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Short-Term Effects of Incorporation Depth of Straw Combined with Manure During the Fallow Season on Maize Production, Water Efficiency, and Nutrient Utilization in Rainfed Regions

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Abstract: Diminishing soil fertility and crop productivity due to traditional intensive cultivation has prompted the use of straw and manure to improve soil health in Northeast China. However, few comparative studies have explored the influence of varying straw and manure incorporation depths on crop growth. A field experiment in the rainfed black soil regions of Gongzhuling and Keshan assessed the effects of deep (30 cm) and shallow (15 cm) incorporations of straw and manure on soil fertility, maize root growth, and maize productivity. Deep incorporations, via subsoiling tillage (DST) and deep-plow (DDT) tillage, enhanced soil water storage of 30–100 cm soil layer during periods of low rainfall, improved the availability of nutrients (nitrogen, phosphorus, and potassium) and soil organic matter content, especially in deeper soil, compared to shallow incorporation using rotary tillage (SRT). Both DST and DDT induced a larger rooting depth and a higher fine root (diameter class 0–0.5 mm) length density by 31.0% and 28.9%, respectively, accompanied by reduced root turnover. Furthermore, the sub-surface foraging strategies of roots under the DST and DDT treatments boosted the total nitrogen, phosphorus, and potassium uptake (6.5–17.9%) and achieved a higher dry mass accumulation during the later growth period, thus leading to notable improvements in the 100-kernel weight and yield (16.1–19.7%) and enhancing water- and nutrient-use efficiencies by 2.5–20.5%. Overall, compared to shallow incorporation, deep incorporation of straw and manure significantly enhances root growth and spatial distribution of soil water and nutrients, which has great potential for increasing maize yield in rainfed agricultural areas.

Keywords: organic waste; soil fertility; root morphology; yield; rainfed agricultural areas

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1. Introduction

Maize (*Zea mays* L.) is a primary cereal crop worldwide, and maximizing the efficient use of water resources and soil nutrients is crucial to enhancing maize productivity and achieving sustainable agricultural development [1]. Northeast China, a principal black soil area among the globe's four major regions, aligns latitudinally with the maize belts of the U.S. and Ukraine [2]. This area is also an important rainfed agricultural area, with maize production accounting for approximately 40% of the national total [3]. However, the uneven spatiotemporal distribution of precipitation in this area has restricted the maize yield production (Figure 1). Additionally, the intensive implementation of heavy machinery has led to a decline in soil fertility. The plow pan caused by shallow tillage machinery further impedes maize yield enhancement [4,5]. Recently, the adoption of straw-returning machines, supported by government initiatives, has made straw returning a prevalent agricultural practice in Northeast China, enhancing soil fertility and ecological protection [6]. Livestock manure in Northeast China annually amounts to 2.87 × 10^8 tons, potentially causing ecological and environmental issues as a significant nonpoint source of pollution if not managed properly [7,8]. Conversely, appropriate incorporation of treated livestock manure can boost soil fertility and improve crop yield potential by incorporating organic matter and essential nutrients into the soil [9,10]. Therefore, the effective utilization of straw and manure is urgently needed to achieve high maize yields while maintaining or improving black soil fertility.

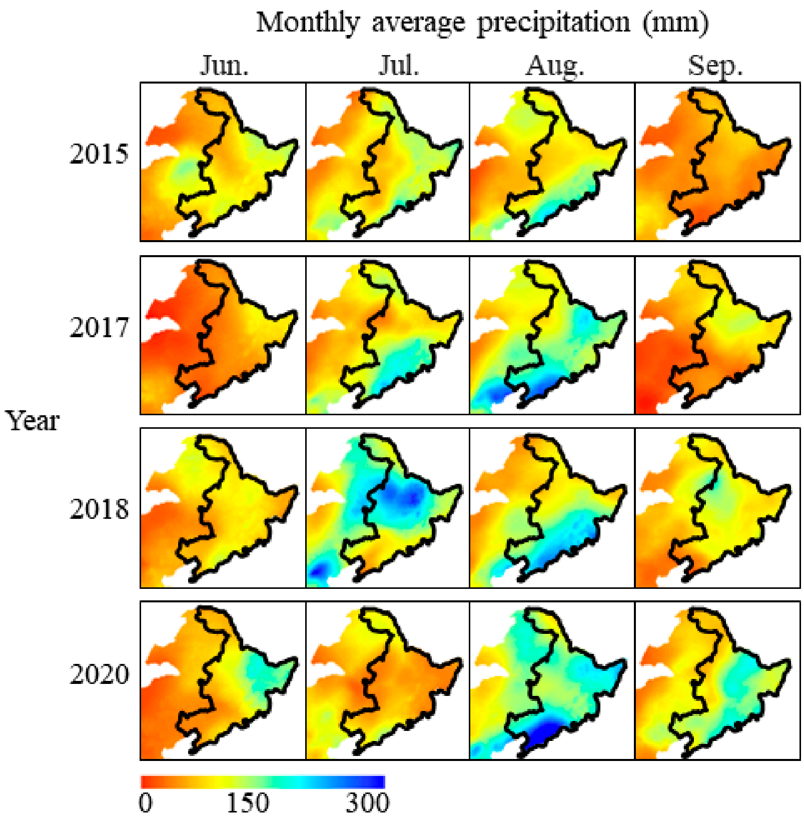

Figure 1. Monthly average precipitation during the main growth period of maize in Northeast China from 2015 to 2020.

In recent years, the productivity effects of co-applying straw and manure have received considerable attention [11,12]. Applying straw and manure reduces fertilizer use, boosts wheat yields, and yields significant economic benefits [13]. Combining poultry manure with straw enhances soil microbial abundance, activity, and biomass, mitigates excess mineral nitrogen accumulation through biological immobilization, and maintains grain crop yields [14]. Straw and manure incorporation together also had significant and positive residual effects on rice yields and N uptake and resulted in savings of 60 kg N ha^{-1} each for wheat and rice [15]. Over three years on coastal salt-affected farmlands, straw and manure co-incorporation raised maize yields by more than 14.3% and enhanced the grain water use index (WUI) by over 14.7%, relative to practices excluding straw and manure [16]. Field experiment results also showed the potential of straw and manure co-incorporation to enhance the yields of sunflower seeds and sugarcane [17,18]. Additionally, co-incorporation of straw and manure can promote crop N uptake [19]. However, these studies were mostly based on traditional shallow tillage, and few have explored the influence of different incorporation depths of straw combined with manure on maize growth. Therefore, it is very important to clarify the effects and mechanisms of various straw and manure incorporation depths on maize production in Northeast China.

The morphological traits and dynamics of the root system are critical for efficient water and nutrient utilization by crops, influencing plant development and crop production [20–22]. Root length density and root surface-area density are vital indicators for evaluating morphological traits and the potential to obtain more water and nutrients [23]. Moreover, root productivity and turnover rate are crucial indicators for assessing root dynamics, which also have a significant impact on a plant's ability to absorb water and nutrients [24,25]. However, traditional shallow tillage practices in Northeast China result in a shallow plowed layer and an imbalanced distribution of water and fertilizer [26]; coupled with uneven rainfall, these conditions restrict root growth and rooting depth, thereby potentially impeding the effective utilization of soil moisture and nutrients in deeper soil layers [27,28]. The shallow incorporation of straw and manure has gained widespread recognition and incorporation because it not only increases soil water storage capacity but also promotes straw decomposition and increases soil nutrients [12], resulting in a significant increase in root length density and root productivity compared to only straw return [26,29]. However, the cold climate in the northeastern region of China suppresses the decomposition of straw when it is applied at shallow soil depths [30]. In addition, shallow tillage results in a thick plow layer, which is unfavorable for improving the sub-surface and promoting rooting depth [31,32]. Therefore, further research is necessary to investigate whether deep straw and manure incorporation, such as based on subsoiling tillage (DST) or deep-plow tillage (DDT), can enhance root growth by enhancing soil fertility and ultimately increase water- and nutrient-use efficiency and crop yield compared to shallow incorporation (SRT). Analyzing crop root distribution poses challenges due to their subterranean nature. The minirhizotron technique facilitates nondestructive, in situ measurements of individual root segments over their lifecycle with high temporal resolution, minimally affecting root processes [33].

