The effects of straw-returning and inorganic K fertilizer on the carbon–nitrogen balance and reproductive growth of cotton

HU Wei¹, YU Chaoran², ZHAO Wenqing¹, LIU Ruixian³, YANG Changqin³ and ZHOU Zhiguo¹*

Abstract

Background: Many studies have indicated that straw-returning could meet part or even all of the potassium (K) demand for crop growth in the field, but few have compared the effects of crop straw as K source and inorganic K fertilizer on carbon–nitrogen (C–N) balance of cotton and the reproductive growth. To address this, field experiments were conducted using the cotton cultivar, Siza 3, under three treatments (CK as control group one, no crop straw and inorganic K fertilizer were applied; K150 as control group two, 150 kg·ha⁻¹ of K₂O was applied; and W9000, 9 000 kg·ha⁻¹ wheat straw, which could provide K₂O about 150 kg·ha⁻¹, was incorporated into soil).

Results: Although the final reproductive organ biomass did not differ between W9000 and K150, W9000 had a higher ratio of reproductive organ biomass to total biomass (RRT), suggesting that straw-returning was more conducive to the allocation of biomass to reproductive organs. The theoretical maximum biomass of reproductive organ was higher, but the average and maximum accumulation rates of reproductive organ biomass were 2.8%~8.3% and 2.5%~8.2% lower under W9000 than K150. Also, the duration of rapid-accumulation period for reproductive organ biomass (T) was 2.0~2.8 d longer under W9000 than K150, which was a reason for the higher RRT under W9000. Straw-returning altered the dynamics of leaf K with the growth period, so that W9000 had a more drastic effect on leaf C metabolism than K150. Consequently, lower soluble sugar/free amino acid and C/N ratios were measured under W9000 than K150 at boll-setting (BSS) and boll-opening (BOS) stages. Higher leaf net photosynthetic rate, sucrose phosphate synthase and sucrose synthase activities, and lower acid invertase activity were observed under W9000 than K150 at BSS and BOS and these were more conducive to sucrose accumulation. However, less sucrose was measured under W9000 than K150 at these stages. This should be because straw-returning promoted the assimilate transport capacity when compared with inorganic K fertilizer application, which also explained the higher RRT under W9000 than K150. The lower acid invertase activity under W9000 inhibited the conversion of sucrose to other sugars, hence lower contents of soluble sugar and starch were measured under W9000 than K150.

Conclusion: Under low K condition, crop straw as K source can increase the assimilate transport from source to sink, leading to lower C/N ratio in leaf and higher allocation of biomass to reproductive organs than inorganic K fertilizer.

Keywords: Gossypium hirsutum L., Crop straw, Inorganic potassium fertilizer, Reproductive growth, C-N balance

Introduction

Cotton (Gossypium hirsutum L.) is a complex structure crop with indeterminate growth habit, which makes it more sensitive to soil potassium (K) status than other field crops (Oosterhuis 2001). Soil K deficiency would decrease boll weight, boll number and lint percentage.
imperative to minimize the dependence on inorganic K for den on cotton producers (Dong et al. 2010). Hence, it is increasing price of K fertilizer has become a heavy bur-

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since after adding K fertilizer, the transport of C assimila-

ratio of soluble sugar to free amino acid in cotton leaves,

K fertilizer application decreased the C/N ratio and the

accumulation of total biomass (Makhdum et al. 2007; Hu

et al. 2015), and the negative effects of K shortage on the
growth of reproductive organs was significantly greater

than that on vegetative organs (Hu et al. 2015). Wang

et al. (2012) and Hu et al. (2017) attributed this result to

the restriction caused by K deficiency on the transport of

carbon (C) and nitrogen (N) assimilation products from

source organs to sink organs, resulting in large amounts

of assimilates accumulating in the leaves, not in the

reproductive organs.

Zhang et al. (2014) observed that leaf C/N ratio was closely related to reproductive growth. Potassium application altered the C metabolism in cotton leaves (Wang et al. 2012), increased the activities of enzymes related to sucrose synthesis, such as sucrose synthase (SuSy) and sucrose phosphate synthase (SPS), and decreased the activity of enzymes responsible for sucrose decomposi-

tion, such as acid invertase, in cotton leaves (Zahoor et al. 2017a). Potassium application also decreased the contents of unstructured sugars (hexose, sucrose, starch) in cotton leaves (Hu et al. 2016a). Also, the N metabolism in cotton leaves was promoted by K fertilizer application. Potassium fertilizer application could improve NO$_3^-$ uptake by roots (Rufty et al. 1981), leading to high N concentration and nitrate content in cotton leaves (Hu et al. 2016b; Zahoor et al. 2017b). Hu et al. (2017) found that K fertilizer application decreased the C/N ratio and the ratio of soluble sugar to free amino acid in cotton leaves, since after adding K fertilizer, the transport of C assimila-
tion products to the reproductive organs was improved more significantly than that of N assimilation products.

