T-FACE studies reveal that increased temperature exerts an effect opposite to that of elevated CO$_2$ on nutrient concentration and bioavailability in rice and wheat grains

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Abstract
Elevated CO$_2$ concentration has been reported to decrease grain nutrient concentrations and thus worsen nutritional deficiency and hidden hunger. One nutritional aspect is mineral content, yet mineral bioavailability can be limited by the presence of phytic acid. Given that future climate scenarios predict elevated global temperature driven by elevated atmospheric CO$_2$ concentrations, we used Temperature by Free-Air CO$_2$ Enrichment (T-FACE) field experiments to investigate whether elevated temperature alters the effects of elevated CO$_2$ on grain mineral concentrations, grain mineral yields, and their bioavailability in a range of wheat and rice genotypes. We found that the negative effects of elevated CO$_2$ were compensated for by positive effects of elevated temperature. As a result, the combined elevated CO$_2$ and elevated temperature increased concentrations of some minerals by up to ~15% in both rice and wheat relative to control conditions. Moreover, the combined elevated CO$_2$ and elevated temperature did not significantly change total yields of some minerals despite lower grain yields. The combined CO$_2$ and temperature elevation increased phytic acid concentration in rice by 18.1% but decreased it in wheat by 3.5%. The mineral bioavailability, estimated as the mole ratio of phytic acid to minerals in rice and wheat grains, was limited by the combined CO$_2$ and temperature elevation in only a few cases. Our results indicate that under future climate conditions of elevated temperature and CO$_2$, the nutritional quality of rice and wheat with respect to minerals may remain unchanged.

Keywords
bioavailability, climate change, mineral concentration, mineral yield, phytic acid, temperature by free-air CO$_2$ enrichment
Over two billion people suffer from protein and mineral (especially Fe and Zn) deficiencies in their diets (Tulchinsky, 2010). These deficiencies can harm immunity, cognitive development, and growth (Stein, 2010) and increase the risks of related diseases, such as coronary heart disease (Appel et al., 2005) and stunting (King et al., 2015). The international community has made some progress in combating hidden hunger caused by nutrient deficiencies, for example, by biofortification programs (Rawat et al., 2013; Rehman et al., 2018, 2020). However, climate change might be eroding that progress, especially for people depending on cereal-based diets in low-income countries (Kennedy et al., 2003; Loladze, 2002). Rice and wheat, two of the world’s most important staple food crops, feed over 90% of the world’s population (Fitzgerald et al., 2009), and their grains are important, albeit inadequate, dietary sources of proteins and minerals (Fageria, 2003; Shewry, 2009).

One of the main features of climate change is the elevation of atmospheric CO\textsubscript{2} concentration (Stocker et al., 2014). Free-Air CO\textsubscript{2} Enrichment (FACE) facilities have been designed to study crop responses to elevated CO\textsubscript{2}, as such facilities mimic future natural conditions very well, despite the claim that they may underestimate crop responses to CO\textsubscript{2} (Allen et al., 2020). Elevated CO\textsubscript{2} has been reported to reduce concentrations of several essential elements in C\textsubscript{3} crops such as rice and wheat (Loladze, 2014; Manderscheid et al., 1995; Myers et al., 2014; Seneweera & Conroy, 1997), thus exacerbating mineral deficiencies in humans. This reduction could be caused by nutrient dilution effects of increased carbohydrate production (Fernando et al., 2014; Gifford et al., 2000; Köhler et al., 2019), reduced uptake associated with elevated CO\textsubscript{2}-induced decreases in transpiration (Ainsworth et al., 2001; McGrath & Lobell, 2013; Wullschleger et al., 2002), or reduced translocation of minerals from the vegetative parts to the grains (Ujiie et al., 2019). Such effects caused by elevated CO\textsubscript{2} have led to the alarming statement that “Increasing CO\textsubscript{2} threatens human nutrition” (Myers et al., 2014). On the contrary, elevated CO\textsubscript{2} increases root biomass and root surface (Kim et al., 2001) and the availability of some minerals from the soil (Wang et al., 2016). This positive effect of elevated CO\textsubscript{2} on uptake may counteract the aforementioned negative effects on nutrient concentration. Indeed, concentrations of K (Manderscheid et al., 1995) and Ca (Seneweera & Conroy, 1997) were found to increase under elevated CO\textsubscript{2} conditions. Lieffering et al. (2004) showed an increase in the concentration of several minerals under elevated CO\textsubscript{2}.

Climate change is associated not only with elevated CO\textsubscript{2} but also with a higher temperature, among other things (Stocker et al., 2014). Elevated temperature may increase or decrease photosynthesis, depending on crop species and baseline temperature values (Kattge & Knorr, 2007; Ruiz-Vera et al., 2015). However, it always increases transpiration (Trenberth et al., 2005; Yamaji & Ma, 2014), thus enhancing the uptake and mass flow of nutrients (Ainsworth et al., 2002; McGrath & Lobell, 2013; Wullschleger et al., 2002). These effects suggest that elevated temperature most likely has an effect on mineral concentrations—an effect that is opposite to that of elevated CO\textsubscript{2}. Very few researchers have addressed the effect of elevated temperature combined with elevated CO\textsubscript{2}, which would better mimic the future climate than elevated CO\textsubscript{2} alone. Using growth chamber facilities, Asif et al. (2019) showed that combined elevated temperature and elevated CO\textsubscript{2} did not change protein and Zn concentrations in wheat grain. Two recent studies using Temperature by Free-Air CO\textsubscript{2} Enrichment (T-FACE) facilities have shown that combined elevated temperature and elevated CO\textsubscript{2} slightly increased protein concentration in rice grain but reduced it in wheat grain (Wang et al., 2019) and reduced some mineral concentrations in soybean seed (Köhler et al., 2019). Using samples from the same experiments reported by Wang et al. (2019), Wang et al. (2020) recently reported some preliminary results demonstrating that combined elevated temperature and elevated CO\textsubscript{2} did not change mineral concentrations in grains of rice and wheat.

