Field Crops Research 319 (2024) 109650

Contents lists available at ScienceDirect

Field Crops Research

journal homepage: www.elsevier.com/locate/fcr

Exogenous melatonin improves peanut field productivity and quality at reduced nitrogen application

Lijie Li [a,b], Xiangguo Cheng [a], Yi Zhang [a], Dana Kohtz [b], Xiaohui Wang [a], Xiaotian Zhang [a], Xiangjun Kong [a], Huiyun Xue [a], Peipei Jia [a], Na Bai [a], Zengqiang Li [a], Peng Xiao [c], Xiaoping Pan [b], Zhiyong Zhang [a,*], Baohong Zhang [b,*]

[a] *Xinxiang Key Laboratory of Crop Root Biology and Green Efficient Production, School of Life Sciences, Henan Institute of Science and Technology, Xinxiang, Henan 453000, China*
[b] *Department of Biology, East Carolina University, Greenville, NC 27858, USA*
[c] *Department of Mathematics, East Carolina University, Greenville, NC 27858, USA*

ARTICLE INFO

Keywords:
Peanut (*Arachis hypogaea* L.)
Melatonin
Nitrogen fertilizer
Yield
Quality
Carbon and nitrogen accumulation and translocation

ABSTRACT

Context: Nitrogen (N) plays integral roles in plant growth and yield. Finding ways to increase plant yield with reduced N usage will promote both agricultural and environmental sustainability. Melatonin acts as a multifunctional regulatory molecule in numerous metabolic processes crucial for plant growth and development as well as response to environmental stresses. The effects of melatonin on the material accumulation and transport, source-sink dynamics, as well as its association with yield and quality formation of peanut (*Arachis hypogaea* L.) remain unclear, especially at different N levels.
Objectives: We aim to investigate the response mechanism of melatonin in peanut plants subjected to reduced N application, in order to confirm the hypothesis that melatonin regulates carbon and N accumulation and transport, and coordinates source-sink relationships to increase production and improve quality.
Methods: This study examined the effects of two seed dressing treatments (with or without 0.5 μM MT) and three N fertilizer levels (90, 135, and 180 kg/ha) using a randomized complete block design with split plots and three biological replications over 2021 and 2022. The evaluation focused on photosynthetic physiology, enzyme activities related to carbon and N metabolism, accumulation and transport of dry matter and N, yield, and quality, while exploring the relationships among these variables.
Results: Melatonin-treated plants had more stable carbon and N metabolism than the untreated ones. This stability was linked to improved photosynthesis, sucrose production, and N assimilation, especially at the reduced N levels (90 and 135 kg/ha). Across three N levels and two years of field tests, MT increased peanut dry matter by 23.49 % from 455.63 g/m^2 to 562.66 g/m^2, enhanced the accumulation and mobilization of dry matter and N to grains by increasing peanut grain mass by 22.41–29.07 % at different N levels. This process appears to subsequently elevate the effective pod rate, leading to an average increase in pod yield, fat and protein content by 12.63 %, 7.95 %, and 10.33 %, respectively, over a two-year period and across three N application levels.
Conclusions: Plants subjected to melatonin treatment exhibited a coordinated source-sink relationship, which is manifested in high photosynthetic capacity and a high proportion of assimilates transported to pods, thus promoting effective proportions and pod fullness to improve peanut yield and quality under reduced N application.
Significance: Our research provided insights into the response mechanism of melatonin on peanut carbon and N metabolism across various N treatments, contributing to a deeper understanding of how melatonin enhances crop yield and quality.

1. Introduction

Peanut (*Arachis hypogaea* L.) is a globally important oil and food crop. In China, peanut is grown annually on 4.7 × 10^6 ha, accounting for about 20 % of the world's total planting area, ranking second, but its yield ranks first with approximately 40 % of the world's total yields (Shi

---

* Corresponding authors.
E-mail addresses: z_zy123@126.com (Z. Zhang), zhangb@ecu.edu (B. Zhang).

https://doi.org/10.1016/j.fcr.2024.109650
Received 8 July 2024; Received in revised form 27 October 2024; Accepted 30 October 2024

et al., 2022). Combining richness in seed oil (~46–58 %) and protein (~22–32 %), peanut's industrial development helps to fight malnutrition, improve the self-sufficiency rate of edible vegetable oil and ensure food security (Shi et al., 2022; Liu et al., 2024).

As a leguminous crop, peanut has the ability to fix atmospheric nitrogen (N), but its growth still requires sufficient N, phosphorus (P), and potassium (K) nutrients through fertilization (Crusciol et al., 2021). According to a recent field trial study conducted in the North China Plain (NCP), the main peanut producing area in China, peanuts require 42.0 kg of N, 4.6 kg of P, and 15.3 kg of K to produce 1000 kg of pods (Hu et al., 2023). However, farmers in the NCP usually apply excessive mineral fertilizers, especially N fertilizer, which can exceed 190 kg per hectare (Yang et al., 2022a), to ensure higher yields (Feng et al., 2020). Excessive fertilization may be due to the following reasons: Firstly, small-scale farming households, which are the core model of traditional agriculture in the NCP for decades, usually blindly invest and overinvest in resources to obtain high yields and higher economic returns from limited arable land (Feng et al., 2020). Secondly, the farmers have limited choice in improving peanut productivity, i.e., no suitable rhizobia inoculation (Bai et al., 2019) or other applicable management methods, but only N fertilizer. Consequently, the overuse of chemical fertilizer has led to lower N use efficiency and biological N fixation, and higher N losses (i.e., gaseous emissions and leaching), and increased economic expenditure thus threatening agricultural green development in the NCP (Zou et al., 2022). Hence, it is imperative to urgently address several critical issues in peanut production, including the reduction of nitrogen fertilizer usage, enhancement of nitrogen fertilizer efficiency, improvement of yield and quality, mitigation of nitrogen losses in agricultural fields, and minimization of environmental impacts (Wan, 2017).

The accumulation and transport of carbon (C) and N are the most critical processes in plant growth and development, and these processes determine the yield and quality of grains (Aluko et al., 2023). It is well known that the grain filling material is partly derived from photo-assimilation directly transferred to grains and partly from the remobilization of assimilates from vegetative tissues, of which approximately 70–90 % originates from the former (Gao et al., 2022, Liu et al., 2024). Non-structural carbohydrates (NSCs) accumulated in vegetative tissues can be redistributed to the grains in the later stage of grain filling, which is an important C source of grains, and is positively correlated with grain yield (Aluko et al., 2023; Wang et al., 2023b). Although the contribution of remobilization of NSCs accumulated in vegetative tissues to yield is relatively low, it can serve as a storage material for grain filling under conditions such as limited photosynthesis and insufficient supply of assimilates during grain filling stage (Agherikia et al., 2019). Moreover, the N in the grain comes from both the redistribution of N accumulated in vegetative tissues before flowering, and the N uptake from the soil during grain filling (Tegeder and Masclaux-Daubresse, 2018). The remobilization of N from vegetative tissues not only forms the major N source and accounts for the greatest proportion of grain N content, but also exhibited a significant positive correlation with grain yield and grain protein concentration (Gao et al., 2022, Sharma et al., 2023). In plants, C and N metabolism are closely interactive: the energy such as ATP and NAD(P)H, and C skeleton from C metabolism are necessary for N utilization, and organic N produced by N assimilation, such as amino acids are the building blocks of proteins, including proteins that are indispensable for C metabolic reactions (Nunes-Nesi et al., 2010). Since both C and N metabolism require C skeleton, ATP and NAD(P)H, there is a competitive relationship between C metabolism and N metabolism (Foyer et al., 2001). In the case of excessive N fertilizer, N assimilation competes with C metabolism, which consumes excessive energy and reduces the non-structural carbohydrates required for crop growth, thus affects C metabolism (Poorter and Nagel, 2000). Hoogmoed and Sadras (2016) reported that the increase in NSC concentration reduced the concentration of N in the shoots in wheat. Therefore, optimizing the relationship between C and N metabolism and coordinating the accumulation and transport of C and N is crucial to improve the yield and quality of grains (Aluko et al., 2023; Gao et al., 2022).

Crop yield depends on not only the production capacity of photosynthetic source assimilates, but also the size and storage capacity of the reservoir (Liu et al., 2024). Coordinating source-sink relationships is of great significance to improving crop yields (Wang et al., 2020); higher biomass yields and optimized source-sink ratios will drive the next great leap in crop yields (Liu et al., 2024). Peanut is a crop with continuous blooming, long flowering period, and a high number of flowers (Liu et al., 2024). However, the ultimate rate of gynophore formation is limited to a range of 30–70 %, the final rate of fully developed pods is confined to 15–30 %, and the variability in the rate of fully developed pods rate range from 20 % to 80 % (Ji et al., 2015). Consequently, the reduction of redundant and ineffective pod sink has emerged as a crucial technical challenge for enhancing both the yield and quality of peanuts (Wang et al., 2020). Furthermore, peanuts possess unlimited growth habits and branching tendencies, which lead to leaf redundancy and premature leaf senescence during the early and late growth stages, respectively; this is also a key issue affecting peanut production (Wan and Zhang, 2019). Therefore, the measures to prevent leaf sources redundancy, and address the issue of abundant flowers but limited pegs formation, as well as numerous pods with insufficient filling, are crucial for enhancing the proportion of effective pods and improving pod fullness, thereby significantly contributing to the improvement of peanut yield and quality (Liu et al., 2024).

Melatonin (N-acetyl-5-methoxytryptamine, MT) is an indoleamine found in different plant tissues (Dubbels et al., 1995). To date, MT has been defined as an essential phytohormone in plants (Arnao and Hernandez-Ruiz, 2019), which has multi-regulatory roles on a large number of physiological, biochemical, and molecular actions in plants (Arnao and Hernandez-Ruiz, (2021); (Yang et al., 2022b). Exogenous MT promoted N uptake and assimilation by improving the activities of N uptake and metabolism related enzymes, and ultimately, improved the plant growth and yield in wheat (Qiao et al., 2019) and cucumber (Xu et al., 2024). Exogenous MT increased the content of non-structural carbohydrates (NSC) and N and promoted their transport to grains by improving the activities of key enzymes involved in sucrose transporter and N uptake (Qin et al., 2023). Recent studies have also shown that MT treatment improved the quality of fruits and vegetables (Wang et al., 2022). Foliar spraying of MT increased the content of soluble sugar in fruit, especially of sucrose, by promoting the sucrose phosphate synthase (SPS) activity in the tomato leaves under salt stress (Jahan et al., 2023) and cotton under drought (Khattak et al., 2023). During pear fruit ripening, MT increased the content of soluble sugars, especially sucrose and sorbitol, which was associated with up-regulation of SPS expression (Liu et al., 2019a). Numerous studies have showed that MT enhanced plant stress resistance and promoted plant growth and yield by regulating C and N metabolism. However, how peanut yield and quality responds to MT treatment under field conditions, along with the underlying mechanisms, remain inadequately understood, particularly in the context of varying N supply levels. In order to make full use of land, light and heat resources and adapt to mechanize operations, summer-peanuts are commonly planted in the NCP. Over a two-year period (2021 and 2022) of field experiments, this study conducted a systematic analysis of the effects of exogenous MT on the yield and quality of summer-peanuts under varying N levels. Additionally, it investigated the responses of photosynthetic physiological characteristics, C and N metabolic enzyme activities, as well as the accumulation and transport of dry matter and N, and explored their intrinsic relationships with yield and quality formation. The results of this study would provide a theoretical basis for the application of MT in high-yield and high-quality summer peanut cultivation as well as environmental suitability.

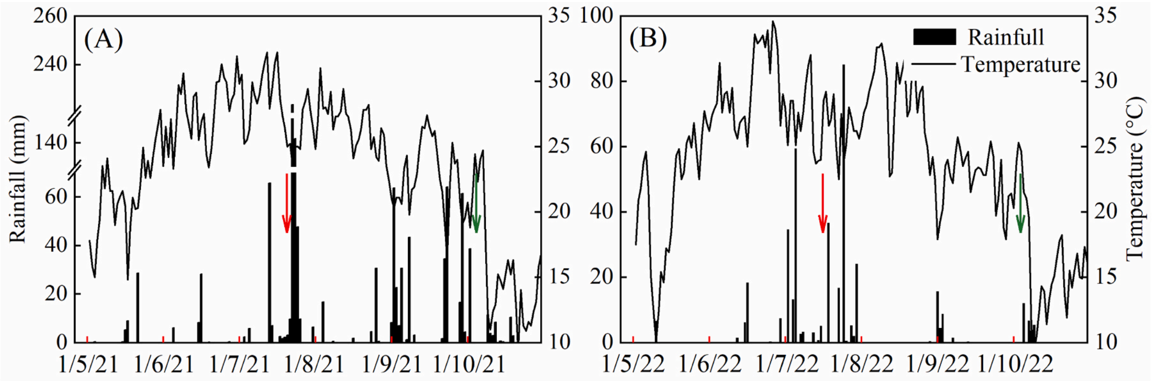

Fig. 1. Daily total rainfall and variation of temperature during the peanut growing season in 2021 (A) and 2022 (B). The dates indicated by the red and green arrows are anthesis and maturity stage, respectively.

Table 1
Experimental design and the treatments #.