In recent years, K deficiency in soil has become one of the main limiting factors for cotton production around the world, and this phenomenon is found in many parts of China (Wang et al. 2012). Thus, the farmers have to apply large amounts of inorganic K to cope with the soil K deficiency. However, the long-term application of inorganic K will result in a series of environmental problems, such as soil crusts, soil acidification and underground water pol-

ylation (Paradelo et al. 2013; Udeigwe et al. 2015). Also, the increasing price of K fertilizer has become a heavy bur-

den on cotton producers (Dong et al. 2010). Hence, it is imperative to minimize the dependence on inorganic K for cotton production. Previous studies have showed that crop straw usually contains high K content (Almaz et al. 2017; Sui et al. 2017) and the K cation in crop straw is more easily released than other nutrients (Li et al. 2014). Wu et al. (2011) reported that K cation in crop residue could be released more than 90% after 90 days post decomposition. Thus, crop straw has been considered as bio-K fertilizer instead of inorganic K fertilizer in many countries (Sui et al. 2015). Some experiments have indicated that straw-returning could meet part or even all of the K demand for crop growth in the field (Tan et al. 2007; Liu et al. 2010; Zhao et al. 2014). Since wheat (Triticum aestivum L.) and cotton double cropping is widely adopted in the Yangtze Valley region, lots of wheat straw can be harvested before planting cotton. Previous studies have showed that 0.9 t·ha$^{-1}$ wheat straw returning to soil could replace about 150 kg·ha$^{-1}$ K$_2$O which is the recommended quantity of K application for cotton production in the Yangtze Valley region (Sui et al. 2015; Yu et al. 2016) in the short term (≤ 3 years). However, the nutrients in the straw are released gradually (Wu et al. 2011), and the nutrient release pattern of straw is similar to that of slow-release fertilizer (Zhang et al. 2018). Hence, the effect of straw returning to the field on the dynamic growth of crops may be different from that of inorganic K fertilizer. However, little is known of the comparative effects of straw-returning and inorganic K fertilizer application on cotton growth.

We hypothesized that crop straw as K source and inor-

ganic K fertilizer would have different effects on C-N balance in cotton, which would influence the dynamic growth of reproductive parts. The objective of the current study was to explore and compare the effects of crop straw-returning and inorganic K fertilizer on the relationship between C and N metabolism, and the dynamics of cotton reproductive growth.

**Materials and methods**

**Experimental materials and treatments**

The cotton cultivar, Siza 3 was planted in the experimen-
tal field of Jiangsu Academy of Agricultural Sciences (clay soil with low available K content) in Nanjing (32° 20‘ N and 118° 52’ E) from 2012 to 2013. The seeds were sown on 25 April in nutrient bowls. On 5 June, healthy and uniform seedlings were selected for transplanting in the field. There treatments [CK, no crop residue and K fertilizer as control group one; K150, 150 kg·ha$^{-1}$ K$_2$O, which is the recommended quantity of K fertilizer in the Yangtze River cotton belt (Hu et al. 2015), was applied into the soil in the form of potassium sulfate before transplanting as control group two; and W9000, 9 000 kg·ha$^{-1}$ wheat straw, which could supply about 150 kg·ha$^{-1}$ of available K for cotton based on final cotton yield (Sui et al. 2015; Yu et al. 2016), was crushed and incorporated in
0~10 cm soil before transplanting] were designed for this experiment. A randomized complete blocks design with three replications was used for this experiment. The area of each plot was 7 × 4 m². The intra- and inter-row spacings were 0.3 m and 1.0 m, respectively. In addition, the placement of each plot was fixed in the 2 years. The nutrients in 0~20 cm soil before transplanting in each year are showed in the Additional file 1: Table S1. The nutrient (N, P and K) analysis of the crop straw performed using the Kjeldahl method (Nelson and Sommers 1972), the molybdenum blue colorimetric method (Rodriguez et al. 1994), and the flame atomic absorption spectrophotometer method (Hu et al. 2015), respectively, showed that the nutrients provided by 9,000 kg wheat straw were 96 kg N, 39 kg P₂O₅ and 161 kg K₂O in 2012 and 79 kg N, 37 kg P₂O₅ and 133 kg K₂O in 2013. In this study, sufficient N (300 kg·ha⁻¹) and P (150 kg·ha⁻¹) were applied to all plots during the growth season (Yu et al. 2016).

Biomass accumulation
Three plants per plot were sampling at 80, 95, 110, 125, 140, 155 days after sowing (DAS) in 2012 and at 80, 95, 110, 125, 140 DAS in 2013 to measure the plant biomass. The plant samples were divided into vegetative organs (root, stem, fruiting branches, petiole and leaves) and reproductive organs including bolls, flowers, and buds. After the samples were oven-dried at 105 °C for 30 min and then at 80 °C to constant weight, the dry weight was recorded.

Sampling and processing
The functional leaf [the third (after tip pruning) or fourth (before tip pruning) from top to bottom of mainstem] was sampled at the peak flowering stage (PFS) on 15 July, 2012 and 18 July, 2013, at the boll-setting stage (BSS) on 15 August, 2012 and 15 August, 2013, and at the boll-opening stage (BOP) on 8 September 2012 and 10 September 2013. The sampled leaves were cleaned with deionized water. Then, the main veins of the leaves were abandoned. Half of the leaves to be used for the measurement of enzyme activities, were quick-frozen using liquid nitrogen before being stored at −80 °C in an ultralow temperature freezer. The remaining leaves to be used for determining the concentrations of N, P and K and the contents of substance related to C and N metabolism, were dried in an 80 °C oven.