The extent of hidden hunger depends not only on nutrient concentration but also on nutrient bioavailability. Phytic acid (PA), an anti-nutritional factor (Bohn et al., 2007), abounds in cereal grain, and its anion (phytate) forms indigestible mixed salts with many mineral cations (Bohn et al., 2007; Maddaiah et al., 1964; Vohra et al., 1965), thus limiting their bioavailability (Brown et al., 2001) and reducing the nutritional value of cereal grain. Elevated CO\textsubscript{2} reduces wheat grain PA concentration (Fernando et al., 2012). Elevated temperature has been found to increase PA concentration in lentil seeds (Thavarajah et al., 2010) and rice grain (Su et al., 2014). Again, elevated CO\textsubscript{2} and elevated temperature appeared to have opposite effects on grain PA concentrations. No study has so far provided information on PA concentration and mineral bioavailability influenced by combined elevated CO\textsubscript{2} and elevated temperature.

We hypothesize that the effects of elevated CO\textsubscript{2} and elevated temperature cancel each other out, resulting in hardly any effects of future climate change on the concentrations of minerals and PA and the bioavailability of essential minerals. To test this hypothesis, we conducted experiments in T-FACE facilities in different years for rice and an experiment with several varieties of wheat to collected data on mineral and PA concentrations in the
grains. We also calculated the mole ratio of PA to minerals as a proxy for the bioavailability of minerals, as advised by Bohn et al. (2007).

2 | MATERIALS AND METHODS

2.1 | Experimental site and weather conditions

A T-FACE facility was established at Kangbo Village, Guli Town, Changshu Municipality, Jiangsu Province, China (31°30′N, 120°33′E). The soil in this area is classified as a Gleyic Stagnic Anthrosol. The soil and agronomic history of all plots were similar. Information on the soil’s chemical properties, which were assessed on the basis of air-dried soil samples, in 2010 (the year of establishment of T-FACE) and 2018 (the last year of experimentation in this study), is provided in Table S1. There was little change in soil mineral contents during the experimental years and among treatments in 2018 (Table S1). Detailed information on the weather conditions, including daily maximum and minimum air temperatures, daily total global radiation, daily mean relative humidity, daily mean wind speed, and daily total precipitation during the experimental period for all rice and wheat experiments, is provided in Figure S1.

2.2 | Experimental design

The T-FACE facility was as described by Cai et al. (2016). The design of the CO₂ exposure system followed the principles described by Okada et al. (2001). The CO₂ concentration in each experimental plot was monitored with 16 CO₂ sensors equally distributed in the plot. The uniformity of CO₂ concentration in the plot was automatically controlled based on wind speed and direction. Infrared heating equipment was designed according to the principles described by Kimball et al. (2008). The canopy temperatures were monitored using six infrared temperature sensors arranged in a regular hexagonal array in the plot (Figure S2). The canopy temperature increase was realized with 12 infrared heaters in each plot and automatically controlled by a computer feedback control system based on the monitored canopy temperature in each plot. The performance of the infrared heaters on a horizontal surface is shown by a thermal image (Figure S2) scanned by a FLIR system (Model T630sc, Stockholm, Sweden), and the performance on a vertical plane is shown in Table S2, which provides nighttime (20:00) air temperatures at canopy height and 0.5 m above canopy for each treatment. The height of both the CO₂ and the infrared temperature sensors was adjusted to plant height. The treatments in our T-FACE system included all four combinations of two levels of CO₂ concentration and two levels of canopy temperature: (1) CT (control), where ambient conditions were maintained; (2) C+T, where 500 μmol/mol was the target atmospheric CO₂ concentration; (3) CT⁺, where 2.0°C was the target canopy temperature increase; and (4) C+T⁺, where the elevated CO₂ was combined with the elevated temperature. The level of CO₂ elevation and temperature elevation was chosen based on the IPCC A2 emission scenario for atmospheric CO₂ concentration and surface temperature around the year 2040 (Solomon et al., 2007).

Each treatment was replicated in three octagonal rings. To create consistent light environments across the four treatments, rings of the CT and C+T treatments had the same number of—but non-functional—heaters, whose height was also adjusted to the same height above the plant canopy as the functional infrared heaters in the CT⁺ and C+T⁺ treatments. Each ring had an area of 50 m², and the shaded area below the 12 (infrared) heaters under the vertical viewing angle was about 2.75 m². The imposition of elevated CO₂ and temperature started on July 19, 2013, and July 8, 2017, for the rice experiments and on March 10, 2018, for the wheat experiment and lasted until the final harvest. Although the temperature was increased during both daytime and nighttime, we sprayed pure CO₂ only during the day, following the protocol in the studies by Hasegawa et al. (2013), Hileman et al. (1994), and Jongen et al. (1995). The average of the actually achieved increase in daytime and nighttime canopy temperature under CT⁺ and C+T⁺ and in daytime CO₂ concentration under C+T and C+T⁺ is shown in Table 1.

2.3 | Crop cultivation practices

We grew rice (2013 and 2017 experiments) using the local rice cv. Changyou 5 and wheat (2018 experiment) using winter wheat cvs Sumai188, Zhenmai9, Xinnong518, and Yangmai16 (abbreviated as SM188, ZM9, XN518, and YM16, respectively). Data from the 2013 rice experiment for agronomic traits and grain yield have already been published (Cai et al., 2016).

For rice, three 3-leaf-stage seedlings were transplanted per hill in all rings that were still exposed to ambient air conditions on June 23 (2013) and June 27 (2017). The spacing of hills was 15.3 cm × 25.4 cm (i.e., 25.7 hills m⁻²; 77.1 plants m⁻²). Wheat seeds were sown on November 10, 2017, at a density of 262.5 kg seed/ha and a row spacing of 30 cm. The four varieties of wheat were arranged in plots in each ring according to the block split-plot design, with CT, C+T, CT⁺, and C+T⁺ as main plots and varieties as the split plots randomized across main plots and three replications.
All net split plots in each ring were surrounded by at least three rows of the same variety of wheat plants receiving the same treatment. Detailed information on nitrogen application is provided in Table S1. Water, weed, disease, and pest management followed local practices.

2.4 Sample collection and chemical analysis

Plant materials from 1 m² ground area were sampled at crop maturity to determine the grain yield of each plot. In addition, a smaller sample of three hills of rice or 30 wheat plants per plot was taken, from which plant organs (leaves, stems, and panicles or ears) were separated, and the panicles or ears were further dissected into branch, glumes, and grains. They were dried at 105°C for 30 min and then at 80°C to a constant weight for further chemical analyses. Based on the moisture percentage and the proportions of grains for the small samples and the weight of plant grains from the 1 m² area, the yield of dry brown-rice grain (i.e., the part after removing the inedible husk from the whole rice grain) and the dry grain yield of wheat per m² ground area were determined. The brown-rice grains from the three hills and the wheat grains from the 30 wheat plants were ground with a ball mill to obtain a fine powder to determine the concentrations of minerals and PA. This procedure was followed to obtain data for the whole grains, because PA and minerals are mainly present in the aleurone layer of rice and wheat grains, and the layer can be removed by milling (Kumar et al., 2017).

