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Nitrogen availability determines the vertical patterns of accumulation, partitioning, and reallocation of dry matter and nitrogen in maize

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ABSTRACT

Context or problem: The reallocation of dry matter (DM) and nitrogen (N) from vegetative tissues to the grains are critical for both yield quantity and protein content of cereal crops. However, it is unclear to which extent the dynamics in DM and N reallocation depend on N availability, and how individual leaves within the maize canopy respond to different N availabilities in terms of these processes.
Objective or research question: This study aimed to quantify DM and N accumulation, partitioning, and reallocation from vegetative to reproductive parts in maize in relation to soil N availability.
Methods: A long-term N fertilizer trial was conducted in Jilin province, Northeast China, growing maize at three N fertilizer levels (low N availability, N0; intermediate N availability, N1; and high N availability, N2). The accumulation, partitioning, and reallocation of DM and N were quantified at the whole-plant, organs, and single-leaf scales in 2015 and 2016.
Results: Although both post-silking DM accumulation and post-silking N uptake increased in response to higher N availability, 8.3–38.8% of grain N still needed to be reallocated from vegetative organs with a larger fraction coming from leaves (10.5 – 36.5%) than from stems (4.4 – 11.6%). This dependency of grain N on N reallocation from vegetative parts increased at lower soil N availability. Furthermore, the vertical patterns of reallocated leaf DM and reallocated leaf N changed with N availabilities. While in general leaves in the middle part of the canopy tended to reallocate more DM and N to the grains than leaves from the upper or lower canopy parts, these most-contributing leaves were higher in the canopy at low than at high N availability.
Conclusions and implications: Grain N relies on reallocation more than does grain DM. At lower N availability, more grain DM and grain N need to be reallocated from vegetative organs. Furthermore, the leaf ranks from which most DM and N were reallocated, gradually increased from the bottom to upper ranks as N availability decreased. Together, our results on the dynamics of N and DM uptake and reallocation during the reproductive phase are important for plant and crop models that require these processes for accurate predictions of maize performance.

1. Introduction

Yields in cereal crops are determined both by dry matter (DM) accumulation through photosynthesis and by the reallocation of photosynthates from vegetative organs to grain during the reproductive stage (Olmedo Pico and Vyn, 2021; Yang et al., 2021). Meanwhile, maize grain quality is associated with grain protein content, which is closely linked to grain nitrogen (N) content (Cliquet et al., 1990; Kumar et al., 2019). Grain N content in turn is the sum of N uptake from soil and N reallocation during the reproductive phase. Hence, understanding the accumulation, partition, and reallocation processes of DM and N is essential to meet the grain demand for both high yield and high quality.

Grain DM production is contributed by leaf area, photosynthesis duration and photosynthetic rates per unit leaf area (Li et al., 2022; Wang et al., 2018). Many studies pointed out that increasing the total DM accumulation or the proportion of post-silking DM is vital for achieving higher grain DM since more than half of the total aboveground DM was accumulated through photosynthesis at the post-silking stage

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(Cliquet et al., 1990; Liu et al., 2022; Wei et al., 2019). Grain DM may come from either current assimilation allocated directly to grains or from the reallocation of assimilates derived from pre-silking photosynthesis that were temporarily stored in vegetative organs (Liu et al., 2017; Molla et al., 2019). However, the relative contributions of photo-assimilations and reallocated DM to grain may differ between maize genotype and depends on soil N availability (Chen et al., 2015; Evans, 1989). For instance, compared to older cultivars, modern stay-green cultivars typically show higher post-silking DM accumulation, with much less DM reallocation from vegetative parts to grains (Liu et al., 2017; Zhang et al., 2022). In addition, at high N availability leaf senescence is typically postponed allowing plants to maintain high LAI longer, and thus to increase post-silking canopy photosynthesis and post-silking DM accumulation (Olmedo Pico and Vyn, 2021; Zhang et al., 2010; Zhao et al., 2019).

Grain N in turn is derived from either post-silking root N uptake or from reallocation from vegetative organs (Molla et al., 2019; Wang et al., 2018). For a given N content, there is a trade-off between leaf N reallocation and leaf photosynthesis rate during post-silking stages, because as more N is reallocated from leaves to grains, less N remains in leaves to support photosynthesis (Brown et al., 2019; Liu et al., 2022; Ning et al., 2017). Although higher N reallocation is important to support grain growth, it may concomitantly result in lower leaf N content which in turn speeds up leaf senescence, leading to reduced leaf N contents, green leaf area and canopy photosynthesis (Evans and Clarke, 2019; Hikosaka, 2016). This trade-off is mediated by the amount of N absorbed from the soil and hence by N fertilizer application. Higher N availability entails that more N needed for grain growth can be taken up from the soil reducing the need for N reallocation such that more N remains in leaves and leaf longevity is prolonged (Boomsma et al., 2009; Li et al., 2022).

Previous studies mostly focused on N distribution, and reallocation during the grain-filling stage at the whole plant level (Chen et al., 2019; Liu et al., 2022). However, it is well known that leaf N distribution is uneven among leaves; the upper leaves generally have higher N contents and specific leaf N than the lower leaves (Archontoulis et al., 2011; Bertheloot et al., 2008; Hirose and Werger, 1987). This entails that more N could be reallocated from upper leaves during the grain-filling stage. However, the upper leaves receiving more light generally have higher photosynthetic rates and nitrogen-use efficiency and thus contribute more to canopy photosynthesis than do lower shaded leaves (Anten et al., 1995; Hikosaka, 2016; Li et al., 2019). Therefore, plants tend to reallocate relatively more N from lower leaves partly through shade-induced senescence (Archontoulis et al., 2011; Liu et al., 2022). The vertical pattern of N reallocation and its effects on post-silking canopy photosynthesis is also affected by N availability from the soil, due to the fact that N availability affects leaf area and thus canopy shading, leaf N content as well as the amount of grain N that can directly be derived from the soil (Li et al., 2022; Perez et al., 2019).

