

Response of Mountain Flue-cured Tobacco to Vertical Deep Rotary Tillage Combined with Organic Carbon Fertilizer

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Abstract [Objectives] To evaluate the effects of deep vertical rotary tillage combined with organic carbon fertilizer on the improvement of acidic mountain soils. [Methods] A field experiment was conducted comparing three treatments: deep vertical rotary tillage, deep vertical rotary tillage combined with liquid organic carbon fertilizer, and traditional cultivation. The impacts of these treatments on the growth, biomass accumulation, yield, and quality of flue-cured tobacco were investigated. [Results] Deep vertical rotary tillage enhanced the growth of tobacco roots and aerial parts, as well as the accumulation of dry matter, nitrogen, phosphorus, potassium, and nicotine. The combination of vertical deep rotary tillage with organic carbon fertilizer promoted tobacco root development, increased leaf area, and facilitated the accumulation of dry matter, nitrogen, phosphorus, potassium, nicotine, and chlorine, thereby leading to an increase in the yield of flue-cured tobacco. The application of liquid organic carbon fertilizer enhanced the potassium and sugar content of flue-cured tobacco, but it also resulted in an increased nicotine content. Therefore, flue-cured tobacco cultivation is recommended under acidic soil conditions in mountainous regions, employing deep vertical rotary tillage. [Conclusions] The application of liquid organic carbon fertilizers at the time of transplanting, combined with rooting solution irrigation, and again 20 d after transplanting combined with topdressing, effectively promotes the growth and dry matter accumulation of flue-cured tobacco.

Key words Flue-cured tobacco, Soil improvement, Deep vertical rotary tillage, Organic carbon fertilizer, Dry matter, Yield, Quality of flue-cured tobacco

0 Introduction

A favorable soil environment is the prerequisite for the production of high-quality tobacco leaves^[1]. Rational cultivation can coordinate the water, fertilizer, air, and heat conditions of the soil for tobacco planting^[2], thereby effectively promoting the sustainable cycle of the tobacco field ecosystem and the efficient utilization of resources. This approach ensures the continuous improvement in the yield, quality, and efficiency of flue-cured tobacco^[3–4]. Currently, the production of flue-cured tobacco employs small rotary tillers for mechanical plowing, which results in issues such as a shallow plow layer (12–16 cm), high plow depth, susceptibility to soil compaction, and uneven distribution of soil ameliorants^[5]. These factors hinder the development of flue-cured tobacco roots, increase the likelihood of premature aging in tobacco plants, and lead to inconsistent yield and quality of tobacco leaves^[3], thereby significantly affecting the production of high-quality tobacco leaves. Vertical deep rotary tillage uses a vertical drill bit within specialized machinery to rapidly disrupt the soil,

facilitating deep loosening and fragmentation^[6]. This process enables thorough and uniform incorporation of soil ameliorants, such as lime and green manure, into both the surface and subsurface soil layers, thereby enhancing the soil environment^[7–8]. Smashing ridge tillage promotes nitrogen uptake by crops and has been shown to increase the yields of wheat and corn^[9]. Yu Shikang *et al.*^[10] found that the fragmentation of parent rocks through integrated cultivation of soil and parent rocks enhanced the water storage substrate. This approach overcomes the limitations of the shallow purple soil vegetated layer, which is characterized by low water capacity, poor water retention, and limited drought resistance. Thus, it is a rapid and effective method to expand water-holding space and improve water storage capacity. Deng Xiaohua *et al.*^[11] proposed that in mountainous regions where tobacco was cultivated, the use of smashing ridges enhanced soil quality, and smashing ridges with appropriate depth were beneficial for increasing both the yield and output value of tobacco leaves. Deng Yongsheng *et al.*^[12] demonstrated that vertical deep rotary tillage deepened the tillage layer, improved the soil's physicochemical properties, enhanced the efficacy of soil ameliorants, and promoted the growth of flue-cured tobacco. In recent years, a large amount of nitrogen fertilizer has been applied to flue-cured tobacco to enhance leaf yield and improve the income of tobacco farmers. However, an imbalance between carbon and nitrogen has emerged as a limitation in the nutrient equilibrium of crops^[13]. The use of organic carbon fertilizers has been shown to improve the uptake efficiency of mineral nutrients such as nitrogen, phosphorus, and po-

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tassium^[14], promote increases in plant height, stem thickness, and fresh biomass per plant^[15], and has thus become an important strategy for supplementing carbon in crop cultivation. Huang Qionghui *et al.*^[16] reported that the application of organic carbon fertilizer enhanced tobacco plant height, increased the number of effective leaves, and promoted the accumulation of dry matter in tobacco leaves, thereby improving both the yield and output value of tobacco leaves. However, studies examining the combined effects of vertical deep rotary tillage and organic carbon fertilizer in flue-cured tobacco cultivation remain limited. To address this gap, experiments were conducted to evaluate the effects of integrating vertical deep rotary tillage with organic carbon fertilizer application on the growth, dry matter and nutrient accumulation, yield, and quality of flue-cured tobacco. This research aims to provide technical guidance for the conservation of acidic soils in the mountainous tobacco-growing regions of Xiangxi Autonomous Prefecture.

1 Materials and methods

1.1 Experimental sites and materials The experiment was conducted from 2024 to 2025 in Huayuan County, Hunan Province, located within the subtropical monsoon mountain humid climate zone (28°31'35" N, 109°27'4" E) at an altitude of 530.0 m. The site experienced an annual sunshine duration of 1 219.2 h, an average annual rainfall of 1 363.8 mm, an average annual temperature of 15.0 °C, and a frost-free period of 279.0 d. The experimental field consisted of yellow soil upland, with green manure having been planted as the preceding crop. Soil analysis indicated a pH of 5.12, with alkali-hydrolyzable nitrogen, available phosphorus, available potassium, and organic matter contents of 82.17 mg/kg, 12.11 mg/kg, 113.34 mg/kg, and 12.50 g/kg, respectively. Organic carbon fertilizer, primarily composed of fulvic acid with a water-soluble organic carbon content of at least 12.5% and a specific gravity between 1.28 and 1.30, was applied at a rate of 45 kg/ha per application. The machinery utilized in the experiment was supplied by Hunan Tianye Modern Intelligent Equipment Co., Ltd.