Nitrogen concentration was assessed using the semi-micro-Kjeldahl method (Bremner, 1960). Samples of 0.5 g each were digested in a graphite tube with 10 ml H₂SO₄ and subsequently digested with H₂O₂ until a clear color was obtained. Finally, the remaining solution was filled up with ultrapure water to 100 ml. Then, 10 ml of the digestion solution were taken out and distilled with 10 ml of 10 mol/L NaOH. The distillate was titrated with 0.02 mol/L H₂SO₄ using brominated alcohol green methyl red as an indicator.

To assess the mass concentration of P, K, Ca, Mg, Fe, Mn, Cu, and Zn in the rice and wheat grains, samples of 0.5 g were digested in a graphite tube with 10 mL HNO₃-HClO₄ mixed acid (3:1, v:v). The digestion temperature was regulated until the last drop of clear solution was obtained. Finally, the solution was filled up with ultrapure water to 25 ml. Minerals in the solution were measured by an inductively coupled plasma (ICP) optical emission spectrometer (OES) 710 (Agilent Technologies, California, USA). We measured the mass concentration of minerals twice using this method, and the data for the two sets of measurements agreed well (Figures S3 and S4). In this study, we analyzed the data of the second measurement, because PA was assessed only in the second measurement, thus using measurements from the same time period while calculating the mole ratio of PA:minerals described below.

To measure the PA concentration, samples of 0.5 g were extracted with 10 ml of 0.2 mol/L HCl for two hours at room temperature while being shaken every 20 min. PA in the extract was measured by an indirect method described by Lantzsch (Haug & Lantzsch, 1983) that uses pink color absorption developed by un-reacted ferric with 2,2'-bi-pyridine and measured at 519 nm with a spectrophotometer (Shimadzu, UV-1201, Kyoto, Japan).

From the above measurements, we calculated the yield of minerals as the product of mineral concentrations and grain yield per m². To investigate the mineral bioavailability, we relied on the conclusion of previous studies (Dong & Saneoka, 2020; Israr et al., 2013; Kumar et al., 2017; Liang et al., 2010; Morris & Ellis, 1985; Vodouhè et al., 2020) that PA to mineral mole ratios are negatively correlated with mineral bioavailability. We thus calculated the mole ratio of PA to minerals using the method described by Gargari et al. (2007):
where \([\text{PA}] / [\text{Mineral}_i]\) means the mole ratio of PA to the \(i\)-th mineral, PA and \(\text{Mineral}_i\) mean PA concentration \((\text{mg} \ (100 \text{ g})^{-1})\) and the \(i\)-th mineral concentration \((\text{mg} \ (100 \text{ g})^{-1})\), 660 is the mole mass of PA (Hotz & Brown, 2004), \(\text{MM}_i\) stands for the mole mass of the \(i\)-th mineral: the MM of P, K, Ca, Mg, Fe, Mn, Cu, and Zn is 31.0, 39.1, 40.0, 24.3, 55.8, 54.9, 63.5, and 65.4 g/mol, respectively.

### 2.5 Data processing and statistical analysis

We first assumed no interaction and calculated the overall relative effect (\%) of elevated CO\(_2\), \(e[\text{CO}_2]\), using the averaged values of two years’ experiments of rice or of four varieties of wheat under the treatments with CO\(_2\) enrichment (CT and C\(^+\)T) divided by those without CO\(_2\) enrichment (CT and CT\(^+\)), i.e.:

\[
e[\text{CO}_2] = \left( \frac{\text{C}^+ \text{T} + \text{C}^+ \text{T}^-}{\text{CT} + \text{C}^+ \text{T}^-} - 1 \right) \times 100\%
\]  

Similarly, the effect of elevated temperature, \(e[\text{Tem}]\), assuming no interaction, was calculated:

\[
e[\text{Tem}] = \left( \frac{\text{CT} + \text{C}^+ \text{T}^-}{\text{CT} + \text{C}^+ \text{T}^-} - 1 \right) \times 100\%\]

To further test our major hypothesis, i.e., whether the effects of elevated CO\(_2\) and elevated temperature can cancel each other out, we introduced so-called dummy variables—using the procedure of first PROC REG and then PROC NLIN of SAS (SAS Institute Inc., Cary, NC, USA) to analyze the datasets that are typically analyzed by ANOVA. This was because our procedure, as described below, enables statistical examination by the \(F\) test of whether the effect of one experimental factor (temperature) cancels out the effect of the other experimental factor (CO\(_2\)). In the model, we introduced a set of dummy variables and assigned values (0 or 1) to separate individual levels of each experimental factor. The effect of elevated CO\(_2\) (C\(^+\)), elevated temperature (T\(^+\)), their interaction (C\(^+\) × T\(^+\)), and the effect of year (Y1 for rice) or of variety (V1, V2, V3 for wheat) and their interactions with C\(^+\) and T\(^+\) were represented by:

**FIGURE 1** Illustration of twelve scenarios where the effects of elevated CO\(_2\) (C\(^+\)) and elevated temperature (T\(^+\)) on a nutrition trait \(y\) are opposite and compensate for each other so that it brings the \(y\) value in the C\(^+\)T\(^+\) treatment back to the value of the CT treatment. \(a_0 \sim a_3\) are parameter values of the model: \(y = a_0 + a_1 \text{C}^+ + a_2 \text{T}^+ + a_3 \text{C}^+ \times \text{T}^+\). (a)–(f): elevated CO\(_2\) generally decreases whereas elevated temperature increases \(y\); (g)–(l): elevated CO\(_2\) generally increases whereas elevated temperature decreases \(y\). The effect of temperature just compensates for the effect of CO\(_2\), i.e., \(y\) value is equal in CT and C\(^+\)T\(^+\) treatments, (a) and (g); via the main effect of temperature; (b) and (h): via the interaction between CO\(_2\) and temperature, where CO\(_2\) has no main effect; (d) and (j): via both the main effects and the non-crossover interactive effects between CO\(_2\) and temperature, where the \(\text{T}^+\) effect decreases with elevated CO\(_2\); (e) and (k): via both the main effects and the non-crossover interactive effects between CO\(_2\) and temperature, where the \(\text{T}^+\) effect increases with elevated CO\(_2\); and (f) and (l): via both the main effects and the crossover interactive effects between CO\(_2\) and temperature. A thirteenth scenario for the equal \(y\) value in CT and C\(^+\)T\(^+\) treatments, in which neither C\(^+\) nor T\(^+\) has any effect \((a_1 = a_2 = a_3 = 0)\) such that \(y\) value stays invariant among four treatments, is not shown in this figure; but this scenario also occurred in our data and is referred to as scenario (m).
TABLE 2: Effects of elevated CO$_2$ (C$^+$), elevated temperature (T$^+$), their interaction (C$^+$·T$^+$), and their interactions with year (Y) on mineral concentrations in brown-rice (cv. CY5) grain in the 2013 (Y1) and 2017 (Y2) experiments.