Net photosynthetic rate \( (P_n) \)
The \( P_n \) of functional leaf was measured between 9:00 and 11:00 using a Li-6400 gas exchange measuring system (Li-COR, Lincoln, NE, USA) in three replications. The leaf chamber condition was set at relative humidity of (65 ± 5)%, leaf temperature of (32 ± 2) °C, CO₂ concentration of (380 ± 5) μmol·mol⁻¹, and quantum flux of 1 500 μmol·m⁻²·s⁻¹.

Leaf C, N, P and K contents
Leaf C content was assayed with the potassium dichromate wet digestion method according to Zhang et al. (2014). A H₂SO₄-H₂O₂ solution was used to digest the leaf tissues. Then, the contents of N, P and K in H₂SO₄-H₂O₂ solution were analyzed with the Kjeldahl method (Nelson and Sommers 1972), the molybdenum blue colorimetric method (Rodriguez et al. 1994), and the flame atomic absorption spectrophotometer method (Hu et al. 2015).

Carbohydrates and N compounds
Leaf tissues (0.1 g) and 80% (v/v) ethanol (5 mL) were placed into a 10 mL centrifuge tube. Then, the tube was heated for 30 min in an 80 °C water bath before centrifuging for 5 min at 4 000 r·min⁻¹. The solid at the bottom of centrifuge tube was extracted twice more using 5 mL 80% (v/v) ethanol. After each centrifugation, the supernatant was collected. The final volume of the supernatant was fixed at 25 mL by adding 80% (v/v) ethanol. The anthrone colorimetric method was used for the assay of soluble sugar content and the resorcinol chromogenic method was used for the measurement of sucrose content in the final supernatant (Hendrix 1993). The final insoluble residue mentioned above was used for starch measurement. The starch in the residue was degraded to glucose using the perchloric acid decomposition method described previously (Hu et al. 2015). Then, the glucose content was assayed using the anthrone reagent (Morris 1948).

Nitrate content was extracted according to Ruiz and Romero (2002). Millipore-filtered water (10 mL) was used to extract nitrate in the dried leaves (0.2 g); 100 μL extract and 0.2 mL salicylic acid (10%) were added into a tube to wait for 20 min. Then, 4.75 mL NaOH (8%) was pipetted into the tube to wait for 30 min. The nitrate was calculated after measuring the absorbance of the mixture at 410 nm. The extraction of carbohydrates described above were used for assaying the free amino acid content using the ninhydrin method according to Yemm et al. (1955).

Enzyme activity
The crude enzyme solution was extracted according to Huber and Israel (1982). SPS (E.C. 2.4.1.14) activity was quantified according to the method of Hu et al. (2015); 200 μL crude enzyme solution and 350 μL reaction
solution prepared with extraction buffer, fructose-6-P (50 mmol·L$^{-1}$), MgCl$_2$ (10 mmol·L$^{-1}$) and UDP-glucose (50 mmol·L$^{-1}$) were incubated for 30 min at 30 °C before adding 2 mol·L$^{-1}$ NaOH (100 μL). After cooling, the mixture was heated again at 80 °C with 0.1% (w/v) resorcin (1 mL) which was prepared with 95% (v/v) ethanol and 30% (w/v) HCl (3.5 mL) for 10 min. The resorcin in the reaction mixture was measured at the wavelength of 480 nm. The assay method of SuSy (E.C. 2.4.1.13) activity was same as the method described above for SPS except that fructose 6-P was replaced with D-fructose.

For acid invertase (E.C. 3.2.1.26) activity assay, the crude enzyme solution of 100 μL was incubated at 30 °C with 2.2 mL acetic acid-NaOH (200 mmol·L$^{-1}$, pH 5.0) and 200 μL sucrose (1 mol·L$^{-1}$) for 30 min. After adding 1 mL 3,5-dinitro salicylic acid, the mixture was boiled for 5 min. The glucose in the reaction mixture was determined at the wavelength of 540 nm (Hu et al. 2019a). The assay method of alkaline invertase activity was similar to that of acid invertase except that sodium acetate-acetic acid (100 mmol·L$^{-1}$, pH 7.5) was used to replace acetic acid-NaOH.

### Statistical analysis

The statistical analysis software SPSS (ver. 22.0, IBM, USA) was used for the analysis of variance (ANOVA) and means were compared using the LSD test at $P=0.05$. The mapping software Origin (Pro 8.0, OriginLab, USA) was chosen to make the figures. Yang et al. (2011) reported that the determination coefficient ($R^2$) for all treatments was higher than 0.95 and the $P$ values were less than 0.01, suggesting that the formula could well describe the dynamic process of reproductive biomass accumulation.