Treatments	N usage	Seed dressing
N90	90 kg/ha	clear water
N90+MT	90 kg/ha	0.5 μM MT
N135	135 kg/ha	clear water
N135+MT	135 kg/ha	0.5 μM MT
N180	180 kg/ha	clear water
N180+MT	180 kg/ha	0.5 μM MT

N: nitrogen; MT: melatonin.

2. Materials and methods

2.1. Experimental sites

Field experiments were conducted in 2021 and 2022 at Yanjin Experimental Station (35°12′N, 114°07′E), Xinxiang, Henan Province, P. R. China. The soil type was sandy loam soil at the experimental sites. The upper 20 cm of the soil contained total N: 0.55 g/kg, total P: 0.60 g/kg, available P: 10.02 mg/kg, available K: 102.70 mg/kg, and organic matter: 10.27 g/kg, pH 7.7. The site was characterized as a warm-temperate continental monsoon climate. During the whole summer-peanut growing periods, the accumulative temperature above 0 ℃ was 4308℃ and 4384 ℃, and the cumulative precipitation was 1115 mm and 392 mm in 2021 and 2022, respectively (Fig. 1).

2.2. Experimental design

The split-plot design was adopted in the field experiment. The main plot consisted of two seed dressing methods (dressed with or without 0.5 μM MT), and the sub-plot comprised three N fertilizer rates [90 (N90), 135 (N135), and 180 kg N ha^{-1} (N180)]. There are a total of six treatments (Table 1), and each treatment comprised three biological replications with a plot area of 56 m^2 (2.8 m × 20 m). All treatments were conducted in fixed locations to account for the legacy effects of N. Additionally, 120 kg ha^{-1} of the P fertilizer (P_2O_5) and K fertilizer (K_2O) inputs were in the summer-peanut growing season, respectively. All N, P and K fertilizers were uniformly applied by broadcast method as basal fertilizers before peanut sowing. The local peanut variety "Yuhua 37″ was planted with a row spacing of 40 cm and an interplant spacing of 10 cm, on June 6th, 2021 and June 8th, 2022, respectively. During the peanut growth period, fungicides (30 % benzoylpropiconazole E.C.) were applied in early July to manage peanut diseases, and pesticides (high-efficiency chlorfenapyr E.C.) were used in late July to control insect pests. Weeds were controlled manually during the growing season. Irrigation was carried out twice in early and late August. The peanuts were harvested on October 8th, 2021 and October 10th, 2022, respectively.

2.3. Sampling and measurements

2.3.1. Peanut sampling

The experimental data of peanut were collected at two stages: anthesis (when 50 % of plants in a plot have begun flowering) and maturity (when 50 % of all developed pods show testa or pericarp coloration) each year. A total of 21 peanut plants were randomly selected from each plot, and 6 peanut plants were placed in liquid N and stored in an −80°C freezer prior to measure the activities of enzymes related to C and N metabolism. Another 15 plants were separated into leaf, stem, and pod, and dried in an oven at 105°C for 30 minutes, and then dried at 80°C to constant weight. After measuring the dry weight, all plant samples were ground with a grinder to determine N content.

2.3.2. SPAD, net photosynthetic rate (P_n), and photosystem II (PSII) maximum photochemical efficiency (F_v/F_m)

At the anthesis and mature stage, a SPAD-502 chlorophyll meter (Konica Minolta, Japan) was used to measure the SPAD, a Li-6800 photosynthetic meter (LI-COR Company, USA) was employed for measuring the net photosynthetic rate (P_n), and a Handy-PEA fluorometer (Hansatech, UK) was used to measure the photosystem II (PSII) maximum photochemical efficiency (F_v/F_m) in the second top peanut leaves of the main stem. All the measurements were conducted between 09:00 and 11:00 on a sunny day. Before measuring chlorophyll fluorescence parameters, peanut leaves needed to be dark-adapted for 20 min. Five plants were selected from each plot to measure SPAD, P_n and F_v/F_m, i.e., each treatment was repeated 15 times.

2.3.3. Determining dry matter as well as N accumulation and translocation

The accumulation and translocation of dry matter (DM) in peanut plants were calculated as follows:

Post-anthesis dry matter (DM) accumulation (DMA, g/m^2) = Total DM accumulation at the maturity stage –Total DM accumulation at the anthesis stage.

Post-anthesis DM translocation (DMT, g/m^2) = Total DM accumulation at the anthesis stage − DM accumulation of vegetative parts at the maturity stage.

Dry matter (DM) translocation efficiency (DMTE, %) = DMT / total DM accumulation at the anthesis stage ×100.

The contribution of post-anthesis DM translocation to pod yield (CDMT, %) = DMT / pod yield ×100.

The N concentration was measured in the leaves, stems, and pods of peanuts using a Kjeltec 2300 Analyzer Unit (Foss Tecator AB, Hoganas, Sweden) (Qiao et al., 2019). The accumulation and translocation of N in peanut plants were calculated as follows:

N accumulation amount (g/m^2) = N concentration (%) × DM accumulation amount (g/m^2).

Post-anthesis N translocation (NT, g/m^2) = Total N accumulation at the anthesis stage – N accumulation of vegetative parts at the maturity stage.

The contribution of NT to pod (CNT, %) =NT / pod N at maturity ×100.

Post-anthesis N accumulation (NA, g/m^2) =Total N accumulation at the maturity stage –Total N accumulation at the anthesis stage.

Contribution of NT to pod (CNT, %) =NT / pod N at maturity ×100.

2.3.4. N assimilation

Nitrate reductase (NR) activity was measured according to the method of Kaiser and Lewis (1984). Enzyme activity was expressed in 1 μmol h^{-1} of nitrite (μg/g FW/h). The activities of glutamine synthetase (GS), glutamate synthetase (GOGAT), and glutamate dehydrogenase (GDH) were determined by individual kits (Solebao Technology Co., LTD., Beijing, China) according to the manufacturer's instructions.

The soluble protein (SP) content in the leaves was determined using the Coomassie brilliant blue G-250 reagent according to a previous report (Bradford, 1976) with bovine serum albumin (BSA) as a standard. The total free amino acid (FAA) content was determined by the ninhydrin method (Yemm et al., 1955) with glycine as the standard.

2.3.5. Carbon metabolism

The activities of sucrose phosphate synthase (SPS), sucrose synthase (SS) and Ribulose-1,5-bisphosphate carboxylase (RuBP) were determined by individual kits (Solebao Technology Co., LTD., Beijing, China) according to the manufacturer's instructions.

The content of sucrose, fructose, and glucose in peanut leaves were determined following the protocols previously reported (Li, 2000). In short, approximately 200 mg of dry samples was first extracted with ethanol and held at 80℃ in a water bath for 30 min; then, the supernatant was used for measuring the contents of glucose, fructose, and sucrose by colorimetric method. The supernatant was obtained by extracting the residue with 10 mL of 52 % $HClO_4$ (v/v) and used for the estimation of the starch contents. The starch content was calculated following the procedure described by Shen et al. (2019).

2.3.6. Yield components

At harvest, a 6 m^2 quadrat was selected from each experimental plot, and all peanut plants in the quadrat were dug out for yield measurements; 10 representative plants were sampled from each quadrat and oven dried at 80 °C to 10 % moisture content with a constant weight. Meanwhile, the number of pegs, full pods, empty pods, total pods and hundred-grain weight (HGW) were recorded. Pod rate (PR, %) = total pods number (TPN) / number of pegs (NP) × 100; Effective pod rate (EPR)= effective pods number (EPN) / TPN × 100. The TPN refers to the sum of the full pods number and the empty pods number. All pods collected from the peanut plants in the quadrat were air-dried and weighed to calculate the pod yield of peanuts.

2.3.7. Determination of fat, protein, soluble sugar content in peanut seeds

After the peanut harvest, all pods were collected from the peanut plants and air-dried. One hundred intact peanut kernels were taken from each plot, and the oleic acid (OA), linoleic acid (LA), fat, protein and soluble sugar contents of peanut kernels in different treatments were measured using a multifunctional grain near-infrared analyzer (Bruker, MATRIX-1).

2.4. Statistical analysis

At least six biological replicates were performed for the indexes related to C and N metabolism, and fifteen biological replicates were performed for photosynthesis related indicators and the indexes related to dry matter and N accumulation and translocation. Duncan's multiple range test ($p \leq 0.05$) and analysis of variance (ANOVA) were performed by using SPSS 19.0 software (SPSS Inc., Chicago, IL, USA) for all traits, with MT, N fertilization and year as fix factors. Graphs including the PCA Biplot were plotted using the Origin 2021 software (Origin Lab Corporation, USA).

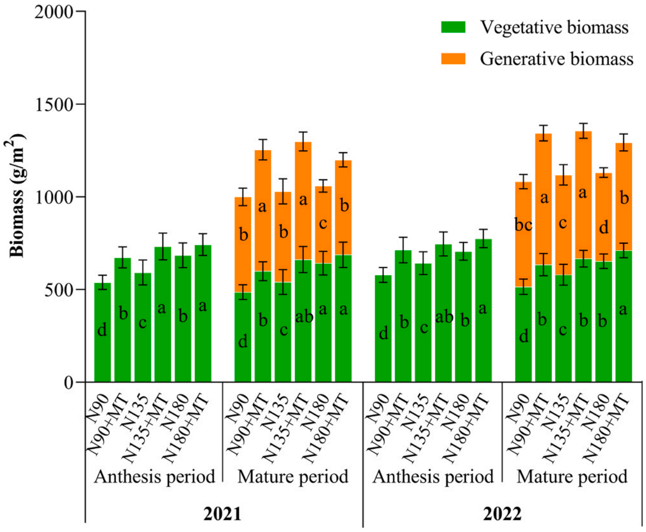

Fig. 2. Vegetative and generative dry matter of peanuts treated with MT in response to three N fertilizer levels in 2021 and 2022. The vegetative dry matter and generative dry matter are shown in green and orange, respectively. Each column represents the mean dry matter of six replicates, and the bar represents the standard error. Different letters represent significant differences between treatments with p-values < 0.05.

3. Results

3.1. Dry matter accumulation and translocation

In both years, the application of MT significantly increased the vegetative dry matter of individual peanut plants at the anthesis stage (Fig. 2). On average over the two years and three N levels, the vegetative dry matter increased with N input in both the plants treated with or without MT and the difference between the plants treated with or without MT was consistent across levels of N input (Fig. 2). At the maturity stage, based on two years' average, compared to the planted without MT treatment, the application of MT significantly increased grain mass per plant by 26.35 %, 29.07 % and 22.41 % under N180, N135, and N90 treatments, respectively (Fig. 2). N application significantly increased vegetative dry matter and reproductive dry matter both in plants treated with or without MT (Fig. 2).

The DMA and DMT were significantly affected by the application of MT, N supply level, and year as well as by the interaction between MT application and N supply level (Table S1). As N levels increased, DMA first increased, then decreased, while DMT first remained unchanged and then declined (Fig. 3 and Table S1). In both years, the application of MT had positive effect on both DMA and DMT. Averaged across three N levels and two years, DMA was significantly increased by 23.49 % from 455.63 g/m^2 in plants treated without MT to 562.66 g/m^2 in plants treated with MT, and DMT increased by 31.36 % from 55.41 g/m^2 in plants treated without MT to 72.34 g/m^2 in plants treated with MT. The application of MT had no effect on DMTE, while it had negligible effects on the contribution of CDMT because of the increased DMA in plant treated with MT. N input significantly decreased CDMT and DMTE in both plants treated with or without MT.

3.2. N accumulation and translocation

The NA and NT were significantly affected by the application of MT,

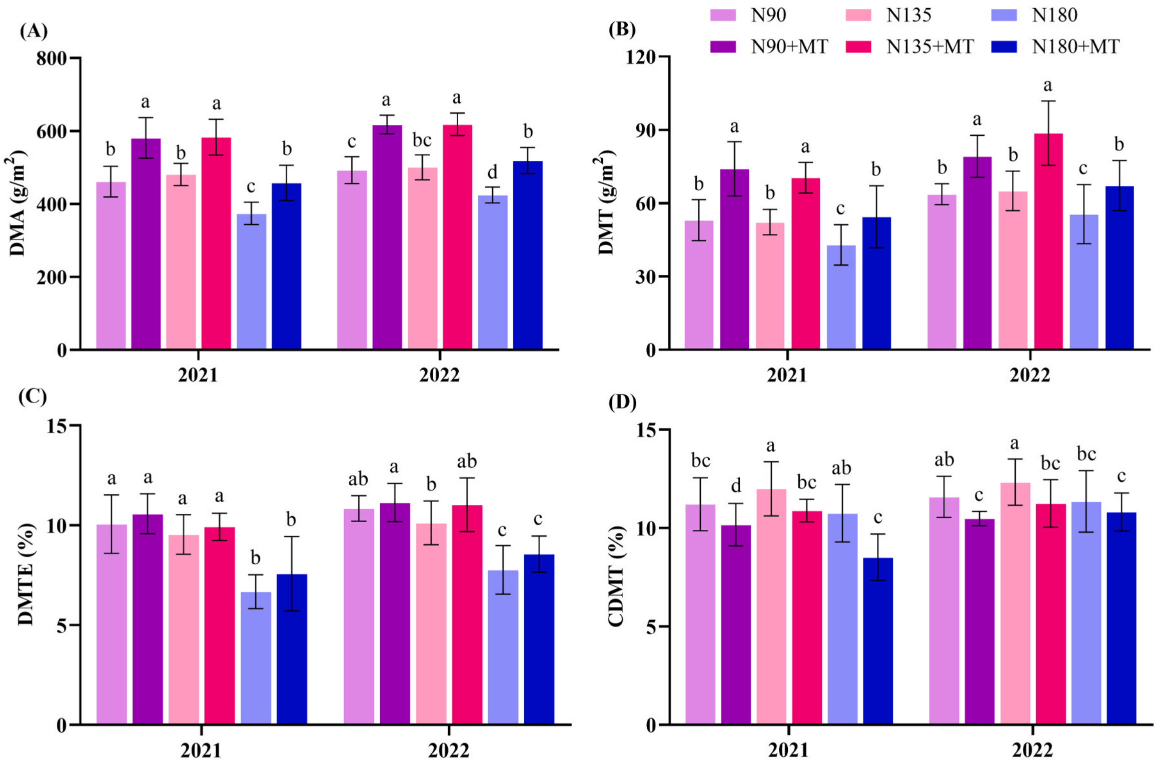

Fig. 3. The effect of MT on post-anthesis dry matter accumulation (DMA, A), post-anthesis dry matter translocation (DMT, B), dry matter translocation efficiency (DMTE, C) and the contribution of post-anthesis dry matter translocation to pod yield (CDMT, D) responses to N input in peanut. Different letters represent significant differences between treatments with *p*-values < 0.05.