1.2 Experimental design A single-factor test was conducted with four treatments: T1, vertical deep rotary tillage without the application of organic carbon fertilizer; T2, vertical deep rotary tillage with a single application of liquid organic carbon fertilizer (mixed with water and applied together with rooting solution); T3, vertical deep rotary tillage with two applications of liquid organic carbon fertilizer (once at transplanting and once 20 d after transplanting, both mixed with water for application); and CK, traditional horizontal rotary tillage without the application of organic carbon fertilizer. A randomized block design was used with three replicates, and each plot measured 100 m². The flue-cured tobacco variety used was Yunyan 87, with a nitrogen application rate of 109.50 kg/ha (the ratio of nitrogen, phosphorus, and potassium was 1 : 1.27 : 2.73). The planting density was 16 650

plants/ha, and 16 to 18 effective leaves were retained per plant. The specific experimental procedure was as follows: approximately 10 d before transplanting, lime was evenly applied to the green manure at a rate of 1 500 kg/ha^[17]. Subsequently, the green manure was turned and pressed, with a fresh grass application rate of 7 500 kg/ha^[18]. The soil was then ploughed, and ridges were formed. Vertical deep rotary tillage was performed in a single operation using a vertical deep rotary tillage ridger, completing rotary tillage to a depth of 35 cm and ridge formation with a ridge width of 120 cm and a ridge height of 35 cm. The traditional horizontal rotary tillage method involved first using a horizontal rotary tiller for tillage, followed by a micro ridger for ridging. Soil ploughing, with a depth of 16 cm, and ridging, with a ridge width of 120 cm and a ridge height of 35 cm, were completed in two separate operations. Other field management practices were conducted in accordance with the technical regulations for producing high-quality flue-cured tobacco in Xiangxi Autonomous Prefecture.

1.3 Research indicators and detection methods

1.3.1 Measurement indicators of root system. Five tobacco plants were sampled from each treatment at 30, 60, and 90 d after transplanting. The selected tobacco plants exhibited uniform and consistent growth. Sampling methods varied at different time points. At 30 d, the entire root system of each plant was carefully dug and rinsed clean for subsequent analysis. At 60 and 90 d, soil columns measuring 10 cm in diameter and 20 cm in height were collected using a plant root sampler for further examination. At 60 d, one soil column was taken from the side edge of the ridge (ridge side) and another from the middle of the ridge (ridge middle), both located 5 cm from the tobacco stem. At 90 d, one soil column was collected from the ridge side and one from the ridge middle, each positioned 8 cm from the tobacco stem. Root length, surface area, volume, average diameter, and number of root tips were determined using the LA-2400 multi-parameter root analysis system (LEGENTSYS, Canada)^[19].

1.3.2 Survey indicators of aerial growth. At 30, 60, and 90 d after transplanting tobacco seedlings, the height, number of leaves, stem girth, leaf length, and leaf width of five selected flue-cured tobacco plants from each treatment were measured according to the standard YC/T142-2010 method^[20]. Leaf area = 0.634 5 × leaf length × leaf width. The selection of tobacco leaves varied at different time points: at 30 d, the largest leaf of the entire flue-cured tobacco plant was measured; at 60 and 90 d, the plants were divided into three sections for measurement—the lower part averaged the 2nd to 4th leaves, the middle part the 6th to 9th leaves, and the upper part the 11th to 13th leaves^[12].

1.3.3 Determination of dry matter, nutrients, nicotine and chlorine. Seventy days after transplanting, five sample plants were selected from each treatment. The roots, stems, and leaves were separated and then blanched in an oven at 105 °C for 30 min. Following blanching, the samples were dried at 80 °C until a constant mass was achieved, after which the dry matter content was meas-

ured. Total nitrogen was quantified using the Kjeldahl method, total phosphorus was determined via the molybdenum-antimony colorimetric method, and total potassium was measured by flame photometry^[21]. Nicotine and chlorine contents were analyzed using a flow analyzer (SKALAR San ++, Netherlands)^[22].

Accumulation of nitrogen (phosphorus, potassium, nicotine, chlorine) (mg/plant) = [Dry matter mass (g) × 10³ × nitrogen (phosphorus, potassium, nicotine, chlorine) concentration]/100 (1)

Organ distribution ratio of dry matter (nitrogen, phosphorus, potassium, nicotine, chlorine) = Amount of dry matter (nitrogen, phosphorus, potassium, nicotine, chlorine) of a certain organ/Total amount of dry matter (nitrogen, phosphorus, potassium, nicotine, chlorine) of the plant × 100% (2)

1.3.4 Investigation on the economic characteristics of flue-cured tobacco. Each processed sample was individually graded and weighed. The yield and the proportion of first-class tobacco were subjected to statistical analysis. The average price and output value were calculated using the purchase price data from the current year^[23].

1.3.5 Determination of chemical composition in tobacco leaves. Tobacco leaf samples of B2F and C3F grades were selected after the roasting process. The contents of total sugar (TS), reducing sugar (RS), total nitrogen (TN), nicotine (Ni), and chlorine (Cl) were determined using a flow analyzer (SKALAR San ++)^[22]. Potassium (K) content in the tobacco leaves was measured by flame photometry. Coordination indices, including the sugar-nicotine ratio (TS/Ni), nitrogen-nicotine ratio (TN/Ni), and potassium-chlorine ratio (K/Cl), were calculated statistically.