The accumulation, partitioning, and reallocation of dry matter (DM) and nitrogen (N) affect the metabolic processes of the crop and are intricately linked to plant physiological events (Fournier and Andrieu, 1999; Zhao et al., 2019). Although aboveground DM or N accumulation, partitioning, and reallocation have been well documented in maize plants (Liu et al., 2022; Molla et al., 2019), little information is available about the vertical distribution and reallocation patterns of DM or N across leaf ranks and the extent to which this distribution is modified by changes in N availability. In this study the objectives were to: (1) quantify the DM and N accumulation, partitioning, and reallocation at the whole plant and organ scales during the reproductive phase, and (2) improve our understanding of the dynamic responses of vertical DM and N reallocation within the maize canopy to different N availabilities. These questions were addressed in a long-term field N fertilizer trial with maize.

2. Materials and methods

2.1. Experimental Site

A long-term field experiment with maize grown under different nitrogen (urea) fertilizer levels was started in 2009 at the Gongzhuling Experimental Station of the Chinese Academy of Agricultural Sciences (43°53 N, 124°81E) in Jilin province, Northeast China. The meteorological data for 2015 and 2016 at the experimental site were recorded daily during the maize growing season (from 1 May to 30 September). The average daily temperature varied from − 1.5–28.0 °C in 2015 and from 1.1 °C to 28.6 °C in 2016 respectively (Fig. 1). The total precipitation was 433.6 mm in 2015 and 652.0 mm in 2016.

2.2. Experimental design

The data in this study was collected in 2015 and 2016, which were the year seven and eight in this long-term trial. Soil samples from the 0–20 cm soil layers were taken before fertilizer application during each growing season at the beginning of the field experiments (Table 1). The total N amount and application stage of the three N fertilizer management practices were as follows: no N fertilizer was applied throughout the whole growth period (N0), 150 kg N ha^{-1} was all applied as base fertilizer before sowing (N1), and 150 kg N ha^{-1} was applied as described for the N1 treatment, and an additional 150 kg N ha^{-1} splits equally at V6 and silking stages (N2). The chemical fertilizer phosphorus (superphosphate) and potassium (potassium chloride) were applied before sowing at a rate of 42.5 kg P_2O_5 ha/year and 42.5 kg K_2O ha/year to all treatments. As the N treatments had already been maintained for 6–7 years before that start of the experiment, soil N at the start of the experimental years was already different and these differences were increased by current-year N fertilizer application (Table 1).

The experiments used a random block design with three replications each. Individual plots were 45.5 m^2, and each contained seven rows of maize, 10 m long with 0.65 m distance between rows. The seeds of widely grown maize hybrid XY335 were sown by hand at 6.75 plants/m^2 on 1 May in 2015 and 29 April in the year 2016. All weeds, diseases, and pests were controlled.

2.3. Plant sampling

Adjacent plants from the same inner row were tagged by applying a dot of red paint to their leaves at the V3 stage. Leaf rank was counted from the bottom (leaf 1) to the top, and tags were placed on leaves 4, 8, and 12 to avoid confusion as lower leaves senesced (Fan et al., 2020). Maize plants were randomly selected among tagged plants, and the whole plant shoots were cut down from the soil surface at silking (R1) and at physiological maturity (R6). Plant samples were separated into individual leaves (leaf blades), stem (including leaf sheath), grain, cob, bracts, and tassels. The dry weight of each sample was determined after drying for 72 h at 85 °C in a forced-air drying oven. The samples were then weighed, ground into a fine powder, and analyzed the N concentration based on the Kjeldahl method (Bremner, 1960). The post-silking DM accumulation and N uptake were calculated at the whole-plant level, and the DM and N allocation to different organs was quantified. The absolute reallocated amounts and the fractions of the reallocated leaf DM and leaf N, and the contributions of both to grain DM and grain N were calculated at organ scale (leaf or stem sections) and individual leaf scale (individual leaf rank), respectively.

2.4. Measurements of accumulation, partitioning, and reallocation processes of dry matter and nitrogen

2.4.1. Whole-plant dry matter and N accumulation during post-silking stage

The post-silking DM accumulation Eq. (1) and N uptake Eq. (2) were calculated as:

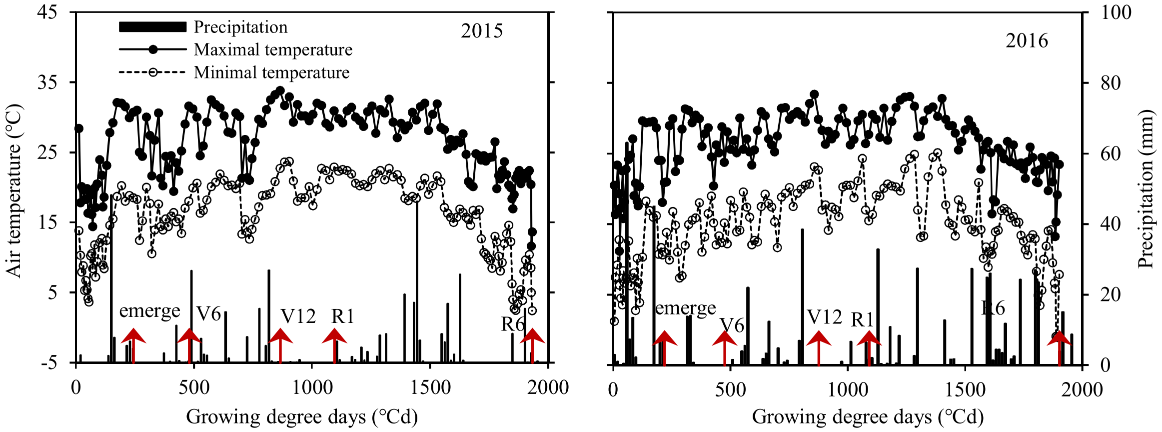

Fig. 1. The mean daily maximum and minimum temperature, and daily precipitation during experimental years. Red arrows indicate key growth stages in maize.

