 ab	1.44 ± 0.04 a	6.80 ± 0.58 a	1.46 ± 0.04 a	6.82 ± 1.89 ab	1.21 ± 0.01 a	6.91 ± 0.17 ab	1.22 ± 0.01 a	6.49 ± 0.02 b

All three experimental sites showed a significant difference in soil and microbial respiration. Site ES1 demonstrated moderate soil moisture, and a significant difference was found between the sites; the highest moisture content was recorded at ES3, whereas the lowest was noted at ES2. There was a significantly high mean rainfall and relative humidity (1607 mm and 80%, respectively) at ES3, followed by ES1 (1084 mm and 76%) and ES2 (950 mm and 58%). Similarly, the microbial and soil respiration was found in the order ES3 > ES1 > ES2 (Table 4). For the site-specific agro-meteorological parameters (soil temperature and bulk density, altitude, rainfall, and relative humidity), our results elucidated a significantly high ($p < 0.05$) TOC (4.05 g kg^{-1}), TN (0.42 g kg^{-1}), and C:N ratio (8.53) at ES3, which was followed by ES1 and ES2.

Table 4. Impact of the experimental site (ES), crop species (CS), sustainable agrobiotechnological practices (SAIs) on soil physico-chemical properties and soil sustainability indicators (microbial and soil respiration).

Individual and Interaction Results	Moisture Content (%)	Total Organic Carbon (g kg^{-1})	Total Nitrogen (g kg^{-1})	C:N Ratio	Microbial Respiration (mg CO$_2$ m^{-2} hrs^{-1})	Soil Respiration (mg CO$_2$ m^{-2} hrs^{-1})
Individual factor results						
p-value	<0.03	<0.01	<0.03	<0.01	<0.01	<0.01
Control	6.31 [d]	4.17 [d]	0.42 [d]	8.37 [d]	111.64 [a]	125.89 [a]
AMF (RT)	7.32 [b]	5.16 [c]	0.51 [c]	10.11 [b]	88.17 [c]	103.77 [c]
Biochar (RT)	8.11 [a]	6.82 [b]	0.58 [a]	8.96 [c]	104.01 [b]	120.13 [b]
AMF + Biochar (RT)	7.96 [c]	6.99 [a]	0.56 [b]	12.96 [a]	77.89 [d]	91.46 [d]
Experimental site (ES)						
p-value	<0.05	<0.04	<0.05	<0.03	<0.03	<0.01
ES1-Varanasi (V)	5.00 [b]	3.92 [a]	0.40 [a]	8.09 [b]	94.52 [b]	121.37 [b]
ES2-Sultanpur (S)	4.09 [c]	3.18 [b]	0.37 [b]	7.01 [c]	88.43 [c]	116.38 [c]
ES3-Gorakhpur (G)	6.23 [a]	4.01 [a]	0.42 [a]	8.53 [a]	101.02 [a]	132.60 [a]
Crop species (CS)						
p-value	0.74	0.04	0.03	0.05	0.04	0.01
Vigna mungo	6.30 [a]	4.31 [a]	0.53 [a]	7.88 [b]	119.28 [a]	167.02 [a]
Brassica juncea	6.28 [b]	4.18 [b]	0.49 [b]	8.91 [b]	113.96 [b]	151.95 [b]
Zea mays	6.23 [b]	4.09 [c]	0.43 [b]	9.18 [a]	110.20 [c]	138.39 [c]
Years (Y)						
p-value	0.05	0.04	0.57	0.95	0.02	0.04
2013	4.98 [b]	5.27 [b]	0.43 [a]	8.02 [a]	109.84 [b]	131.92 [a]
2014	5.03 [a]	6.01 [a]	0.47 [a]	8.09 [a]	117.31 [a]	133.74 [a]
Interaction results (*p*-value)						
SAIs × ES	0.0382 *	0.0301 *	0.6143 ns	0.5106 ns	0.0203 **	0.0324 *
SAIs × CS	0.0291 **	<0.0112 **	0.0274 **	0.0351 *	0.0006 ***	0.0159 **
SAIs × Y	<0.0226 **	0.0035 ***	<0.0371 *	0.0475 *	0.0121 **	<0.0248 **
ES × CS	ns	ns	ns	ns	ns	ns
ES × Y	ns	ns	ns	ns	ns	ns
CS × Y	ns	0.0489 *	0.0371 *	ns	0.0485 *	ns

Multivariate analysis of variance (MANOVA) for multiple comparisons analysis showing *p*-value in bold indicates statistical significance; for the Duncan post hoc test, mean values (*n* = 18) followed by different letters ([a–d]) within a column show significant difference ($p \leq 0.05$). The asterisk symbols (ns, *, **, ***) in each interactions result based on p-value represent no significant differences, significant difference ($p > 0.05$), ($p < 0.05$), ($p < 0.03$), and ($p < 0.01$), respectively.

The obtained field dataset from each experimental site (ES1 (V), ES2 (S), and ES3 (G)) during the period of the study indicated a significant difference in microbial ($p < 0.03$) and soil respiration ($p < 0.01$). The distinct rate of microbial (101, 94, and 88 mg CO_2 m^{-2} hrs^{-1}) and soil respiration (132, 121, and 116 mg CO_2 m^{-2} hrs^{-1}) were monitored at ES3, ES1, and ES2, respectively. In general, the rates of respiration observed were higher during the second year of study. In the interaction of the factors (shown in interaction result), significant interactions were found between the SAIs and study sites for the moisture content, TOC, and rate of microbial and soil respiration.

3.2. Crop Species-Specific Factors

The rates of microbial and soil respiration under different cropping systems (expressed in mg CO_2 m^{-2} hrs^{-1}) monitored in our study significantly varied with each cropping system; specifically, with the highest TOC (4.31 g kg^{-1}) and TN (0.53 g kg^{-1}) and lowest C:N ratio (7.88) under *V. mungo,* followed by *B. juncea* and *Z. mays* (Table 2). Higher carbon (3–5%) and nitrogen (8–23%) profiles were recorded in *V. mungo* over *B. juncea* and *Z. mays*. Consequently, the rate of microbial (4–8%) and soil (9–20%) respiration were also high under *V. mungo* cropping systems, followed by *B. juncea* and *Z. mays*. The C:N ratio trend was in the order *Z. mays* > *B. juncea* > *V. mungo* (Table 2), which also supported the response the of microbial and soil respiration dataset. We observed a lower rate of respiration in monocot (*Z. mays*) in comparison to dicot (*V. mungo* and *B. juncea*) crops. The impact of crop species on soil respiration was more highly significant ($p < 0.01$) than the microbial respiration ($p < 0.01$). The interaction of factors (shown in interaction result) suggested a higher impact of crop species under SAIs (Table 4).