| Treatment | N (mg/g) | P (mg/g) | K (mg/g) | Fe (µg/g) | Mg (µg/g) | Cu (µg/g) | Zn (µg/g) | Ca (mg/g) | Mn (µg/g) |
|-----------|----------|----------|----------|-----------|-----------|-----------|-----------|-----------|-----------|
| e[CO$_2$] | -7.7%    | 1.9%     | -3.1%    | 9.1%      | 1.2%      | 8.3%      | -7.0%     | -5.4%     | -4.5%     |
| e[Tem]    | 12.5%    | 14.3%    | 9.3%     | 10.9%     | 10.0%     | 16.4%     | 7.0%      | 5.9%      | 28.8%     |

Regression analysis:

- $a_0$ = 13.2
- $a_{1(Y1)} = -1.1$ ns
- $a_{2(T^+)} = 1.6^*$
- $a_{3(C^+·T^+)} = 3.65$
- $a_{4(Y1)} = -0.36$
- $a_{5(C^+·Y1)} = 7.5$
- $a_{6(T^+·Y1)} = 0.13$
- $a_{7(C^+·T^+·Y)} = -0.19$
- $R^2 = 0.35$
- $F$-value = 21.12
- $F_{0.05}$ critical value = 4.38
- Scenario:
  - (a)
  - (m)
  - (f)
  - (a)
  - NA

Note: e[CO$_2$] and e[Tem] represent the relative main effects (%) of C$^+$ and T$^+$ assuming no interaction; these main effects are calculated by Equations (2) and (3), respectively, described in the Section 2.5.

For rice:

$$y = a_0 + a_1C^+ + a_2T^+ + a_3C^+ \times T^+ + a_4Y1 + a_5C^+ \times Y1 + a_6T^+ \times Y1 + a_7C^+ \times T^+ \times Y1$$  (4)

For wheat:

$$y = a_0 + a_1C^+ + a_2T^+ + a_3C^+ \times T^+ + a_4V1 + a_5V2 + a_6V3 + a_7C^+ \times V1 + a_8C^+ \times V2 + a_9C^+ \times V3 + a_{10}T^+ \times V1 + a_{11}T^+ \times V2 + a_{12}T^+ \times V3 + a_{13}C^+ \times T^+ \times V1 + a_{14}C^+ \times T^+ \times V2 + a_{15}C^+ \times T^+ \times V3$$  (5)

Note that, for this type of analysis, a level of each experimental factor has to be set as the reference level. Ambient CO$_2$, ambient temperature, Y2 (for rice) or V4 (for wheat), were set by the REG model as the reference, and so, they are not shown in Equations (4) and (5). Thus, the coefficient $a_0$ represents the estimate for Y2 or V4 in the CT treatment. In the REG procedure, initially, all coefficients were included in the model, and then, the least significant coefficients were removed, one at a time, until the remaining coefficients were all significant ($p < 0.05$). However, the coefficients for the main effects of C$^+$ and T$^+$ were always maintained (regardless of their significance) in order to show the direction of their effects.

We then ran PROC NLIN of SAS with the GAUSS method, rebuilt from the final step of the REG model ( needless to say, estimates of the coefficients from the PROC NLIN were the same as those from the PROC REG). The residual sum of squares of this model was compared with the residual sum of squares of the model where $a_2$ was fixed to $-(a_1 + a_3)$. The F-value calculated using these two residual sums of squares and the equivalent degrees of freedom allowed us to test whether...
any effect of the elevated temperature would compensate for the opposite effect of elevated CO₂ on mineral concentrations, yields, and bioavailability in grains. The logic for this test is as follows: as long as the effect of elevated temperature, either via its main effect or via its interaction with elevated CO₂, or via both, just compensates for the effect of elevated CO₂, it brings the y value in the C+T+ treatment for the reference year or variety back to its value in the CT treatment. Mathematically, this would mean:

\[ a_0 + a_1 C^+ + a_2 T^+ + a_3 C^+ \times T^+ = a_0 \]  

(6)

As stated earlier, C+ and T+ were coded in the model as 1, and equation (6) gives \( a_2 = -(a_1 + a_3) \). Twelve possible scenarios for an equal y value in CT and C+T+ treatments are illustrated in Figure 1. For this F test, again, we took the level of \( p < 0.05 \) to judge the significance threshold, and a significant F-value suggests that the elevated temperature and elevated CO₂ effects cannot compensate for each other.
RESULTS

The signs of the effect of CO₂, e[CO₂], and the effect of temperature, e[Tem], calculated using Equations (2–3) assuming no interaction between CO₂ and temperature, agreed well with those identified by regression analysis that included interaction terms, unless the CO₂-temperature interaction term was significant (Tables 2–7). Also, the effects of interactions with year (for rice) or with varieties (for wheat) on nutrient concentrations were significant only occasionally. In the following text, we describe the main effects of CO₂ and temperature only in terms of the regression results and evaluate whether the effects of elevated temperature could cancel out the effects of elevated CO₂.

3.1 Mineral concentrations

For brown-rice grains, elevated CO₂ did not significantly affect mineral concentrations, with the exception of a decline (4.5%) in Mn concentration (Figure 2, Table 2). Elevated temperature significantly increased the concentrations of N, P, K, Mg, and Mn (by 12.5%, 14.3%, 9.3%, 10.0%, and 28.8%, respectively) (Figure 2, Table 2). The positive effect of elevated temperature overrode the (non-significant) negative or positive effects of elevated CO₂ on P and Mg concentrations (Figure 2, Table 2) and the concentrations of K and Mn in 2013 and Fe and Zn in 2017 (Table S3). Furthermore, year effects were significant for all mineral concentrations, except for N concentration (Figure 2, Table 2).