Table 1
The soil N content (g/kg) before sowing, the N fertilizer applications, and the different N availabilities in this study.

Year	Treatments	Soil N content (g/kg)	N fertilizer application (kg/ha)	N availability (g/kg)
2015	N0	1.09 c	0	1.09 c
	N1	1.25 b	150	1.29 b
	N2	1.40 a	300	1.49 a
2016	N0	1.08 c	0	1.08 c
	N1	1.29 b	150	1.33 b
	N2	1.46 a	300	1.55 a

Note: Different letters in the same column indicate significant differences at $P < 0.05$ ($n = 10$). The soil bulk density was $1.68/\text{cm}^3$ in this study, and the nutrient availability of P and K were applicated the same for all N management treatments.

$$\text{Post-silking DM (g/plant)} = \text{whole plant DM}_{R6}\text{-whole plant DM}_{R1} \qquad (1)$$

$$\text{Post-silking N uptake (g/plant)} = \text{whole plant N}_{R6}\text{-whole plant N}_{R1}(2)$$

where DM_{R1} and DM_{R6} represent the whole-plant DM in R1 (silking) and R6 (maturity) stages, and N_{R1} and N_{R6} represent the whole-plant N content in the R1 and R6 stages.

2.4.2. Harvest index

Harvest index (HI) and N harvest index (NHI) were calculated as follows:

$$\text{HI=Grain DM/ whole-plant DM}_{R6} \qquad (3)$$

$$\text{NHI=Grain N /whole-plant N}_{R6} \qquad (4)$$

where Grain DM and Grain N are the grain dry matter (g plant^{-1}) and grain N (g plant^{-1}) content at the R6 stage, respectively.

2.4.3. DM and N partitioning

$$\text{DM partitioning (\%)=DM of specific vegetative organ /whole-plant DM} \times 100\% \qquad (5)$$

$$\text{N partitioning (\%) =N of specific vegetative organ /whole-plant N} \times 100\%(6)$$

DM partitioning and N partitioning were defined as the fractions of DM or N of each organ to whole-plant DM or N. Where the specific vegetative organs represent of leaf, stem, cob, brace, tassel, and grain, respectively.

2.4.4. The reallocated amount and reallocation fraction of DM and N

$$\text{Reallocated DM amount (g)= DM}_{R1}\text{-DM}_{R6} \qquad (7)$$

$$\text{Reallocated N amount (g)= N}_{R1}\text{-N}_{R6} \qquad (8)$$

$$\text{DM reallocation fraction (\%) =Reallocated DM amount /DM}_{R1} \qquad (9)$$

$$\text{N reallocation fraction (\%)=Reallocated N amount /N}_{R1} \qquad (10)$$

We calculated the reallocated amount and fraction of DM and N at organ level (leaf or stem) and leaf level (individual leaf rank) in this study, respectively. When calculating at the organ level, the DM_{R1}, N_{R1}, DM_{R6}, and N_{R6} represent the dry matter or N contents of the leaf or stem fractions of the plant at the R1 and R6 stages, respectively. When calculated at leaf level for individual leaves, the DM_{R1}, N_{R1}, DM_{R6}, and N_{R6} represent the dry matter or N contents of individual leaves at each leaf rank at the R1 and R6 stages, respectively.

2.4.5. Contribution of DM and N reallocation to grain

$$\text{Contribution of reallocated DM to grain DM (\%)=Reallocated DM amount/ Grain DM} \qquad (11)$$

$$\text{Contribution of reallocated N to grain N (\%)=Reallocated N amount/Grain N} \quad (12)$$

Where DM_{R1} and DM_{R6} represent the dry matter of each leaf rank at the R1 and R6 stages. Grain DM represents the dry matter of grain at the R6 stage. N_{R1} and N_{R6} represent the N content of each leaf rank at the R1 and R6 stages, and Grain N represents the N content of grain at the R6 stage.

2.5. Statistical analysis

Statistical analysis was performed using R software (version 3.6.1). One-way analysis (ANOVA) was used to evaluate the effect of N availability on the accumulation, partitioning, and reallocation of dry matter and N content. Differences were compared using the least significant difference test at a 0.05 level of probability. Due to substantial year-by-treatment interactions for most of the assessed variables, the treatment effect was evaluated separately for each year.

3. Results

3.1. Whole-plant dry matter and N content accumulation

The whole-plant DM and N increased with N availability in both years regardless of developmental stage, and the effect of N availability on whole-plant N content was stronger than the effects on whole-plant DM (Table 2). The post-silking DM increase was slightly larger than the grain DM, in all cases including the N0 treatment. This indicates that net assimilation during the reproductive phase was consistently more than sufficient to meet DM demand for grain filling. However, the N uptake from the soil during the post-silking stage was not sufficient for the amount of N in grains, as indicated by post N/grain N values being $< 100\%$. This fraction increased with the level of fertilizer. This indicates that part of the N in grains must have been made available through reallocation from vegetative organs to grain and that this reallocation requirement was larger at low N availability (Table 2).