3.3. Sustainable Agrobiotechnological Intervention (SAI)-Based Response

SAIs significantly altered the overall soil quality; for example, the biochar + RT treated field demonstrated the highest value of moisture content (8.11%, $p \leq 0.03$) and TN (0.58 g kg^{-1}, $p \leq 0.03$), whereas the maximum value of TOC (6.99 g kg^{-1}, $p \leq 0.01$) and C:N ratio (12.96, $p \leq 0.01$) was recorded for the AMF + biochar + RT field in comparison to the control (Table 2). In addition to the aforesaid physico-chemical properties, the soil biological characteristics also responded significantly to the adopted SAIs (specific results are presented in the next section). The highest increase in moisture and total nitrogen content was found for the field treated with biochar + RT (28% and 38%, respectively), followed by AMF + biochar + RT (26% and 33%) and AMF + RT (16% and 21%). Field data demonstrated significant 68%, 63%, and 24% improvements in TOC under AMF + biochar + RT, AMF + RT, and biochar + RT, respectively. A higher C:N ratio was recorded against the control under all three aforesaid practices, and the trend was in the order AMF + biochar + RT > AMF + RT > biochar + RT. The control field (RT) showed a higher rate of microbial and soil respiration for the entire study duration. In SAI-amended field plots, a significantly ($p < 0.01$) reduced rate of microbial (30%, 21%, and 7%) and soil respiration (27%, 18%, and 5%) were monitored for AMF + biochar + RT, AMF + RT, and biochar + RT, respectively. Overall, the rate of microbial and soil respiration was therefore in the order control > biochar + RT > AMF + RT > AMF + biochar + RT. Among all adopted SAIs, the AMF + biochar + RT was noted as the most promising practice, owing to improved soil fertility parameters and the reduced rate of soil respiration (Table 2, Figure 2, Figure 3). Correlation of the factors (interaction results) indicated SAIs had the highest significant impact with crop species (CS) ($p < 0.05$), over the years (Y) ($p < 0.01$) at each experimental site (ES) for the moisture content, TOC, and TN plus C:N ratio (Table 2).
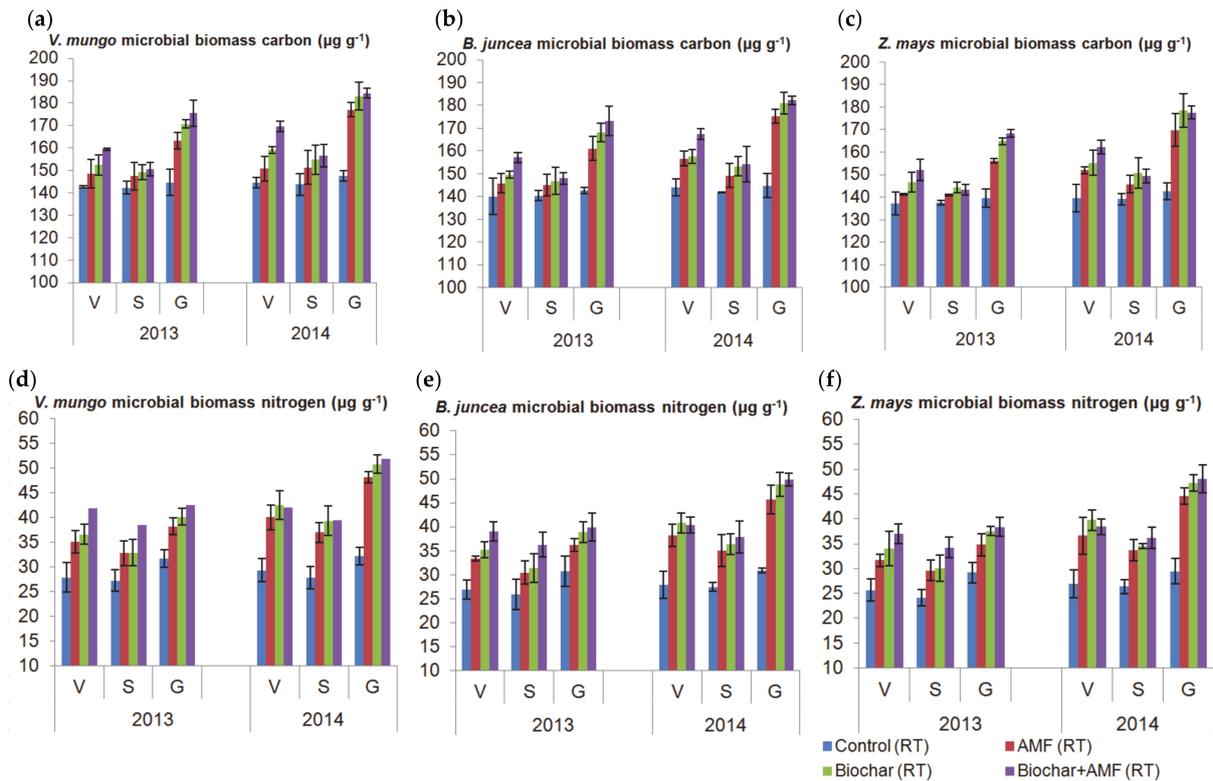
Figure 2. Soil microbial biomass carbon (MBC) and microbial biomass nitrogen (MBN) results showing significant increases under integrated SAI-managed fields in comparison to control at the three experimental sites (ES1-Varanasi (V), ES2-Sultanpur (S) and ES3-Gorakhpur (G)) for three crops, i.e., MBC (**a**–**c**) and MBN (**d**–**f**) for V. mungo, B. juncea, and Z. mays, respectively. A total of four given treatments included one control (RT), i.e., reduced tillage with no biochar and AMF amendments; and three SAIs, i.e., AMF + RT (arbuscular mycorrhizal fungi amendment with reduced tillage), biochar + RT (biochar amendment with RT), and AMF + biochar + RT (integration of all aforementioned SAIs). Data values represent mean ± SD of different treatments (*n* =18) with significant difference (*p* = 0.05).
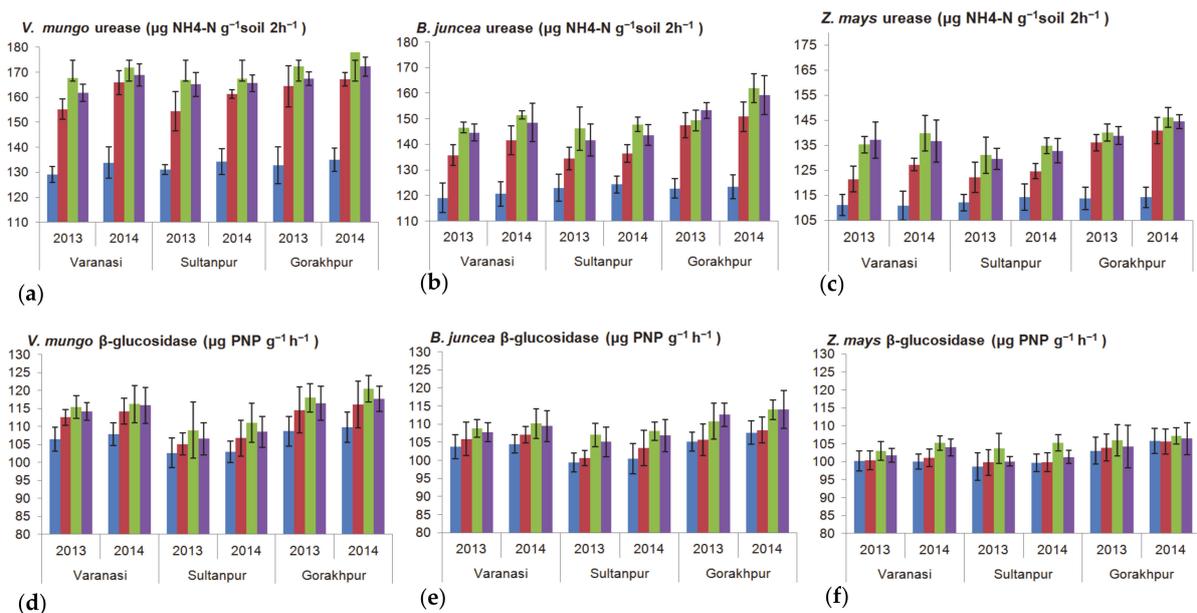