For wheat grains, elevated CO₂ significantly decreased N and Mg concentrations (by 8.7% and 5.9%, respectively) and significantly enhanced Zn and Mn concentrations (by 7.9% and 18.3%, respectively) (Figure 3, Table 3). Elevated temperature significantly increased the concentrations of N, Fe, Mg, Cu, Zn, and Mn (by 14.6%, 10.0%, 8.5%, 8.3%, 13.5%, and 64.8%, respectively) did not affect the concentrations of P and Ca, and significantly lowered the concentration of K (by 12.1%) (Figure 3, Table 3). The effects of elevated temperature and elevated CO₂ could not cancel out each other for the concentrations of N of variety XN518, Fe of variety ZM9, and P, K, Mg, Ca, and Mn.

| Treatment | Grain (g/m²) | N (g/m²) | P (g/m²) | K (g/m²) | Fe (mg/m²) | Mg (g/m²) | Cu (mg/m²) | Zn (mg/m²) | Ca (g/m²) | Mn (mg/m²) |
|-----------|-------------|----------|----------|----------|------------|----------|------------|------------|----------|----------|
| e[CO₂]    | 11.5%       | 2.2%     | 13.6%    | 7.5%     | 18.3%      | 13.1%    | 15.6%      | 3.9%       | 7.6%     | −0.8%    |
| e[Tem]    | −23.7%      | −13.3%   | −12.7%   | −16.8%   | −17.1%     | −16.1%   | −12.2%     | −18.2%     | −20.9%   | −3.6%    |

Regression analysis

| Parameter | Estimate | SE | t-value | p-value |
|-----------|----------|----|---------|---------|
| a₀        | 807      |    | 11.0    | 2.07    |
| a₁(C⁺)    | 76**     | 0.2ns| 0.23**  | 0.2ns   |
| a₂(T⁺)    | −189**   | −1.3* | −0.25** | −0.6**  |
| a₃(C⁺·T⁺) |          |     |         |         |
| a₄(Y₁)    | −97      | −2.0 | −0.52   | −0.3    |
| a₅(C⁺·Y₁) |          |     |         |         |
| a₆(T⁺·Y₁) |          |     |         |         |
| a₇(C⁺·T⁺·Y₁)| 3.3**    |     |         |         |

Note: e[CO₂] and e[Tem] represent the relative main effects (%) of C⁺ and T⁺ assuming no interaction; these main effects are calculated by Equations (2) and (3), respectively, described in the Section 2.5.

3 | RESULTS

The signs of the effect of CO₂, e[CO₂], and the effect of temperature, e[Tem], calculated using Equations (2–3) assuming no interaction between CO₂ and temperature, agreed well with those identified by regression analysis that included interaction terms, unless the CO₂-temperature interaction term was significant (Tables 2–7). Also, the effects of interactions with year (for rice) or with varieties (for wheat) on nutrient concentrations were significant only occasionally. In the following text, we describe the main effects of CO₂ and temperature only in terms of the regression results and evaluate whether the effects of elevated temperature could cancel out the effects of elevated CO₂.

3.1 Mineral concentrations

For brown-rice grains, elevated CO₂ did not significantly affect mineral concentrations, with the exception of a decline (4.5%) in Mn concentration (Figure 2, Table 2).
Mn of variety SM188 (Table S4). In wheat, the Mn concentration showed many significant interactions (Figure 3 and Table 3).

### 3.2 Grain yield and mineral yields

In rice, elevated CO₂ increased grain yield by 11.5% and the yields of P and Mg by 13.6% and 13.1%, respectively, while having no significant effect on the yields of N, K, Fe, Cu, Zn, and Mn (Figure 4, Table 4). Elevated temperature reduced grain yield by 23.7% and the yields of N, P, K, Mg, Cu, and Ca by 13.3%, 12.7%, 16.8%, 16.1%, 12.2%, and 20.9%, respectively, but enhanced the yield of Mn (by 16.4%) while having no significant effect on the yields of other minerals (Figure 4, Table 4). The effects of elevated CO₂ and elevated temperature could not cancel each other out for the grain yield and the K yield (Figure 4 and Table 4) and the yield of N and Zn in the 2013 experiment (Table S5). Year effects were significant for grain yield and the yields of N, P, K, Mg, Cu, Zn, and Ca (Figure 4, Table 4).

In wheat, elevated CO₂ enhanced grain yield by 13.8% and the yield of Fe, Cu, Zn, and Mn by 18.6%, 6.6%, 25.1%, and 35.7%, respectively, but did not significantly affect the

| Treatment | Grain (g/m²) | N (g/m²) | P (g/m²) | K (g/m²) | Fe (mg/m²) | Mg (g/m²) | Cu (mg/m²) | Zn (mg/m²) | Ca (g/m²) | Mn (mg/m²) |
|-----------|-------------|----------|----------|----------|------------|-----------|------------|------------|-----------|------------|
| e[CO₂]    | 13.8%       | 4.1%     | 4.8%     | 7.4%     | 18.6%      | 7.4%      | 6.6%       | 25.1%      | 3.8%      | 35.7%      |
| e[Tem]    | −16.0%      | −3.8%    | −16.0%   | −25.7%   | −8.3%      | −8.7%     | −10.1%     | −18.3%     | −17.3%    | −18.6%     |

Regression analysis

| a0        | 754 | 17 | 2.67 | 3.56 | 45.3 | 1.14 | 4.74 | 44.4 | 0.51 | 15.7 |
| a1(C⁺)    | 63**| 1ns| 0.11| 0.21ns| 5.7**| 0.07ns| 0.69*| 13.3**| 0.02ns| 3.4** |
| a2(T⁺)    | −113**| −1ns| −0.40**| −0.85**| −3.7*| −0.09*| −0.02ns| −2.0 ns| −0.08*| −3.4** |
| a3(C⁺·T⁺) |      |    |    |     |    |     |    |     |     |     |
| a4(V1)    | −71 |    |     |     |     |     |    | 0.85 |     | −13.3 |
| a5(V2)    | −190| −4 | −0.44| −1.00| −7.6 | −0.27| −0.86| −10.7 | −0.14 |
| a6(V3)    | −92 | −3 | −0.42| −0.45| −8.8 | −0.23| −1.15| −10.5 | −0.09 |
| a7(C⁺·V1) | 83  |    |     |     |     |     |    |     |     |     |
| a8(C⁺·V2) |     |    |     |     |     |     |    |     |     |     |
| a9(C⁺·V3) |     |    |     |     |     |     |    |     |     |     |
| a10(T⁺·V1)|     |    |     |     |     |     |    |     |     |     |
| a11(T⁺·V2)|     |    |     |     |     |     |    |     |     |     |
| a12(T⁺·V3)|     |    |     |     |     |     |    |     |     |     |
| a13(C⁺·T⁺·V1)|     |    |     |     |     |     |    |     |     |     |
| a14(C⁺·T⁺·V2)|     |    |     |     |     |     |    |     |     |     |
| a15(C⁺·T⁺·V3)|     |    |     |     |     |     |    |     |     |     |