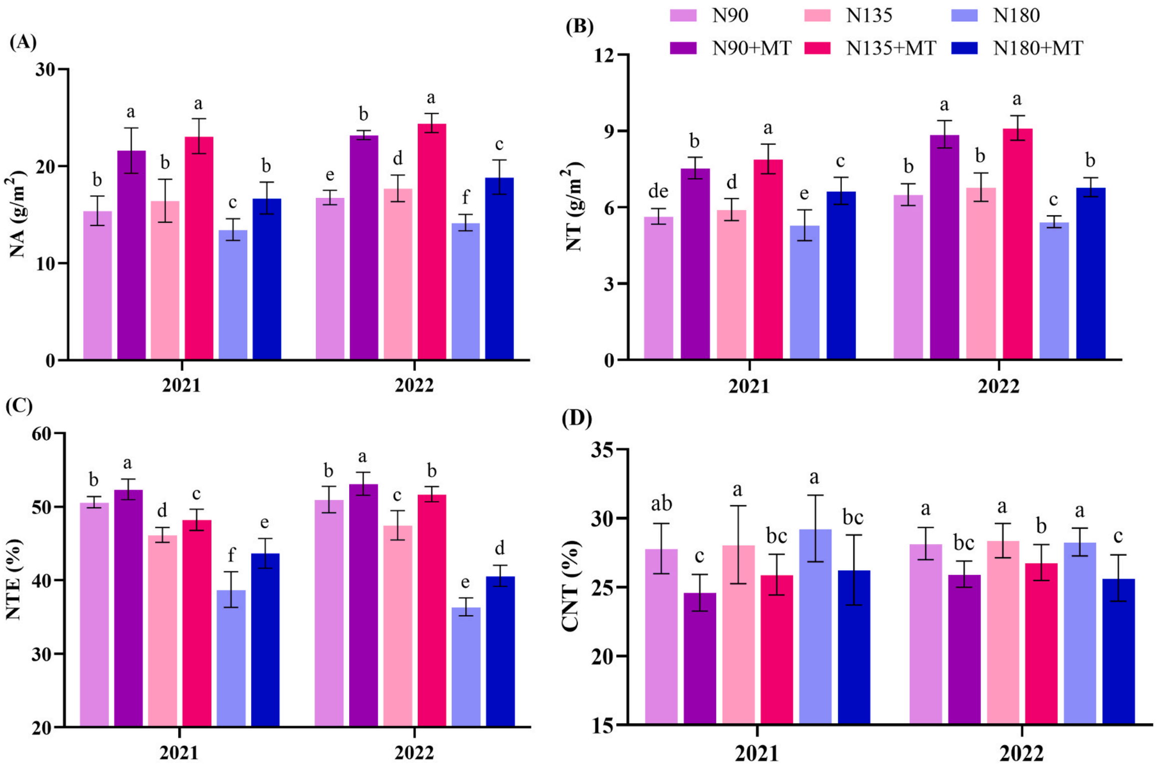

Fig. 4. The effect of melatonin on post-anthesis N accumulation (NA, A), post-anthesis N translocation (NT, B), N translocation efficiency (NTE, C) and the contribution of post-anthesis N translocation to pod N (CNT, D) responses to N input in peanut. Different letters represent significant differences between treatments with *p*-values < 0.05.

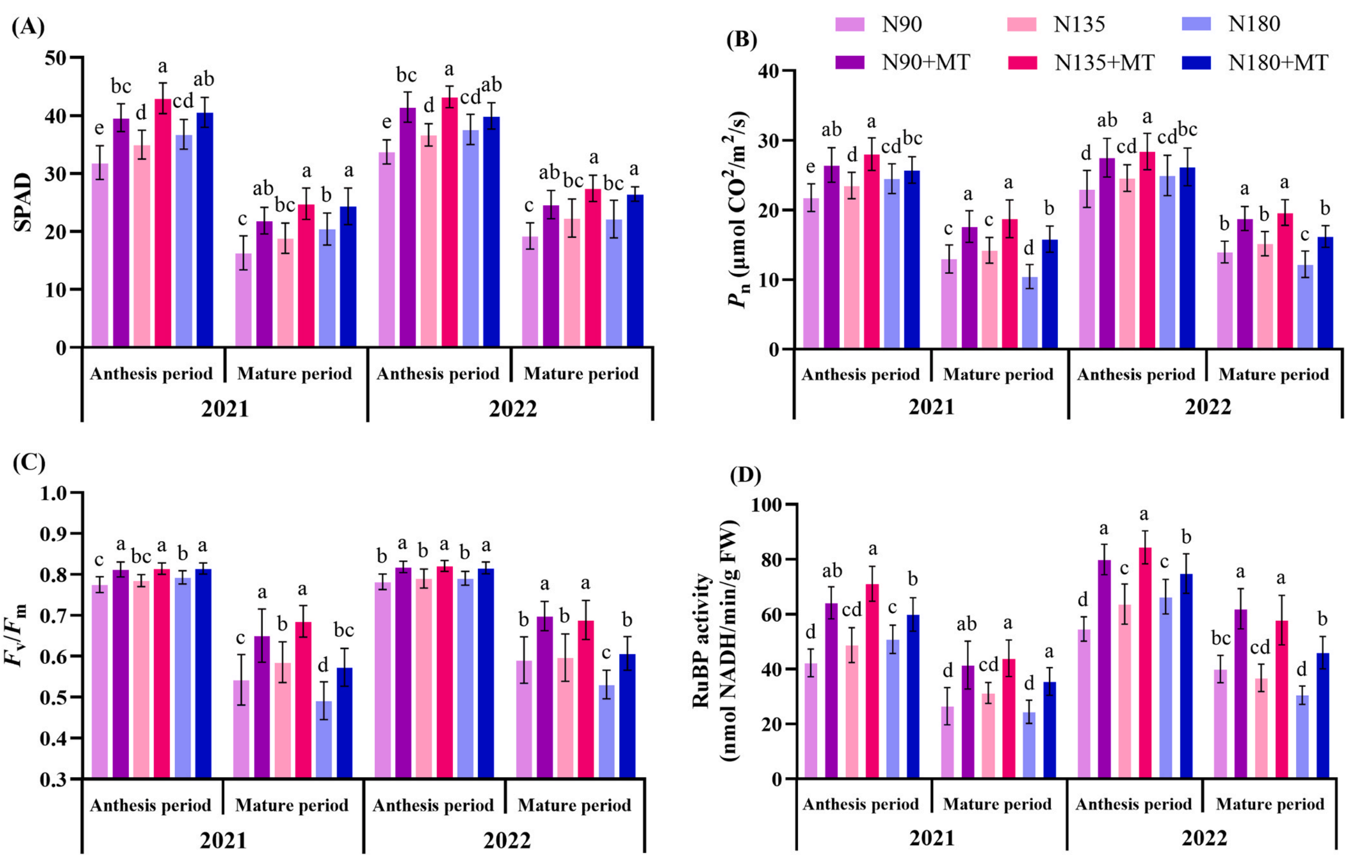

Fig. 5. The effect of melatonin on the SPAD (A), net photosynthetic rate (P_n, B) and rubisco ribulose-1, 5 bishosphate carboxylase (RuBP, C) in the leaves of peanut under different N supply conditions. Different letters represent significant differences between treatments with p-values < 0.05.

N supply level, and year as well as by the interaction between MT application and N supply level and the interaction between N supply level and year (Table S2). As N input increased, both NA and NT increased, then decreased, and reached maximum values under N135 treatment (Fig. 4, Table S2). The application of MT had an impact on NA and NT in both years, and the impact on the former was greater than the latter. Compared with the plants without MT treatment, the NT increased by 28.60 % while the NA increased by 36.18 % in plants treated with MT, respectively, averaged over two years and three N levels. The application of MT had positive effect on NTE, and the NTE increased by 7.78 % in plants treated with MT, respectively, averaged over two years and three N levels. The increased NA in plants treated with MT decreased the contribution of CNT by 9.31 %, averaged across three N levels and two years. Increasing N input had no effect on CNT, while it had negligible effects on NTE, especially in plants without MT treatment.

3.3. SPAD, P_n, F_v/F_m and RuBP activity

At the anthesis and mature stages, the value of SPAD, P_n and RuBP activities were significantly affected by the application of MT, N supply, and the year, and F_v/F_m was only significantly affected by the application of MT (Table S3). As displayed in Fig. 5 and Table S3, at both anthesis and mature stages, the application of MT increased the value of SPAD, P_n, F_v/F_m and RuBP activities under three N supply conditions in both years, and this positive effect of MT was more obvious under the N90 treatment. Compared with N90 treatment, the value of SPAD, P_n, F_v/F_m and RuBP activity treated with N90 + MT increased by 23.62 %, 20.54 % 4.72 %, and 49.03 % at the anthesis stage, and increased by 31.06 %, 35.07 %, 19.05 % and 55.85 % at the mature stage, respectively, averaged over two years.

3.4. The content of sucrose, starch, fructose, and glucose and the activities of C metabolism-related enzymes

MT treatments and N supply levels significantly affected the activities of SPS and SuS and the contents of sucrose, starch, fructose, and glucose at both anthesis and mature stages, except the starch at the anthesis stage (Table S4 and Fig. 6). The year significantly affected the SuS activity, and the contents of sucrose, and glucose at both anthesis and mature stages, and affected the SPS activity at the anthesis stage. The interaction between MT application and the year significantly affected SPS activity at the mature stage and affected the SuS activity at both anthesis and mature stages. The interaction between MT application and the year significantly affected SPS activity at the anthesis stage and affected the SuS activity at the mature stage. Under the same N level, the application of MT increased the activities of SPS and SuS at both anthesis and mature stages in both years. Averaged over two years, compared to N90, N135, and N180 alone, the actives of SPS and SuS under the treatment of N90 + MT, N135 + MT and N180 + MT increased by 61.11 %, 43.46 %, 54.32 %, and 50.88 %, 59.67 %, 52.68 % at the anthesis stage, and by 60.69 %, 64.47 %, 94.82 % and 65.36 %, 49.89 %, 40.96 % at the maturity stage. In both years, MT-treated plants consistently exhibited higher sucrose, fructose, and glucose levels across all N supply conditions at the anthesis stage, and under N135 and N90 at the maturity stage. Additionally, MT also boosted starch content at both stages under N135 and N90 conditions.