Figure 3. *Cont.*

(g) **(h)** **(i)**

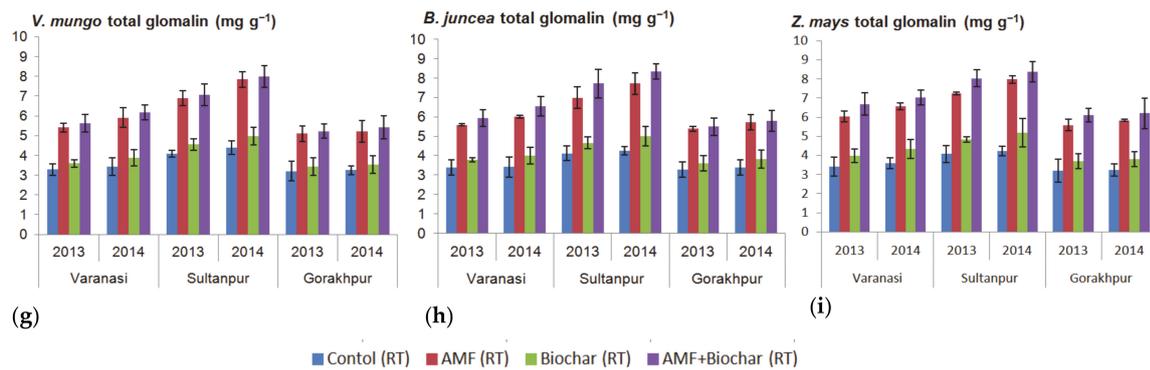■ Contol (RT) ■ AMF (RT) ■ Biochar (RT) ■ AMF+Biochar (RT)

Figure 3. Soil enzymes, i.e., urease, β-glucosidase, and glomalin, activity results showing significant improvement under sustainably integrated SAI-managed fields in comparison to the control at three experimental sites (ES1-Varanasi (V), ES2-Sultanpur (S), and ES3-Gorakhpur (G)) for three crop fields, i.e., urease (**a–c**), β-glucosidase (**d–f**), and glomalin (**g–i**) for *V. mungo*, *B. juncea*, and *Z. mays*, respectively. Data values represent mean ± SD of different treatments (*n* = 18) with significant difference (*p* = 0.05).

3.4. Soil Microbial Biomass and Soil Enzymes Response

Soil MBC, MBN, and soil enzymes like β-glucosidase (µg g^{-1}), urease (µg g^{-1}), and total glomalin (mg g^{-1}) estimated in the study significantly ($p < 0.05$) varied with the ES, CS, and SAIs. The increase in MBC (7–25%) and MBN (37–63%) were found under AMF + biochar + RT practiced for *V. mungo*, *B. juncea*, and *Z. mays* at each experimental site (Figure 2). The increase in MBC and MBN followed the order, SAI-wise, of: AMF + biochar + RT > biochar + RT > AMF + RT > control; crop species (CS)-wise: *V. mungo* > *B. juncea* > *Z. mays*; and experimental site (ES)-wise: ES3 > ES1 > ES2. Interestingly, we monitored a reduced rate of microbial and soil respiration with improved MBC and MBN under AMF + biochar + RT.

Results indicated enhanced enzymatic activity in urease (27–31%) and β-glucosidase (8–22%) under biochar+RT practiced *V. mungo*, *B. juncea,* and *Z. mays* crops (Figure 3). The increase for urease and β-glucosidase followed the order, SAI-wise, of: biochar + RT > AMF + biochar + RT > AMF + RT > control, crop species (CS)-wise: *V. mungo* > *B. juncea* > *Z. mays*; and experimental site (ES)-wise: ES2 > ES1 > ES3. The improvement for glomalin activity followed the order, SAI-wise, of: AMF + biochar + RT > AMF + RT> biochar+RT>control, crop species (CS)-wise: *Z. mays* > *B. juncea* > *V. mungo*; and experimental site (ES)-wise: (S2 > ES1 > ES3. As per correlation results, soil respiration was negatively correlated with TG activity (r = −0.85, r= −0.82, and r= −0.62) under the adopted SAIs practices. Among all studied enzymes, the maximum increase was recorded in glomalin (7–98%), followed by urease (16−32%) and β-G activity (1−10%) under biochar + RT, followed by AMF + biochar + RT > AMF + RT at each experimental site (Figure 3). The observed dataset showed higher β-G and urease activity in biochar + RT treated plots, and soil and microbial respiration data were also observed in accordance with this (Figure 3).

4. Discussion

Soil carbon management (enhancing carbon content and reducing carbon emissions) in agroecosystems has been one of the daunting sustainability challenges worldwide in recent years. In relation to this, scientific studies are revealing sustainable ways to address the two aforesaid carbon management strategies either as separate subjects or through non-integrated agrobiotechnological interventions. For example, SAIs such as reduced tillage (RT), biochar, and biofertilizers, viz. AMF applications, are three carbon management strategies demonstrated to hold certain potential [20,23,25]. Interestingly, in the present study, the integrated impact of SAIs (AMF + biochar + RT) on the simultaneous increase in soil carbon and fertility and reduced soil and microbial respiration was validated at the field scale (Figure 4). In coalition with SAIs, our study also disentangled the effects of distinct

experimental sites (ES1, ES2, ES3) and crop species (*Vigna mungo, Brassica juncea,* and *Zea mays*), thereby aligning the study as the first of its kind, to the best of our knowledge.
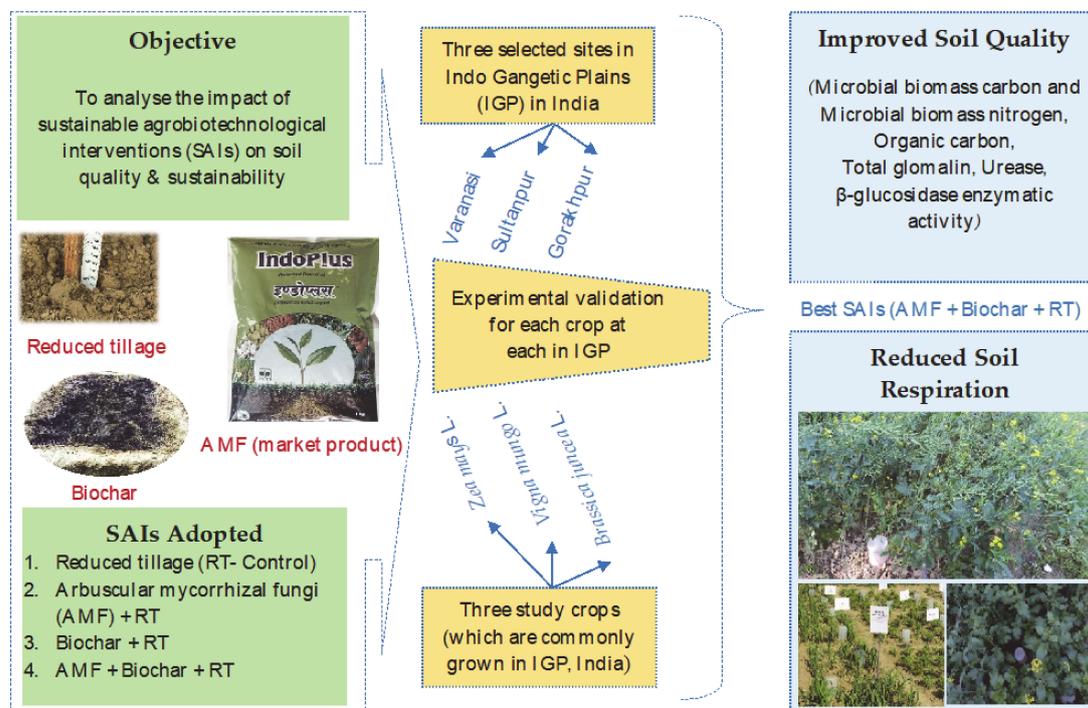

Figure 4. Flowchart of overall practices employed for three different crops grown at three different sites of the Indo-Gangetic Plain, North India as well as the effects of sustainable agrobiotechnological interventions on soil quality and respiration.