| R²        | 0.80 | 0.59 | 0.36 | 0.53 | 0.67 | 0.57 | 0.41 | 0.76 | 0.35 | 0.47 |
| F-value   | 4.05 | 0.00 | 2.94 | 7.01 | 0.90 | 0.14 | 0.38 | 0.57 | 2.12 | 0.00 |
| F₀.05 critical value | 4.08 | 4.07 | 4.07 | 4.07 | 4.07 | 4.07 | 4.07 | 4.07 | 4.07 | 4.06 |
| Scenario⁶ | NA   | (g)  | (g)  | NA   | (g)  | (k)  | NA   | (g)  | NA   | NA  |

Note: e[CO₂] and e[Tem] represent the relative main effects (%) of C⁺ and T⁺ assuming no interaction; these main effects are calculated by equation (2) and equation (3), respectively, described in the Section 2.5.

a₀ ∼ a₁₅ represent the parameter estimation values of equation (5), based on the method of stepwise regression analysis described in the Section 2.5 (values of a₁ and a₂: *p < 0.05; **p < 0.01; ns, not significant. Values other than a₁ and a₂, p < 0.05; cells without values, not significant).

F-values are for testing whether the effect of T⁺ can compensate for the effect of C⁺, via either its main effect or/and their interactive effect, where the null hypothesis (H₀) is a₂ = −(a₁ + a₃). The F-values shown in bold refer to those ≥the F₀.05 critical value, meaning that a₂ ≠ −(a₁ + a₃), i.e., the effect of T⁺ cannot compensate for the effect of C⁺; see the text in the Section 2.5 for more details.

⁶ Lowercase letters in brackets wherever the F-value was insignificant correspond to the compensation scenario type illustrated in Figure 1; NA: not applicable in any scenario because of interactions of C⁺·V and/or T⁺·V and/or C⁺·T⁺·V.

SM188, ZM9, XN518, and YM16 are abbreviations for winter wheat cvs Sumai188, Zhenmai9, Xinnong518, and Yangmai16, respectively.
yield of the other minerals. Elevated temperature, however, significantly decreased grain yield by 16.0% and the yields of P, K, Fe, Mg, Ca, and Mn by 16.0%, 25.75%, 8.3%, 8.7%, 17.3%, and 18.6%, respectively, but did not significantly affect the yields of N, Cu, or Zn (Figure 5, Table 5). Interactions between environmental factors and variety were often significant. Elevated CO₂ and elevated temperature could not cancel each other out for the yield of K (Figure 5, Table 5) and the grain yield of variety XN518 (Table S6). Variety effects were significant on many nutrient yields (those of N, P, K, Fe, Mg, Cu, Zn, and Ca) (Figure 5, Table 5).

### 3.3 Phytic acid and the phytic acid to mineral mole ratio

In rice, elevated CO₂ significantly increased the grain phytate (PA) concentration by 8.0% and significantly increased the PA/K, PA/Mg, PA/Zn, and PA/Mn mole ratios (by 12.2%, 6.6%, 13.6%, and 14.3%, respectively). Elevated temperature significantly enhanced the PA concentration by 10.1% but reduced the PA/Mn mole ratio by 15.3% (Table 6). Other PA/mineral mole ratios remained unaffected by either elevated CO₂ or elevated temperature (Figure 7, Table 7, and Table S8). There were hardly any effects of variety observed on PA concentration or PA/mineral mole ratios (Figure 7 and Table 7).

| Treatment     | PA (mg/g) | PA/K | PA/Fe | PA/Mg | PA/Cu | PA/Zn | PA/Ca | PA/Mn |
|---------------|-----------|------|-------|-------|-------|-------|-------|-------|
| [CO₂]         | 8.0%      | 12.2%| 2.4%  | 6.6%  | 0.5%  | 13.6% | 9.7%  | 14.3% |
| [Tem]         | 10.1%     | 0.1% | −14.2%| 0.5%  | 1.4%  | 0.0%  | 4.8%  | −15.3%|

Regression analysis

| Parameter | Coefficient | p-value |
|-----------|-------------|---------|
| a₀        | 4.3         | 0.06    |
| a₁ (C⁺)   | 0.6**       | < 0.05  |
| a₂ (T⁺)   | 0.3**       | < 0.05  |
| a₃ (C⁺·T⁺) | −0.3        | ns      |
| a₄ (Y1)   | −0.8        | ns      |
| a₅ (C⁺·Y1) | −0.8        | ns      |
| a₆ (T⁺·Y1) | 1.13        | 0.02    |

R² | F-value |
---|---------|
| 0.96 | 28.00 |

F₀.05 Critical value | 4.45 |
Scenarios

| Scenario | PA/K | PA/Fe | PA/Mg | PA/Cu | PA/Zn | PA/Ca | PA/Mn |
|----------|------|-------|-------|-------|-------|-------|-------|
| a₀       | 4.6  | 0.16  | 0.82  | 0.75  | 0.75  | 0.49  | 0.09  |

Note: [CO₂] and [Tem] represent the relative main effects (%) of C⁺ and T⁺ assuming no interaction; these main effects are calculated by Equations (2) and (3), respectively, described in the Section 2.5.

α₁ − α₂ represent the parameter estimation values of Equation (4), based on the method of stepwise regression analysis described in the Section 2.5 (values of α₁ and α₂: *p < 0.05; **p < 0.01; ns, not significant. Values other than α₁ and α₂, p < 0.05; cells without values, not significant).

F-values are for testing whether the effect of T⁺ can compensate for the effect of C⁺, via either its main effect or/and their interactive effect, where the null hypothesis (H₀) is α₂ = −(α₁ + α₃). The F-values shown in bold refer to those ≥ the F₀.05 critical value, meaning that α₂ ≠ −(α₁ + α₃), i.e., the effect of T⁺ cannot compensate for the effect of C⁺; see the text in the Section 2.5 for more details.