2 Results and analysis

2.1 Effects of different treatments on flue-cured tobacco growth

2.1.1 Effects on the growth of subterranean root systems. Table 1 shows the root morphological indicators at various time points. Thirty days after transplanting, root length, surface area, volume, and the number of root tips followed the order T3 > T2 > T1 > CK, but no significant differences were observed in the average root system diameter. At 60 d after transplanting, root length, surface area, volume, and the number of root tips on both ridge middle and ridge side were significantly greater in treatments T1, T2, and T3 compared to CK. No significant differences were detected in the average root diameter on the ridge middle, whereas the average root diameter on the ridge side in treatments T1 and T2 was significantly larger than that in CK. At 90 d after transplanting, analysis from the ridge middle perspective revealed that root length, surface area, and the number of root tips in treatments T1, T2, and T3 were significantly greater than those in CK. Root volume in treatments T2 and T3 was significantly higher than in T1 and CK, while no significant differences were observed in the average root diameter among the treatments. From the ridge side perspective, root length and the number of root tips in treatments T1, T2, and T3 were significantly greater than those in CK. In addition, root volume in treatments T2 and T3 was significantly larger than in T1 and CK. The average root diameter in CK was significantly greater than in treatments T1, T2, and T3. No significant differences in root surface area were detected among various treatments.

Table 1 Comparison of root morphological indicators in flue-cured tobacco under various treatments

Days after transplanting//d	Treatment	Root length//cm	Root surface area//cm ²	Root volume//cm ³	Root average diameter//mm	Number of root tips
30	T1	508.05 ± 53.51 c	407.94 ± 15.64 c	20.02 ± 1.24 ab	2.08 ± 0.09 a	794 ± 34 c
	T2	725.16 ± 39.51 b	557.34 ± 38.91 b	21.78 ± 2.57 a	1.94 ± 0.27 a	1 275 ± 69 b
	T3	913.01 ± 24.42 a	717.50 ± 18.73 a	25.21 ± 2.81 a	1.93 ± 0.18 a	1 504 ± 48 a
	CK	475.31 ± 29.49 d	280.63 ± 28.75 d	18.74 ± 1.07 b	2.15 ± 0.12 a	517 ± 57 d
60	T1-ridge middle	663.79 ± 28.14 a	237.98 ± 16.47 a	4.79 ± 1.73 a	1.14 ± 0.16 a	998 ± 74 a
	T2-ridge middle	649.73 ± 12.19 a	198.17 ± 10.51 a	4.34 ± 0.90 a	0.92 ± 0.42 a	805 ± 57 a
	T3-ridge middle	632.99 ± 28.68 a	197.57 ± 19.55 a	3.83 ± 1.77 a	0.82 ± 0.13 a	841 ± 48 a
	CK-ridge middle	554.74 ± 22.65 b	170.39 ± 17.73 b	2.55 ± 1.15 b	0.88 ± 0.47 a	514 ± 23 b
	T1-ridge side	388.04 ± 11.78 a	166.64 ± 16.74 a	4.88 ± 0.21 a	1.13 ± 0.77 a	472 ± 55 a
	T2-ridge side	404.70 ± 60.72 a	157.10 ± 66.58 a	4.85 ± 2.11 a	1.24 ± 0.07 a	550 ± 42 a
	T3-ridge side	335.08 ± 69.97 a	158.30 ± 48.45 a	4.30 ± 1.70 a	0.93 ± 0.18 ab	463 ± 49 a
	CK-ridge side	249.20 ± 20.77 b	74.14 ± 23.53 b	1.31 ± 2.18 b	0.82 ± 0.05 b	343 ± 38 b
90	T1-ridge middle	399.15 ± 93.46 a	77.49 ± 12.64 a	0.79 ± 0.67 b	0.77 ± 0.17 a	865 ± 26 a
	T2-ridge middle	456.61 ± 19.96 a	96.68 ± 24.08 a	1.74 ± 0.65 a	0.72 ± 0.12 a	986 ± 33 a
	T3-ridge middle	428.38 ± 17.50 a	87.75 ± 19.45 a	1.54 ± 0.55 a	0.70 ± 0.12 a	939 ± 23 a
	CK-ridge middle	247.11 ± 19.03 b	53.69 ± 8.42 b	0.61 ± 0.89 b	0.73 ± 0.13 a	480 ± 32 b
	T1-ridge side	345.74 ± 52.23 a	66.72 ± 7.98 a	0.92 ± 0.30 b	0.55 ± 0.09 b	621 ± 34 b
	T2-ridge side	386.66 ± 20.36 a	78.01 ± 12.34 a	1.35 ± 0.59 a	0.69 ± 0.38 b	909 ± 49 a
	T3-ridge side	357.86 ± 34.67 a	68.26 ± 5.84 a	1.17 ± 0.46 a	0.69 ± 0.35 b	841 ± 48 a
	CK-ridge side	266.70 ± 23.01 b	64.95 ± 3.82 a	1.01 ± 0.56 b	1.24 ± 0.45 a	410 ± 26 c

NOTE The data in the table are expressed as mean ± standard deviation. Values of the same type in the same column that are followed by different lowercase letters denote a statistically significant difference ($P < 0.05$). The same below.

At 60 and 90 d after transplanting, only partial root samples were collected. The sampling conducted at 60 d did not demonstrate the benefits of deep rotary tillage combined with organic carbon fertilizer. However, at 90 d after transplanting, the root volume in the ridge middle and ridge side in treatments T2 and T3 was significantly greater than that observed in T1, indicating a higher root number in these treatments. The average root diameter is influenced not only by root development but also by soil compaction. Therefore, no significant differences were observed in the average root diameter of tobacco plants at 30 and 60 d after transplanting, nor in the ridge middle at 90 d after transplanting. Notably, the average root diameter on the ridge side in CK was highest at 90 d, possibly due to a greater proportion of thick roots and fewer thin roots. These results manifest that vertical deep rotary tillage promotes the growth and development of flue-cured tobacco roots, and that its combination with organic carbon fertilizer further enhances root development and increases root number.