4.1. Role of Site-Specific Factors in Spatiotemporal Variations in Microbial and Soil Respiration

Observed field data validated the variation in agro-meteorology of each study site falling under different agro-climatic zones (Table 1), which contributed to the spatiotemporal variation of the microbial ($p < 0.05$) and soil respiration during both the years of study ($p < 0.07$) (Table 4). The present study indicated higher temperature, rainfall, mean relative humidity, and soil moisture; and lower bulk density at ES3-Gorakhpur (G), and these parameters demonstrated opposite trends at ES2-Sultanpur (S). Increased temperature can increase the root respiration, through greater root biomass, exudation of carbon (sugars, amino acids, organic acids, etc.) [43] and decomposition of soil organic matter via enhaced soil microbial activity. A study of elevated soil temperature (top 100 cm) found a significant increase (34−37%) in soil respiration [44]. Recent research highlighted the ~10% increase in the rate of soil microbial respiration due to changes in global mean temperature over the preceding 25 years [6]. The present study assigned significant differences ($p < 0.05$) in site-specific agro-meteorology of the studied sites (ES1, ES2, ES3) as key factors that regulated the spatiotemporal variation of microbial and soil respiration.

The relatively lower bulk density (Table 4) at the ES3 site in comparison to ES1 and ES2 was probably responsible for enhancing the rate of respiration via improved soil porosity, moisture, and resultant microbial activity at ES3. Differences in moisture possibly owe to the differences in soil texture, bulk density, and total organic carbon noted at sites ES1, ES2, and ES3. For one example, carbon-rich fine silty soil (largely at ES3) demonstrated greater moisture content over sandy loam soil [45,46]. Irrespective of site, the soil temperature, moisture, and carbon content were crucial factors in determining the soil carbon dynamics, depending on rate regulation of organic matter decomposition and microbial activity [5,47], as also confirmed via the agro-meteorological results of our study (Table 1). In addition, the present study highlights the obvious site-specific significant changes in the microbial (ES3, 101; ES1, 94; and ES2, 88 mg CO_2 m^{-2} hrs^{-1}) and soil respiration (ES3, 132; ES1, 121; and

ES2, 116 mg CO_2 m^{-2} hrs^{-1}) as cumulative consequences of altered altitude, rainfall, and relative humidity. The enhanced rate of respiration found at site ES3, which had the highest relative humidity (80%), is in line with a previous study that indicated humidification-based enhancement of litter inputs and a 28% rise in the microbial respiration rate [48]. Besides agro-meteorological datasets, the present study also elucidated a higher TOC, TN, and C:N ratio as the key regulatory factors behind the spatiotemporal variation in microbial and soil respiration. The noted increase in rate of microbial activity (enhanced due to increased soil organic carbon [49]) and soil respiration at the ES3 site was anticipated because of its high soil moisture, carbon, temperature, and relative humidity (Tables 1 and 2) [50]. Enhanced respirations were also due to the increased soil microbial biomass having the potential to increase the soil organic matter decomposition. As in this study, the spatiotemporal variation in the rate of soil respiration also was identified in a global modeling study that reported the region of study (tropical/temperate), temperature, and precipitation as key regulating factors in soil respiration [51].

4.2. Impact of Crop Species on Microbial and Soil Respiration

The observed results for soil-fertility parameters, i.e., TOC ($p < 0.03$), TN ($p < 0.02$), and C:N ratio ($p < 0.04$), showed significant differences in magnitude for each studied crop species, i.e., *V. mungo*, *B. juncea*, and *Z. mays* (Table 2). The C and N content found were in the order *V. mungo* > *B. juncea* > *Zea mays*. *V. mungo* demonstrated the highest carbon and nitrogen content, but the lowest C:N ratio. *V. mungo* had relatively high crop density (cropping distance), capability of nitrogen fixation, fine root growth, and frequent leaf-litter addition in comparison to *B. juncea* and *Z. mays* crops. The cumulative impact of the witnessed crop-specific attributes of *V. mungo* was assumed to be the likely cause of organic matter availability, thereby enhancing microbial activity and organic carbon breakdown; which consequently resulted in high microbial and soil respiration rates [15,27,52,53]. Furthermore, the *Vigna* sp. characteristics (such as intense crop density and fine roots) were anticipated to increase the root-exudate secretion (a primary carbon and energy source for microbes), resultant microbial activity, and the rate of carbon mineralization and respiration [54]. The crop-specific distinct rate of microbial and soil respiration followed the order *V. mungo* > *B. juncea* > *Zea mays*. The significant variation in the rate of microbial ($p < 0.0381$) and soil respirations ($p < 0.01$) noted for the *V. mungo*, *B. juncea*, and *Z. mays* crops in present study was in support of the impact of crop-specific root exudation and the type of rhizodeposits. The crop root exudates and rhizodeposits had cues of recruiting specific rhizospheric microbiome, which in turn results in varied nutrient turnover (rate and type) via specific soil enzymatic processes and secretions [55]. This study examined the lower rate of respiration in the monocot (*Z. mays*) over the dicots (*V. mungo* and *B. juncea*). In relation to this, Innes et al. and Orio et al. reported that distinct microbial community shifts in monocots and dicots could be the plausible reason behind variation in rates of respiration [56,57]. The crop-specific C:N ratio is also significant parameter in determining the rate of carbon turnover. For example, a low soil C:N ratio could also augment the organic matter decomposition and hasten the rate of respiration [32,58], as witnessed in the *V. mungo* crop in the present study. The highest ($p < 0.01$) impact of crop species (CS) on soil respiration over the microbial respiration ($p < 0.03$) signifies the importance of crop-specific root traits (root growth, density, root exudation) and root respiration in contributing to rate of soil respiration. In corroboration with this study, a previous research study done with *Brassica campestris* L. crop plant suggested > 44% contribution of root respiration to the soil respiration [59]. Interaction results validated a higher impact of crop species (CS) with sustainable agrobiotechnological interventions (SAIs), but not for the experimental sites (ES) and years of study (Y) (Table 2). Overall, our study indicated the promising role of crop-specific attributes in contributing to any specified rate of respiration.

4.3. Sustainable Agrobiotechnological Interventions (SAIs) Response to Soil Quality, and Microbial and Soil Respiration

SAIs significantly improved the only key soil properties such as moisture content ($p < 0.01$), TOC ($p < 0.01$), TN ($p < 0.03$), and C:N ratio ($p < 0.01$) under AMF + RT, biochar + RT, and AMF + biochar + RT treated fields in comparison to the control (Table 2). Interaction results indicated the biochar + RT treated plot retained the highest moisture at each experimental site (ES), crops (CS) and year of study (Y), and N content at each CS and Y. Nutrient richness in biochar led to N release in soil, thereby improving soil TN, while high moisture could be due to biochar's capability to enhance soil hydraulic characteristics (soil porosity and water-holding capacity), macropores, and bioturbation activity [40,60]. Biochar-based interventions demonstrated an insignificant change in pH and EC.