Lowercase letters in brackets wherever the F-value was insignificant correspond to the compensation scenario type illustrated in Figure 1; letter (m) refers to the extreme scenario where α₁ = α₂ = α₃ = 0, which is not illustrated in that figure; NA: not applicable in any scenario because of interactions of C⁺·Y and/or T⁺·Y and/or C⁺·T⁺·Y.
DISCUSSION

Earlier reports have raised the alarm about climate change threatening grain nutrition quality (Manderscheid et al., 1995; Myers et al., 2014; Seneweera & Conroy, 1997). These studies were based on conventional FACE studies, without taking into account the other important climate change variable—elevated temperature. Only recently has a new generation of FACE facilities, T-FACE, been developed, making it possible to examine the combined effect of elevated CO\textsubscript{2} and elevated temperature under field conditions (Cai et al., 2016; Köhler et al., 2019; Ruiz-Vera et al., 2015; Wang et al., 2019). With the data collected from T-FACE experiments, we analyzed the nutrient concentration, nutrient yields, and nutrient bioavailability of whole grains of rice and wheat. These data allowed us not only to examine the individual effects of elevated CO\textsubscript{2} and elevated temperature, but also more importantly to investigate whether the effects of elevated temperature can compensate for the effects of elevated CO\textsubscript{2} on grain nutritional values in light of our analytical framework for compensation scenarios (Figure 1).

| Treatment | PA (mg/g) | PA/K | PA/Fe | PA/Mg | PA/Cu | PA/Zn | PA/Ca | PA/Mn |
|-----------|-----------|------|-------|-------|-------|-------|-------|-------|
| e[CO\textsubscript{2}] | −1.3% | 7.4% | 3.1% | 6.2% | −6.2% | 11.9% | 7.7% | 1.2% |
| e[Tem] | −2.2% | −6.7% | 15.5% | 6.9% | 1.8% | 14.5% | 4.3% | 8.1% |

Regression analysis

| Parameter | Value |
|-----------|-------|
| a\textsubscript{0} | 6.6 |
| a\textsubscript{1}(C+) | −0.3* |
| a\textsubscript{2}(T+) | −0.4** |
| a\textsubscript{3}(C+·T+) | 0.4 |
| a\textsubscript{4}(V1) | −0.3 |
| a\textsubscript{5}(V2) | −0.3 |
| a\textsubscript{6}(V3) | −0.3 |
| a\textsubscript{7}(C+·V1) | |
| a\textsubscript{8}(C+·V2) | |
| a\textsubscript{9}(C+·V3) | |
| a\textsubscript{10}(T+·V1) | |
| a\textsubscript{11}(T+·V2) | |
| a\textsubscript{12}(T+·V3) | |
| a\textsubscript{13}(C+·T+·V1) | |
| a\textsubscript{14}(C+·T+·V2) | |
| a\textsubscript{15}(C+·T+·V3) | |

| R\textsuperscript{2} | 0.15 |
| F-value | 6.07 |
| F\textsubscript{0.05} Critical value | 4.06 |

Note: e[CO\textsubscript{2}] and e[Tem] represent the relative main effects (%) of C\textsuperscript{+} and T\textsuperscript{+} assuming no interaction; these main effects are calculated by Equation (2) and (3), respectively, described in the Section 2.5.

Insert Table 7

SM188, ZM9, XN518, and YM16 are abbreviations for winter wheat cvs Sumai188, Zhenmai9, Xinnong518, and Yangmai16, respectively.
Elevated CO₂ (by 200–400 µmol mol⁻¹) has been reported to decrease grain mineral concentrations (Manderscheid et al., 1995; Myers et al., 2014; Seneweera & Conroy, 1997). Previous FACE studies also found that elevated CO₂ (150 µmol mol⁻¹ increment) induced decreases in the concentrations of Ca, Fe, Cd, and Mg, but increased those of K, B, Zn, Pb, and Se in wheat grain (Hogy et al., 2009, 2013), at least trend-wise. Here, our data show that significant declines in mineral concentrations were relatively infrequent (Figures 2 and 3; Tables 2 and 3), and we also found significant increases (Zn and Mn in wheat grain) (Figure 3 and Table 3). This may be associated with the relatively low CO₂ increment (by 105–130 µmol mol⁻¹) in our C+T treatment (Table 1).

Elevated temperature has been reported to increase the concentrations of B, Ca, and Fe in soybean seeds (Köhler et al., 2019) and those of Fe and Zn in rice and wheat grain (Wang et al., 2020). In line with those reports, our data show that elevated temperature increased some mineral concentrations in rice and wheat grains (Figures 2 and 3; Tables 2 and 3; Tables S3 and S4). These main positive effects of elevated temperature mostly exceeded the small negative effects of elevated CO₂ on these nutrient concentrations (Figures 2 and 3; Tables 2 and 3; Tables S3 and S4).

We observed that the combination of elevated CO₂ and elevated temperature increased some mineral concentrations in rice and wheat grain (e.g., N, P, K, and Mg in rice; N and Fe in wheat) (Figures 2 and 3; Tables 2 and 3). These results differ from the few previous reports based on T-FACE that showed only increased protein concentration in rice grain (Wang et al., 2019) but no effect on...
concentrations of minerals in rice and wheat grain (Wang et al., 2020) and reduced levels of some mineral concentrations in soybean seed (Köhler et al., 2019). There were also cases where no effect of the combined elevated CO₂ and elevated temperature was observed on mineral concentrations. These cases occurred because there were no responses, or because the effect of elevated temperature compensated for the effect of elevated CO₂ via either their main effects or their specific interactions (Figures 2 and 3; Tables 2 and 3; Tables S3 and S4).

Our data show that elevated CO₂ enhanced grain yields of rice and wheat but that elevated temperature decreased grain yield (Figures 4 and 5; Tables 4 and 5), partly because the temperature increases on some days in our experiments were in the supra-optimal range of temperatures (Figure S1). The elevated temperature-induced decrease in grain yield could not be cancelled out by the elevated CO₂-induced increase and resulted in a lower grain yield in response to the combined elevated CO₂ and elevated temperature (Figures 4 and 5; Tables 4 and 5). However, most of the negative effects of elevated temperature on grain mineral yields could be cancelled out by the positive effect of elevated CO₂ (Figures 4 and 5; Tables 4 and 5; Tables S5 and S6), as shown in scenario (g) in Figure 1, resulting in no change in the grain mineral yields in response to the combined elevated CO₂ and elevated temperature.