### Results

#### Reproductive organ biomass accumulation

The reproductive organ biomass was higher under K150 and W9000 than CK, with no obvious difference was found between K150 and W9000 (Table 1). The ratio of reproductive organ biomass to total biomass (RRT) increased by 4.8%~7.8% and 8.8%~13.6% under K150 and W9000, and being significantly higher under W9000 than K150. The reproductive organ biomass had a highly significant positive correlation with seed cotton yield ($P<0.01$) (Fig. 1).

#### Simulation of reproductive organ biomass accumulation

Equation (1) was used to fit the reproductive organ biomass accumulation, and the results in Tables 2 showed that the determination coefficient ($R^2$) for all treatments was higher than 0.95 and the $P$ values were less than 0.01, suggesting that the formula could well describe the dynamic process of reproductive biomass accumulation.

### Table 1 Reproductive organ biomass accumulation and ratio of reproductive organ biomass to total biomass (RRT) in 2012 and 2013

| Treatment | 2012 Reproductive organ biomass (kg·ha$^{-1}$) | RRT (%) | 2013 Reproductive organ biomass (kg·ha$^{-1}$) | RRT (%) |
|-----------|---------------------------------------------|--------|---------------------------------------------|--------|
| CK        | 1 959.4 b                                   | 43.4 c | 2 181.7 b                                   | 46.4 c |
| K150      | 3 679.4 a                                   | 45.2 b | 4 403.4 a                                   | 50.0 b |
| W9000     | 3 691.6 a                                   | 47.2 a | 4 480.0 a                                   | 52.2 a |

CK, no crop straw and K fertilizer were applied as the blank control treatment; K150, 150 kg ha$^{-1}$ K$_2$O was applied before transplanting the cotton seedlings; and W9000, 9000 kg ha$^{-1}$ wheat straw was crushed and incorporated in 0~10 cm soil before transplanting. Values followed by different letters within the same column are significantly different at $P=0.05$. Each value represents the mean of three replications.
The Biom under K150 and W9000 was significantly increased relative to CK, and it was higher under W9000 than K150. The DAS1 and DAS2 were earliest under CK, followed by K150 and W9000; T and VT increased under K150 and W9000 relative to CK, and T was longer under W9000 than K150, although VT was lower under W9000. The variations in DASm and Vm among treatments followed the same trend as T and VT, respectively.

Concentration of N, P and K in functional leaf
Leaf K level under K150 and W9000 was significantly higher than that under CK at all sampling dates in 2012 (except for PFS) and 2013 (Table 3). At PFS and BOS, leaf K concentration was similar under K150 and W9000, but it was lower under K150 than W9000 at BSS. Leaf N concentration under K150 and W9000 increased significantly at the BSS in 2013 and at the BOS in both years (Table 3). No significant difference was found in leaf P content across treatments and sampling dates (Table 3).

**Table 2** The fitting of reproductive organ biomass accumulation and main eigenvalues of reproductive organ biomass accumulation for different treatments in 2012 and 2013

| Year | Treatment | Regression equations | $R^2$ | Fast accumulation period | Fastest accumulation point |
|------|-----------|----------------------|-------|--------------------------|---------------------------|
|      |           |                      |       | DAS1/d | DAS2/d | T/d | Vm/(kg d$^{-1}$) | Vm/(kg d$^{-1}$) | DASm/d |
| 2012 | CK        | Bio = 1 979.8/(1 + 563 429.6e$^{-0.136 5DAS}$) 0.991 3** | 87.4 | 106.7 | 19.3 | 59.6 | 67.5 | 97.0 |
|      | K150      | Bio = 3 718.6/(1 + 722 502.4e$^{-0.113 0DAS}$) 0.987 8** | 107.7 | 131.0 | 23.3 | 92.8 | 105.0 | 119.4 |
|      | W9000     | Bio = 3 829.4/(1 + 238 847.9e$^{-0.100 8DAS}$) 0.999 2** | 109.8 | 136.0 | 26.1 | 85.1 | 96.4 | 122.9 |
| 2013 | CK        | Bio = 2 220.2/(1 + 4 127 754.6e$^{-0.138 5DAS}$) 0.992 6** | 100.5 | 119.5 | 19.0 | 67.9 | 76.9 | 109.9 |
|      | K150      | Bio = 4 617.7/(1 + 667 404.8e$^{-0.117 0DAS}$) 0.991 6** | 103.3 | 125.8 | 22.5 | 119.3 | 135.1 | 114.6 |
|      | W9000     | Bio = 4 926.3/(1 + 321 420.9e$^{-0.107 0DAS}$) 0.996 2** | 106.2 | 130.7 | 24.5 | 116.3 | 131.8 | 118.5 |