2.1.2 Effects on aerial growth. Table 2 displays the growth in-

dicators of the aerial parts of flue-cured tobacco at various time points. Thirty days after transplanting, plant height, stem girth, leaf number, and maximum leaf area were significantly greater in treatments T2 and T3 compared to T1 and CK. At 60 d after transplanting, plant heights in treatments T2 and T3 remained significantly higher than those in T1 and CK. Stem girth and upper leaf area in treatments T1, T2, and T3 were significantly larger than in CK, while the middle leaf area in treatments T2 and T3 was also significantly greater than in CK. No significant differences were observed in leaf number or lower leaf area among various treatments. By 90 d after transplanting, no significant differences were detected in plant height, stem girth, or leaf number across treatments, but the middle and upper leaf areas in treatments T2 and T3 remained significantly larger than those in T1 and CK. These findings confirm that vertical deep rotary tillage promotes the growth of flue-cured tobacco, and its combination with liquid organic carbon fertilizer further enhances growth and development.

Table 2 Comparison of aerial growth indicators in flue-cured tobacco under various treatments

Days after transplanting//d	Treatment	Plant height//cm	Stem girth//cm	Leaf number	Area//cm ²		
					Lower leaf	Middle leaf	Upper leaf
30	T1	38.24 ± 2.27 b	6.22 ± 0.36 bc	9.20 ± 0.45 ab	—	578.36 ± 47.31 b	—
	T2	49.98 ± 3.71 a	7.24 ± 0.67 ab	9.60 ± 0.55 a	—	771.35 ± 47.84 a	—
	T3	49.66 ± 1.62 a	7.68 ± 0.47 a	9.60 ± 0.55 a	—	741.42 ± 80.71 a	—
	CK	37.64 ± 1.12 b	6.06 ± 0.27 c	8.60 ± 0.55 b	—	583.71 ± 38.86 b	—
60	T1	109.53 ± 3.52 b	9.30 ± 0.17 a	20.00 ± 1.00 a	890.70 ± 157.20 a	1 232.36 ± 89.87 bc	1 286.22 ± 43.25 a
	T2	115.37 ± 2.32 a	9.37 ± 0.15 a	18.33 ± 1.53 a	871.63 ± 119.14 a	1 366.52 ± 70.63 b	1 246.87 ± 67.79 a
	T3	116.93 ± 4.55 a	9.23 ± 0.31 a	19.00 ± 1.00 a	936.19 ± 98.72 a	1 667.75 ± 64.84 a	1 229.32 ± 53.80 a
	CK	106.70 ± 2.00 b	8.43 ± 0.31 b	20.67 ± 0.58 a	843.76 ± 137.45 a	1 161.45 ± 47.74 c	1 043.36 ± 79.35 b
90	T1	111.53 ± 4.58 a	9.47 ± 0.35 a	17.00 ± 1.00 a	—	1 175.79 ± 59.45 b	910.72 ± 57.81 b
	T2	108.17 ± 2.58 a	9.73 ± 0.21 a	16.00 ± 1.00 a	—	1 450.35 ± 69.55 a	1 208.56 ± 61.89 a
	T3	107.60 ± 3.83 a	10.10 ± 0.62 a	16.00 ± 1.00 a	—	1 416.85 ± 20.31 a	1 184.24 ± 54.92 a
	CK	106.17 ± 1.94 a	9.27 ± 0.38 a	16.00 ± 1.00 a	—	1 030.79 ± 92.85 b	823.57 ± 57.77 b

2.2 Effects of different treatments on the accumulation and distribution of dry matter in flue-cured tobacco As illustrated in Fig. 1A, the total dry matter mass of tobacco plants in various treatments followed the order T3 > T2 > T1 > CK. However, the distribution patterns across different plant organs were not uniform. Root dry matter mass in treatments T1, T2, and T3 was significantly higher than in CK; stem dry matter mass was significantly greater in treatments T2 and T3 compared to CK; and leaf dry matter mass in treatments T2 and T3 exceeded that of both T1 and CK significantly. As shown in Fig. 1B, over half of the dry matter in flue-cured tobacco was mainly distributed to the leaves, with the distribution ratio of dry matter in the roots of treatments T1, T2, and T3 surpassing that of CK. These findings suggest that vertical deep rotary tillage enhances dry matter accumulation in flue-cured tobacco and increases dry matter allocation to the roots. Moreover, the combined application of this tillage method with organic carbon fertilizer further promotes dry matter accumulation in

flue-cured tobacco.

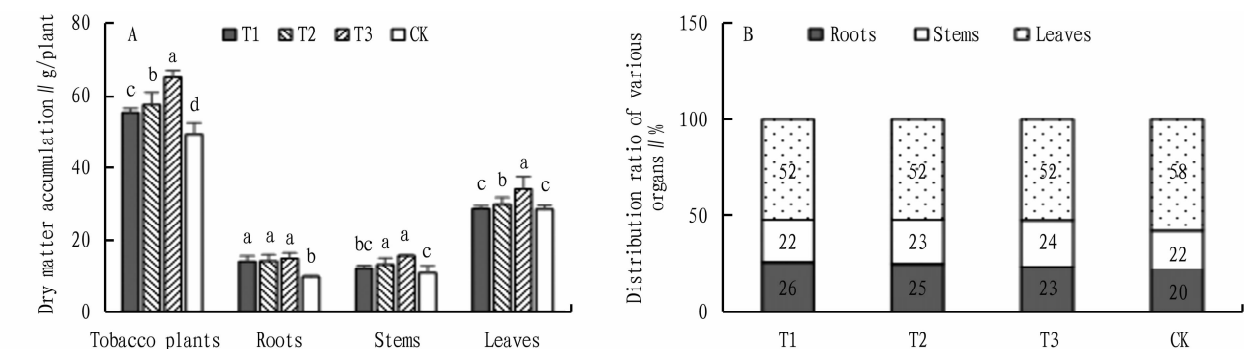
2.3 Effects of different treatments on the accumulation and distribution of nitrogen, phosphorus and potassium in flue-cured tobacco

2.3.1 Accumulation and distribution of nitrogen. As shown in Fig. 2A, nitrogen accumulation in tobacco plants was greater in treatments T1, T2, and T3 compared to CK. Meantime, nitrogen accumulation in treatments T2 and T3 exceeded that observed in T1. The distribution of nitrogen accumulation varied among different plant organs. Specifically, nitrogen accumulation was relatively high in the root systems in treatments T1, T2, and T3, while stem nitrogen accumulation was notably elevated in T2 and T3. In the leaves, nitrogen accumulation followed the order T3 > T2 > T1 > CK. As presented in Fig. 2B, the majority of nitrogen accumulated in tobacco plants was localized in the leaves. These findings indicate that vertical deep rotary tillage enhances nitrogen accumulation in tobacco plants, and that its combined application with

liquid organic carbon further improves this effect.