3.2. Fraction of DM and N content in different organs

At the R1 stage, the largest fraction of DM was in the stem (60%) followed by leaves (30%) and the remaining organs (cob, bract, and tassel) together accounting for about 10%. This DM distribution pattern was not significantly affected by N fertilizer treatments. At the R6 stage, maize plants tended to allocate less DM to grains and more to vegetative parts at low than at high N availability. For instance, the proportions of grain DM to whole-plant DM decreased from 0.49 in the N2 treatment to 0.42 in the N0 treatment (Fig. 2a). The distribution pattern of N was different. At the R1 stage, around 50% of N was in leaves and 40% in the stem, without a significant effect of N availabilities. At the R6 stage, about two-thirds of the total N was in the grains, with a nitrogen harvest index (NHI) of 0.61–0.67. Increased N availability decreased N allocation to the grains, but caused a considerable increase in the N allocation to other organs (Fig. 2b). Overall, the soil N availability did not affect partitioning of DM and N among organs at the R1 stage. However, at the R6 stage, more DM was allocated to leaf and stem, and more N was allocated to grain in response to low N availability.

3.3. Reallocation of DM and N in leaf versus stem organs

Overall plants reallocated relatively more N than DM from leaves to grains and grain N hence also depended relatively more on this reallocation than gain DM (Table 3). More specifically, leaves reallocated up to 60% of their N, but only 15% of their DM. The reallocated N from both leaf and stem contributed 10.5–35.6% to grain N, but the reallocated DM only account for 1.6–10.9% to grain DM. Furthermore, the amount of N reallocation from leaves was larger than that from stem, and this was consistent across all treatments. N availability significantly influenced DM and N reallocation, reallocation fractions, as well as the contribution of reallocation to grain DM and N (Table 3). Notably, in absolute terms, the highest DM and N reallocation amounts were obtained under the intermediate N1 treatment. The reallocation fractions of reallocated DM or N and their contributions to grain DM or grain N however were greater as N availability became less. This indicates that in terms of relative contributions, the accumulation of DM and N in grains becomes increasingly dependent on DM and N reallocation from vegetative parts when external N availability goes down.

3.4. Distribution and reallocation of DM and N from individual leaves

Leaf DM and leaf N content (i.e., the total amounts of DM or N in a leaf) of individual leaves increased with leaf rank up to the ear leaf (rank 14) and then decreased towards the top leaf (Fig. 3). The maximum leaf DM was around 6 g/leaf at the R1 stage, whereas the maximum leaf N content was 0.15 g N/leaf. N deficiency significantly reduced DM and N content particularly for the leaves closer to the ear. The reduction in leaf N content with decreasing N availability was much greater than the reduction in leaf DM (Fig. 3). Furthermore, the differences of leaf N among N availabilities became greater at the R6 stage.

The amount of reallocated DM from individual leaves increased with leaf rank up to the ear leaf and then decreased towards the top leaf (Fig. 4a). The negative value of the reallocated DM in the upper leaves indicates there was net DM accumulation rather than reallocation between the R1 and R6 stages in these leaves (Fig. 4a). By contrast, the reallocation of leaf N content was largely from middle and upper leaves within the canopy, especially near the ear leaf. This indicates that the leaf N reallocation was largely from living leaves via re-translocation of N reserves and less from senescing leaves (Fig. 4b). The reallocation of DM and N was affected by N availability, with the highest amount of DM and N reallocation occurring in the N1 treatment. The maximum reallocation values of leaf DM and leaf N content were 0.9 g and 0.05 g, respectively (Fig. 4). The general pattern of N accumulation across leaf ranks was an initial increase with leaf rank at low leaf ranks and a decline at higher ones, with the highest values being at somewhat intermediate leaf ranks. But this maximum shifted upwards with decreasing N availability, indicating that as plants are more N limited, N reallocation occurs progressively more from upper leaves (Fig. 4b).

The lower the N availability, the higher the reallocation fraction (the ratio of reallocated DM or N per leaf to whole leaf DM or leaf N) of leaf DM or leaf N. For instance, the average value of reallocated leaf DM fractions during the two experimental years were 5%, 17%, and 16% (N2, N1, and N0), and the reallocated leaf N fractions were 23%, 46%, and 53% (Fig. 5). Individual leaves reallocated up to 60% of their own N but only up to 30% of their DM. The reallocation fractions of leaf DM and leaf N differed among leaf ranks and these distribution patterns were affected by N availability (Fig. 5). The lowest-ranked leaves at the bottom of the canopy exhibited the highest reallocated leaf DM fraction, and the decline in fractions with leaf rank was strongest in the N2 treatment (i.e., where values became negative at the top) (Fig. 5a). The reallocated leaf N fraction under N0 and N1 treatments was much bigger than that in N2 treatments, but the range difference among leaf ranks within the maize canopy was much smaller compared with N2 treatments (Fig. 5b).

Table 2

Whole-plant dry matter (DM) and N content (N) at silking (R1) and maturity (R6), harvest index (HI), nitrogen harvest index (NHI), dry matter accumulation during post-silking (Post DM), and N accumulation during post-silking (Post N) in 2015 and 2016.