**Table 3** Changes in leaf N, P and K concentrations for different treatments in 2012 and 2013

| Year | Treatment | Peak flower stage (PFS) | Boll-setting stage (BSS) | Boll-opening stage (BOS) |
|------|-----------|-------------------------|-------------------------|-------------------------|
|      |           | K/(mg kg$^{-1}$) | N/(mg kg$^{-1}$) | P/(mg kg$^{-1}$) | K/(mg kg$^{-1}$) | N/(mg kg$^{-1}$) | P/(mg kg$^{-1}$) | K/(mg kg$^{-1}$) | N/(mg kg$^{-1}$) | P/(mg kg$^{-1}$) |
| 2012 | CK        | 29.45 a | 32.01 a | 3.17 a | 13.59 c | 25.02 a | 2.95 a | 8.84 b | 16.38 b | 2.25 a |
|      | K150      | 31.24 a | 31.65 a | 2.98 a | 18.55 b | 27.04 a | 3.23 a | 16.09 a | 20.29 a | 2.54 a |
|      | W9000     | 34.05 a | 29.97 a | 3.16 a | 21.66 a | 27.84 a | 3.18 a | 18.51 a | 21.95 a | 2.54 a |
| 2013 | CK        | 33.26 b | 37.04 a | 3.54 a | 10.48 c | 26.46 b | 2.65 a | 9.09 b | 15.94 b | 2.25 a |
|      | K150      | 32.83 a | 36.80 a | 3.40 a | 17.42 b | 30.33 a | 2.82 a | 15.81 a | 18.68 a | 2.31 a |
|      | W9000     | 30.45 a | 35.57 a | 3.32 a | 21.89 a | 29.06 a | 2.68 a | 16.49 a | 20.11 a | 2.38 a |

CK, no crop straw and K fertilizer were applied as the blank control treatment; K150, 150 kg ha$^{-1}$ K$_2$O was applied before transplanting the cotton seedlings; and W9000, 9 000 kg ha$^{-1}$ wheat straw was crushed and incorporated in 0–10 cm soil before transplanting. Values followed by different letters within the same column are significantly different at $P = 0.05$. Each value represents the mean of three replications.
Net photosynthetic rate of functional leaf
The $P_n$ of functional leaf under K150 and W9000 was noticeably higher than that under CK across sampling dates for both years (except for PFS in 2012, Fig. 2). Compared with K150, the $P_n$ of functional leaf increased significantly under W9000 at the BSS (16.1%~21.6%) for both years and at the BOS (24.3%) in 2013.

Carbohydrates and N compounds in functional leaf
Soluble sugar content was markedly decreased under K150 and W9000 at BSS and BOS (Fig. 3), and the decrease was larger under W9000 than K150 in 2012. Sucrose content did not differ among treatments at PFS (except for 2013, Fig. 3), but was obviously decreased under K150 and W9000 at BSS and BOS in both years. Similar to soluble sugar content, a larger decrease in sucrose was observed under W9000 than K150 (except for BOS in 2013). At PFS, there were no differences in starch content among treatments (Fig. 3), but the starch content was significantly decreased under K150 and W9000 at BSS (18.6%~24.9% under K150; 21.4%~29.3% under W9000) and BOS (26.0%~35.2% under K150; 31.9%~44.5% under W9000).

At PFS, no significant differences in nitrate content were detected among treatments (Fig. 4). At BSS, the nitrate content increased significantly by 28.0%~32.1% under K150 and 25.0%~48.7% under W9000 in the 2 years. Similar increases in nitrate content were observed under K150 and W9000 at BOS in the 2 years (except for K150 in 2012). No significant change in nitrate content was observed between K150 and W9000 in the 2 years. The free amino acid concentration under K150 and W9000 significantly decreased at BSS (19.9%~21.8% under K150; 23.7%~25.0% under W9000) and BOS (17.3%~24.9% under K150; 20.4%~29.6% under W9000) in the 2 years (Fig. 4). There was no significant difference between K150 and W9000.

The soluble sugar/free amino acid ratio did not differ among treatments at PFS in 2012 (Fig. 5), but was significantly decreased under K150 and W9000 at BSS and BOS in 2012 and at all sampling dates in 2013, and the decrease was larger in W9000 than K150 (apart from BOS in 2012). Similar to soluble sugar/free amino acid ratio, the C/N ratio significantly decreased under K150 and W9000 at all sampling dates (apart from PFS in 2012, Fig. 5), and compared with K150, W9000 had a lower C/N ratio (apart from BOS in 2012).
Enzyme activities related to C metabolism
Sucrose phosphate synthase activity was little affected by K150 and W9000 at PFS, but was significantly increased under K150 and W9000 at BSS and BOS (Fig. 6), and the increase was more significant under W9000 (45.2%–116.8%) than K150 (27.7%–62.3%). The activity of SuSy was markedly influenced by K150 at PFS (Fig. 6). An increase of 27.7%–123.8% was observed in SuSy activity under K150 at BSS and BOS; an increase of 54.1%–170.2% was found under W9000 at BSS and BOS. Besides, the increase under W9000 was greater than that under K150 (apart from BSS in 2013).

Acid invertase was markedly reduced under K150 and W9000 at BSS and BOS in 2012 and at all sampling dates in 2013 (Fig. 7). Also, the decrease under W9000 was more obvious than that under K150 (except at the PFS in 2013). There were no significant differences in alkaline invertase among treatments in 2012 (Fig. 7). In 2013, the alkaline invertase was much lower under K150 at BOS and under W9000 at BSS and BOS.