2.3.2 Accumulation and distribution of phosphorus. Phosphorus accumulation in tobacco plants was greater in treatments T1, T2, and T3 compared to CK (Fig. 3A). Meanwhile, phosphorus accumulation in T3 exceeded that observed in T2 and T1. Analysis by plant organ revealed that phosphorus accumulation in roots and leaves was higher in treatments T1, T2, and T3 than in CK.

Stems in treatments T2 and T3 exhibited greater phosphorus accumulation than those in T1 and CK. As illustrated in Fig. 3B, phosphorus was predominantly allocated to the leaves of tobacco plants. These results show that vertical deep rotary tillage enhances phosphorus accumulation in tobacco plants. Moreover, when combined with liquid organic carbon fertilizer, applying the fertilizer twice further promotes phosphorus accumulation in these plants.



NOTE In the same group of bar charts, bars labeled with different lowercase letters denote a statistically significant difference ($P < 0.05$). The same below.

Fig. 1 Comparison of dry matter accumulation and distribution in flue-cured tobacco under various treatments

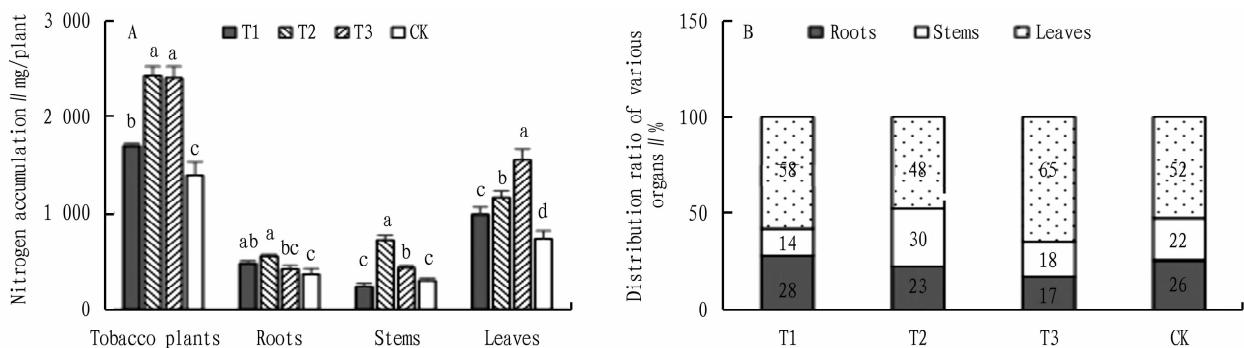


Fig. 2 Comparison of nitrogen accumulation and distribution in flue-cured tobacco under various treatments

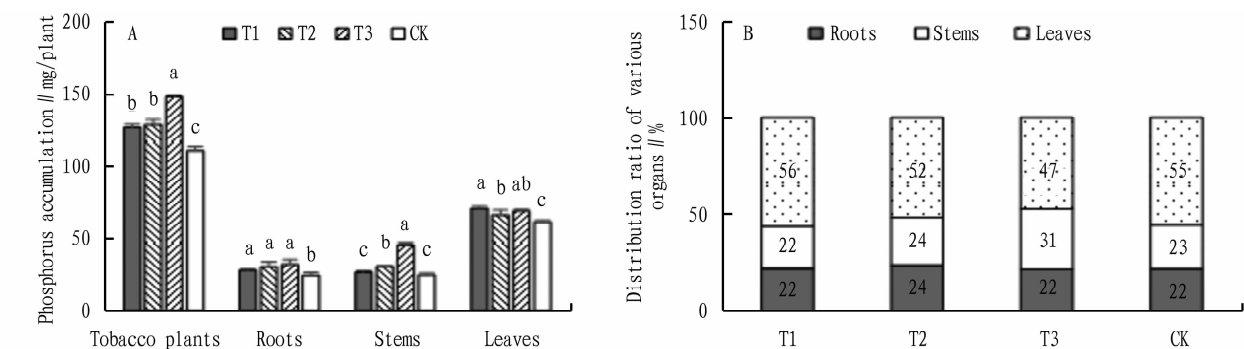


Fig. 3 Comparison of phosphorus accumulation and distribution in flue-cured tobacco under various treatments

2.3.3 Accumulation and distribution of potassium. As shown in Fig. 4A, potassium accumulation in tobacco plants under various treatments followed the order $T3 > T2 > T1 > CK$. Among the different plant organs, potassium accumulation was relatively high in the root system in treatment T1, whereas stems exhibited higher potassium accumulation in treatment T3. Potassium accumulation in tobacco leaves across all treatments also followed the pattern $T3 > T2 > T1 > CK$. Fig. 4B indicates that the majority of accumula-

ted potassium in tobacco plants was allocated to the leaves. The potassium distribution ratio in the leaves in treatments T3 and T2 exceeded 50%, which was higher than that observed in treatments T1 and CK. These findings demonstrate that vertical deep rotary tillage enhances potassium accumulation in tobacco plants. Moreover, when combined with liquid organic carbon fertilizer, this practice further facilitates potassium accumulation and increases potassium content in tobacco leaves.