		Dry matter per plant (DM)						N content per plant (N)					
Year	N availability	DM_{R1} (g)	DM_{R6} (g)	Post DM (g)	Grain DM (g)	HI	Post DM/ grain DM (%)	N_{R1} (g)	N_{R6} (g)	Post N (g)	Grain N (g)	NHI	Post N/grain N (%)
2015	N0	117.0 c	230.1 c	113.1 c	98.3 c	0.43 b	114.9 a	0.97 c	1.77 c	0.80 c	1.18 c	0.67 a	68.1 c
	N1	151.1 b	322.9 b	171.8 b	159.6 b	0.49 a	107.7 b	2.17 b	4.43 b	2.26 b	2.91 b	0.66 a	77.7 b
	N2	166.6 a	381.0 a	214.3 a	183.6 a	0.48 a	116.7 a	2.65 a	5.91 a	3.26 a	3.60 a	0.61 b	90.6 a
2016	N0	119.4 c	217.3 c	97.9 c	90.8 c	0.42 b	107.8 a	0.83 c	1.56 c	0.73 c	1.04 c	0.67 a	70.6 b
	N1	161.4 b	341.9 b	180.5 b	164.5 b	0.48 a	109.7 a	2.12 b	4.29 b	2.16 b	2.77 b	0.65 b	78.1 ab
	N2	175.2 a	384.7 a	209.5 a	187.0 a	0.49 a	112.0 a	2.68 a	6.10 a	3.42 a	3.73 a	0.61 c	91.7 a

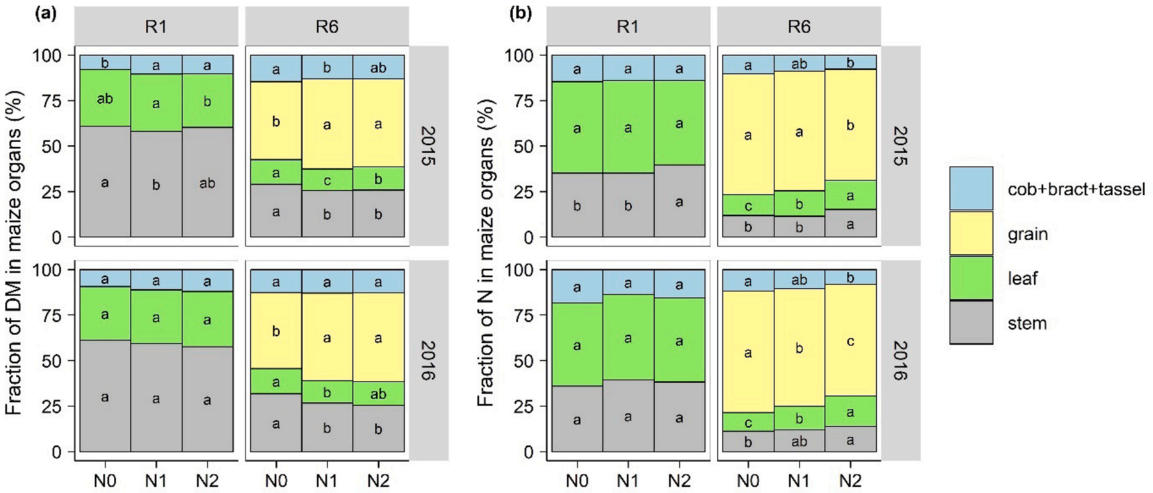

Fig. 2. The proportion of DM (a) and N (b) in different plant organs to total DM and total N of the whole plant under different N availabilities at the R1 and R6 stages in 2015 and 2016. Different letters indicate significant ($P < 0.05$) difference between N treatments.

Table 3
The reallocation amount of DM and N in leaf and stem, reallocation fraction (%) of leaf and stem is the percentage of reallocated DM or N amount to total DM or N at the R1 stage, and the contribution of DM and N reallocation in leaf and stem to grain DM and grain N in 2015 and 2016.

		DM reallocation in leaf versus stem						N reallocation in leaf versus stem					
Year	N availability	amount (g)		fraction (%)		Contribution to grain DM (%)		amount (g)		fraction (%)		Contribution to grain N (%)	
		Leaf	Stem	Leaf	Stem	Leaf	Stem	Leaf	Stem	Leaf	Stem	Leaf	Stem
2015	N0	5.5 b	5.2 a	15.0 a	7.2 a	5.6 a	5.3 a	0.3 b	0.1 b	59.1 a	38.1 a	24.5 a	11.1 a
	N1	8.8 a	5.3 a	18.7 a	6.0 a	5.5 a	3.3 ab	0.5 a	0.3 a	43.7 b	33.4 a	16.5 b	8.8 a
	N2	0.8 c	2.0 b	1.7 b	2.0 b	0.5 b	1.1 b	0.3 b	0.2 ab	22.1 c	15.0 b	7.6 c	4.4 b
2016	N0	5.2 ab	4.0 a	14.8 a	5.4 a	5.8 a	4.4 a	0.2 b	0.1 b	57.5 a	40.6 a	20.9 a	11.6 a
	N1	6.1 a	4.3 a	12.7 a	4.5 a	3.7 b	2.6 a	0.4 a	0.3 a	44.6 a	38.2 a	16.0 ab	11.6 a
	N2	2.7 b	3.2 a	5.0 b	3.2 a	1.4 c	1.7 a	0.2 b	0.2 b	17.4 b	17.1 b	5.8 b	4.7 b