3.5. The contents of SP and FAA and the activities of N metabolism-related enzymes

Both the application of MT and N supply levels significantly affected the activities of NR, GS, GOGAT and GDH, and the contents of SP and

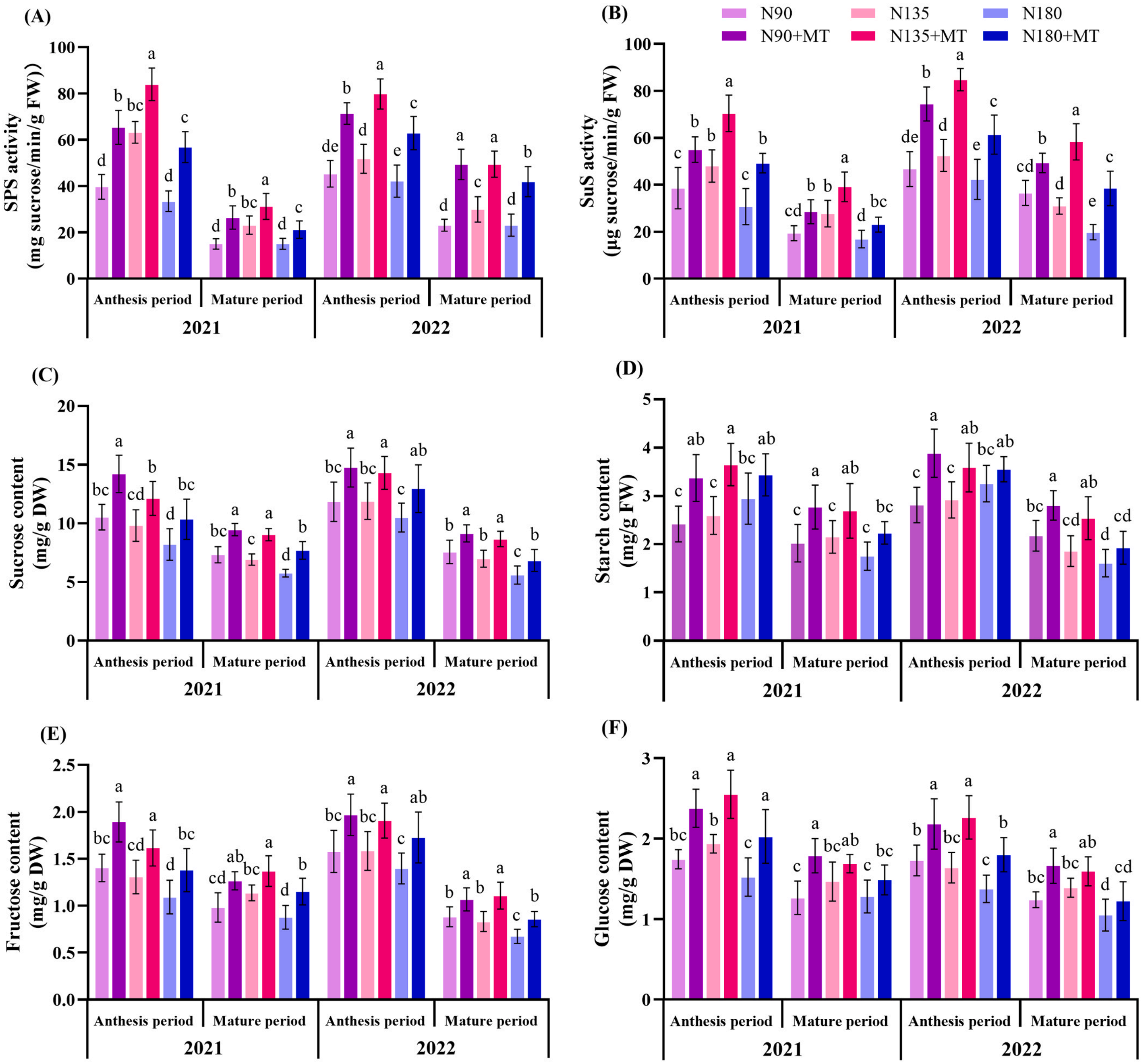

Fig. 6. The effect of melatonin on (A) sucrose phosphate synthase (SPS) activity, (B) sucrose synthase (SuS) activity, (C) sucrose content, (D) starch content, (E) fructose content, and (F) glucose content in the leaves of peanut at the anthesis and mature periods under different N supply conditions. Different letters represent significant differences between treatments with p-values < 0.05.

FAA (Table S5 and Fig. 7). The year also significantly affected these indexes except GS activity. The interaction between MT treatment and N supply levels significantly affected the activities of NR, GS, GOGAT and GDH at both anthesis and mature stages, and affected GS activity and FAA content at the mature stage. Both the interaction between MT application and the year, and the interaction between N supply levels and the year significantly affected NR activity. In both years, the application of MT increased the activities of NR, GS, GOGAT, and GDH and the contents of SP and FAA on both anthesis and mature stages, especially under N90 supply condition. Compared with N90 treatment, the NR, GS, GOGAT, and GDH and the contents of soluble protein and free amino acids in leaves treated with N90 + MT increased by 38.70 %, 31.77 %, 47.69 %, 38.23 %, 12.71 %, and 18.20 % at the anthesis stage, and increased by 45.29 %, 51.30 %, 93.93 %, 37.68 %, 25.64 %, and 72.14 % at the mature stage, respectively, averaged over two years.

3.6. Sink characteristics and yield

MT treatment significantly increased NP, ENP, EPR although it did not affect PR in 2020 and 2021 under all three N supply conditions (Table 2). The NP shows a gradual increase trend in both years as increasing N levels, while the PR, EPN, EPR showed a gradual decrease trend.

The application of MT had a positive effect on pod yield in both years under all three N supply conditions (Table 2). In 2021, the pod yields decreased in the following orders of treatments: N135+MT > N90+MT > N135 > N90 > N180+MT > N180. In 2022, the pod yields were similar to that in 2021 but a little bit of difference as following: N90+MT > N135+MT > N180+MT > N135 > N90 > N180. Compared with N90, N135 and N180 treatment, the pod yields were increased by 13.51 %, 13.06 %, and 11.32 % with N90+MT, N135+MT, and N180 + MT, respectively, averaged over two years. In 2021, compared with different

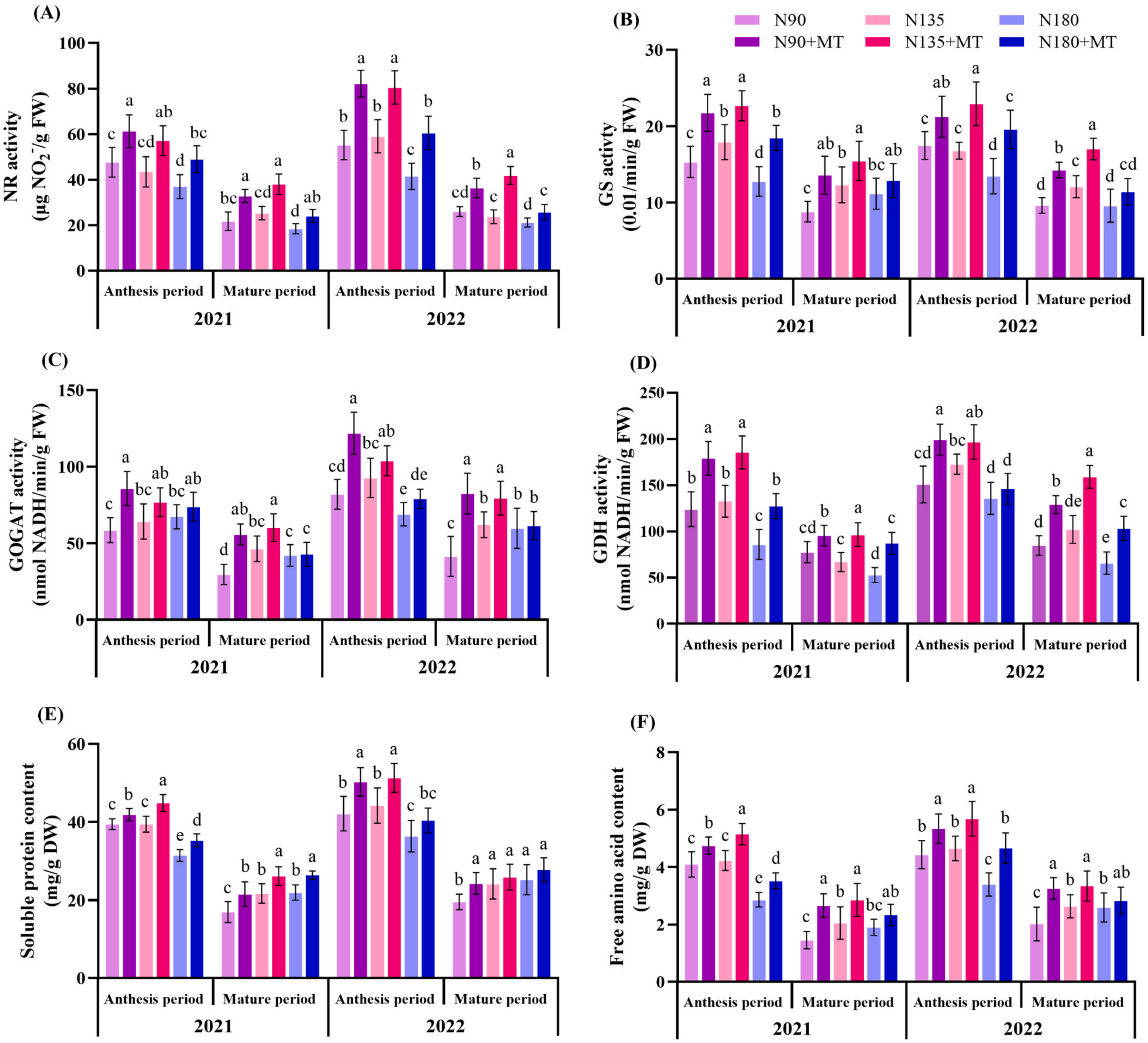

Fig. 7. The effect of melatonin on the activities of N metabolism-related enzymes including nitrate reductase (NR, A), glutamine synthetase (GS, B), glutamate synthase (GOGAT, C), glutamate dehydrogenase (GDH, D), soluble protein content (E), and free amino acid content (F) in peanut at the anthesis and mature periods under different N supply conditions. Different letters represent significant differences between treatments with p-values < 0.05.

levels of N fertilizer alone, the application of MT increased hundred-pod weight but not to a significant level. In 2022, compared with N90 and N135, the application of MT significantly increased the hundred-pod weight by 9.64 % and 9.86 %, respectively.

3.7. Grain quality

Both MT application and N supply levels significantly affected the content of fat, protein, OA, and LA, and the interaction between MT application and N supply levels significantly affected fat and protein content (Table 3). In both years, the application of MT had no effect on the soluble sugar content while it had a positive effect on fat content and protein content, especially under N90 supply conditions (Table 3). Compared with N90 treatment, the fat content and protein treated with N90+MT increased by 11.90 % and 11.96 %, respectively, averaged over two years. MT treatment significant enhanced the content of OA and LA, which enhance the peanut oil quality.

3.8. Overall effects of exogenous MT on peanut growth and development as well as yield and quality

A principal components analysis (PCA) was conducted using the data from the application of MT under three N supply conditions to elucidate the enzyme activities and substance contents related to C and N metabolism at both anthesis (Fig. 8A) and mature stages (Fig. 8B), the dry matter and N accumulation and transport, sink characteristics, and grain yield and quality (Fig. 8C). According to Fig. 8A and B, the first axis separated N180 from N180+MT, N135+MT and N90 +MT. Additionally, N90 +MT and N135+MT were closely grouped, indicating similar value of the enzyme activities and substance content related to the C and N metabolism in these treatments. The score plot also reveals that the points representing N180 always in the rightmost, and the point representing the N90 +MT and N135+MT were always on the right and far from the point representing N180, which means the treatment of N90 +MT and N135+MT could alleviate the negative effects of on these

Table 2
The effect of MT on the sink characteristics and yield in peanut under different N supply conditions [#].

Treatment	NP (No/m^2)		PR (%)		EPN (No/m^2)		EPR (%)		HPW (g)		PY (g/m^2)	
	2021	2022	2021	2022	2021	2022	2021	2022	2021	2022	2021	2022
N90	1060.8 ±120.7d	1276.6 ±127.9d	54.7±6.1a	50.5 ±4.9ab	13.6 ±2.6b	15.7 ±2.8b	59.9 ±4.9bc	66.6 ±3.6bc	148.4±8.0ab	155.1±10.1b	392.9 ±32.2b	506.0 ±29.9bc
N90+MT	1286.7 ±140.0c	1536.8 ±188.0c	57.5±7.8a	52.7±7.1a	16.2 ±3.0a	18.5 ±2.4a	67.2±5.5a	72.7±5.1a	157.2±9.6a	170.0±12.1a	440.5 ±28.2a	581.3±35.6a
N135	1236.3 ±135.7c	1496.5 ±206.1c	53.2±4.7a	47.5 ±5.5bc	13.8 ±2.8b	15.6 ±2.7b	56.4±5.7c	62.9±6.5c	150.4 ±11.2ab	153.7±9.4bc	397.6 ±28.7b	513.9 ±38.7bc
N135+MT	1466.2 ±169.9b	1712.3 ±143.6b	56.0±4.8a	48.3 ±5.6ab	16.8 ±2.3a	18.0 ±2.8a	63.8 ±7.2ab	67.3±6.8b	158.9±10.4a	168.9±11.6a	452.4 ±36.7a	577.4±40.4a
N180	1423.9 ±116.3b	1752.6 ±161.5b	41.3±4.7b	44.8±6.9c	11.7±1.9c	13.2±1.9c	46.5±6.2d	48.8±6.7e	141.2±10.3c	145.0±12.6c	307.1 ±22.9d	476.2±28.2c
N180+MT	1637.6 ±199.6a	1938.2 ±214.4a	45.7±6.3b	45.2±2.5c	13.9 ±2.4b	15.5 ±1.2b	55.9±3.4c	55.4±7.6d	147.6±8.1bc	152.0 ±10.4bc	347.2±29.6c	521.8±31.4b
NMT	1240.3±66.8b	1508.6 ±103.3b	52.5±7.5a	47.3±3.7a	13.0 ±1.8b	14.8 ±1.5b	53.9±3.1b	58.7±4.4b	146.8±6.19b	150.9±7.5b	366.3 ±34.1b	498.7±25.8b
MT	1463.5 ±134.8a	1729.1 ±104.9a	57.8 ±11.8a	48.1±4.7a	15.6 ±1.7a	17.3 ±1.0a	63.3±4.9a	64.8±4.2a	154.6±4.4a	162.7±8.1a	410.4 ±13.3a	560.2±23.6a
N90	1173.8±94.4c	1406.7 ±109.1c	59.0 ±11.4a	52.1±5.9a	14.9 ±1.8a	17.1 ±1.9a	64.7±6.0a	71.1±4.7a	151.8±6.5a	163.5±10.7a	332.3 ±27.0b	499.0±21.4b
N135	1351.3±81.0b	1604.4 ±134.0b	57.8 ±12.0a	47.8±4.3b	15.3 ±1.7a	16.8 ±1.9a	60.6±5.8a	65.6±5.4b	154.87±8.3a	160.3±9.7a	419.0 ±34.7a	545.6±30.7a
N180	1530.8 ±104.5a	1845.4 ±126.7a	46.9 ±10.3b	45.4±4.7b	12.8 ±1.2b	14.4 ±0.9b	52.6±4.9c	53.1±6.4c	143.77±6.8b	148.5±7.3b	424.6 ±26.5a	543.7±27.6a
NAOVA												
MT	***		N.S.		***		***		***		***	
N	***		***		***		***		**		***	
Y	***		**		***		***		*		***	
MT*N	N.S.		N.S.		N.S.		N.S.		N.S.		N.S.	
MT*Y	N.S.		N.S.		N.S.		N.S.		N.S.		N.S.	
N*Y	N.S.		N.S.		N.S.		N.S.		N.S.		**	
MT*N*Y	N.S.		N.S.		N.S.		N.S.		N.S.		N.S.	