At present, many regions employ shallow straw and manure incorporation (SRT) methods to improve topsoil fertility. We hypothesized that deep incorporation of straw combined with manure (DST and DDT) can improve the spatial matching of roots with soil water and nutrients, optimize root growth strategies, and ultimately increase water- and nutrient-use efficiency and maize yield. A field positioning experiment in the black soil region of Northeast China was conducted to (1) clarify the distribution of soil water and nutrients under DST and DDT; (2) compare the effects of various straw and manure incorporation depths on the spatial matching between soil water and nutrients and the root foraging strategy (e.g., rooting depth, proportion of fine roots, and root turnover); and (3) explore mechanisms of different straw and manure incorporation depths on water and nutrient utilization and yield of maize in rainfed agricultural areas.

2. Materials and Methods

2.1. Experimental Site

Field positioning experiments commenced in October 2015 at Gongzhuling (GZL, 43°21′ N, 124°43′ E) and Keshan (KS, 48°02′ N, 125°34′ E) Experimental Stations in Northeast China (Figure 2a). The area of GZL has a temperate continental monsoon climate with an annual precipitation of 572 mm, an average annual temperature of 4.5 °C, and a thin-layer black soil (humus layer ≤ 30 cm). The area of KS has a cold temperate continental monsoon climate with an average annual precipitation of 499 mm, an average annual temperature of 2.4 °C, and a thick-layer black soil (humus layer ≥ 60 cm). Data for this study, collected in 2020, show that the annual precipitation at GZL was 584.1 mm with a daily maximum temperature of 28 °C, while KS received 569.8 mm of rainfall and had a daily maximum temperature of 26 °C (Figure 2b,c). Prior to maize planting in April 2015, the soil properties within the upper 30 cm were as follows: soil organic carbon (SOC) of 11.02 g kg^{-1}, total nitrogen (TN) of 1.35 g kg^{-1}, available phosphorus (AP) of 21.25 mg kg^{-1}, and available potassium (AK) of 188 mg kg^{-1} in GZL; and SOC of

13.3 g kg^{-1}, TN of 1.25 g kg^{-1}, AP of 39.65 mg kg^{-1}, and AK of 197 mg kg^{-1} in KS. Both experimental sites had undergone long-term shallow rotary tillage before the experiment.

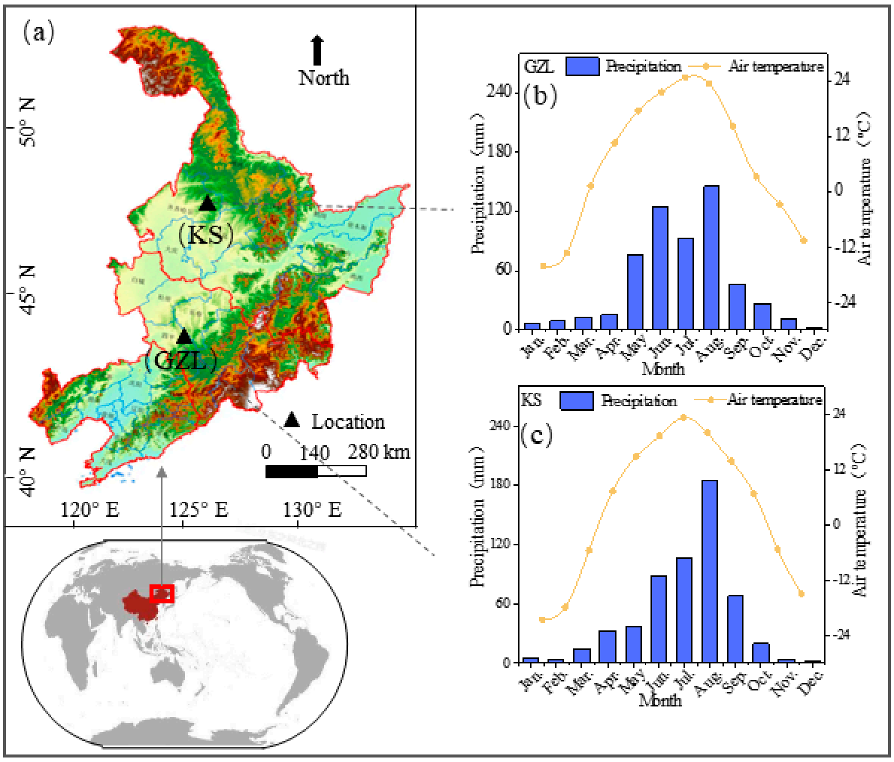

Figure 2. Location of the experimental site (**a**); the precipitation and air temperature for 2020 in Gongzhuling (GZL, (**b**)) and Keshan (KS, (**c**)).

2.2. Experimental Design and Field Management

The field study employed a randomized block design, consisting of three treatments and four replicates: DST, deep incorporation of straw combined with manure by subsoiling tillage (soil depth of approximately 30 cm); DDT, deep incorporation of straw combined with manure by deep-plow tillage (soil depth of approximately 30 cm); and SRT, shallow incorporation of straw combined with manure by rotary tillage (soil depth of approximately 15 cm). Subsoiling tillage, deep-plow tillage, and rotary tillage are the most widely used cultivation methods in Northeast China, respectively. Subsoiling machinery utilizes subsoiling shanks or tines to penetrate the soil, cutting and lifting it; deep-plowing machinery employs deep-plow blades or shears to break and invert the hardened sub-surface layers; rotary tillers are equipped with rotating blades or tines that break up and mix the soil, typically operating at shallower depths. In this study, for the DST treatment, subsoiling was first carried out to loosen the deeper soil layers, followed by rotary tillage to ensure the straw and manure were mixed as thoroughly as possible with the deeper soil layers. The area of each plot was 198 m^2 (22 m × 9 m). The planting density in GZL was 67,500 plants ha^{-1}, with a row spacing of 60 cm and plant spacing of 24.7 cm; the planting density in KS was 90,000 plants ha^{-1}, with a row spacing of 60 cm and plant spacing of 18.5 cm. After the maize harvest, the maize straw was crushed to approximately 5 cm using a straw chopper and spread evenly on the surface of the plots. The maize straw was crushed to approximately 5 cm using a straw chopper and spread evenly on the surface of the plots. The average SOC, TN, TP, and TK content of the straw was 408.2 g C kg^{-1}, 8.9 g N kg^{-1}, 1.6 g P kg^{-1}, and 17.0 g K kg^{-1}, respectively. The amount of maize straw returned to the soil was approximately 9500 kg ha^{-1}. Simultaneously, the cow manure was applied at a rate of 7500 kg ha^{-1}, which its SOC, TN, TP, and TK content was 369.0 g C kg^{-1}, 7.8 g N kg^{-1}, 3.6 g P kg^{-1}, and 4.6 g K kg^{-1}, respectively. Subsequently, rotary tillage was carried out using a MASCHIO rotary tillage machine (Maschio Gaspardo, Imola, Italy); subsoiling tillage was conducted using a Chinese ISQ-340 full-range deep loosening machine (Zhenxing Machinery Manufacturing, Tianjin, China), and the maize straw combined with manure was mixed with the soil using a rotary tillage machine. Deep-plow tillage was performed using a Norwegian KVERNELAND hanging flip plow (Kverneland, Stavanger, Norway), and the maize straw combined with manure was buried in the soil.