2.4 Effects of different treatments on the accumulation and distribution of nicotine and chlorine in flue-cured tobacco

2.4.1 Accumulation and distribution of nicotine. As can be seen from Fig. 5A, nicotine accumulation in tobacco plants in treatments T2 and T3 was greater than that observed in T1 and CK. Nicotine levels in plants in treatment T3 exceeded those in T2. Among the various plant organs, nicotine accumulation was nota-

bly higher in the roots and leaves of plants in treatments T2 and T3, while stems exhibited relatively high nicotine levels in treatment T3. Fig. 5B indicates that nicotine was mainly localized in the tobacco leaves, followed by the roots. These findings illustrate that the application of vertical deep rotary tillage combined with liquid organic carbon fertilizer enhances nicotine accumulation in tobacco plants, particularly in the leaves.

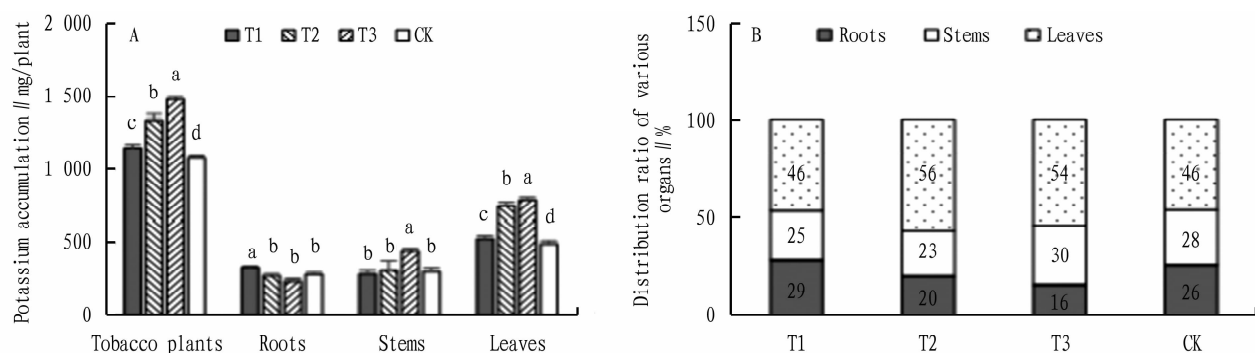


Fig. 4 Comparison of potassium accumulation and distribution in flue-cured tobacco under various treatments

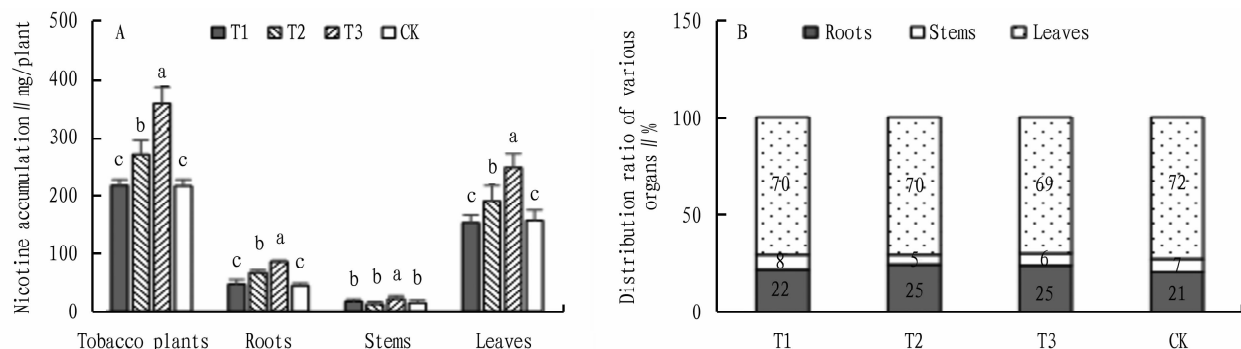


Fig. 5 Comparison of nicotine accumulation and distribution in flue-cured tobacco under various treatments

2.4.2 Accumulation and distribution of chlorine. The accumulation of chlorine in tobacco plants was greater in treatments T3 and CK compared to T1 and T2 (Fig. 6A). Besides, chlorine accumulation in treatment T2 exceeded that observed in T1. Chlorine accumulation varied among different plant organs. The root systems of treatments T1 and T3 exhibited relatively higher chlorine concentrations. Stem chlorine accumulation was relatively high in

treatment T3, while leaf chlorine accumulation was comparatively high in both T3 and CK. Overall, chlorine was mainly distributed in the tobacco leaves, followed by the stems (Fig. 6B). The relatively high chlorine accumulation in CK plants is attributed to the presence of chlorine in the topsoil plough layer and the shallow root distribution. The high chlorine accumulation in T3 plants is associated with their larger biomass and well-developed root systems.

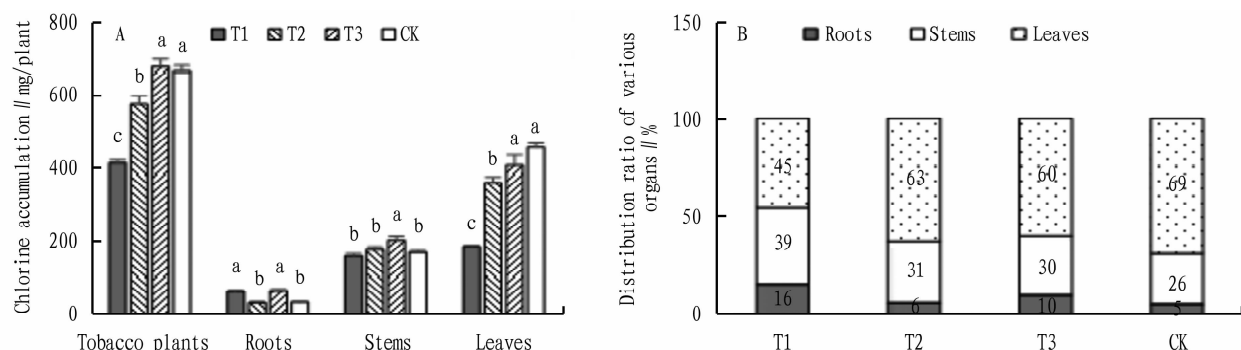


Fig. 6 Comparison of chlorine accumulation and distribution in flue-cured tobacco under various treatments

2.5 Effects of different treatments on the economic traits of flue-cured tobacco Economic trait indicators, including the proportions of first-class and medium-quality tobacco as well as the average price, did not exhibit significant differences among various treatments (Table 3). However, the yield and output value of flue-cured tobacco in treatments T1, T2, and T3 (vertical rotary tillage) were significantly higher than those observed in CK. These findings manifest that vertical deep rotary tillage effectively enhances both the yield and output value of tobacco leaves.