EPN: Effective pods number; EPR: Effective pod rate; HPW: Hundred-pod weight; N: N application; NMT: no melatonin application; NP: Number of pegs; MT: melatonin application; PR: Pod rate; PY: Pod yield; Y: year. ***: $p < 0.001$; **: $p < 0.01$; *: $p < 0.05$; N.S.: $p > 0.05$, based on three-way ANOVA considering the factors "melatonin" (2 levels), "N fertilizer" (3 levels), and "year" (2 levels).

Table 3
The effect of MT on the grain quality of peanut under different N supply conditions [#].

Treatment	Fat (%)		Pro (%)		SS (%)		OA (%)		LA (%)		O/L	
	2021	2022	2021	2022	2021	2022	2021	2022	2021	2022	2021	2022
N90	41.6 ±1.7c	43.9 ±2.9b	21.2 ±1.2d	22.6 ±1.6d	5.2 ±0.7b	5.2 ±0.5d	73.2 ±1.9c	74.9 ±2.9b	10.3 ±0.8c	10.9 ±1.0c	7.1 ±0.6a	6.9 ±0.8ab
N90+MT	45.3 ±3.0b	50.4 ±3.2a	24.3 ±1ab	25.7 ±1.6b	5.5 ±0.6b	5.6 ±0.6 cd	75.8 ±2.6ab	77.2 ±2.3a	11.1 ±1.0b	11.8 ±1.0ab	6.9 ±0.6ab	6.6 ±0.6ab
N135	43.6 ±2.6b	46.1 ±3.3b	22.7 ±1.2c	24.1 ±0.8c	5.6 ±0.9b	5.6 ±0.5 cd	76.0 ±2.4ab	76.0 ±2.9ab	11.1 ±0.7b	11.6 ±0.8bc	6.9 ±0.5a	6.6 ±0.5b
N135+MT	47.4 ±3.0a	51.1 ±3.6a	25.0 ±1.2a	27.5 ±1.1a	6.0 ±0.7b	5.9 ±0.5bc	77.1 ±2.5a	78.0 ±2.8a	12.0 ±1.0a	12.4 ±1.1a	6.4 ±0.6b	6.3 ±0.6b
N180	43.2 ±2.4bc	45.4 ±1.9b	22.9 ±1.2c	24.4 ±0.8c	6.4 ±0.7a	6.1 ±0.3ab	74.7 ±3.5bc	76.5 ±2.8ab	11.1 ±0.8b	10.8 ±0.9c	6.8 ±0.6ab	7.1 ±0.7a
N180+MT	44.4 ±2.0b	46.2 ±2.4b	23.9 ±1.4b	25.6 ±1.5b	6.6 ±0.8a	6.4 ±1.0a	76.7 ±2.5a	77.9 ±2.9a	12.4 ±0.9a	12.2 ±0.9ab	6.2 ±0.5b	6.4 ±0.6b
NMT	45.5 ±2.1b	45.1 ±1.6b	22.3 ±0.6b	23.7 ±0.7b	5.5 ±0.8a	5.7 ±0.4a	74.7 ±1.6b	75.6 ±1.3b	10.8 ±0.4b	11.1 ±0.6b	7.0 ±0.3a	6.9 ±0.4a
MT	51.8 ±3.0a	49.2 ±1.7a	24.4 ±0.7a	26.3 ±0.8a	6.0 ±0.6a	6.0 ±0.7a	76.6 ±1.2a	77.7 ±1.5a	11.9 ±0.6a	12.1 ±0.4a	6.5 ±0.3a	6.4 ±0.2a
N90	43.2 ±1.7b	46.7 ±1.6b	22.7 ±0.7b	23.9 ±1.0c	5.2 ±0.6b	5.4 ±0.5b	74.2 ±1.6b	75.8 ±1.9b	10.7 ±0.6b	11.1 ±0.7b	7.0 ±0.4a	6.9 ±0.5a
N135	45.5 ±2.5a	48.6 ±2.1a	23.9 ±0.8a	25.8 ±0.8a	5.8 ±0.8ab	5.8 ±0.5b	76.6 ±1.6a	77.3 ±1.8a	11.6 ±0.5a	12.0 ±0.8a	6.6 ±0.3a	6.5 ±0.4a
N180	41.3 ±1.5c	46.0 ±1.4b	23.4 ±0.9a	25.0 ±0.9b	6.3 ±0.7a	6.3 ±0.6a	76.2 ±1.7a	77.5 ±1.8a	11.8 ±0.6a	11.6 ±0.6ab	6.5 ±0.4a	6.6 ±0.3a
NAOVA												
MT	***		***		N.S.		***		***		N.S.	
N	***		***		***		**		***		N.S.	
Y	***		***		N.S.		**		N.S.		N.S.	
MT*N	***		***		N.S.		N.S.		N.S.		N.S.	
MT*Y	N.S.		N.S.		N.S.		N.S.		N.S.		N.S.	
N*Y	N.S.		N.S.		N.S.		N.S.		N.S.		N.S.	
MT*N*Y	N.S.		N.S.		N.S.		N.S.		N.S.		N.S.	

[#] LA: linoleic acid; N: N application; NMT: no melatonin application; MT: melatonin application; OA: oleic acid; O/L: oleic acid /linoleic acid; Pro: protein; SS: soluble sugar; Y: year. ***: $p < 0.001$; **: $p < 0.01$; *: $p < 0.05$; N.S.: $p > 0.05$, based on three-way ANOVA considering the factors "year" (2 levels), "melatonin" (2 levels), and "N fertilizer" (3 levels).

indicators at both anthesis and mature period. According to Fig. 8C, N180+MT, N135 +MT and N90+MT treatments were distinguished from the N180 by higher PR, DMTE, DNTE, DMT, EPR, Fat, HGW, DNT, OA, NA, EPN, DMA, PY, and TPN. N90 treatments were distinguished from the N180 by higher CDNT, CDMT, PR, DMTE, DNTE, DMT, EPR, Fat, HGW, DNT, and OA.

Correlation analysis of C and N metabolism related indicators at the anthesis and mature stages with dry matter and N accumulation and transport, pod sink characteristics, yield, and quality were performed, respectively (Fig. S1). Pod yield was significantly ($p<0.01$) correlated with the RuBP, SPS, SuS, NR, GOGAT, GDH, SP, and FAA at the anthesis stage, and was significantly ($p< 0.01$) correlated with the P_n, RuBP, SPS, SuS, NR, starch, sucrose, and glucose at the mature stage. Pod yield was significantly ($p< 0.01$) correlated with DMT, DMA, NA, DNT, EPN, and HGW. Fat content was significantly ($p< 0.01$) correlated with P_n, RuBP, SPS, SuS and NR at the anthesis stage, and was significantly ($p< 0.01$) correlated with P_n, SPS, SuS, GDH, and FAA at the mature stage. Fat content was significantly ($p< 0.01$) correlated with DMA, NA, and HGW. Pro was significantly ($p< 0.01$) correlated with SPAD, RuBP, SPS, SuS, NR, GDH, SP, FAA and starch at the anthesis stage, and was significantly ($p< 0.01$) correlated with SPS, NR, GOGAT, and FAA at the mature stage. Protein was always significantly ($p< 0.01$) correlated with DMA, NA and EP. linoleic acid (LA) was significantly ($p< 0.01$) correlated with RuBP, SPS, SuS, GS, GOGAT, GDH, SP, and FAA at the anthesis stage, and was significantly ($p< 0.01$) correlated with SPAD, RuBP, and GOGAT at the mature stage. Oleic acid (OA) was significantly ($p< 0.01$) correlated with DMA, NA, EPN, and Fat.

4. Discussion

A recent study analyzed changes in peanut yields over the period from 1993 to 2018, and revealed that 65 % of the experimental fields in the NCP region exhibited peanut yields ranging from 4000 to 6000 kg/ha, with an average yield of 5138 kg/ha (Zhao et al., 2021). In this study, the peanut pod yields under varying N levels were observed to be between 3071 and 3976 kg/ha and between 4762 and 5139 kg/ha in 2021 and 2022, respectively. In contrast, yields following MT treatment were recorded to range from 3472 to 4524 kg/ha and from 5218 to 5813 kg/ha, respectively. These findings indicate that, without MT application, peanut yields at the experimental sites were within the medium to lower range typical of the NCP. However, the application of MT significantly enhances peanut yields, elevating them to the upper-middle range in this region. In this study, the peanut yield in 2021 was significantly lower than that in 2022, due mainly to a rare heavy rain that occurred locally on July 20, 2021 (Fig. 1), resulting in water accumulation and a reduction in production. In addition, the application of MT increased peanut oil and protein contents, especially under low N supply conditions.

4.1. MT treatment promoted post-anthesis dry matter accumulation and translocation

Chlorophyll is responsible for photon absorption, transmission, and transportation, and its content is directly related to the photosynthetic capacity, and the length of the green period of the leaves (Gao et al., 2024). According to a previous study, dry matter accumulation rates were highest from the anthesis stage to maturity stage, but this is also the process of gradual aging of peanut leaves (Liu et al., 2024). In this study, the application of MT significantly increased the content of chlorophyll, especially under reduced N supply conditions (Fig. 5). We also founded that there were increases in dry matter accumulation at both anthesis and maturity stages in plants treated with MT (Fig. 2), which primarily due to less leaf senescence (relatively high chlorophyll content) and an enhanced photosynthesis. As N level rise, the P_n value