Maize was sown in early May using a no-tillage maize planter in flat cultivation mode. The locally dominant maize varieties were Fumin 985 in GZL and Ruifuer 1 in KS. Fertilization and sowing operations were completed in one step using a no-tillage maize planter. For the GZL treatment, a maize-specific compound fertilizer with an N: P2O5: K2O ratio of 26:11:13 was applied at a rate of 600 kg ha^{-1} (sum of N, P_2O_5 and K_2O). For the KS treatment, the compound fertilizer ratio was 26:12:12, also applied at 600 kg ha^{-1} (sum of N, P_2O_5, and K_2O). Other field management measures, such as pest control, were kept consistent with local farming practices, and the maize was harvested around October 10th.

2.3. Sampling and Measurements

2.3.1. Soil Water, Nutrients, and Organic Carbon

Soil volumetric water content (SWC, %) in the 0–100 cm soil layer was recorded using a TDR meter (time-domain reflectometry, Soil Moisture Equipment Corp., Germany) with four replicates during the seeding, jointing, tasseling, filling, and maturing stages at two sites. Soil water storage (SWS, mm) dynamics were determined by multiplying the volumetric water content by the horizon depth, yielding water in millimeters [34]. Assuming uniformity within each depth interval, SWS was calculated per 10 cm intervals and aggregated to derive the total profile (1 m) SWS.

Water-use efficiency (WUE, kg ha^{-1} mm^{-1}) was determined using the following equation [35]: WUE = Y/ET, where Y (kg ha^{-1}) and ET (mm) are the grain yield and the total evapotranspiration during the maize growing season, respectively.

The ET was calculated using the soil water balance equation [36]: $ET = SWS_0 - SWS_t + P + I - R$, where SWS0 is soil water storage at sowing, SWSt is soil water storage at maturity in the 0–100 cm soil layer (mm), P is precipitation (mm), I is the irrigation level (mm) (I = 0 because, in the absence of irrigation, the study region depends on rainfall throughout the growing season), and R is the surface runoff (mm), which was set to zero because the slope of the experimental plot was less than 5° and each plot was surrounded by border ridges.

The sampling method involved randomly selecting four points within each treatment plot during the mature stage in 2020 from the central row of each plot after removing surface litter. At each point, soil samples were collected using an auger from three depth intervals: 0–15 cm, 15–30 cm, and 30–45 cm. Each soil sample was air-dried and then ground to pass through a 2 mm sieve for chemical analysis. The soil available nitrogen was calculated as the sum of the NH_4^+–N and NO_3^-–N contents by a continuous flow analyzer (SkalarSan++ System, Skalar, Breda, The Netherlands). Soil available phosphorus content was measured by the molybdenum blue colorimetric method using a UV-Visible Spectrophotometer [37]. Soil available potassium content was measured by a flame photometer (Waters, MA, USA). The soil organic carbon content was analyzed with potassium dichromate oxidation-ferrous sulfate titrimetry.

2.3.2. Minirhizotron Installation and Root Image Analysis

The CI-600 root monitoring system (CID Bio-Science, Camas, WA, USA) was used to monitor root growth characteristics during the maize growing season. Following Yan et al. [38], minirhizotron tubes (external diameter, 7 cm; internal diameter, 6.4 cm; length, 90 cm) were installed. All tubes were buried at an angle of 45° to vertical to a depth of 64 cm (equal to a vertical soil depth of 45 cm). The aboveground part was wrapped in black tin foil, and the tube opening was sealed with a black lid. A CI-600 scanner was used to capture root images (image size: 21.59 cm × 19.56 cm) at three tube depths (0–15 cm, 15–30 cm, and 30–45 cm soil depth).

Root morphological traits and dynamics were analyzed using WinRHIZOTron 2015HR software [39]. Live and dead roots within each image were distinguished by color and morphology. The identification of new roots was primarily based on their color and morphological characteristics, such as the lighter color (whiter) and smoother surface, while black, wrinkled roots were classified as dead. Measurements of length and surface area for both live and dead roots were recorded independently. Subsequently, root length density (RLD) and root surface-area density (RSD) were calculated using formulas $RLD = (L\sin\theta)/A$ and $RSD = (S\sin\theta)/A$, respectively. In the formulas, L and S represent the length and surface area of live roots, respectively. A represents the analyzed image area (cm^2), and θ represents the angle between the minirhizotron tubes and vertical direction.

Root production was evaluated mainly according to Yan et al. [38]. The increase in the lengths of existing roots within each observation interval was calculated as root production (RP, cm cm^{-2}). Root turnover (RT, per growth period) can be estimated by the length-based method [40]. RT = GPRP/RLDmax, where GRRP represents the sum of the fine root length that is produced within the growth period, and RLDmax represents the maximum value of LRL during the growth period.

2.3.3. Dry Matter Weight, Nutrient Uptake, Nutrient-Use Efficiency, and Maize Yield

At two sites, four representative maize plants from each plot were collected at the seeding, jointing, tasselling, filling, and mature stages. Then, the plant samples were separated into leaves, stems and sheaths, husks, cobs, and grains before drying to a constant weight at 80 °C to calculate the accumulated biomass. After drying, the samples were crushed and passed through a 100-mesh sieve to determine the N, P, and K contents for each plant fraction. The total N concentration was obtained by the micro-Kjeldahl method. The samples were digested using sulfuric acid and hydrogen peroxide. The digestion was carried out in a digestion furnace, which was gradually heated to approximately 180 °C until the solution became clear, indicating that the sample had been fully digested. Then, the total P was estimated using the molybdenum antimony–d-iso-ascorbic acid colorimetry (MADAC) method, and the total K concentration was determined by flame photometry. The uptake of N, P, and K was calculated as the dry weight multiplied by the total N, P, and K content.

Nitrogen-use efficiency (NUE, kg kg^{-1}), phosphorus-use efficiency (PUE, kg kg^{-1}), and potassium-use efficiency (KUE, kg kg^{-1}) were calculated by the following equations [41]: NUE = Grain yield/total N uptake; PUE = Grain yield/total P uptake; KUE = Grain yield/total K uptake.

2.4. Statistical Analyses

Analysis of variance (ANOVA) assessed the effects of varying straw and manure incorporation depths, soil layer depths, and their interactions on grain yield, maize growth, and soil properties using SPSS 27.0 (SPSS Inc., Chicago, IL, USA). Duncan's test at $p < 0.05$ significance level compared mean variable values. Origin Pro 2021 produced the figures. Grain yield influencers were analyzed with a random forest (RF) model via R's "Random Forest" package (R Core Team 2018). A structural equation model (SEM), built with AMOS 21.0 (AMOS Development Corporation, Meadville, PA, USA), elucidated the regulatory pathways impacting maize yield, incorporating only variables significantly related to yields.