2.6 Effects of different treatments on the chemical composition of flue-cured tobacco leaves From the perspective of B2F grade tobacco leaves, significant differences were observed in the contents of nicotine, total nitrogen, potassium, as well as in the

sugar-nicotine ratio and potassium-chlorine ratio among tobacco leaves under various treatments (Table 4). The nicotine, total nitrogen, and potassium contents were relatively higher in treatments T2 and T3, with treatment T3 exhibiting the highest levels. Regarding C3F grade tobacco leaves, the application of organic carbon fertilizer was found to increase the sugar, nicotine, and potassium contents. For both B2F and C3F grade tobacco leaves, no significant differences in chemical composition were detected between vertical deep rotary tillage and traditional cultivation. These results prove that vertical deep rotary tillage combined with liquid organic carbon fertilizer enhances the potassium and sugar content of tobacco leaves, but the concomitant increase in nicotine content warrants careful consideration.

Table 3 Comparison of economic traits of flue-cured tobacco under various treatments

Treatment	Yield//kg/ha	First-class tobacco//%	Medium-quality tobacco//%	Output value//yuan/ha	Average price//yuan/kg
T1	1 950.00 ± 45.87 a	40.16 ± 1.04 a	51.75 ± 2.01 a	42 321.65 ± 256.06 a	21.70 ± 0.96 a
T2	1 949.72 ± 25.55 a	38.98 ± 2.11 a	47.63 ± 1.36 a	42 434.55 ± 350.88 a	21.76 ± 0.80 a
T3	1 993.06 ± 20.43 a	39.76 ± 1.08 a	46.29 ± 2.20 a	39 674.51 ± 152.24 b	19.91 ± 1.16 a
CK	1 762.11 ± 35.51 b	41.18 ± 1.53 a	44.12 ± 1.96 a	38 357.01 ± 240.01 c	21.77 ± 1.45 a

Table 4 Comparison of chemical composition of flue-cured tobacco leaves under various treatments

Grade	Treatment	TS//Ni	TN//Ni	K//Cl	TS//%	RS//%	Ni//%	TN//%	K//%	Cl//%
B2F	T1	7.07 ± 0.79 a	0.78 ± 0.04 a	3.81 ± 0.23 b	25.48 ± 1.39 a	19.56 ± 0.54 a	3.62 ± 0.24 c	2.83 ± 0.07 b	1.45 ± 0.01 c	0.38 ± 0.03 a
	T2	7.17 ± 0.31 a	0.84 ± 0.07 a	5.38 ± 0.69 a	26.75 ± 0.56 a	21.46 ± 1.79 a	3.74 ± 0.17 b	3.13 ± 0.12 a	1.83 ± 0.20 b	0.34 ± 0.10 a
	T3	5.90 ± 0.38 b	0.78 ± 0.03 a	5.32 ± 0.19 a	23.91 ± 2.06 a	18.46 ± 0.92 a	4.05 ± 0.13 a	3.22 ± 0.05 a	2.24 ± 0.02 a	0.42 ± 0.12 a
	CK	6.69 ± 0.44 ab	0.75 ± 0.03 a	3.28 ± 0.81 b	24.34 ± 1.53 a	17.81 ± 1.51 a	3.64 ± 0.18 c	2.72 ± 0.09 b	1.37 ± 0.20 c	0.43 ± 0.09 a
C3F	T1	9.05 ± 0.22 a	1.04 ± 0.15 a	4.31 ± 0.66 b	22.94 ± 0.96 b	17.17 ± 0.31 b	2.54 ± 0.17 c	2.64 ± 0.26 b	1.55 ± 0.12 b	0.36 ± 0.09 a
	T2	7.87 ± 0.32 b	0.87 ± 0.12 b	6.41 ± 0.48 a	25.71 ± 1.05 a	21.42 ± 1.48 a	3.27 ± 0.27 a	2.82 ± 0.20 a	2.20 ± 0.13 a	0.35 ± 0.13 a
	T3	6.84 ± 0.46 b	0.73 ± 0.04 b	6.44 ± 0.34 a	25.29 ± 0.53 a	21.14 ± 0.68 a	3.70 ± 0.19 a	2.69 ± 0.17 b	2.64 ± 0.11 a	0.41 ± 0.11 a
	CK	6.99 ± 0.17 b	0.79 ± 0.03 b	3.36 ± 0.39 c	23.07 ± 0.58 b	19.13 ± 0.58 b	3.30 ± 0.16 b	2.62 ± 0.09 b	1.47 ± 0.13 b	0.44 ± 0.05 a

3 Discussion

Vertical deep rotary tillage has been shown to enhance the development of both the root system and aerial parts of flue-cured tobacco, as well as to promote the accumulation of dry matter, ultimately leading to increased tobacco leaf yield. In the present study, tobacco plants subjected to vertical deep rotary tillage exhibited accelerated growth during the early and middle stages. This finding contrasts somewhat with the results reported by Lai Hongmin *et al.* [24], who studied acidic tobacco planting soil in Baise, Guangxi. The discrepancy may be attributed to differences in soil management practices; in Huayuan, Hunan, the acidic tobacco soil in this study was improved through the incorporation of green manure, lime application, and organic carbon fertilizer. This study showed that vertical deep rotary tillage enhanced root development in tobacco plants and significantly increased dry matter accumulation, corroborating the findings of Deng Xiaohua *et al.* [11] and Lai Hongmin *et al.* [24]. In the context of improving tobacco-growing soils in mountainous regions, the integration of

vertical deep rotary tillage with practices such as lime application and green manure incorporation effectively increased the depth of the soil plough layer, increased soil organic matter content, and improved soil pH. These improvements facilitated root penetration in flue-cured tobacco, promoted root and aerial growth, increased dry matter accumulation, and consequently enhanced tobacco leaf yield.