AMF + biochar + RT was found to be the most promising SAI, as it demonstrated the maximum improvement in carbon content (67%), with a reduced carbon emission rate (microbial, 30% and soil respiration, 27%). The likely reason was the integrated nature of the agro-biotechnological interventions that utilized AMF along with biochar and reduced tillage. In the integration, AMF + biochar + RT practices added more biochar-based recalcitrant carbon, providing better protection to soil carbon via secreting the soil-aggregation-specific glomalin enzyme and maintaining a less-oxidative environment, changing the fungal-to-bacterial ratio and trapping more carbon in fungal biomass. Such conditions induce overall biological activity with the reduced rate of respirations. Based on previous results for independent intervention, the improved soil carbon and reduced carbon emissions under integrated SAIs may be due to the biochar-based carbon input and AMF-based carbon protection, supplemented by a reduced rate of oxidation under reduced tillage practices [19,21,32]. In particular, AMF has the highest carbon use efficiency and induces soil aggregation, whereas biochar impacts soil physicochemical properties, nutrient availability, and soil enzymes, which help in improving the soil carbon [61]. A high C:N ratio under each SAI (AMF + RT, biochar + RT, and AMF + biochar + RT) indicated the importance of biochar, AMF, and reduced-tillage-mediated carbon stabilization, and a reduced rate of respiration (Table 2), as also supported by existing findings [27,53,58,62].

Integration of SAIs and soil biochemistry are overall key factors that control soil quality, and microbial and soil respiration, as evident from the present research (Figures 2 and 3, Table 2). First, rice-husk biochar utilized in integrated SAIs (AMF + biochar + RT) enhanced the proportion of soil-recalcitrant-carbon and stabilized rhizodeposits, sequestering C by inducing micro-aggregation through organo-mineral interactions and a fungi:bacteria ratio with higher carbon-use efficiency [20,22,60]. Second, AMF enhanced the overall soil quality, with a reduced rate of microbial (21%) and soil (18%) respiration, which was likely due to massive fungal hyphal-mediated nutrient availability (23% and 21% increases in TOC and TN, respectively) and a change in soil aggregation via AMF-based enzymes (discussed further in a later section) [23,63]. Third, reduced-tillage practices also improved soil aggregation, SOM, and microbial biomass, and thus captured more carbon in soil aggregates and microbial biomass [64]. Correlation of the factors suggested the highest significant impact of SAIs on the crop species and soil properties (moisture content, TOC, TN, and C:N ratio) during the study period at each experimental site (Table 2), while interaction results demonstrated a higher significant ($p < 0.06$) response of microbial respiration. Overall, correlation and interaction results cross-validated the integration of each SAI as the most suitable practice in agroecosystem sustainability (improved soil quality with a reduced rate of microbial and soil respiration).

4.4. Soil MBC, MBN, and Soil Enzymes' Response to Microbial and Soil Respiration

Carbon (MBC, β-glucosidase, and total glomalin) and nitrogen (MBN and urease)-related soil microbial biomass and soil enzymes were analyzed to understand the belowground factors of the microbial and soil respiration. The obtained results showed a significant increase ($p < 0.05$) in MBC and MBN in all integrated SAIs, irrespective of the experimental sites and crop species (Figure 2). The highest response was recorded for AMF

+ biochar + RT. The higher accumulation of MBC and MBN was attributed to the integration of SAIs, where AMF physiology (massive hyphal growth and soil aggregation through fungal biomass), biochar impact, and RT practices (which cause minimum soil disturbance) showed combined impacts. As in this study, an increase in MBC and enzymatic activity under reduced tillage was also reported in other regional and global studies [13,14]. A plausible explanation for the higher MBN could be the improved nitrogen content and N use efficiency through biochar- and AMF-induced changes in soil properties, including nitrogen availability [61,64,65]. Along with improved MBC and MBN, a reduced rate of microbial and soil respiration was monitored under AMF + biochar + RT (Figure 2, Table 2). This was attributed to the high microbial biomass leading to the alteration in the microbial community (preferably fungal biomass), which utilizes the soil carbon for self-biomass accumulation. Improvements in microbial biomass and soil carbon were also reported by Kallenbach et al. and Bowles et al., who described a higher rate of carbon allocation from soil to microbial biomass [64,66].

Soil enzymes are one of the most sensitive parameters governing soil quality, nutrient cycling, and rate of respiration. Two carbon-related (β-Glucosidase and glomalin) and one nitrogen-related (urease) soil enzymes were chosen for the assessment of soil biochemical response to the rate of respiration with the adopted SAIs.

In addition to microbial biomass, the soil enzymes also demonstrated significant changes under SAIs. Selected enzymes like β-Glucosidase played a crucial role in plant-derived SOM decomposition through cellulose degradation that induced the organic-matter variation, whereas urease was responsible for nitrogen mineralization and cycling. Notably, enhanced urease (27–31%) and β-glucosidase (8–22%) activity was noted in all test crops maintained under biochar + RT in the present study. These results were in accordance with previous reports that noted 15% and 19% increases in urease and β-glucosidase activities, respectively, in organically managed soil, and around 1.5 times increase in undervegetated soil [67,68]. Among the studied enzymes, the highest (98%) increase in glomalin enzymatic response and its negative correlation with soil respiration were noted in the present research. Accordingly, a slower rate of respiration was recorded under AMF + biochar + RT (with high glomalin activity) compared to AMF + RT and biochar + RT among the crops and sites [69]. Glomalin enzyme has also is related to soil aggregation and carbon sequestration [42,63]. Therefore, a higher glomalin activity under AMF + biochar + RT possibly could have improved the soil carbon and thus reduced the rate of microbial and soil respiration. The other enhanced soil enzymatic (β-glucosidase and urease) activity under biochar + RT indicated the higher soil carbon and nitrogen dynamics, which resulted in an increased rate of respiration in comparison to other given SAIs [68]. For AMF-based integrated practices (AMF + biochar + RT and AMF + RT), the activated AMF and its carbon-capturing enzyme (glomalin) may be responsible for the improved soil aggregation, C:N ratio, and carbon-use efficiency, consequently reducing the respiration rate. Previous studies have reported similar results, and suggested the role of AMF in soil aggregation and carbon capture [23,63]. Furthermore, any changes in enzyme activities and rates of respiration could also be due to alteration of the aerobic and anaerobic microbial population and a poor oxidative environment for microbial activity under reduced-tillage practices [11,13].