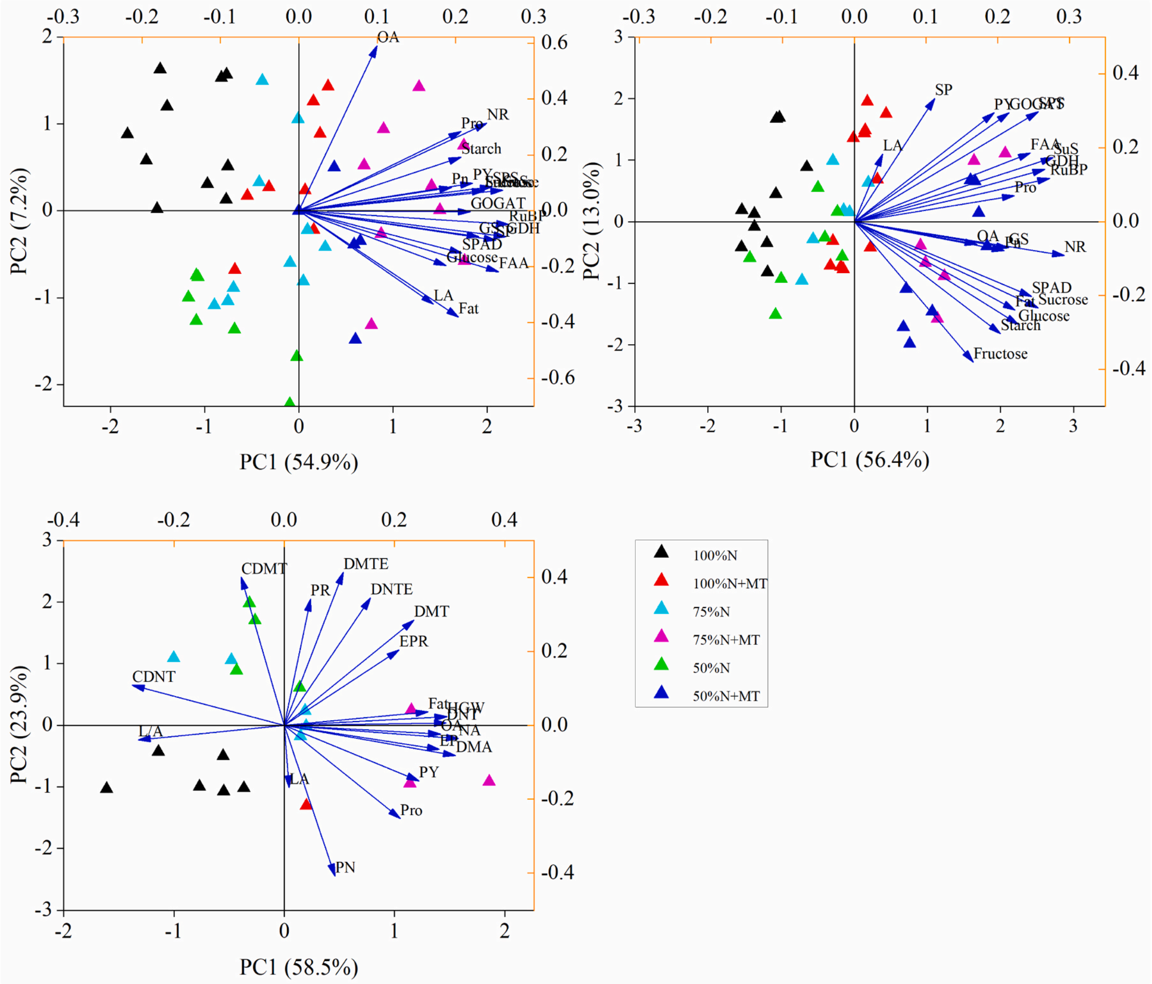

Fig. 8. Principal components analysis (PCA) biplot showing the differences in enzyme activities and substance content related to carbon and nitrogen metabolism among treatments at the anthesis (A) and mature (B) stages and the differences in the dry matter and nitrogen accumulation and transport, sink characteristics, and grain yield and quality under MT treatment the relationship among the related indicators that describe them. The relevant indicators are shown by arrows, while the different treatments are indicated as triangles of different colors. CDMT: contribution of post-anthesis dry matter translocation to pod yield; CNT: contribution of post-anthesis nitrogen translocation to pod yield; DMA: post-anthesis dry matter accumulation; DMT: post-anthesis dry matter translocation; DMTE: dry matter translocation efficiency; EPN: effective pod number; EPR: effective pod rate; FAA, free amino acid; GDH: glutamate dehydrogenase; GOGAT: glutamate synthase; GS: glutamine synthetase; HPW: hundred-pod weight; LA: linoleic acid; NA: post-anthesis nitrogen accumulation; NEP: number of effective pods per plant; NP: Number of pegs per plant; NR: nitrate reductase; NT: post-anthesis nitrogen translocation; NTE: nitrogen translocation efficiency; TPN: total pods number; OA: oleic acid; O/L: oleic acid /linoleic acid; P_n, net photosynthetic rate; PR: pod rate; Pro: protein; PY: pod yield; SPS: sucrose phosphate synthase; SP: soluble protein; SuS: sucrose synthase.

increased during the anthesis stage, promoting vigorous vegetative growth early in flowering. However, at the mature stage, P_n initially rises and then falls (Fig. 5). This pattern affects pod development, as insufficient photosynthetic capacity in the late growth period limits dry matter accumulation in peanut plants under high N levels. Melatonin-treated plants maintain higher P_n in both the early and late growth stages, which was beneficial to the accumulation of carbohydrates. Rubisco is a rate-limiting enzyme for C fixation in photosynthesis in plants (Li et al., 2023). Moreover, the application of MT also significantly increased the Rubisco activity, and both effects on chlorophyll content and Rubisco activity ultimately contributed to an improvement in P_n, resulting in greater photosynthetic assimilation capacity, and increased dry matter accumulation.

Non-structural carbohydrates (NSC), including soluble sugars and starch, are the main photosynthetic products of plants, which can usually reflect the overall C supply status of the plant (Mathan et al., 2021). Before flowering, NSC mainly exists in the form of starch in stems and leaves. After entering the pod-setting stage, the sucrose produced by photosynthesis of functional leaves and the sucrose hydrolyzed from stored starch are both transported to the grain via the phloem, where it is synthesized into starch (Yang et al., 2020). A higher sucrose supply level in the leaves is a prerequisite for a higher starch accumulation rate in the grain, which is beneficial to increasing grain yield (Liu et al., 2024). Previous studies have shown that N has a great impact on the accumulation and transport of NSC in crops (Wu et al., 2018). In this study, we also found that the content of soluble sugar and starch were decreased with increasing N supply levels (Fig. 6), which may be because more assimilates are used for the formation of structural carbohydrates and N assimilation (Wang et al., 2023). While the application of MT increased the content of soluble sugar and starch, and the activity of SPS and SuS in leaves at both anthesis and mature periods under different N supply conditions, especially under reduced N conditions, suggesting that MT application benefits growth and promotes sugar metabolism and sucrose uptake into the phloem (Khattak et al., 2023). Qin et al. (2023) found that the foliar spraying of MT promoted the formation of photosynthetic products by promoting photosynthesis and by enhancing the enzymatic activities of SPS, AI, NI and SSs (synthesis direction), and it also could promote sucrose transport by upregulating gene expression of OsSUT family, thereby accelerating the synthesis, transformation and material transport of NSC into rice grains (Qin et al., 2023). The higher sugar content may be due to the higher photosynthetic capacity as indicated by the P_n, which meaning more CO_2 was fixed to form carbohydrates in plants treated with MT (Teng et al., 2022). It can be identified from the correlation coefficient analysis that the DMA were significantly positively correlated with sucrose and starch contents. Therefore, MT may increase the supply capacity of photosynthetic products and promote the storage and redistribution of carbohydrates to the grains, thereby achieving high yields.

Photosynthetic products for crop grain filling not only come from both post-anthesis photosynthesis that are transferred directly to the grains, but also from the remobilization of the assimilate that are temporarily stored in the vegetative part of the plants (Okamura et al., 2018). In peanut, vegetative growth continues after flowering, causing competition between young vegetative tissues and developing seeds for the supply of assimilates (Turner et al., 2005). Post-anthesis dry matter accumulation is a major source for grain filling, and post-anthesis dry matter transfer can serve as a supplement to the grain fillings (Gao et al., 2022). Although the contribution of pre-anthesis dry matter transport to yield is relatively low, assimilates accumulated before flowering are preferentially activated during the grain fillings, and its transport plays an important role in improving sink activity and promoting grain filling (Wada et al., 2017). In our study, we also found that both the dry matter accumulation and dry matter translocation significantly correlated to grain yield, but the former showed the highest correlation coefficients, which suggests that the dry matter accumulation after anthesis than that before anthesis plays a greater role in increased grain yield. As estimated, with the application of MT, the dry matter accumulation after anthesis increased by 35.39 %, while post-anthesis translocation of dry matter increased by 31.06 % on average over the two years and three N levels (Fig. 3). Correlation analysis indicated a strong positive link between post-anthesis accumulation and P_n, RuBP activity and chlorophyll content. This suggests that enhanced dry matter accumulation in MT-treated plants may stem from improved photosynthetic efficiency and prolonged green leaf duration. Ahmad et al. (2021) also reported that MT significantly promoted C and N metabolism in maize, leading to increased dry matter accumulation. A study conducted by Qin et al. (2023) found that MT enhances substance transport in rice, boosting NSC to grains by upregulating auxin target genes like OsSUT, OsAMT, and OsNRT. Melatonin (MT) also increases the dry matter accumulation before anthesis, which is crucial for flower bud differentiation due to its reliance on the energy provided by carbohydrate metabolism and dry matter accumulation (Chen et al., 2018; Qin et al., 2023). This increase in flower bud differentiation likely contributes to a higher grain. A greater number of grain is beneficial to promote dry matter accumulation during grain filling and provide a sink for the transfer of dry matter to the grains (Egli, 2015), which may be an important reason for the rise in that peanut yield with MT treatments.

4.2. Melatonin treatment promoted post-anthesis N accumulation and translocation

Grain N is mainly derived from soil N uptake after anthesis and the translocation of N stored in vegetative tissues, as well as from the recycling and degradation of macromolecules (protein) into amino acids (or ureides) for reproductive sink development (Tegeder and Masclaux-Daubresse, 2018). Plants generally take up inorganic N as nitrate and ammonium, and assimilate it in the form of amino acids through N assimilation enzymes, such as NR, GS, and GOGAT (Tegeder and Masclaux-Daubresse, 2018). After conversion, FAA were transported via the xylem to the vegetative organs (source leaves), where they are partly used for various metabolic processes and protein synthesis, and the rest is redistributed to storage organs (sink organs) via the phloem (Tegeder, 2014; Aluko et al., 2023). Soluble protein represents the largest N pools in all plant tissues (Tegeder and Masclaux-Daubresse, 2018). Up to 75 % of N in leaves is located in chloroplasts in the form of proteins like RuBP and the light-harvesting complex II (LHCII), which are also the main source of N for mobilization and reuse (Wu et al., 2018). Thus, post-anthesis leaf N is vital for maintaining the stay-green state, sustaining photosynthesis, and supporting grain N demands by aiding remobilization and sink construction (Tegeder and Masclaux-Daubresse, 2018). In this study, MT application increased the activities of NR, GS, GOGAT, and GDH enzymes (Fig. 7). This increase in N-metabolism enzymes may enhance N uptake and assimilation at the post-anthesis stage, aligning with the higher content of amino acid and protein, which help leaves to stay green longer. Increased GS activity may promote N translocation to the grains, by re-assimilating ammonium from amino acid catabolism, supporting N needs during grain filling (Masclaux et al., 2000). Additionally, GS is found to promote phloem loading for translocation to sinks (Lothier et al., 2011), and its activity positively correlates with the amount of N remobilization to grains (Kichey et al., 2007). Thus, MT could enhance N absorption by enhancing the activity of N assimilation-related enzymes, benefiting photosynthesis duration for grain filling and promoting N activation and transport for construction of the sink.

N fertilization significantly affects dry matter accumulation and translocation, particularly during early stages of grain development when amino acids are essential for growth and sink establishment (Gao et al., 2022). In this study, plants increased N accumulation with rising N levels, but it decreased at high N levels, which may be related to the reduced NR, GS, and GDH activities (Fig. 7), similar to a previous report (Li et al., 2024a), resulting in lower N use efficiency. In addition, excess N inhibits root nodule formation and rhizobium activities, limiting potential symbiotic N fixation (Huang et al., 2023; Liu et al., 2023). In the present study, MT treatment increased post-anthesis N accumulation and translocation in peanut under varying N supply conditions, similar to a previous report (Qin et al., 2023), who showed that under the same N conditions, MT treatment increased N accumulation and total N concentration, and improved the material transport volume and material transport rate to grains in rice. Our results also suggest that MT treatment is beneficial to root development and nodule development (data not shown), which is in line with Wang et al.'s findings that it increased N uptake and N fixation capacity in plants, while promoting N assimilation by enhancing N assimilation-related enzymes (such as GR, GOGAT and GDH), increasing the synthesis of chlorophyll and amino acids, and enhancing photosynthetic capacity, thereby increasing dry matter accumulation under different N levels (Wang et al., 2022). Enhanced post-anthesis N accumulation and translocation was attributed to the prolonged lifespan of green leaves, boosting photosynthesis and dry matter accumulation (Qin et al., 2023), and improved grain growth and sink capacity (Aluko et al., 2023), and facilitating carbohydrates transport to grains. These factors contributed greatly to the yield of peanuts treated with MT.

4.3. MT synergistically improved peanut yield and quality

Our two-year study with three N levels found that MT application increased the number of effective pods and hundred-pod weight by 18.25 % and 6.72 %, respectively. Similar results were also observed in rice, where MT treatments increased the number of grains per spike under reduced N supply condition (Qin et al., 2023), and in wheat, where MT pre-soaking significantly increased grain number and thousand-grain weight with under N deficient condition (Qiao et al., 2019). Research indicates that post-anthesis N transport was crucial for grain development, as grains require large amounts of amino acids early to support their growth and establish sink capacity (Tegeder and Masclaux-Daubresse, 2018; Aluko et al., 2023). Under MT application, N accumulations increased by 36.18 % and N translocation by 28.60 % compared to sole N supply, potentially enhancing sink capacity and resource redistribution from the leaves to grains (Aluko et al., 2023).