The liquid organic carbon fertilizer, primarily composed of fulvic acid, exhibits an elemental composition of 54.82% carbon, 2.29% hydrogen, 41.14% oxygen, 0.66% nitrogen, and 1.09% sulfur. Application of this fertilizer increases the soil carbon-nitrogen ratio, thereby creating favorable conditions for the proliferation of soil microorganisms, enhancing both the biological and physical fertility of the soil, improving the efficiency of chemical fertilizer utilization, and overall soil quality [25]. When combined with vertical deep rotary tillage, the liquid organic carbon fertilizer effectively supplements soil carbon deficiency, improves soil structure, activates soil microbial communities, promotes root growth in

crops, and enhances both crop stress resistance and chemical fertilizer use efficiency^[26–27]. This study showed that the fine soil fragmentation and deepening of the plough layer achieved through vertical deep rotary tillage, coupled with the carbon-nitrogen balance facilitated by the combined application of organic carbon fertilizer, significantly promoted the growth and development of flue-cured tobacco root systems. More importantly, this approach increased the number of tobacco roots, which in turn facilitated nutrient uptake, supported the aerial growth of plants, and promoted dry matter accumulation. Furthermore, it enhanced the accumulation of nitrogen, phosphorus, and potassium in tobacco plants, thereby contributing to increased tobacco leaf yield.

The soil analyzed in this study exhibited an organic matter content of 12.50 g/kg and an alkali-hydrolyzable nitrogen content of 82.17 mg/kg, characterizing it as a typical soil with "high nitrogen and low carbon levels". Given the pronounced carbon deficiency in this soil, the exogenous application of organic carbon fertilizer may serve as a critical amendment. Such fertilization can restore the balance between soil carbon and nitrogen. The water-soluble low-molecular-weight carbon compounds present in the fertilizer can be directly absorbed and utilized by crop roots and soil microorganisms, thereby promoting the growth of tobacco leaves, enhancing starch accumulation, and increasing the sugar content of flue-cured tobacco leaves. Vertical deep rotary tillage has been shown to stimulate soil nutrient availability^[6,28], while the application of organic carbon fertilizer can improve the efficiency of chemical fertilizer utilization^[25]. The combined use of these practices can enhance the soil's nitrogen supply capacity. Vertical deep rotary tillage facilitates the development of the root system in flue-cured tobacco^[8,12], and the application of organic carbon fertilizer similarly promotes more extensive root growth^[25]. Together, these interventions enhance the nitrogen uptake capacity of flue-cured tobacco. The combined effects of enhancing soil nitrogen supply and increasing the nitrogen absorption capacity of flue-cured tobacco positively influence nicotine synthesis, resulting in a higher accumulation of nicotine in the leaves. This phenomenon primarily explains the high nicotine content observed in tobacco leaves subjected to vertical deep rotary tillage combined with organic carbon fertilizer application. Fulvic acid in organic carbon fertilizer contains 0.66% nitrogen, and its impact is likely modest, although it promotes nicotine synthesis in flue-cured tobacco. However, excessive nitrogen accumulation in tobacco leaves can hinder timely ripening and yellowing, thereby adversely affecting the quality of tobacco leaf curing. This, in turn, may lead to a reduction in the average market price and the proportion of first-class tobacco leaves. Therefore, when implementing this cultivation technique in flue-cured tobacco production, it is advisable to appropriately reduce the amount of nitrogen fertilizer applied.

Chlorine exhibits surface accumulation in the plough layer of soil due to the combined effects of natural environmental factors and human influences, with its concentration gradually decreasing

as soil depth increases^[29]. This study demonstrated that the application of liquid organic carbon enhanced the development of the root system in flue-cured tobacco, resulting in a greater number of roots concentrated in the soil surface layer. Since the root systems of conventionally cultivated tobacco plants are mainly distributed within the surface soil, the accumulation of chlorine in tobacco leaves subjected to both twice-applied organic carbon fertilizer and traditional cultivation is comparatively higher. Nevertheless, the chlorine content in the tobacco leaves after roasting remained within acceptable limits.

In addition, this experiment revealed that, despite the relatively low application of organic carbon fertilizer, nitrogen accumulation in tobacco stems was the highest. Conversely, when the application of organic carbon fertilizer was relatively high, the proportion of phosphorus allocated to the leaves decreased. The underlying mechanisms responsible for these observations require further comprehensive investigation.

4 Conclusions

In the improvement of acidic soils in mountainous regions, vertical deep rotary tillage, including deep plowing, deep loosening, and soil fragmentation, facilitates the development of both the root system and aerial parts of flue-cured tobacco. This practice also promotes the accumulation of dry matter, nitrogen, phosphorus, potassium, and nicotine in flue-cured tobacco, thereby enhancing leaf yield. When combined with the application of organic carbon fertilizer, vertical deep rotary tillage can improve the carbon-nitrogen balance within the soil, which benefits root development, increases leaf area, and supports the accumulation of dry matter. Additionally, this combination promotes the accumulation of nitrogen, phosphorus, potassium, nicotine, and chlorine in flue-cured tobacco, contributing to high sugar and potassium content in tobacco leaves, but it also results in an increased nicotine concentration. In the tobacco production of Xiangxi mountainous tobacco-growing areas, vertical deep rotary tillage is employed alongside the application of liquid organic carbon fertilizers at the time of transplanting, combined with rooting solution irrigation, and again 20 d after transplanting combined with topdressing. At the same time, nitrogen fertilizer application should be appropriately reduced.

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