3. Results

3.1. Soil Water, Available Nitrogen (N), Phosphorus (P), Potassium (K), and Organic Carbon Content

Compared with the SRT, both the DST and DDT significantly increased the soil water content, especially below the 30 cm soil layer (Figure 3). The DST and DDT treatments increased the soil water content in the 30–100 cm soil layer by 11.0% and 10.1%, respectively, compared to that in the SRT. The surface soil moisture content at the KS experimental site was higher than that at the GZL site. The trend of soil water storage variation under different co-incorporation modes of straw and manure was similar to that of soil water content, which increased first and then decreased with the growth stage. The DST and DDT treatments varied only slightly from the SRT in water storage capacity in the 0–30 cm soil layer but significantly increased water storage capacity in the 30–100 cm soil layer (Figure 4). During the early growth stages of maize, the soil water storage at KS exceeded that of GZL; however, in the remaining mid to late growth stages, the difference in soil water storage between the two sites was not significant. Notably, during the filling and maturity stages, the DST treatment at the GZL site exhibited higher soil moisture content compared to other treatments.

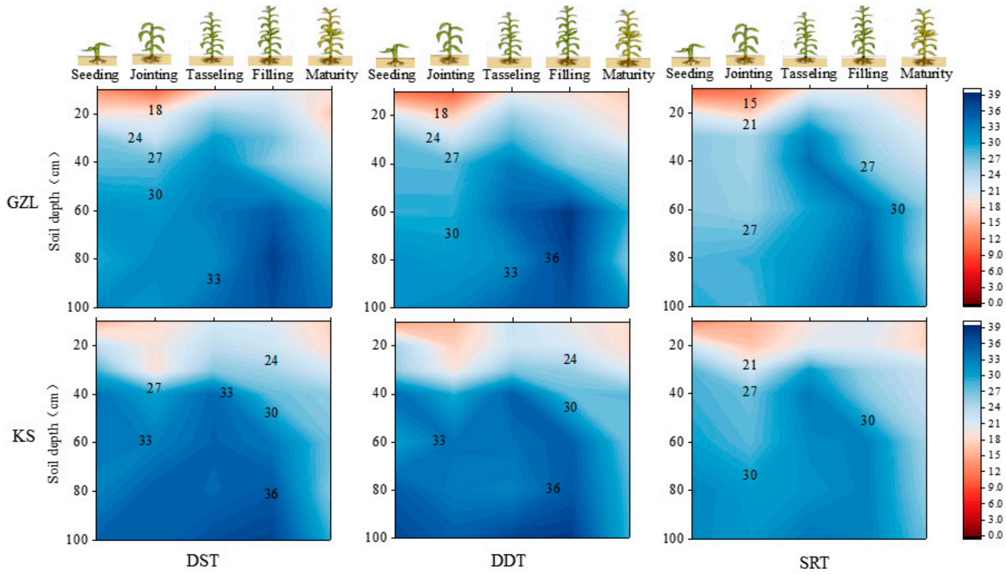

Figure 3. Two-dimensional distributions of soil water content (%) at different growth stages in soil layers of 0–100 cm under various incorporation depths of straw combined with manure at GZL and KS sites. DST and DDT represent deep incorporation of straw combined with manure by subsoiling tillage and deep-plow tillage, respectively. SRT represents shallow incorporation of straw combined with manure by rotary tillage. The data analysis and visualization were performed using Kriging spatial analysis.

The co-incorporation method used for straw and manure, soil depth, and their interaction significantly affected the soil available NPK content. The soil available phosphorus content in the surface layer at the KS site was significantly higher than that at the GZL site. The trends of soil available nitrogen, phosphorus, and potassium under different treatments were similar at both experimental sites. With increasing soil depth, the DST and DDT treatments significantly increased the available NPK content compared to the SRT treatment (Figure 5). In the 15–45 cm soil layer, the DST and DDT treatments led to an average increase of 6.7 mg kg^{-1}, 5.3 mg kg^{-1}, and 28.9 mg kg^{-1} in soil available N, P, and K contents, respectively, compared to the SRT treatment.

The incorporation of straw combined with manure at different depths had a significant effect on soil organic carbon content, with variations observed across soil layers, particularly in the 15–45 cm soil layer (Table 1). In the 0–15 cm soil layer, the soil organic carbon content did not differ significantly under different treatments in GZL and was highest under the DST treatment in KS. In the 15–45 cm soil layer, the average soil organic carbon content across both sites was highest under the DST and DDT treatments. The organic matter content at the KS experimental site was significantly higher than that at the GZL site.

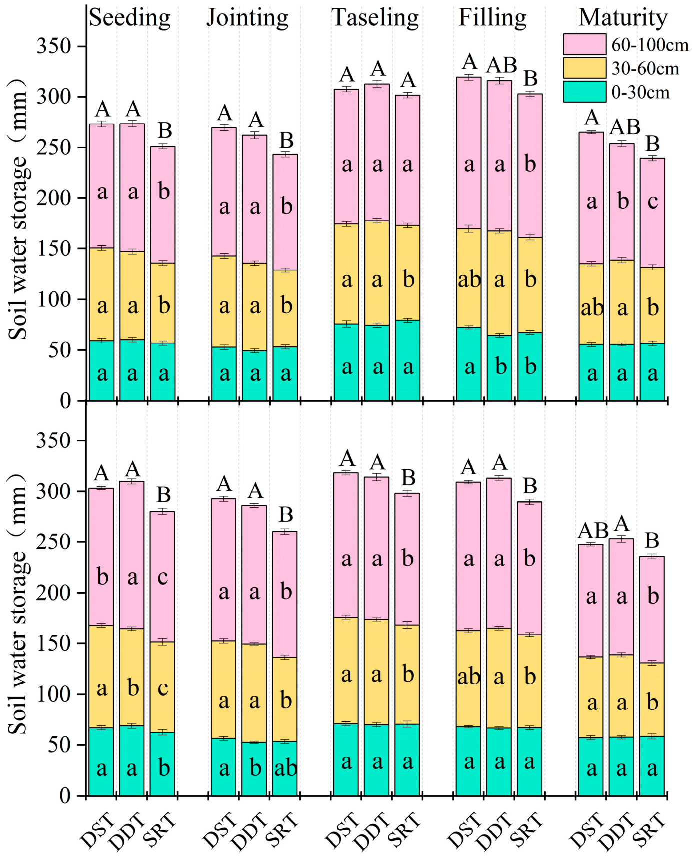

Figure 4. Soil water storage at different growth stages in soil depths of 0–30 cm, 30–60 cm, and 60–100 cm under various incorporation depths of straw combined with manure at GZL and KS sites. DST and DDT represent deep incorporation of straw combined with manure by subsoiling tillage and deep-plow tillage, respectively. SRT represents shallow incorporation of straw combined with manure by rotary tillage. Error bars represent the standard error. Different lowercase letters indicate significant differences between treatments in the same soil layer; different capital letters indicate significant differences in the sum of all soil layers between treatments ($p < 0.05$, $n = 4$).

Table 1. Effects of different incorporation depths of straw combined with manure on soil organic carbon.

Layer (cm)	Treatment	Soil Organic Carbon (g kg^{-1})	
		GZL	KS
0–15 cm	DST	13.08 a A	30.02 a A
	DDT	11.85 a A	25.59 c AB
	SRT	12.56 a A	27.76 b A
15–30 cm	DST	12.06 a AB	24.96 a B
	DDT	11.41a A	26.56 a A
	SRT	9.06 b B	22.18 b B
30–45 cm	DST	11.56 a B	24.06 a B
	DDT	10.93 a A	23.56 a B
	SRT	9.22 b B	20.18 b C
ANOVA			
Layer (L)		42 **	117 **
Straw + manure (SM)		37 **	48 **
L × SM		9 **	41 **

DST and DDT represent deep incorporation of straw combined with manure by subsoiling tillage and deep-plow tillage, respectively. SRT represents shallow incorporation of straw combined with manure by rotary tillage. Different lowercase letters indicate significant differences between treatments within the same soil layer, while different uppercase letters denote significant differences between soil layers within the same treatment ($p < 0.05$, $n = 4$). ** indicate significance levels of $p < 0.01$.