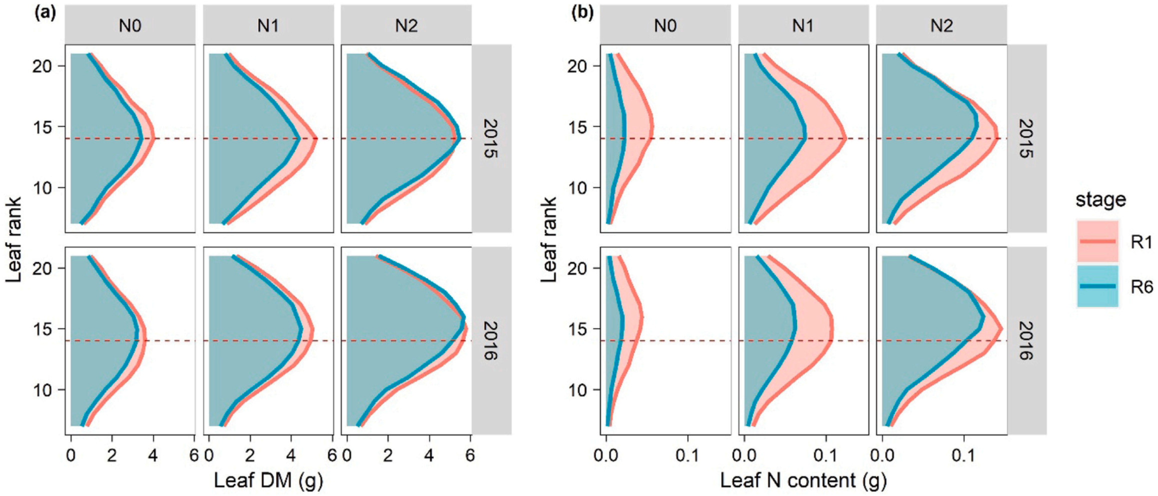

Fig. 3. The vertical distribution patterns of leaf DM (a) and leaf N content (b) at each leaf rank under different N availabilities during the R1 and R6 stages in 2015 and 2016. The dashed red line represents the rank of the ear leaf within the maize canopy.

3.5. *The contribution of DM and N reallocation from individual leaves to grain DM and N*

The DM and N reallocated from individual leaves under lower N availabilities contributed more to grain DM and grain N than that under high N availability (Fig. 6). For instance, the average contributions of total leaf DM reallocation during the two experimental years under N0, N1, and N2 treatments were 5.7%, 4.6%, and 0.9%, respectively (Fig. 6a). Grain N depended more on the reallocation from vegetative organs compared with grain DM. The leaf N reallocations in 2015 and 2016 under N0, N1, and N2 treatments averaged to account for 22.7%, 16.2%, and 6.7%, respectively (Fig. 6b). Furthermore, the vertical distribution of individual leaf contributions of DM and N was affected by N availability, shifting from it coming more from bottom leaves at N2 treatment via middle leaves at N1 to top leaves at N0 (Fig. 6).

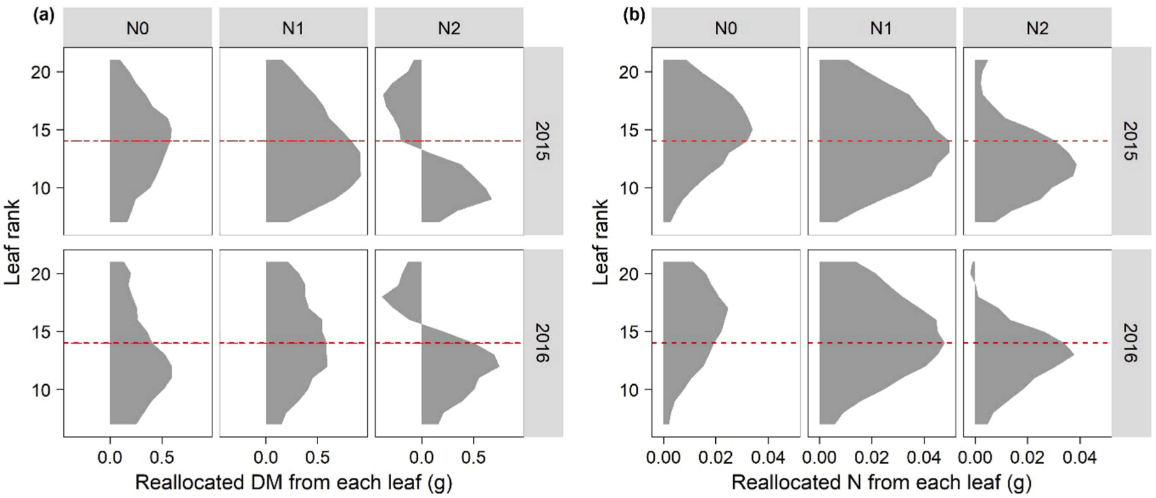

Fig. 4. The amount of leaf DM (a) and leaf N (b) reallocated from individual leaves under different N availabilities. The dashed red line represents the rank of the ear leaf within the maize canopy. Negative values (< 0) indicate that on-balance leaves had accumulated DM or N between the R1 and R6 stages.