5. Conclusions

The present research highlights the significant impacts of integrated SAIs on soil-quality parameters and key soil-sustainability indicators, i.e., microbial and soil respiration, across varied tropical agroecosystems on the Indo-Gangetic Plain of India) for three commonly grown cereal, pulse and oilseed crops. The SAIs applied included microbial inoculum (AMF), organic input (biochar), and reduced tillage. Results observed in the study suggested that important soil-quality parameters (SOC, MBC, MBN, soil enzymes, viz. urease, β-glucosidase, glomalin) and rate of microbial and soil respiration were all intrinsically governed by administered ES, CS, and SAIs during the period of study. The

complex belowground soil biochemistry remained as the major driver of these governances, as noticed in field experiments. Specifically, SAIs AMF + biochar + RT for *V. mungo* recorded the highest soil organic carbon and MBC at each experimental site. Over control (RT), the AMF + biochar + RT for *Z. mays* was noted to have the maximum reduction in microbial and soil respiration. Enhanced glomalin enzyme activity was noted in *Z. mays* cropping for each SAI. Therefore, exploiting *Z. mays* cropping in the IGP region is a promising climate-resilient agriculture option. The observed significant negative correlation between soil respiration and glomalin activity under AMF + biochar + RT, AMF + RT, and biochar + RT was indicative of the role glomalin played under a reduced rate of soil respiration. Overall, our study concludes, in comparison to control, that each applied SAI was demonstrated to improve soil carbon and reduce rates of respiration. Based on explored soil sustainability indicators and owing to enhanced soil physio-chemical and biological properties, the integrated SAI i.e., AMF + biochar + RT is proposed as promising in adaptive agriculture practices. Such climate-adaptive practices are essential for improving crop productivity, health, and socioeconomic conditions of farmers, as well as for attaining the targets of UN Sustainable Development Goals (UN-SDGs) and the UN Decade on Ecosystem Restoration (UN-DER) [70]. Furthermore, our study also encourages rice-husk biochar usage as a vital impetus for a cleaner/climate-smart way of agro-residue utilization. Besides spatiotemporal variations in microbial and soil respiration, our study also revealed the carbon estimates for cereal, legume, and oilseed cropping in North India. Conclusively, under the current status of global soil carbon loss and the negative impacts of changing climate, assessing the important soil-sustainability indicators (microbial and soil respiration) could provide a better carbon-management strategy for agro-environmental sustainability.

Author Contributions: Conceptualization, R.K.D. and P.C.A.; methodology, R.K.D., P.K.D., R.C.; validation, R.K.D., P.K.D., R.C., and P.C.A.; formal analysis, R.K.D., C.S.R., and P.C.A.; investigation, R.K.D. and P.C.A.; resources, P.C.A.; data curation, R.K.D., C.S.R., and P.C.A.; writing—original draft preparation, R.K.D., P.K.D., and R.C.; writing—review and editing, R.K.D., P.K.D., R.C., C.S.R. and P.C.A.; supervision, P.C.A. All authors have read and agreed to the published version of the manuscript.

Conflicts of Interest: The authors declare no conflict of interest.

References

1. Smith, K.A.; Ball, T.; Conen, F.; Dobbie, K.E.; Massheder, J.M.; Rey, A. Exchange of greenhouse gases between soil and atmosphere: Interactions of soil physical factors and biological processes. *Eur. J. Soil Sci.* **2003**, *54*, 779–791. [CrossRef]
2. Abhilash, P.C.; Dubey, R.K. Integrating aboveground-belowground responses to climate change. *Curr. Sci.* **2014**, *106*, 1637–1638.
3. Abhilash, P.C.; Dubey, R.K. Root system engineering: Prospects and promises. *Trends Plant Sci.* **2015**, *20*, 408–409. [CrossRef]
4. Lal, R. Soil Carbon sequestration impacts on global Climate Change and Food Security. *Sustainability* **2004**, *304*, 1623–1627. [CrossRef]
5. Bond-Lamberty, B.; Thomson, A.M. Temperature-associated increases in the global soil respiration record. *Nat. Cell Biol.* **2010**, *464*, 579–582. [CrossRef] [PubMed]
6. Bond-Lamberty, B.; Bailey, V.L.; Chen, M.; Gough, C.M.; Vargas, R. Globally rising soil heterotrophic respiration over recent decades. *Nat. Cell Biol.* **2018**, *560*, 80–83. [CrossRef]
7. INCCA. *India: Greenhouse Gas Emissions 2007*; Indian Network for Climate Change Assessment (INCCA) the Ministry of Environment & Forests Government of India: New Delhi, India, 2010; p. 63.
8. Dubey, P.K.; Singh, G.S.; Abhilash, P.C. Agriculture in a changing climate. *J. Clean. Prod.* **2016**, *113*, 1046–1047. [CrossRef]
9. Dubey, R.K.; Tripathi, V.; Abhilash, P.C. Book review: Principles of plant-microbe interactions: Microbes for sustainable agriculture. *Front Plant Sci.* **2015**, *6*, 986. [CrossRef]
10. Dubey, R.K.; Tripathi, V.; Edrisi, S.A.; Bakshi, M.; Dubey, P.K.; Singh, A.; Verma, J.P.; Singh, A.; Sarma, B.K.; Rakshit, A.; et al. Role of plant growth promoting microorganisms in sustainable agriculture and environmental remediation. In *Advances in PGPR Research*; Singh, H.B., Sharma, B., Kesawani, C., Eds.; CABI Press: Boston, MA, USA, 2017.

11. Dubey, R.K.; Tripathi, V.; Prabha, R.; Chaurasia, R.; Singh, D.P.; Rao, C.S.; El-Keblawy, A.; Abhilash, P.C. *Unravelling the Soil Microbiome*; Springer International Publishing: Berlin/Heidelberg, Germany, 2020.

12. Sarkar, D.; Kar, S.K.; Chattopadhyay, A.; Shikha; Rakshit, A.; Tripathi, V.K.; Dubey, P.K.; Abhilash, P.C. Low input sustainable agriculture: A viable climate-smart option for boosting food production in a warming world. *Ecol. Indic.* **2020**, *115*, 106412. [CrossRef]

13. Melero, S.; López-Garrido, R.; Madejón, E.; Murillo, J.M.; Vanderlinden, K.; Ordóñez, R.; Moreno, F.; Ordóñez-Fernández, R. Long-term effects of conservation tillage on organic fractions in two soils in southwest of Spain. *Agric. Ecosyst. Environ.* **2009**, *133*, 68–74. [CrossRef]

14. Neogi, S.; Bhattacharyya, P.; Roy, K.S.; Panda, B.B.; Nayak, A.K.; Rao, K.S.; Manna, M.C. Soil respiration, labile carbon pools, and enzyme activities as affected by tillage practices in a tropical rice–maize–cowpea cropping system. *Environ. Monit. Assess.* **2014**, *186*, 4223–4236. [CrossRef]

15. Mathew, I.; Shimelis, H.; Mutema, M.; Chaplot, V. What crop type for atmospheric carbon sequestration: Results from a global data analysis. *Agric. Ecosyst. Environ.* **2017**, *243*, 34–46. [CrossRef]

16. Abhilash, P.C.; Dubey, R.K.; Tripathi, V.; Gupta, V.K.; Singh, H.B. Plant Growth-Promoting Microorganisms for Environmental Sustainability. *Trends Biotechnol.* **2016**, *34*, 847–850. [CrossRef]

17. Abhilash, P.C.; Tripathi, V.; Edrisi, S.A.; Dubey, R.K.; Bakshi, M.; Dubey, P.K.; Singh, H.B.; Ebbs, S.D. Sustainability of crop production from polluted lands. *Energy Ecol. Environ.* **2016**, *1*, 54–65. [CrossRef]

18. Dubey, R.K.; Tripathi, V.; Dubey, P.K.; Singh, H.B.; Abhilash, P.C. Exploring rhizospheric interactions for agricultural sustainability: The need of integrative research on multi-trophic interactions. *J. Clean. Prod.* **2016**, *115*, 362–365. [CrossRef]