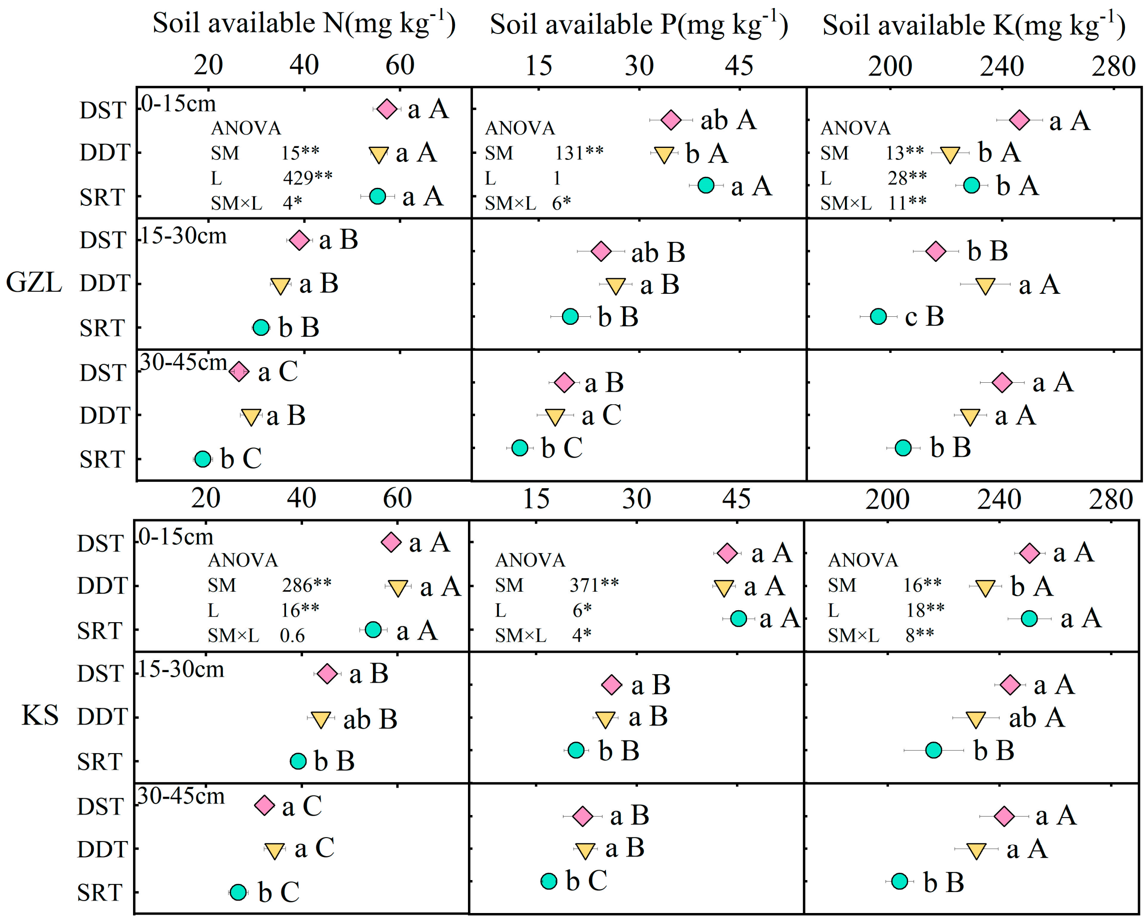

Figure 5. Soil available NPK content in different soil layers under various incorporation depths of straw combined with manure at GZL and KS sites. DST and DDT represent deep incorporation of straw combined with manure by subsoiling tillage and deep-plow tillage, respectively. SRT represents shallow incorporation of straw combined with manure by rotary tillage. Error bars represent the standard error. Different lowercase letters indicate significant differences between treatments within the same soil layer, while different uppercase letters denote significant differences between soil layers within the same treatment ($p < 0.05$, $n = 4$). * and ** indicate significance levels of $p < 0.05$ and $p < 0.01$, respectively.

3.2. Root Morphological Traits and Dynamics

Deep incorporation (DST and SRT) promoted the proliferation of deeper roots and finer roots (0–0.5 mm diameter), and the improvement in the root system varied by growth stage, with the best effect observed during the tasselling and grain filling stages (Figure 6 and Figure S1). During these stages, the fine root length density in the 15–45 cm soil layer increased by 31.0% and 28.9%, and the root surface-area density increased by 34.4% and 31.5%, respectively, in the DST and DDT treatments compared to those in the SRT treatment at both experimental sites. The improvement effect of the DST treatment in the 15–30 cm soil layer at the GZL experimental site was superior to that of the DDT treatment.

Considering the root system over the whole growth period, deep incorporation significantly increased root productivity and decreased the root turnover rate compared to shallow incorporation (Table 2). Compared with the SRT treatment, the DST and DDT treatments significantly increased root productivity at the two experimental sites by 13.2% and 9.5%, respectively. Regarding the root turnover rate during the growth period, the SRT treatment resulted in a higher turnover rate in the 15–45 cm soil layer, which was 6.8% and 8.1% higher than that of the DST and DDT treatments, respectively.