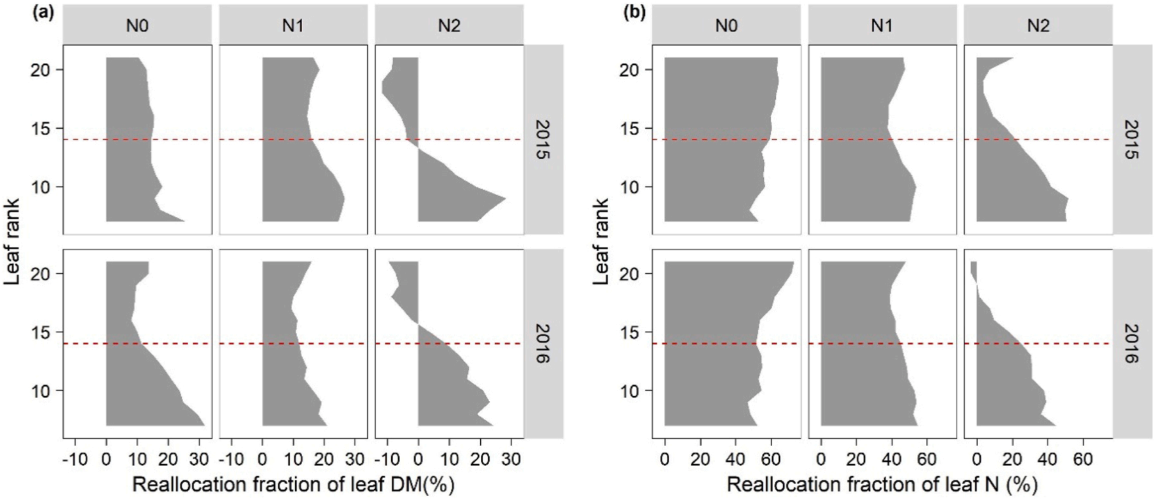

Fig. 5. The reallocation fraction of leaf DM (%) and leaf N content (%) (i.e., the net percentage of DM and N in leaves that were retranslated) at individual leaves under different N availabilities in 2015 and 2016. The dot red line represents the rank of the ear leaf within the maize canopy.

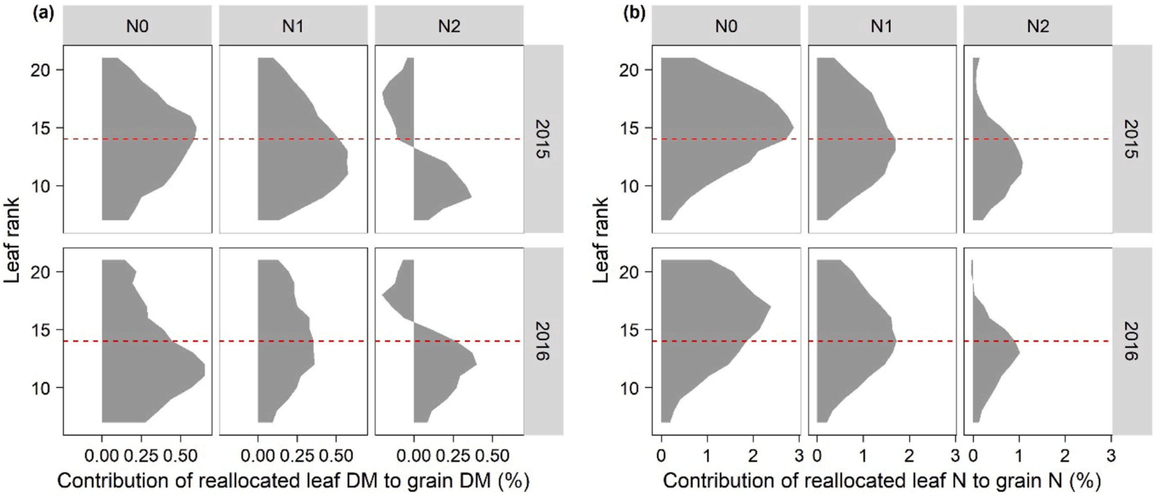

Fig. 6. The contribution (%) of reallocated leaf DM and leaf N to grain DM or grain N at individual leaves under different N availabilities in 2015 and 2016. The dashed red line represents the rank of the ear leaf within the maize canopy.

4. Discussion

4.1. Grain N relies on reallocation more than grain DM

A high post-silking DM accumulation is vital for achieving high maize grain yields, since the majority of the grain DM was found to come from the photosynthates produced during the post-silking stage (Liu et al., 2023; Ning et al., 2013; Parco et al., 2022). In this study, the post-silking DM accounted for 107.7–116.7% of the grain DM regardless of the N availability (Table 2), which was probably because the accumulated post-silking DM derived by canopy photosynthesis provides sufficient assimilates for grain DM (Koutroubas et al., 2012; Liu et al., 2023). The situation was different for nitrogen, where 8.3%−38.8% of grain N was reallocated from vegetative organs during the post-silking stage, and this dependency on N reallocation increased with decreasing N availability (Table 2). Since N uptake depends on the continuous carbohydrate supply from shoot to roots, longer duration of post-silking photosynthesis is beneficial for post-silking N uptake of the maize plants, which in turn increases canopy photosynthesis duration and final grain DM (Gallais et al., 2007; Liu et al., 2023; Winterhalter et al., 2012). The delayed leaf senescence, larger leaf area, and longer photosynthesis duration under high N availability contribute to improving canopy photosynthesis capacity and ultimately, greater post-silking N uptake (Liu et al., 2022; Riha et al., 2013). However, as we found, increasing N availability may simultaneously decrease the N reallocation fraction of the N accumulated at the pre-silking stage (Table 3), and therefore increase the amount of residual N in vegetative organs, since less reallocated N is needed to meet grain N requirements with increasing N availability (Chen et al., 2015; Ning et al., 2013).

4.2. Trade-off between post-silking photosynthesis and N reallocation in leaves in response to N availability

Leaf and stem are the main organs from which DM or N is reallocated to grains, when the accumulated post-silking DM or post-silking N uptake cannot meet the grain requirements (Chen et al., 2015; Liu et al., 2021). The largest amount of DM and N reallocation was at intermediate N availability (Table 3), indicating both N deficiency and very high N availability can restrain the DM and N reallocation from vegetative organs to grain (Wei et al., 2019). On the other hand, the relative contribution of DM or N reallocation from stems and leaves to the total accumulation of DM and N in grains increased with decreasing N availability (Liu et al., 2022; Molla et al., 2019). The amount of N reallocated from leaves was larger than the amount reallocated from stems consistent across all three treatments (Table 3). Since the reallocated leaf N mainly comes from the breakdown of proteins and chlorophyll (Evans and Clarke, 2019; Ma et al., 2022), there is a trade-off between N reallocation and leaf photosynthesis rate. The more N is reallocated from leaf to grain during post-silking stage, the less N will be left to support photosynthesis (Li et al., 2020; Onoda et al., 2017). This trade-off was mediated by N availability (Chen et al., 2015; Ning et al., 2013). A high N availability entails that more N needed for grain growth can be taken up from the soil reducing the need for N reallocation such that more N remains in leaves and leaf longevity is prolonged (Boomsma et al., 2009; Li et al., 2022).