The pod yield of peanuts consists only of the weight of full and

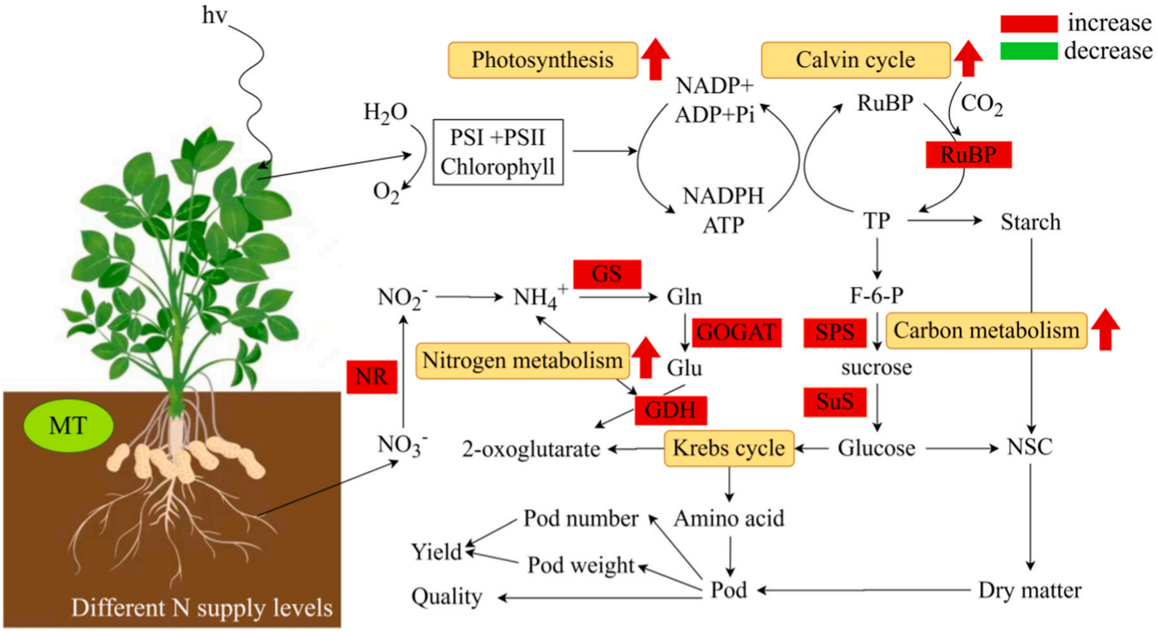

Fig. 9. A model for melatonin-enhanced peanut yield and quality by regulating C and N accumulation and translocation, source-sink dynamics as well as its association with yield and quality formation under different N supply conditions. Metabolism and parameters that increased or decreased it are indicated by the red or green boxes in melatonin-treated plants when compared with the plants without melatonin treatment. The red arrows in the figure denote increased or enhanced processes. F-6-P, fructose-6-phosphate; GDH, glutamate dehydrogenase; Gln, glutamine; Glu, glutamate; GS, glutamine synthetase; GOGAT, glutamate synthetase; NH_4^+, ammonia; NSC, Non-structural carbohydrate; NO_2^-, nitrite; NR, nitrate reductase; RuBP, ribulose-l,5-disphosphate; SPS, sucrose phosphate synthetase; SuS, sucrose synthase; TP, triose phosphate.

blighted pods, which are also called effective pods, but does not include young pods because it has no economic value (Liu et al., 2024). The effective pod setting rate and pod filling rate of peanut plants treated with high N were significantly lower than those of plants with lower levels of N supply (Table 2). Ineffective pods and pod needles may consume too much of the available nutrients, which may be one of the important reasons for limiting the peanut yield under excessive N fertilizer supply conditions (Yang et al., 2024). In this study, MT-treated plants had more mature pods at harvest season, which probably due to higher P_n and higher chlorophyll content, ensuring a steady flow of photo-assimilates. Meanwhile, these plants showed higher pod weight compared with non-treated ones. Therefore, the higher pod yield of MT-treated plants was mainly due to a higher pod number and improved pod filling rate, linked to a greater allocation of photosynthate to the pods.

The soils of the NCP are primarily classified as fuvo-aquic soils (Chinese Soil Taxonomy, entisols in US Soil Taxonomy), which exhibit low fertility, attributable to limited humus accumulation and suboptimal soil structure, characterized by either heavy-textured or heavy sandy compositions (Du et al., 2018). Balanced nutrient management, particularly of N, P, and K, is crucial for sustainable peanut yields in the NCP (Zhao et al., 2021; Hu et al., 2023; Xie et al., 2020), while excessive single nutrient may lead to over-uptake (Wang et al., 2020). In this study, we found that as N levels increased, yields stabilized and then declined. Similarly, Huang et al., (2023b) found through a meta-analysis of post-1990 Chinese studies that the optimal N application rate for peanut yield was 90–135 kg/ha. Excess nitrogen fertilizer can reduce yield due to nutrient imbalance (Zhao et al., 2021; Xie et al., 2020) and the limited growing season of summer peanuts, which leads to insufficient grain filling (Yang et al., 2024). The results of this study showed that MT improved peanut yield under different N levels, especially under low N application levels (Table 2). It has been reported that soybeans and rice treated with MT had a higher yield potentials under different N levels, respectively, according to two recent studies (Wang et al., 2022; Qin et al., 2023). The application of MT promoted the accumulation and transport of substances to the grains, increased the number of effective pods, and thus improved the peanut yield under different N levels.

Peanut grains contain approximately 50 % oil and 25 % protein, which are essential indicators for evaluating peanut quality (Liu et al., 2019b). The amount of N applied not only affected the yield of peanuts, but also the quality of peanuts. Our findings align with a previous study (Yang et al., 2024), showing that oil, amino acids and crude protein levels in peanut grains were increased first and then stabilized with rise in N application, with 135 kg/ha being optimal. The application of MT increased the contents of oil, amino acids, and protein in peanut grain at different N levels, especially at lower N levels (Table 3). Correlation analysis showed that oil content was positively correlated with P_n, SPS, SuS, GDH, and FAA, while protein content was positively correlated with SPS, NR, GOGAT, and FAA, and both the content of oil and protein was positively correlated with SPS, FAA, and post-anthesis dry matter accumulation and N accumulation. Previous studies indicate that the formation of oil is closely related to the activities of SPS and SuS activities, while the formation of protein is closely related to the activities of NR, GS, GOGAT, and GDH in peanut (Liu et al., 2024; Zhang et al., 2013; Li et al., 2024a). The plant treated with MT had higher grain protein mainly due to higher NR, GOGAT, and GDH activities (Fig. 7), that could generate various amino acids used in protein biosynthesis (Gutierrez, 2012). SPS and SuS activities were significantly elevated in the N135+MT treatment compared to that in the N180 treatment, resulting in increased sucrose metabolism in the leaves of N135+MT plants. This enhanced sucrose metabolism may lead to an increase in NSC content and transport rate, thereby providing ample substrates for lipid synthesis and ultimately elevating the fat content in the grains.

5. Conclusion

This study evaluated the effects of MT application and N fertilization on C and N accumulation and translocation, source-sink dynamics, as well as its association with yield and quality formation in peanut. Post-anthesis dry matter accumulation from anthesis to maturity greatly determined the formation of peanut yield. Greater N input decreased dry matter accumulation and translocation in peanut, while MT application had a positive effect on post-anthesis accumulation and translocation of both dry matter and N, especially under reduced N supply conditions, by improving photosynthesis, enhancing the synthesis and transformation of C metabolism and the absorption and utilization of N metabolism, thereby increasing both peanut yield and quality (Fig. 9). The solved molecular mechanism may provide gene targets for using CRISPR/Cas genome editing to improve peanut yield and quality, including oil production (Li et al., 2021, 2024b). As a result of this study, new insights are gained into how exogenous substances can stabilize yield by affecting the C and N cycle in plants at reduced N rates, which is crucial for promoting stable yields in agriculture and environmentally sustainable development.

Funding

This work was financially supported by the Henan Province High-Level Talent International Training Funding Project (Yuke [2023] No. 25), the International Science and Technology Cooperation Project of Henan Province (242102521050), the Henan Center of Outstanding Overseas Scientists (GZS2024018), the Leading Talent Project in Science and Technology Innovation of Central Plain of China (214200510021), the Program for Innovative Research Team (in Science and Technology) at the University of Henan Province (21IRTSTHN023), and the Key Science and Technology Special Project of Xinxiang City of China (ZD2020004). This study is also partially supported by the National Science Foundation (Award # 1658709) and the United States Department of Agriculture (Award # 2023–67037–40312).

CRediT authorship contribution statement

Xiangjun Kong: Investigation, Formal analysis, Data curation. **Xiaotian Zhang:** Investigation, Formal analysis, Data curation. **Peipei Jia:** Investigation, Formal analysis, Data curation. **Huiyun Xue:** Investigation, Formal analysis, Data curation. **Zengqiang Li:** Investigation, Formal analysis, Data curation. **Na Bai:** Investigation, Formal analysis, Data curation. **Xiaoping Pan:** Writing – review & editing, Supervision, Methodology, Funding acquisition, Formal analysis. **Lijie Li:** Writing – review & editing, Writing – original draft, Validation, Investigation, Formal analysis, Data curation. **Peng Xiao:** Software, Methodology, Investigation, Formal analysis. **Xiangguo Cheng:** Writing – original draft, Validation, Investigation, Formal analysis. **Baohong Zhang:** Writing – review & editing, Writing – original draft, Supervision, Methodology, Funding acquisition, Conceptualization. **Yi Zhang:** Investigation, Formal analysis, Data curation. **Zhiyong Zhang:** Writing – review & editing, Supervision, Project administration, Methodology, Conceptualization. **Dana Kohtz:** Writing – review & editing, Formal analysis. **Xiaohui Wang:** Investigation, Formal analysis, Data curation.

Declaration of Competing Interest

The authors declare no any conflict of interest.

Acknowledgements

NO

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.

Appendix A. Supporting information

Supplementary data associated with this article can be found in the online version at doi:10.1016/j.fcr.2024.109650.

Data availability

Data will be made available on request.

References

Agherikia, S., Pahlevani, M., Yamchi, A., Zaynalinezhad, K., Mostafaie, A., 2019. Transcript profiling of genes encoding fructan and sucrose metabolism in wheat under terminal drought stress. J. Plant Growth Regul. 38, 148–163.

Ahmad, S., Kamran, M., Zhou, X., Ahmad, I., Meng, X., Javed, T., Iqbal, A., Wang, G., Su, W., Wu, X., Ahmad, P., Han, Q., 2021. Melatonin improves the seed filling rate and endogenous hormonal mechanism in grains of summer maize. Physiol. Plant. 172, 1059–1072.

Aluko, O.O., Liu, Z., Sun, X., 2023. The interplay of carbon and nitrogen distribution: Prospects for improved crop yields. Mod. Agric. 1, 57–75.

Arnao, M.B., Hernandez-Ruiz, J., 2019. Melatonin: A new plant hormone and/or a plant master regulator? Trends Plant Sci. 24, 38–48.

Arnao, M.B., Hernandez-Ruiz, J., 2021. Melatonin Against environmental plant stressors: A review. Curr. Protein Pept. Sci. 22, 413–429.

Bai, H., Zhang, Y., Yu, H., Irfan, M., Huang, Y., Han, M., Yang, J., Liu, N., Wang, H., Han, X., 2019. Phylogenetic diversity and cross-inoculation of indigenous isolated Bradyrhizobium from nodules of peanut in Liaoning province of China. Mol. Biol. Res Commun. 8, 59–68.

Bradford, M.M., 1976. A rapid and sensitive method for the quantitation of microgram quantities of protein utilizing the principle of protein-dye binding. Anal. Biochem 72, 248–254.

Chen, D., Shao, Q., Yin, L., Younis, A., Zheng, B., 2018. Polyamine function in plants: metabolism, regulation on development, and roles in abiotic stress responses. Front Plant Sci. 9, 1945.

Crusciol, C.A.C., Portugal, J.R., Bossolani, J.W., Moretti, L.G., Fernandes, A.M., Garcia, J. L.N., Garcia, G.Ld.B., Pilon, C., Cantarella, H., 2021. Dynamics of macronutrient uptake and removal by modern peanut cultivars. Plants 10, 2167.

Du, Z., Xiao, Y., Qi, X., Liu, Y., Fan, X., Li, Z., 2018. Peanut-shell biochar and biogas slurry improve soil properties in the North China Plain: A four-year field study. Sci. Rep. 8, 13724.

Dubbels, R., Reiter, R.J., Klenke, E., Goebel, A., Schnakenberg, E., Ehlers, C., Schiwara, H.W., Schloot, W., 1995. Melatonin in edible plants identified by radioimmunoassay and by high performance liquid chromatography-mass spectrometry. J. Pineal Res 18, 28–31.

Egli, D.B., 2015. Is there a role for sink size in understanding maize population–yield relationships. Crop Sci. 55, 2453–2462.

Feng, Y.P., Zhang, Y.Y., Li, S., Wang, C., Yin, X.G., Chu, Q.Q., Chen, F., 2020. Sustainable options for reducing carbon inputs and improving the eco-efffciency of smallholder wheat-maize cropping systems in the Huanghuaihai Farming Region of China. J. Clean. Prod. 244, 118887.

Foyer, C.H., Ferrario-Méry, S., Noctor, G., 2001. Interactions between carbon and nitrogen metabolism. In: Lea, P.J., Morot-Gaudry, J. (Eds.), Plant nitrogen. Springer, Berlin, Heidelberg, pp. 237–254.

Gao, F., Guo, J., Shen, Y., 2024. Advances from chlorophyll biosynthesis to photosynthetic adaptation, evolution and signaling. Plant Stress 12, 100470.

Gao, H., Zhang, C., van der Werf, W., Ning, P., Zhang, Z., Wan, S., Zhang, F., 2022. Intercropping modulates the accumulation and translocation of dry matter and nitrogen in maize and peanut. Field Crops Res 284, 108561.

Gutierrez, R.A., 2012. Systems biology for enhanced plant nitrogen nutrition. Science 336, 1673–1675.

Hoogmoed, M., Sadras, V.O., 2016. The importance of water-soluble carbohydrates in the theoretical framework for nitrogen dilution in shoot biomass of wheat. Field Crop Res 193, 196–200.