In general, our data show that elevated CO₂ combined with elevated temperature reduced grain yield, whereas the combination did not change grain mineral yields, leading to earlier-mentioned increases in mineral concentrations in rice and wheat grain (Figures 2 and 3; Tables 2 and 3). These effects hold promise for people who regard rice and wheat grain as staple foods and consume over 50% of their per capita dietary energy by eating rice and wheat (Food & Agriculture Organization, 2017). Our study contradicts the fear that climate change will lead to lower nutritional quality (Manderscheid et al., 1995; Myers et al., 2014; Seneweera & Conroy, 1997). These previous studies took only elevated CO₂ into account. Our study based on T-FACE facilities suggests that the risk of most mineral
deficiencies may not increase, as the positive effect of increased temperature on mineral concentrations will compensate for the negative effect, or strengthen the slight, positive effect, of elevated CO₂ (Figures 2 and 3; Tables 2 and 3; Tables S3 and S4).

4.2 | Concentrations of phytic acid and bioavailability of minerals

Elevated CO₂ (Fernando et al., 2012) or elevated temperature (Ren et al., 2009; Su et al., 2014; Thavarajah et al., 2010) alters the PA concentration in grains/seeds of crop species, but no study has addressed the effect of elevated CO₂ combined with elevated temperature on PA concentration in rice and wheat grains. Our results show that elevated CO₂ and elevated temperature both increased PA concentration in rice grain (Figure 6 and Table 6) but decreased PA concentration in wheat grain (Figure 7 and Table 7). We also observed that the interaction between elevated CO₂ and elevated temperature led to an even stronger increase in PA concentration in rice grain in the 2013 experiment and to a lesser decline in PA concentration in wheat grain (Figures 6 and 7; Tables S7). These results indicate that future climate may enhance PA concentration in rice grain but reduce it in wheat grain.

PA to mineral mole ratios are negatively correlated with mineral bioavailability (Kumar et al., 2017), but no study has addressed grain mineral bioavailability in response to elevated CO₂, elevated temperature, and their combination. We found that elevated CO₂ either increased or did not affect the mole ratio of PA to minerals in rice grain (Figure 6; Table 6 and Table S7), meaning that it reduced or did not affect the bioavailability of minerals. Elevated temperature increased or had no effect on the bioavailability of most minerals in rice, but the effects were not consistent between the two years (Table S7). Our results illustrate that the slight, negative effect of elevated CO₂ on mineral bioavailability in rice grain was canceled out by the elevated temperature in most cases. In wheat grain, the effects of elevated CO₂ and elevated temperature on the bioavailability of minerals were similar trend-wise, whereas the consistent effect of elevated temperature and elevated CO₂ resulted in no change in the bioavailability of most minerals in wheat grain (Figure 7; Table 7 and Table S8). These results indicate that climate change will hardly limit the bioavailability of minerals in brown rice and wheat grain.

It is noteworthy that the aleurone layer—where PA and minerals are mainly present—of rice and wheat grains can be removed by milling (Brier et al., 2015; Brinch-Pedersen et al., 2007), resulting in lower PA and mineral concentrations in polished rice (without the aleurone layer) and in wheat flour (Brier et al., 2015; Mahgoub & Elhag, 1998). Therefore, the responses of mineral bioavailability in polished rice and wheat flour to climate change may be different from those in whole rice and wheat grains.
FIGURE 5  Grain yields and mineral yields of wheat in four treatments in the 2018 experiment involving four varieties (cvs. SM188, ZM9, XN518, and YM16). SM188, ZM9, XN518, and YM16 are abbreviations for winter wheat cvs Sumai188, Zhenmai9, Xinnong518, and Yangmai16, respectively. Error bars represent standard errors of the means (n = 3). The significant differences among treatments and CT were shown in Table 5. CT, C+T, CT+ and C+T+ refer to ambient conditions, elevated CO2, elevated temperature, and the combination of elevated CO2 and elevated temperature, respectively.

FIGURE 6  Phytic acid (PA) concentrations and the PA to mineral mole ratios in four treatments in brown-rice (cv. CY5) grain in the 2013 and 2017 experiments. Error bars represent standard errors of the means (n = 3). The significant differences among treatments and CT were shown in Table 6. CT, C+T, CT+ and C+T+ refer to ambient conditions, elevated CO2, elevated temperature, and the combination of elevated CO2 and elevated temperature, respectively.
4.3 | Concluding remarks

Conventional FACE studies reported that climate change had a negative effect on grain nutrition quality (Myers et al., 2014). A few previous T-FACE reports (Köhler et al., 2019; Wang et al., 2019, 2020) studied protein and mineral concentrations in soybean, rice, and wheat. Our study complemented these reports by analyzing not only mineral concentrations and their yields but also the bioavailability of these minerals, which ultimately matters for human health.

Our study shared with a few recent studies using T-FACE (Wang et al., 2019, 2020) that combined elevated temperature and elevated CO₂ generally decreased grain yields but not changed grain mineral yields, leading to higher grain mineral concentrations. This suggests that climate change would seem to result in more, not less, nutritious brown-rice grain and wheat grain. This corrects the earlier prevailing claim based on conventional FACE data for the effect of only elevated CO₂ that climate change has a negative effect on grain nutrition quality (Myers et al., 2014).

Our study uniquely analyzed the effects of elevated CO₂ and elevated temperature on the bioavailability of minerals in rice and wheat grains. Combined elevated CO₂ and elevated temperature increased mineral concentration and had limited effect on bioavailability of minerals. Therefore, at least under our experimental conditions, mineral deficiencies will hardly be exacerbated by climate change.

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CONFLICT OF INTEREST

The authors declare no competing interests.

AUTHOR CONTRIBUTIONS

W.L., X.Y., and P.C.S. conceived the study; C.C. and G.L. set up the field experiments under the supervision of W.L., G.P., and X.L.; X.G., B.H., H.Z., C.C., H.L., Y.Z., Z.L., M.D., and R.N. performed the field experiments with contributions from G.L. and W.C.; X.G. analyzed the data and...
drafted the manuscript with contributions from G.L. and C.C.; X.Y., P.C.S., and W.L. made substantial contributions to revising and editing the manuscript.

DATA AVAILABILITY STATEMENT
The authors declare that all data supporting the findings of this study are available within the paper and its supplementary information.

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