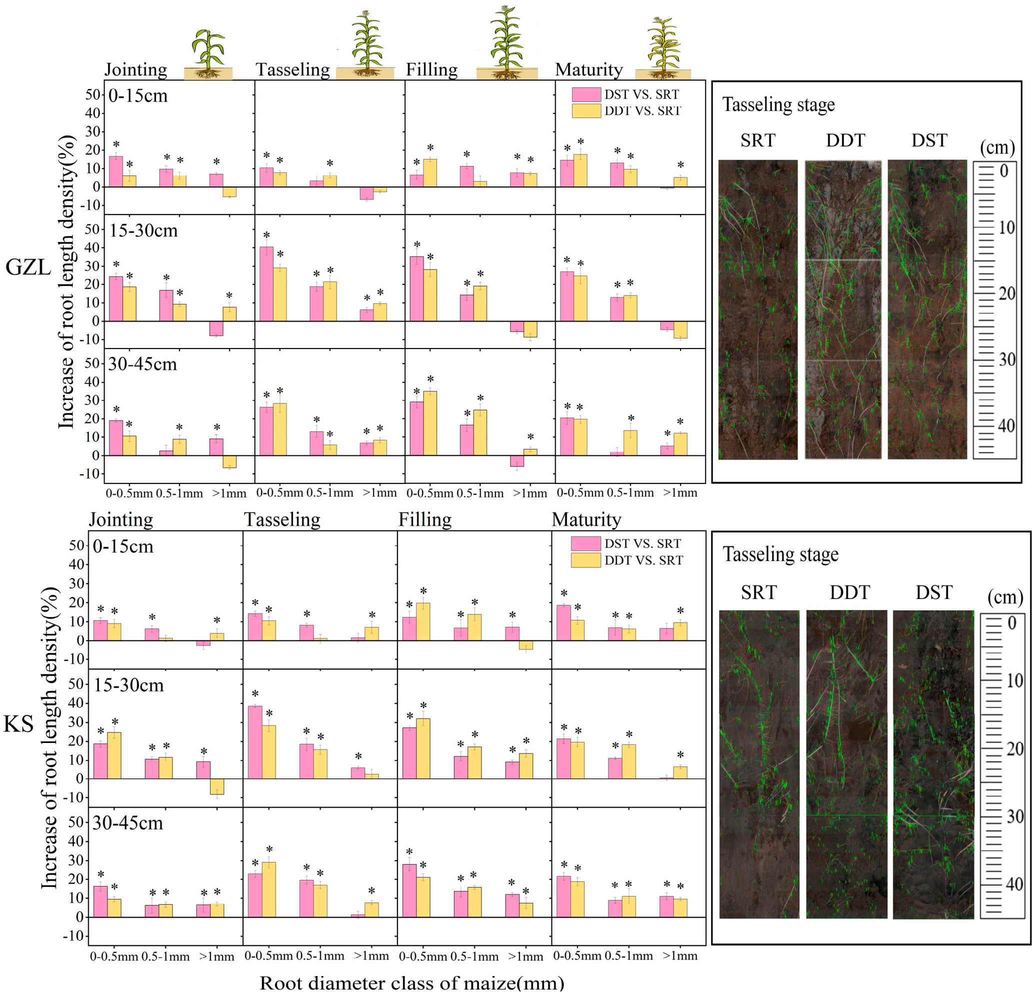

Figure 6. Increase percentage of root length density of deep incorporation treatment (DST and DDT) compared to shallow incorporation (SRT) at different growth stages at GZL and KS sites. The image in the box is the root distribution map of different treatments during the tasseling period scanned and analyzed by the minirhizotron technology. DST VS. SRT: indicates the increase percentage in DST compared to SRT; DDT VS. SRT: indicates the increase percentage in DDT compared to SRT. * Indicates significant differences between DST, DDT, and SRT ($p < 0.05$, $n = 4$).

Table 2. Effects of different incorporation depths of straw combined with manure on root productivity and turnover rate.

Layer (cm)	Treatment	Root Productivity (cm cm^2)		Turnover Rate (gp^{-1})	
		GZL	KS	GZL	KS
0–15	DST	1.65 ± 0.05 a A	1.64 ± 0.04 a A	1.28 ± 0.02 a B	1.26 ± 0.02 a B
	DDT	1.62 ± 0.06 a A	1.59 ± 0.02 ab A	1.27 ± 0.02 a A	1.28 ± 0.05 a B
	SRT	1.56 ± 0.03 b A	1.51 ± 0.05 b A	1.31 ± 0.03 a B	1.32 ± 0.04 a B
15–30	DST	1.47 ± 0.04 a B	1.46 ± 0.03 a B	1.29 ± 0.01 b B	1.34 ± 0.01 b A
	DDT	1.42 ± 0.07 a B	1.39 ± 0.06 a B	1.31 ± 0.03 b A	1.35 ± 0.04 b A
	SRT	1.20 ± 0.06 b B	1.23 ± 0.05 b B	1.40 ± 0.03 a A	1.43 ± 0.05 a A
30–45	DST	0.85 ± 0.04 a C	0.82 ± 0.03 a C	1.39 ± 0.04 b A	1.42 ± 0.03 b A
	DDT	0.80 ± 0.02 a C	0.81 ± 0.03 a C	1.33 ± 0.02 b A	1.38 ± 0.05 ab A
	SRT	0.72 ± 0.03 b C	0.73 ± 0.02 b C	1.47 ± 0.04 a A	1.50 ± 0.03 a A

Table 2. *Cont.*

Layer (cm)	Treatment	Root Productivity (cm cm^2)		Turnover Rate (gp^{-1})	
		GZL	KS	GZL	KS
ANOVA					
Layer (L)		710 **	955 **	33 **	35 **
	Straw + manure (SM)	28 **	35 **	24 **	14 **
L × SM		4 *	3 ns	3 *	0.9 ns

Gp^{-1} represents per growth period, which indicates the frequency of root biomass replacement over a unit time. DST and DDT represent deep incorporation of straw combined with manure by subsoiling tillage and deep-plow tillage, respectively. SRT represents shallow incorporation of straw combined with manure by rotary tillage. Different lowercase letters indicate significant differences between treatments within the same soil layer, while different uppercase letters denote significant differences between soil layers within the same treatment ($p < 0.05$, $n = 4$). * Indicates significance at $p < 0.05$; ** indicates significance at $p < 0.01$. ns indicates not significant.

3.3. Utilization of Water and Nutrients and Maize Yield

Compared to the SRT treatment, the synergistic deep incorporation of straw and manure (DST and DDT) increased the total nutrient (N, P, K) uptake and grain nutrient (N, P, K) accumulation (Figure 7). The DST and DDT treatments at both experimental sites increased the total nutrient (N, P, K) uptake by 6.5–17.9% and grain nutrient (N, P, K) accumulation by 15.3–25.9%, with the largest increase in N and P.

GZL

KS

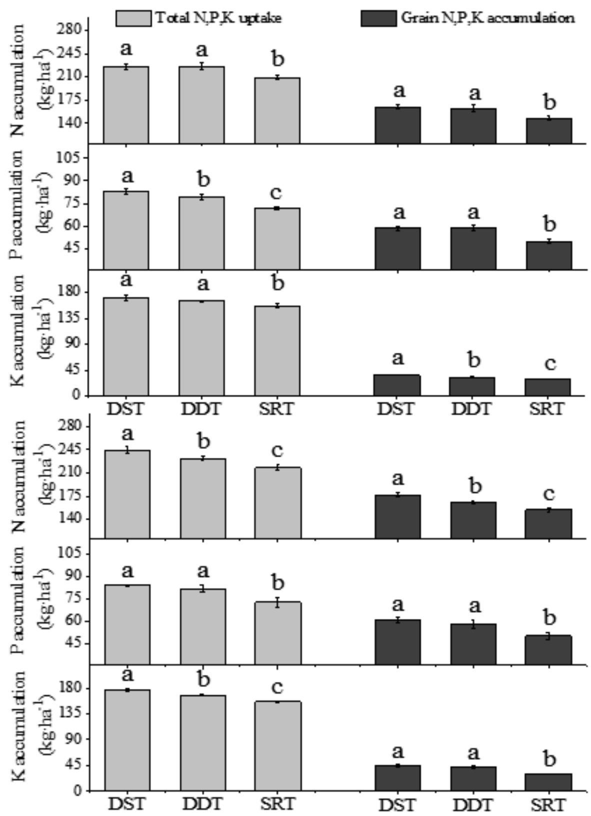

Figure 7. Total nutrient uptake and grain nutrient accumulation of NPK in maize under various incorporation depths of straw combined with manure at GZL and KS sites. DST and DDT represent deep incorporation of straw combined with manure by subsoiling tillage and deep-plow tillage, respectively. SRT represents shallow incorporation of straw combined with manure by rotary tillage. Error bars represent the standard error. Different lowercase letters indicate significant differences between treatments ($p < 0.05$, $n = 4$).

There were significant differences in dry matter yield, grain yield, and 100-kernel weight among the different treatments (Table 3). Compared to the SRT treatment, the DST and DDT treatments increased dry matter yield by an average of 2552–2930 kg ha^{-1} (13.2%) and grain yield by 1876–2180 kg ha^{-1} (17.7%).

Table 3. Effects of different incorporation depths of straw combined with manure on aboveground dry matter weight, grain yield, and 100-kernel weight.