19. Krauss, M.; Ruser, R.; Müller, T.; Hansen, S.; Mäder, P.; Gattinger, A. Impact of reduced tillage on greenhouse gas emissions and soil carbon stocks in an organic grass-clover ley—winter wheat cropping sequence. *Agric. Ecosyst. Environ.* **2017**, *239*, 324–333. [CrossRef]

20. Weng, Z.; Van Zwieten, L.; Singh, B.P.; Tavakkoli, E.; Joseph, S.; Macdonald, L.M.; Rose, T.J.; Rose, M.T.; Kimber, S.W.L.; Morris, S.; et al. Biochar built soil carbon over a decade by stabilizing rhizodeposits. *Nat. Clim. Chang.* **2017**, *7*, 371–376. [CrossRef]

21. Yang, X.; Meng, J.; Lan, Y.; Chen, W.; Yang, T.; Yuan, J.; Liu, S.; Han, J. Effects of maize stover and its biochar on soil CO_2 emissions and labile organic carbon fractions in Northeast China. *Agric. Ecosyst. Environ.* **2017**, *240*, 24–31. [CrossRef]

22. Zhou, H.; Zhang, D.; Wang, P.; Liu, X.; Cheng, K.; Li, L.; Zheng, J.; Zhang, X.; Zheng, J.; Crowley, D.; et al. Changes in microbial biomass and the metabolic quotient with biochar addition to agricultural soils: A meta-analysis. *Agric. Ecosyst. Environ.* **2017**, *239*, 80–89. [CrossRef]

23. Zhang, B.; Li, S.; Chen, S.; Ren, T.; Yang, Z.; Zhao, H.; Liang, Y.; Han, X. Arbuscular mycorrhizal fungi regulate soil respiration and its response to precipitation change in a semiarid steppe. *Sci. Rep.* **2016**, *6*, 19990. [CrossRef]

24. Dubey, P.K.; Singh, G.S.; Abhilash, P.C. *Adaptive Agricultural Practices: Building Resilience under Changing Climate*; Springer: Berlin/Heidelberg, Germany, 2020; ISBN 978−3-030−15518-6.

25. Roldán, A.; García, M.D.M.A.; Alguacil, M.; Caravaca, F. Changes in soil enzyme activity, fertility, aggregation and C sequestration mediated by conservation tillage practices and water regime in a maize field. *Appl. Soil Ecol.* **2005**, *30*, 11–20. [CrossRef]

26. Nan, W.; Yue, S.; Li, S.; Huang, H.; Shen, Y. The factors related to carbon dioxide effluxes and production in the soil profiles of rain-fed maize fields. *Agric. Ecosyst. Environ.* **2016**, *216*, 177–187. [CrossRef]

27. Gao, B.; Ju, X.; Su, F.; Gao, F.; Cao, Q.; Oenema, O. Comparison of soil respiration in typical conventional and new alternative cereal cropping systems on the North China Plain. *PLoS ONE* **2013**, *8*, 80887. [CrossRef]

28. Shukla, S.K.; Yadav, R.L.; Awasthi, S.K.; Gaur, A. Soil microbial biomass nitrogen, *in situ* respiration and crop yield influenced by deep tillage, moisture regimes and N nutrition in Sugarcane-based system in subtropical India. *Sugar Tech* **2016**, *19*, 125–135. [CrossRef]

29. Bhatia, A.; Pathak, H.; Jain, N.; Singh, P.; Singh, A. Global warming potential of manure amended soils under rice–wheat system in the Indo-Gangetic plains. *Atmos. Environ.* **2005**, *39*, 6976–6984. [CrossRef]

30. Ghosh, S.; Wilson, B.C.D.; Ghoshal, S.K.; Senapati, N.; Mandal, B. Organic amendments influence soil quality and carbon sequestration in the Indo-Gangetic plains of India. *Agric. Ecosyst. Environ.* **2012**, *156*, 134–141. [CrossRef]

31. Pathak, H.; Byjesh, K.; Chakrabarti, B.; Aggarwal, P.K. Potential and cost of carbon sequestration in Indian agriculture: Es-timates from long-term field experiments. *Field Crops Res.* **2011**, *12*, 102–111. [CrossRef]

32. Dubey, R.K.; Dubey, P.K.; Abhilash, P.C. Sustainable soil amendments for improving the soil quality, yield and nutrient content of *Brassica juncea* (L.) grown in different agroecological zones of eastern Uttar Pradesh, India. *Soil Tillage Res.* **2019**, *195*, 104418. [CrossRef]

33. McLean, E.O. Soil pH and lime requirement. In *Methods of Soil Analysis: Part 2 Chemical and Microbiological Properties*; Page, A.L., Ed.; American Society of Agronomy Soil Science Society of America Madison: Madison, WI, USA, 1982; pp. 199–224.

34. Blake, G.R.; Hartge, K.H. Bulk density. In *Methods of Soil Analysis. Part 1. Physical and Mineralogical Methods*; Klute, A., Ed.; American Society of Agronomy and Soil Science Society of America: Madison, WI, USA, 1986; pp. 363–375.

35. Walkley, A. A critical examination of a rapid method for determining organic carbon in soils: Effect of variations in digestion conditions and of organic soil constituents. *Soil Sci.* **1947**, *63*, 251–263. [CrossRef]

36. Bremner, J.; Mulvaney, C. Nitrogen-Total. In *Sorghum: State of the Art and Future Perspectives*; Wiley: Hoboken, NJ, USA, 2015; pp. 595–624.

37. Olsen, S.R.; Cole, C.V.; Watanabe, F.S.; Dean, L.A. *Estimation of Available Phosphorus in Soils by Extraction with Sodium Bicarbonate*; Government Printing Office: Washington, DC, USA, 1954.

38. Vance, E.; Brookes, P.; Jenkinson, D. An extraction method for measuring soil microbial biomass C. *Soil Biol. Biochem.* **1987**, *19*, 703–707. [CrossRef]

39. Rong, Y.; Maa, L.; Johnson, D.A.; Yuanc, F. Soil respiration patterns for four major land-use types of the agro-pastoral region of northern China. Agric. *Ecosyst. Environ.* **2015**, *213*, 142–150. [CrossRef]

40. Dubey, R.K.; Dubey, P.K.; Chaurasia, R.; Singh, H.B.; Abhilash, P.C. Sustainable agronomic practices for enhancing the soil quality and yield of *Cicer arietinum* L. under diverse agroecosystems. *J. Environ. Manag.* **2020**, *262*, 110284. [CrossRef]

41. Kandeler, E.; Gerber, H. Short-term assay of soil urease activity using colorimetric determination of ammonium. *Biol. Fertil. Soils* **1988**, *6*, 68–72. [CrossRef]

42. Wright, S.; Upadhyaya, A. A survey of soils for aggregate stability and glomalin, a glycoprotein produced by hyphae of arbuscular mycorrhizal fungi. *Plant Soil* **1998**, *198*, 97–107. [CrossRef]

43. Liu, Y.-R.; Delgado-Baquerizo, M.; Wang, J.-T.; Hu, H.-W.; Yang, Z.; He, J.-Z. New insights into the role of microbial community composition in driving soil respiration rates. *Soil Biol. Biochem.* **2018**, *118*, 35–41. [CrossRef]