Discussion
Previous studies showed that additional inorganic K fertilizer increased cotton productive organ biomass and yield under K deficiency (Makhdum et al. 2007; Hu et al. 2015). It also influenced the physiological metabolism in cotton, which was beneficial to C (Hu et al. 2015) and N (Drosdoff et al. 1947; Wang et al. 2012) metabolism. Hu et al. (2017) further showed that inorganic K fertilizer had a more significant effect on C metabolism than N metabolism. Hence, a lot of results on the effect of inorganic K fertilizer on cotton have been obtained. In this study, we focused on comparing the effects of wheat straw as K source and inorganic K fertilizer on cotton under low K condition.

Comparative effects of straw and inorganic K fertilizer on cotton reproductive biomass under low K condition
Previous studies have found that under low K, the seed-cotton yield and lint yield were much higher under the straw-returning and K fertilizer treatments compared with blank control (Sui et al. 2015; Yu et al. 2016), since both straw-returning and K fertilizer application could increase available K in soil and promote root growth, which in turn facilitated nutrient absorption. Moreover, straw-returning could also alter soil C and N characteristics and microbial activities, which improved the growth environment of root, and was beneficial to the growth of
cotton plants (Hu et al. 2019b). The yield formation of cotton is closely related to the biomass of reproductive organs (Li et al. 2020). The results of correlation analysis in the current study showed that the reproductive organ biomass had a positive correlation with seed cotton yield ($P<0.01$) (Fig. 1), which confirmed the previous conclusion of Li et al. (2020). Although no significant difference in the reproductive organ biomass was measured between K150 and W9000, the W9000 had a significantly higher RRT than K150 (Table 1), indicating that straw-returning was more conducive to the allocation of biomass to reproductive organs than inorganic K fertilizer.

Liu et al. (2018) reported that the K release rate of wheat straw was much slower in dry land than in paddy field, and about 90 days were needed for K ion to release completely in dry land. Thus, the wheat straw returning to the dry land was similar to a kind of slow-release fertilizer which could change the dynamic accumulation and allocation of crop biomass by changing nutrient supply intensity and rate (Fan and Li 2009). As shown in Table 2, the eigenvalues of reproductive organ growth ($\text{Bio}_m, V_T, V_m, T$ and $\text{DAS}_m$) were increased in both K150 and W9000, meaning that both inorganic K fertilizer and straw-returning changed the reproductive organ accumulation pattern. Meanwhile, $\text{Bio}_m$ was higher in W9000 than K150, which indicated that crop straw had greater potential to accumulate the biomass of reproductive organs than inorganic K fertilizer. The $V_T$, $V_m$, $T$ and $\text{DAS}_m$ are the important parameters determining biomass accumulation (Yang et al. 2012; Du et al. 2016). The $V_T$ and $V_m$ were lower under W9000 than K150, suggesting that inorganic K fertilizer could better promote the rate of accumulation of reproductive organs than crop straw. However, $T$ was $2.0 \sim 2.8$ d longer under W9000 than K150, meaning that the duration of rapid reproductive growth period was extended by straw-returning relative to inorganic K fertilizer application, and the extension of rapid reproductive growth period was beneficial for yield formation (Yang et al. 2011). Consequently, $T$ should be dominant compared with $V_T$, leading to the higher RRT under W9000 than K150. Moreover, $\text{DAS}_1$, $\text{DAS}_2$ and $\text{DAS}_m$ were late by $2.1 \sim 2.9$ d, $4.9 \sim 5.0$ d and $3.5 \sim 3.9$ d, respectively, under W9000 compared with K150, indicating that crop straw-returning could delay the start and end times of rapid reproductive growth in relation to inorganic K fertilizer, which might be because crop straw return to soil can effectively retard the senescence of crops and prolong the growth period (Mu et al. 2012).
Comparative effects of straw-returning and inorganic K fertilizer on cotton C–N balance under low K condition

Song et al. (2015) reported that straw-returning could replace inorganic K fertilizer under low K condition to increase the cottonseed K concentration. Similar results were observed in the current study. Leaf K level was much higher under K150 and W9000 than CK (Table 3). Compared with K150, W9000 did not change leaf K concentration at PFS and BOS, but increased leaf K concentration at BSS, suggesting that crop straw incorporation altered the dynamics of leaf K during growth period. A change in leaf K concentration would affect the C–N balance in cotton (Hu et al. 2017). Thus, the leaf C/N and soluble sugar/free amino acid ratios were significantly decreased under K150 and W9000 at BSS and BOS (Fig. 5). And compared with K150, W9000 caused greater reductions in both years (apart from BOS in 2012). A change in the C–N balance in leaves could influence the reproductive growth of crops (Zhang et al. 2014). Hence, the difference in C/N ratios between K150 and W9000 might be one of the reasons for the inconsistency in dynamic accumulation of reproductive organ biomass which was manifested through different accumulation eigenvalues (VT, Vm, T, DAS1, DAS2 and DASm). Greater decreases in C/N ratio and soluble sugar/free amino acid ratio also led us to speculate that crop straw as K source might have a more drastic effect on C or N metabolism than inorganic K fertilizer. Zahoor et al. (2017b) and Hu et al. (2016b) found that extra K fertilizer could obviously increase leaf N and nitrate–N contents. Similarly, leaf N concentration and the content of nitrate as the substrate for N assimilation were higher in K150 and W9000 than CK at BSS in 2012 and at BOS in both years, but did not differ between K150 and W9000 (Table 3, Fig. 4). Also, the content of free amino acid, one of the main products of N assimilation, did not differ between K150 and W9000 (Fig. 4). These indicated that the impacts of straw-returning and K fertilizer application on N metabolism in cotton leaf were consistent, and did not support the above speculation that crop straw-returning had a more drastic effect on N metabolism than inorganic K fertilizer application. Previous studies showed that leaf K status could influence soluble sugar, sucrose and starch accumulation in leaves (Hu et al. 2015; Zahoor et al. 2017a). In support of these reports, we measured lower soluble sugar, as well as sucrose and starch contents under K150 and W9000 relative to CK at BSS and BOS in both years (Fig. 3), and the decrease was larger in W9000 than K150, suggesting that straw-returning had different effects on C metabolism than inorganic K fertilizer at some stages, which supported our above speculation that crop straw-returning had a more drastic effect on C metabolism.