4.2.1. N availability determines the vertical N reallocation patters within maize canopy

Increasing N availability does not necessarily result in a proportional increase in the distribution and reallocation of DM and N from individual leaves (Figs. 3 and 4). It is well-known that the N distribution and photosynthetic rate N are uneven among leaves at different positions in the canopy (Anten et al., 1995; Archontoulis et al., 2011). Leaf DM and leaf N content of individual leaves increased with leaf rank up to the ear leaf and then decreased towards the top leaf, roughly following a bell-shaped function of leaf rank (Fig. 3), which was consistent with previous findings (Archontoulis et al., 2011; Ciganda et al., 2008; Winterhalter et al., 2012). The largest amount of reallocated leaf DM and leaf N came from the middle part of the canopy (Fig. 4). This is mainly because the middle leaves have higher N concentration and bigger leaf area, which entails more DM and N could be reallocated (Fan et al., 2022; Li et al., 2022). However, the bottom leaves reallocated the largest fraction of their DM and N to grains (Fig. 5). Furthermore, the vertical N reallocation pattern across leaves within the canopy was affected by N availability. On the one hand, N reallocation from lower leaves has the smallest opportunity costs in terms of photosynthesis as these leaves are shaded. On the other hand, leaf N contents in upper leaves are higher, hence there being more N available for reallocation. When N availability is limited, maize plants tend to reallocate N to the upper leaves to maximize light interception and photosynthesis, while reducing investment in lower leaves (Fig. 4b). The upper leaves have a higher N concentration and photosynthetic rates than the lower leaves, and this distribution pattern positively affects canopy photosynthesis (Ellsworth and Reich, 1993; Yao et al., 2016). However, when N availability is high, the plants also reallocate more N to lower leaves to increase N uptake and maximize DM production (Fig. 4b). As N availability from the soil declines, N uptake during the reproductive stage supports a smaller fraction of grain N accumulation and thus plants progressively needed to reallocate from upper leaves even if that entailed a relatively large opportunity cost in terms of photosynthesis.

4.3. Implications for N fertilizer management practice and crop models

Increasing grain yield and grain quality, especially protein content, are important for meeting food demand, improving human nutrition, and ensuring a sustainable food system (Duvick, 2005; Ray et al., 2019; Tokatlidis and Koutroubas, 2004). Proper N management can help to ensure that crops receive adequate N for optimal growth and yield, maintain high grain protein content (Hou et al., 2012; Van Oosterom et al., 2001), while preventing losses of N to the environment. In practice, farmers often apply N at excessive rates to ensure a good maize yield (Li et al., 2022; Ning et al., 2013). An N supply in excess of that required for the highest grain DM may increase grain N to an extent (Table 2), but also lead to high levels of residual N in straw at maturity, since less reallocated N from vegetative organs to grain (Table 3). Previous studies also demonstrated that postponed N fertilizer topdressing could enhance yields, nitrogen-use efficiency and effectively avoid the environmental problems caused by excessive fertilization during field production (Xu et al., 2023; Zhai et al., 2022). However, we suggest to pay attention to the soil N availability, and pre-silking N fertilizer application, since a relatively high pre-silking DM accumulation provides a strong base for post-silking growth, which in our study contributed to lower DM reallocation and high grain yield. Moreover, the pre-silking N accumulation in vegetative organs contributed to grain N, which has a positive effect to grain quality.

The current crop models, while rather good at simulating yield amounts (Van Ittersum et al., 2003; Wang et al., 2019) are much less capable of simulating yield quality. This is in part because the physiological processes underlying yield quality are not so well understood and quantified. For instance, as pointed out in this paper, the balance between post-silking N reallocation to support grain N demand and canopy photosynthesis to support assimilate supply is mediated by variation in soil N availability, which is not well described yet in crop models (Van Ittersum et al., 2003; Vos et al., 2010). In this paper, we focused on DM and N reallocation which is an important process driving the balance between post-silking canopy photosynthesis (supporting yield quantity) and N reallocation (mostly, grain protein content). We showed how the balance between N uptake and N reallocation is mediated by N availability, and how this dynamic plays off across different leaves in the canopy providing important data to develop plant models (Barillot et al., 2016; Faverjon et al., 2019). Such models could be used to explore the extent to which vertical DM and N reallocation patterns in maize plant canopies can be optimized in terms of both high grain DM and grain N.

5. Conclusions

Adequate soil N availability is essential to ensure higher DM and N accumulation and higher yield in maize. While post-silking leaf photosynthesis was capable of providing sufficient assimilates for grain DM, 8.3–38.8% of grain N were reallocated from vegetative organs accumulated during pre-silking stage. Moreover, the lower the N availability, the higher the contribution of reallocated N to grain N. The vertical patterns of reallocated leaf DM and leaf N within the maize canopy was mediated by N availability. With decreasing N availability, N reallocation comes increasingly from upper leaves in the canopy and hence comes at a relatively greater cost in terms of photosynthesis. This is important information to improve N management in precision farming and to provide data support for the plant and crop models N treatment effect assessments.

Author Contributions

SL and RX conceived the project and provided funding; PF and BM designed the research; PF and YL performed the experiments; PF analyzed the data and wrote the draft; NA and JBE edited and revised the manuscript.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Data Availability

Data will be made available on request.

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