Treatment	Dry Matter Weight (kg ha^{-1})		Grain Yield (kg ha^{-1})		100-Kernel Weight (g)	
	GZL	KS	GZL	KS	GZL	KS
DST	22515 a	24060 a	13252 a	14089 a	38.4 a	38.0 a
DDT	21706 a	23712 a	12948 a	13869 a	37.9 ab	37.6 a
SRT	20094 b	21429 b	11432 b	11941 b	36.9 b	36.3 a
ANOVA						
Site (S)	23 **		32 **		0.01 ns	
Straw + manure (SM)	53 **		78 **		7 *	
S × SM	0.7 ns		0.03 ns		0.07 ns	

DST and DDT represent deep incorporation of straw combined with manure by subsoiling tillage and deep-plow tillage, respectively. SRT represents shallow incorporation of straw combined with manure by rotary tillage. Mean values followed by different letters are significantly different at $p < 0.05$. * Indicates significance at $p < 0.05$; ** indicates significance at $p < 0.01$. ns indicates not significant.

The NPK utilization efficiency under the deep incorporation was improved compared to that under the shallow incorporation (Table 4). Compared to the SRT treatment, the DST and DDT treatments had greater increases in N and K utilization efficiency, with average increases of 8.3% and 8.1%, respectively, followed by P utilization efficiency, which increased by 3.9%. The synergistic deep incorporation of straw and manure (DST and DDT) also improved the water-use efficiency, with an average increase of 15.3% at both experimental sites compared to that in the SRT treatment.

Table 4. Effects of different incorporation depths of straw combined with manure on nutrient and water-use efficiency.

	Treatment	Nutrient-Use Efficiency (kg kg^{-1})			WUE (kg ha^{-1} mm^{-1})
		NUE	PUE	KUE	
GZL	DST	63.0 a	168.0 a	81.4 a	24.9 a
	DDT	61.4 a	171.8 a	82.5 a	23.9 a
	SRT	56.9 b	161.8 b	74.4 b	20.7 b
KS	DST	57.7 ab	167.4 ab	79.7 b	26.3 a
	DDT	59.8 a	169.1 a	82.7 a	25.9 a
	SRT	54.7 b	164.8 b	77.6 b	22.6 b
ANOVA					
Site(S)		36 **	0.8 ns	0.1 ns	134 **
Straw + manure(SM)		38 **	213 **	42 **	249 **
S × SM		5 *	19 **	3 ns	2 ns

NUE: nitrogen-use efficiency; PUE: phosphorus-use efficiency; and KUE: potassium-use efficiency. DST and DDT represent deep incorporation of straw combined with manure by subsoiling tillage and deep-plow tillage, respectively. SRT represents shallow incorporation of straw combined with manure by rotary tillage. Mean values followed by different letters are significantly different at $p < 0.05$. * Indicates significance at $p < 0.05$; ** indicates significance at $p < 0.01$. ns indicates not significant.

3.4. Linkages Between Maize Yield and Soil Physio-Chemical Properties, Root Growth Variables

The random forest model (RFM) showed that fourteen indicators were considered for explaining the response of maize yield to the different treatments (overall model: R2 = 0.92). The most important indicators were root length density, soil water storage, and total N uptake, followed by soil available N and total P uptake (Figure 8).

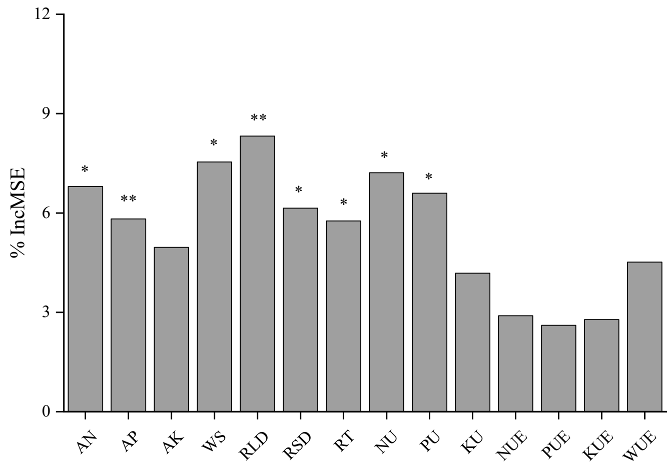

Figure 8. Relative contribution of predictors to the response of maize yield under different treatments using a random forest algorithm. AN: soil available N; AP: soil available P; AK: soil available K; WS: soil water storage; RLD: root length density; RSD: root surface area density; NU: total N uptake; PU: total P uptake; KU: total K uptake; NUE: nitrogen-use efficiency; PUE: phosphorus-use efficiency; KUE: potassium-use efficiency. * Indicates significance at $p < 0.05$; ** indicates significance at $p < 0.01$.

The conceptual diagram illustrates that the deep incorporation improves the soil moisture and nutrient conditions in the deep soil layer, facilitates fine root penetration, and enhances root productivity, thereby increasing nutrient accumulation and water- and nutrient-use efficiency, and ultimately leading to an increase in maize yield (Figure 9).

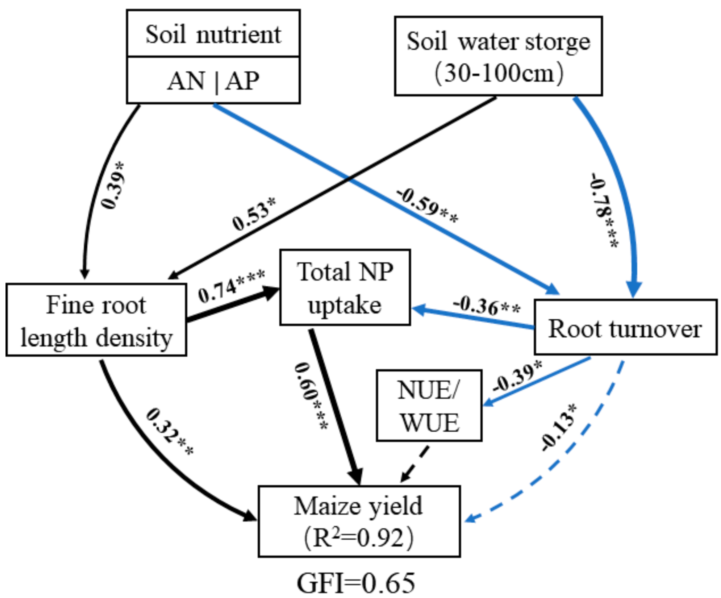

Figure 9. Structural equation model reveals the direct and indirect effects of soil moisture, nutrients, and root growth status on maize yield. Black and blue solid arrows show positive and negative associations between latent variables, respectively ($p < 0.05$, marked *; $p < 0.01$, marked **; $p < 0.001$, marked ***). Dashed lines indicate non-significant pathways. Arrow width corresponds to the strength of the relationship. Numbers adjacent to the arrows are standardized path coefficients. The value of R2 alongside maize yield indicates the variance explained by the model (R2). The model fit statistics are as follows: Chi-square(χ^2) = 105.6, goodness-of-fit index (GFI) = 0.65, comparative fit index (CFI) = 0.77.

4. Discussion

4.1. Deep Incorporation (DST and DDT) Improves the Utilization of Water and Nutrients and Changes Root Growth Strategies

The root morphology exhibits high plasticity and is greatly affected by the living environment [42]. This study demonstrates that deep incorporation can significantly alter root growth patterns and strategies by improving the utilization of water and nutrients in deeper soil layers. This is because both DST and DDT can break the plow pan using agricultural machinery, loosening compacted deep soil. This process facilitates the thorough mixing of straw and manure with the deep soil and the release of nutrients, thereby enhancing nutrient utilization [43,44]. The increased nutrient availability promotes root growth into deeper soil, as evidenced by the significant increase in fine root numbers in these layers. Additionally, the improvement in the plow pan structure enhances soil water storage (Figure 4) and mobility, ensuring sufficient water supply during the critical growth periods when maize has high water demands [31]. The noteworthy findings of this study indicate that this effect is evident in the early stages of maize root growth and becomes particularly pronounced during the tasselling and grain-filling stages (Figure 6), suggesting that the improvement in water and nutrient availability from deep incorporation is sustained over time. Based on the results from both experimental sites, the surface soil moisture content and early-stage water storage at the KS site were higher than those at the GZL site. This may be attributed to the fact that the KS site is in a thick black soil region, whereas the GZL site is in a thin black soil region. The significantly higher organic matter content at the KS site enhances its water retention capacity.