Previous results of inorganic K fertilizer affecting C metabolism showed that K application increased Pn, SPS, and SuSy activities, decreased acid invertase (SAI) activity and had no effect on alkaline invertase activity in cotton leaves (Hu et al. 2015; Zahoor et al. 2017a). Same results were confirmed in this study (Figs. 2, 6, 7). And, compared with K150, the Pn significantly increased under W9000 at BSS (16.1%~21.6%) for both years and at BOS in 2013 (24.3%), indicating that straw-returning can increase C assimilation capacity at the middle and later stages of cotton development. SPS and SuSy as the two key enzymes controlling sucrose biosynthesis (Hendrix and Huber 1986) were much higher under W9000 than K150 at BSS and BOS (except for SPS at BOS in 2012 and SuSy at BSS in 2013), indicating that straw-returning was more conducive to sucrose production at the middle and later stages of cotton development. Acid and alkaline invertases are the main enzymes that catalyze the decomposition of sucrose in leaves (Loka et al. 2020). In the current study, alkaline invertase did not differ between K150 and W9000, but acid invertase was lower under W9000 at BSS and BOS, which could lead to a decrease in sucrose hydrolysis. Overall, the above results suggested that straw-returning would favor a higher sucrose accumulation than inorganic K application at the middle and later stages of cotton development. However, sucrose content was markedly lower under W9000 than K150. Wang et al. (2012) and Hu et al. (2017) have reported that high leaf K concentration was beneficial to increasing the sucrose transfer rate in phloem. Thus, the higher leaf K content under W9000 than K150 at BSS suggested that the lower leaf sucrose under the straw-returning treatment than inorganic K fertilizer treatment was associated with the different assimilate transport capacity. Also, an inhibited sucrose hydrolysis due to lower invertase activity would reduce the conversion of sucrose to other sugars, such as hexose and starch (Loka and Oosterhuis 2016). Thus, lower contents of soluble sugars and starch were measured under W9000 relative to K150.

Conclusion

Although the reproductive organ biomass did not differ between straw-returning and inorganic K fertilizer treatments, straw-returning better increased the RRT. Inconsistent eigenvalues of reproductive growth (VT, Vm, T, DAS1, DAS2 and DASm) were observed between straw-returning and inorganic K fertilizer treatments, and the larger T was the main reason for the larger allocation of biomass to reproductive organs in straw-returning treatment than inorganic K fertilizer treatment. Straw-returning altered the dynamics of leaf K concentration with the growth period, resulting in a more drastic effect on C metabolism in relation to inorganic K application.
Consequently, lower soluble sugar/free amino acid and C/N ratios at BSS and BOS were measured in straw-returning treatment than inorganic K fertilizer treatment. Further analysis of C metabolism (Pn, SPS, SuSy and acid invertase activities) indicated that straw-returning was more conducive to sucrose accumulation than inorganic K fertilizer at the middle and later stages. However, sucrose content was markedly lower in straw-returning treatment than inorganic K application treatment at BSS and BOS. This might be because straw-returning led to higher assimilate transport capacity than inorganic K fertilizer, which also could explain the larger RRT in straw-returning treatment than inorganic K fertilizer treatment. Also, compared with inorganic K fertilizer, straw-returning inhibited the conversion of sucrose to other sugars, resulting in lower contents of soluble sugars and starch.

**Abbreviations**

RRT: Ratio of reproductive organ biomass to total biomass; Bio<sub>T</sub>: Theoretical maximum biomass of reproductive organs; DAS: Days after sowing; DAS<sub>i</sub>: Initiation DAS of rapid-accumulation period of reproductive organ biomass; DAS<sub>T</sub>: Termination DAS of rapid-accumulation period of reproductive organ biomass; T: Duration of rapid-accumulation period of reproductive organ biomass; V<sub>i</sub>: Average accumulation rate of reproductive organ biomass; V<sub>i</sub>: Maximum accumulation rate of reproductive organ biomass; T<sub>RRT</sub>: Occurrence time of V<sub>i</sub>; PFS: Peak flowering stage; BSS: Boll-setting stage; BOS: Boll-opening stage; SPS: Sucrose phosphate synthase; Pr: Net photosynthetic rate; C/N ratio: Carbon/nitrogen ratio; SuSy: Sucrose synthase.