44. Pries, C.E.H.; Castanha, C.; Porras, R.C.; Torn, M.S. The whole-soil carbon flux in response to warming. *Science* **2017**, *355*, 1420–1423. [CrossRef]

45. Lohila, A.; Aurela, M.; Regina, K.; Laurila, T. Soil and total ecosystem respiration in agricultural fields: Effect of soil and crop type. *Plant Soil* **2003**, *251*, 303–317. [CrossRef]

46. Grand, S.; Rubin, A.; Verrecchia, E.P.; Vittoz, P. Variation in soil respiration across soil and vegetation types in an Alpine valley. *PLoS ONE* **2016**, *11*, e0163968. [CrossRef]

47. Salazar, A.; Sulman, B.N.; Dukes, J.S. Microbial dormancy promotes microbial biomass and respiration across pulses of drying-wetting stress. *Soil Biol. Biochem.* **2018**, *116*, 237–244. [CrossRef]

48. Kukumägi, M.; Ostonen, I.; Kupper, P.; Truu, M.; Tulva, I.; Varik, M.; Aosaar, J.; Sober, J.; Löhmus, K. The effects of elevated atmospheric humidity on soil respiration components in a young silver birch forest. *Agric. For. Meteorol.* **2014**, *194*, 167–174. [CrossRef]

49. Wang, X.; Ren, T. Spatial and temporal variability of soil respiration between soybean crop rows as aeasured continuously over a growing season. *Sustainability* **2017**, *9*, 436. [CrossRef]

50. Bai, C.; Ling, Y.; Zhu, Y.; Yaoxiang, G.E.; Lin, X.; Jla, W. The temporal and spatial variation of soil respiration in pepper (*Capsicum annuum* L.), eggplant (*Solanum melongena* L.) and maize (*Zea mays* L.) agro-ecosystems in Northwest of China. *Aust. J. Crop Sci.* **2012**, *6*, 1565–1571.

51. Hashimoto, S.; Carvalhais, N.; Ito, A.; Migliavacca, M.; Nishina, K.; Reichstein, M. Global spatiotemporal distribution of soil respiration modeled using a global database. *Biogeosciences* **2015**, *12*, 4121–4132. [CrossRef]

52. Srinivasarao, C.; Venkateswarlu, B.; Lal, R.; Singh, A.K.; Kundu, S.S.; Vittal, K.P.R.; Patel, J.J.; Patel, M.M. Long-term manuring and fertilizer effects on depletion of soil organic carbon stocks under pearl millet-cluster bean-castor rotation in western india. *Land Degrad. Dev.* **2011**, *25*, 173–183. [CrossRef]

53. Bilandžija, D.; Željka, Z.; Kisić, I. Influence of tillage practices and crop type on soil CO_2 emissions. *Sustainability* **2016**, *8*, 90. [CrossRef]

54. Tripathi, V.; Dubey, R.; Singh, H.B.; Singh, N.; Abhilash, P.C. Is *Vigna radiata* (L.) R. Wilczek a suitable crop for lindane contaminated site? *Ecol. Engineering.* **2014**, *73*, 219–223. [CrossRef]

55. Zhu, Z.; Ge, T.; Liu, S.; Hu, Y.; Ye, R.; Xiao, M.; Tong, C.; Kuzyakov, Y.; Wu, J. Rice rhizodeposits affect organic matter priming in paddy soil: The role of N fertilization and plant growth for enzyme activities, CO_2 and CH_4 emissions. *Soil Biol. Biochem.* **2018**, *116*, 369–377. [CrossRef]

56. Innes, L.; Hobbs, P.J.; Bardgett, R.D. The impacts of individual plant species on rhizosphere microbial communities in soils of different fertility. *Biol. Fertil. Soils* **2004**, *40*, 7–13. [CrossRef]

57. Orio, A.G.A.; Brücher, E.; Ducasse, D.A. Switching between monocot and dicot crops in rotation schemes of Argentinean productive fields results in an increment of arbuscular mycorrhizal fungi diversity. *Appl. Soil Ecol.* **2016**, *98*, 121–131. [CrossRef]

58. Truong, T.H.H.; Marschner, P. Respiration, available N and microbial biomass N in soil amended with mixes of organic materials differing in C/N ratio and decomposition stage. *Geoderma* **2018**, *319*, 167–174. [CrossRef]

59. Hao, Q.; Jiang, C. Contribution of root respiration to soil respiration in a rape (*Brassica campestris* L.) field in Southwest China. *Plant, Soil Environ.* **2014**, *60*, 8–14. [CrossRef]

60. Abujabhah, I.S.; Bound, S.A.; Doyle, R.; Bowman, J.P. Effects of biochar and compost amendments on soil physico-chemical properties and the total community within a temperate agricultural soil. *Appl. Soil Ecol.* **2016**, *98*, 243–253. [CrossRef]

61. Cavagnaro, T.R. Impacts of compost application on the formation and functioning of arbuscular mycorrhizas. *Soil Biol. Biochem.* **2014**, *78*, 38–44. [CrossRef]

62. Morales-Romero, D.; Campo, J.; Alvarez, H.G.; Freaner, F.M. Soil carbon, nitrogen and phosphorus changes from conversion of thorn scrub to buffelgrass pasture in northwestern Mexico. *Agric. Ecosyst. Environ.* **2015**, *199*, 231–237. [CrossRef]

63. Wang, P.; Wang, Y.; Wu, Q.S. Effects of soil tillage and planting grass on arbuscular mycorrhizal fungal propagules and soil properties in citrus orchards in southeast China. *Soil Tillage Res.* **2016**, *155*, 54–61. [CrossRef]

64. Bowles, T.M.; Acosta-Martínez, V.; Calderón, F.; Jackson, L.E. Soil enzyme activities, microbial communities, and carbon and nitrogen availability in organic agroecosystems across an intensively-managed agricultural landscape. *Soil Biol. Biochem.* **2014**, *68*, 252–262. [CrossRef]
65. Wright, A.L.; Hons, F.M.; Matocha, J.E., Jr. Tillage impacts on microbial biomass and soil carbon and nitrogen dynamics of corn and cotton rotations. *Appl Soil Ecol.* **2005**, *29*, 85–92. [CrossRef]
66. Kallenbach, C.M.; Grandy, A.S.; Frey, S.D.; Diefendorf, A.F. Microbial physiology and necromass regulate agricultural soil carbon accumulation. *Soil Biol. Biochem.* **2015**, *91*, 279–290. [CrossRef]
67. Tejada, M.; Hernandez, M.T.; Garcia, C. Application of two organic amendments on soil restoration: Effects on the soil biological properties. *J. Environ. Qual.* **2006**, *35*, 1010–1017. [CrossRef]
68. Mayor Ángeles, G.; Goirán, S.B.; Vallejo, V.R.; Bautista, S. Variation in soil enzyme activity as a function of vegetation amount, type, and spatial structure in fire-prone Mediterranean shrublands. *Sci. Total. Environ.* **2016**, *573*, 1209–1216. [CrossRef]
69. Borie, F.; Rubio, R.; Rouanet, J.; Morales, A.; Borie, G.; Rojas, C. Effects of tillage systems on soil characteristics, glomalin and mycorrhizal propagules in a Chilean Ultisol. *Soil Tillage Res.* **2006**, *88*, 253–261. [CrossRef]
70. Abhilash, P.C. Restoring the unrestored: Strategies for restorung global land duing the UN Decade on Ecosystem Restoration (UN-DER). *Land* **2021**, *2*, 201. [CrossRef]