Hu, J., Yang, Y., Zhang, H., Li, Y., Zhang, S., He, X., Huang, Y., Ye, Y., Zhao, Y., Yan, J., 2023. Reduction in nitrogen rate and improvement of nitrogen use efficiency without loss of peanut yield by regional mean optimal rate of chemical fertilizer based on a multi-site field experiment in the North China Plain. Plants 12, 1326.

Huang, M., Liu, Y., Sun, W., Zou, X., 2023b. Meta-analysis on the effect of nitrogen application on growth, yield and quality of peanut. J. Peanut Sci. 3, 63–72.

Huang, W., Yang, Y., Zheng, H., Olesen, J.E., Rees, R.M., Zou, J., Zhang, L., Hu, S., Qiao, B., Wang, X., Shen, S., Yang, B., Bai, Z., Zheng, A., Li, W., Song, Z., Wen, X., Chen, F., Yin, X., 2023. Excessive N applications reduces yield and biological N fixation of summer-peanut in the North China Plain. Field Crop Res 302, 109021.

Jahan, M., Li, G., Xie, D., Farag, R., Hasan, M., Alabdallah, N., Al-Harbi, N., Al-Qahtani, S., Zeeshan, M., Nasar, J., Altaf, M., Rahman, M., 2023. Melatonin mitigates salt-induced growth inhibition through the regulation of carbohydrate and nitrogen metabolism in tomato seedlings. J. Soil Sci. Plant Nut 23, 4290–4308.

Ji, J., Wan, Y., Liu, F., Zhang, K., 2015. The study on the relationship of the peanut pods spatial distribution and drought resistance ability. J. Peanut Sci. 44, 28–34.

Kaiser, J.J., Lewis, O.A.M., 1984. Nitrate reductase and glutamine synthetase activity in leaves and roots of nitrate-fed *Helianthus annuus* L. Plant Soil 77, 127–130.

Khattak, W., He, J., Sun, J., Ali, I., Bilal, W., Zahoor, Khan, K., Wang, Y., Zhou, Z., 2023. Foliar melatonin ameliorates drought-induced alterations in enzyme activities of sugar and nitrogen metabolisms in cotton leaves. Physiol. Plant. 175, e14011.

Kichey, T., Hirel, B., Heumez, E., Dubois, F., Le Gouis, J., 2007. In winter wheat (*Triticum aestivum* L.), post-anthesis nitrogen uptake and remobilisation to the grain correlates with agronomic traits and nitrogen physiological markers. Field Crop Res 102, 22–32.

Li, H.S., 2000. Principles and techniques of plant physiological experiment. Higher education press, Beijing, China, pp. 194–203.

Li, C., Brant, E., Budak, H., 2021. CRISPR/Cas: a Nobel Prize award-winning precise genome editing technology for gene therapy and crop improvement. J. Zhejiang Univ. Sci. B 22, 253–284. https://doi.org/10.1631/jzus.B2100009.

Li, G., Guo, X., Sun, W., et al., 2024a. Nitrogen application in pod zone improves yield and quality of two peanut cultivars by modulating nitrogen accumulation and metabolism. BMC Plant Biol 24, 48. https://doi.org/10.1186/s12870-024-04725-1.

Li, R., He, Y., Chen, J., Zheng, S., Zhuang, C., 2023. Research progress in improving photosynthetic efficiency. Int J. Mol. Sci. 24, 9286.

Li, L., Zhang, D., Zhang, Z., Zhang, B.H., 2024b. CRISPR/Cas: a powerful tool for designing and improving oil crops. Trends Biotechnol. https://doi.org/10.1016/j.tibtech.2024.09.007.

Liu, N., Chen, H., Huai, D., Xia, F., Huang, L., Chen, W., Wu, B., Ren, X., Luo, H., Zhou, X., Chen, Y., Lei, Y., Liao, B., Jiang, H., 2019b. Four QTL clusters containing major and stable QTLs for saturated fatty acid contents in a dense genetic map of cultivated peanut (*Arachis hypogaea* L.). Mol. Breed. 39, 23.

Liu, Z., Gao, F., Li, X., Zhang, J., 2024. Source-sink coordinated peanut cultivar increases yield and kernel protein content through enhancing photosynthetic characteristics and regulating carbon and nitrogen metabolisms. Plant Physiol. Bioch 206, 108311.

Liu, Y., Yan, Z., Wang, J., Zhao, J., Liu, Y., Zou, J., Li, L., Zhang, J., Wan, S., 2023. Optimizing initial nitrogen application rates to improve peanut (*Arachis hypogaea* L.) biological nitrogen fixation. Agronomy 13, 3020.

Liu, J., Yue, R., Si, M., Wu, M., Cong, L., Zhai, R., Yang, C., Wang, Z., Ma, F., Xu, L., 2019a. Effects of exogenous application of melatonin on quality and sugar metabolism in'zaosu'pear fruit. J. Plant Growth Regul. 38, 1161–1169.

Lothier, J., Gaufichon, L., Sormani, R., Lemaitre, T., Azzopardi, M., Morin, H., Chardon, F., Reisdorf-Cren, M., Avice, J.C., Masclaux-Daubresse, C., 2011. The cytosolic glutamine synthetase GLN1;2 plays a role in the control of plant growth and ammonium homeostasis in Arabidopsis rosettes when nitrate supply is not limiting. J. Exp. Bot. 62, 1375–1390.

Masclaux, C., Valadier, M., Brugiere, N., Morot-Gaudry, J., Hirel, B., 2000. Characterization of the sink/source transition in tobacco (*Nicotiana tabacum* L.) shoots in relation to nitrogen management and leaf senescence. Planta 211, 510–518.

Mathan, J., Singh, A., Ranjan, A., 2021. Sucrose transport and metabolism control carbon partitioning between stem and grain in rice. J. Exp. Bot. 72, 4355–4372.

Nunes-Nesi, A., Fernie, A.R., Stitt, M., 2010. Metabolic and signaling aspects underpinning the regulation of plant carbon nitrogen interactions. Mol. Plant 3, 973–996.

Okamura, M., Arai-Sanoh, Y., Yoshida, H., Mukouyama, T., Adachi, S., Yabe, S., Nakagawa, H., Tsutsumi, K., Taniguchi, Y., Kobayashi, N., Kondo, M., 2018. Characterization of high-yielding rice cultivars with different grain-filling properties to clarify limiting factors for improving grain yield. Field Crop Res 219, 139–147.

Poorter, H., Nagel, O., 2000. The role of biomass allocation in the growth response of plants to different levels of light, CO_2, nutrients and water: a quantitative review. Aust. J. Plant Physiol. 27, 1191. –1191.

Qiao, Y., Yin, L., Wang, B., Ke, Q., Deng, X., Wang, S., 2019. Melatonin promotes plant growth by increasing nitrogen uptake and assimilation under nitrogen deficient condition in winter wheat. Plant Physiol. Bioch 139, 342–349.

Qin, B., Zou, J., Cao, L., Wang, M., Zhang, Y., 2023. Melatonin regulates material transport to reduce carbon emissions and increase yield under different nitrogen in rice. Agr., Ecosyst. Environ. 342, 108235.

Sharma, S., Kumar, T., Foulkes, M.J., Orford, S., Singh, A.M., Wingen, L.U., Karnam, V., Nair, L.S., Mandal, P.K., Griffiths, S., Hawkesford, M.J., Shewry, P.R., Bentley, A.R., Pandey, R., 2023. Nitrogen uptake and remobilization from pre- and post-anthesis stages contribute towards grain yield and grain protein concentration in wheat grown in limited nitrogen conditions. CABI Agric. Biosci. 4, 12.

Shen, J., Wang, Y., Shu, S., Jahan, M., Zhong, M., Wu, J., Sun, J., Guo, S., 2019. Exogenous putrescine regulates leaf starch over accumulation in cucumber under salt stress. Sci. Hortic. -Amst. 253, 99–110.

Shi, L., Wang, B., Hu, H.Z., Hongguang, Y., 2022. Mechanism and experiment of full-feeding tangential-flow picking for peanut harvesting. Agriculture 12, 1448.

Tegeder, M., 2014. Transporters involved in source to sink partitioning of amino acids and ureides: opportunities for crop improvement. J. Exp. Bot. 65, 1865–1878.

Tegeder, M., Masclaux-Daubresse, C., 2018. Source and sink mechanisms of nitrogen transport and use. N. Phytol. 217, 35–53.

Teng, Z., Zheng, W., Jiang, S., Hong, S., Zhu, Z., Zang, Y., 2022. Role of MT in promoting plant growth by regulating carbon assimilation and ATP accumulation. Plant Sci. 319, 111276.

Turner, N.C., Davies, S.L., Plummer, J.A., Siddique, K.H.M., 2005. Seed filling in grain legumes under water deficits, with emphasis on chickpeas. Adv. Agron. 87, 211–250.

Wada, H., Masumoto-Kubo, C., Tsutsumi, K., Nonami, H., Tanaka, F., Okada, H., Erra-Balsells, R., Hiraoka, K., Nakashima, T., Hakata, M., Morita, S., 2017. Turgor-responsive starch phosphorylation in *Oryza sativa* stems: A primary event of starch degradation associated with grain-filling ability. PLoS One 12, e181272.

Wan, S., 2017. Some problems of peanut production under the structural reform of agricultural supply side. J. Peanut Sci. 46, 60–63.

Wan, S.B., Zhang, J.L., 2019. Discussion on new ways to reduce cost and increase efficiency of peanut industry in China. Chin. J. Oil Crop Sci. 41, 657–662.

Wang, J., Geng, Y., Zhang, J., Li, L., Guo, F., Yang, S., Zou, J., Wan, S., 2023. Increasing calcium and decreasing nitrogen fertilizers improves peanut growth and productivity by enhancing photosynthetic efficiency and nutrient accumulation in acidic red soil. Agronomy 13, 1924.

Wang, Y., Pang, Y., Chenc, K., Zhai, L., Shenc, C., Wang, S., Xu, J., 2020. Genetic bases of source-, sink-, and yield-related traits revealed by genome-wide association study in Xian rice. Crop J. 8, 119–131.

Wang, K., Xing, Q., Ahammed, G.J., Zhou, J., 2022. Functions and prospects of melatonin in plant growth, yield, and quality. J. Exp. Bot. 73, 5928–5946.

Wang, T., Xing, L., Song, H., Wei, Y., Li, P., Lu, Q., Hu, N., Liu, Y., Zhao, Y., Liu, J., Zhang, B., Peng, R., 2023b. Large-scale metabolome analysis reveals dynamic changes of metabolites during foxtail millet grain filling. Food Res Int 165, 112516. https://doi.org/10.1016/j.foodres.2023.112516.

Wu, H., Xiang, J., Zhang, Y., Zhang, Y., Peng, S., Chen, H., Zhu, D., 2018. Effects of post-anthesis nitrogen uptake and translocation on photosynthetic production and rice yield. Sci. Rep. -UK 8, 12891.

Xie, M., Wang, Z., Xu, X., Zheng, X., Liu, H., Shi, P., 2020. Quantitative estimation of the nutrient uptake requirements of peanut. Agronomy 10, 119.

Xu, Y., Xu, R., Li, S., Ran, S., Wang, J., Zhou, Y., Gao, H., Zhong, F., 2024. The mechanism of melatonin promotion on cucumber seedling growth at different nitrogen levels. Plant Physiol. Bioch 206, 108263.

Yang, W.Q., Shi, C.R., Kang, T., Ding, H., Xu, Y., Dai, L.X., Zhang, Z.M., 2020. Effects of water stress on non-structural carbohydrates contents in different peanut organs. Chin. Agric. Sci. Bull. 36, 28–35.

Yang, S., Zhao, Y., Qin, X., Ding, C., Chen, Y., Tang, Z., Huang, Y., Reiter, R.J., Yuan, S., Yuan, M., 2022b. New insights into the role of melatonin in photosynthesis. J. Exp. Bot. 73, 5918–5927.

Yang, Y., Zou, J., Huang, W., Manevski, K., Olesen, J.E., Rees, R.M., Hu, S., Li, W., Kersebaum, K.C., Louarn, G., Ferchaud, F., Si, J., Xiong, S., Wen, X., Chen, F., Yin, X., 2022a. Farm-scale practical strategies to increase nitrogen use efficiency and reduce nitrogen footprint in crop production across the North China Plain. Field Crops Res 283, 108526.

Yemm, E.W., Cocking, E.C., Ricketts, R.E., 1955. The determination of amino acids with ninhydrin. Analyst 80, 209–214.

Zhang, J., Gao, F., Lin, Y., Wang, Y., Yang, C.T., Zhang, F., Li, Y., Li, X., 2013. Differences in seed kernel quality and related enzyme activities of different quality type peanut cultivars. Chin. J. Appl. Ecol. 24, 481–487.

Zhao, S., Lü, J., Xu, X., In, X., Luiz, M.R., Qiu, S., Ciampitti, L., He, P., 2021. Peanut yield, nutrient uptake and nutrient requirements in different regions of China. J. Integr. Agr. 20, 2502–2511.

Zou, J., Yang, Y., Shi, S., Li, W., Zhao, X., Huang, J., Zhang, H., Liu, K., Harrison, M.T., Chen, F., Yin, X., 2022. Farm-scale practical strategies to reduce carbon footprint and emergy while increasing economic benefits in crop production in the North China plain. J. Clean. Prod. 359, 131996.