**Supplementary Information**

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**Authors’ contributions**

Conceived, designed and performed the experiments: Hu W, Yu CR, Zhao WQ, and Zhou ZG. Analyzed the data: Hu W and Yu CR. Contributed reagents/materials/analysis tools: Yang CQ, Liu RX and Zhou ZG. Wrote the paper: Hu W and Yu CR. All authors read and approved the final manuscript.

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**Availability of data and materials**

All relevant data are within this article.

**Declarations**

**Ethics approval and consent to participate**

Not applicable.

**Consent for publication**

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**Competing interests**

The authors have declared that no competing interests exist.

**Author details**

1College of Agriculture, Nanjing Agricultural University, Nanjing 210095, China.

2Vegetable Research Institute, Guangdong Academy of Agricultural Sciences, Guangdong 510640, China.

3Institute of Industrial Crops, Jiangsu Academy of Agricultural Sciences, Nanjing 210014, China.

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**References**

Almaz MG, Halim RA, Yusoff MM, et al. Effect of incorporation of crop residue and inorganic fertilizer on yield and grain quality of maize. Indian J Agric Res. 2017;51:574–9. https://doi.org/10.18805/IJARE-A-264.

Bange MP, Milroy SP. Growth and dry matter partitioning of diverse cotton genotypes. Field Crop Res. 2004;87:73–87. https://doi.org/10.1016/j.fcr.2003.09.007.

Chen J, Guo Z, Chen H, et al. Effects of different potassium fertilizer types and dosages on cotton yield, soil available potassium and leaf photosynthesis. Arch Agron Soil Sci. 2021;67:275–87. https://doi.org/10.1080/03650340.2020.1723005.

Dong H, Kong X, Li W, et al. Effects of plant density and nitrogen and potassium fertilization on cotton yield and uptake of major nutrients in two fields with varying fertility. Field Crop Res. 2010;119:106–13. https://doi.org/10.1016/j.fcr.2010.06.019.

Drosdoff M, Sell HM, Gilbert SG. Some effects of potassium deficiency on the nitrogen metabolism and oil synthesis in the tung tree (Aleurites fordii). Plant Physiol. 1947:22:538. https://doi.org/10.1104/pp.22.4.538.

Du X, Chen B, Meng Y, et al. Effect of cropping system on cotton biomass accumulation and yield formation in double-cropped wheat-cotton. Int J Plant Prod. 2016;10:29–44. https://doi.org/10.22069/IJPUP.2016.2551.

Fan XH, Li YC. Effects of slow-release fertilizers on tomato growth and nitrogen leaching. Commun Soil Sci Plan. 2009;40:3452–68. https://doi.org/10.1080/00103620903326016.

Hendrix DL. Rapid extraction and analysis of nonstructural carbohydrates in plant tissues. Crop Sci. 1993;33:1306–17. https://doi.org/10.2135/crops ci1993.001833390033000600377.

Hendrix DL, Huber SC. Diurnal fluctuations in cotton leaf carbon export, carbohydrate content, and sucrose synthesizing enzymes. Plant Physiol. 1986;81:584–6. https://doi.org/10.1104/pp.81.2.584.

Hu W, Yang J, Meng Y, et al. Potassium application affects carbohydrate metabolism in the leaf subtending the cotton (Gossypium hirsutum L.) boll and its relationship with boll biomass. Field Crop Res. 2015;179:120–31. https://doi.org/10.1016/j.fcr.2015.04.017

Hu W, Jiang N, Yang J, et al. Potassium (K) supply affects K accumulation and photosynthetic physiology in two cotton (Gossypium hirsutum L.) cultivars with different K sensitivities. Field Crop Res. 2016a;196:51–63. https://doi.org/10.1016/j.fcr.2016.06.005.

Hu W, Zhao W, Yang J, et al. Relationship between potassium fertilization and nitrogen metabolism in the leaf subtending the cotton (Gossypium hirsutum L.) boll during the boll development stage. Plant Physiol Bioch. 2016b;101:113–23. https://doi.org/10.1111/plphys.2016.01.019.

Hu W, Coomer TD, Loka DA, et al. Potassium deficiency affects the carbon-nitrogen balance in cotton leaves. Plant Physiol Biochem. 2017;115:408–17. https://doi.org/10.1016/j.plaphy.2017.04.005.

Hu W, Liu Y, Loka DA, et al. Drought limits pollen tube growth rate by altering carbohydrate metabolism in cotton (Gossypium hirsutum) pistils. Plant Sci. 2019a;286:108–17. https://doi.org/10.1016/j.plantsci.2019.06.003.

Hu W, Sui N, Yu C, et al. Comparative effects of crop residue incorporation and inorganic potassium fertilization on soil C and N characteristics and microbial activities in cotton field. J Cotton Res. 2019b;2:24. https://doi.org/10.1186/s42397-019-0040-3.