Some studies have shown that improper straw incorporation can negatively affect early crop growth due to the consumption of soil oxygen and nitrogen during decomposition, leading to slow growth [45]. For instance, the rice root growth rate decreased by 5.1–9.3% during 0–36 d after rice was transplanted [30]. However, when straw incorporation is properly managed, such as by spacing it appropriately with planting or incorporating stabilized straw/manure compost, it can mitigate these negative effects and promote crop growth. Our study found that deep incorporation of straw combined with manure promotes straw decomposition, improves soil structure, and increases nutrient availability, significantly enhancing early maize root growth. This indicates that deep incorporation not only alleviates the competition for oxygen and nitrogen but also improves the soil environment, enabling maize to adopt deeper root growth strategies and thereby enhancing overall growth performance [46,47].

In terms of root dynamics, a key finding of our study is that both DST and DDT treatments significantly increased root productivity and reduced root turnover by enhancing the availability of water and nutrients. This likely occurs because favorable soil conditions allow roots to extend further to acquire more nutrients, thereby increasing root lifespan and reducing turnover [48,49]. Otherwise, plants allocate more photosynthetic products to fine root production, shortening fine root lifespan and accelerating turnover in unfavorable soil conditions [49]. Additionally, shallow incorporation leads to loosened upper soil and compact lower soil, reducing nutrient availability in the sub-surface and increasing root turnover. This finding not only reveals the adaptive mechanisms of roots in rainfed regions but also provides new perspectives for soil management and agricultural practices. By optimizing the spatial match between maize roots and soil water and nutrient utilization, the health and stability of plant roots can be effectively improved, thereby enhancing crop productivity and achieving sustainable agriculture [38].

Notably, previous studies have shown that water stress inhibits maize root growth, leading to a significant decrease in root length and productivity [50,51]; when soil moisture levels exceed the maize water stress threshold, water supply does not significantly impact root growth [52]. However, our study in rainfed regions indicates that although the deep soil moisture content in all treatments was above the water stress level, the increase in deep soil moisture following deep incorporation was still a crucial factor in promoting root growth in rainfed areas. Contribution analysis and structural equation modeling in this study confirmed this finding. This may be attributed to the high temperatures and intermittent rainfall in Northeast China during the maize growing season, which often lead to surface soil moisture deficits. The increase in moisture after deep incorporation provides more water to the surface soil [52]. In contrast, shallow incorporation creates a compacted plow pan, significantly reducing water supply capacity and hindering root growth [31,53,54].

4.2. The Improvement in Root Growth Strategies Enhances Maize Productivity

Our research findings indicate that root length density is closely related to maize uptake of key nutrients (N, P, K). Deep incorporation promotes deeper root penetration and expansion, supporting robust crop growth during periods of high water and nutrient demand, particularly during critical growth stages such as tasselling and grain filling [43,55]. This strategy enhances crop resilience and overall yield [46,47,56]. The similar results observed between the two sites also indicate that, compared to shallow mixing of straw and manure using rotary tillage, deep tillage measures are more effective, and this has a certain level of general applicability in this experiment.

Additionally, our findings indicate that deep incorporation reduces the frequency of root turnover, potentially lowering the metabolic costs associated with root growth. This allows crops to allocate more energy and resources to above-ground growth and yield enhancement [48,57]. This process makes resource allocation more efficient, promoting overall plant health and increasing yield [58,59]. By optimizing root structure, deep incorporation enables better adaptation to the environment in deeper soil layers, improving water- and nutrient-use efficiency. This method not only reduces the reliance on new root growth during maize development but also reduces energy consumption and metabolic stress associated with frequent root renewal [60]. In contrast, shallow incorporation leads to loose upper soil and compact lower soil, hindering effective root penetration and utilization of deep water and nutrients [61,62]. Such soil structure forces plants to constantly produce new roots to meet their growth demands, increasing root turnover and significantly raising metabolic costs [28]. The frequent production of new roots consumes substantial energy and resources, inhibiting above-ground growth and affecting overall crop yield and quality. Moreover, shallow incorporation can result in uneven nutrient distribution, restricting root access to shallow resources and exacerbating energy and resource waste [22,63]. Therefore, deep incorporation has significant advantages in improving crop growth efficiency and yield, while shallow incorporation, due to its soil structure limitations, leads to high root turnover and metabolic costs, adversely affecting long-term crop health and growth.

4.3. Future Research on the Long-Term Effects of Different Straw Incorporation Depths on Maize Productivity

The improvement in soil fertility and crop productivity through no-/mini-tillage or with straw mulching may require a long period [64,65]. A study in South Africa reported reduced maize yields in the short term with no-/mini-tillage plots [66], which is consistent with our previous findings in the black soil region of northeast China (Figure S2). This is presumably attributed to topsoil compaction and limited root proliferation [67]; conversely, short-term deep cultivation with straw and manure has the advantage of loosening the upper soil and improving the nutrients in the lower soil. Other positive management practices, such as crop rotations that include deep-rooting crops, should also be considered, as they can improve soil structure, enhance water infiltration, and promote long-term soil health and productivity. Currently, the long-term effects of the incorporation depth of straw combined with manure on maize production have not been studied. It is necessary to conduct long-term experiments to compare and analyze the effects of long-term no/mini-tillage combined with straw and manure incorporation on maize growth and yield in rainfed areas.

5. Conclusions

This six-year field experiment demonstrated that deep incorporation of straw combined with manure (DST and DDT) is a promising agricultural practice for preserving black soil fertility and enhancing maize production in rainfed regions. Deep co-incorporation of straw and manure, as compared to shallow incorporation, increased soil water storage and nutrient availability in deeper soil layers, thereby stimulating deep root growth, increasing fine root length density, and reducing root turnover. The sub-surface foraging strategies of roots under the DST and DDT treatments boosted the total nutrient uptake, accumulation of dry matter during the later growth period, and grain yield while maintaining higher water- and nutrient-use efficiencies.

Supplementary Materials: The following supporting information can be downloaded at: https://www.mdpi.com/article/10.3390/agronomy14112504/s1, Figure S1: Increase percentage of root surface-area density of deep incorporation treatment (DST and DDT) compared to shallow incorporation (SRT) at different growth stages at GZL and KS sites. Figure S2: Effects of tillage and straw returning combinations on maize yield at GZL and KS.

Author Contributions: Conceptualization, C.L., S.H. and G.C.; methodology and software, C.L., S.H., Q.Z., T.X., W.Y. and S.W.; formal analysis, visualization, and validation, S.H., S.W., W.Y., Q.Z., H.W., T.X., Y.J., G.C. and C.L.; investigation and data curation and resources, S.H., S.W., W.Y., Q.Z., H.W. and T.X.; original draft preparation, review, editing, and supervision, S.H., S.W., W.Y., Q.Z., H.W., T.X., Y.J., G.C. and C.L.; project administration and funding acquisition, C.L., S.H. and S.W. All authors have read and agreed to the published version of the manuscript.

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Data Availability Statement: The original contributions presented in the study are included in the article/Supplementary Materials, further inquiries can be directed to the corresponding authors.

Conflicts of Interest: The authors declare that they have no conflicts of interest.

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