Received: 7 April 2022 | Revised: 21 September 2022 | Accepted: 5 October 2022

DOI: 10.1111/jac.12624

CLIMATE CHANGE

Journal of Agronomy and Crop Science WILEY

Effect of N and CO_2 supply on source size per grain at anthesis and its relationship with grain growth in wheat

Remy Manderscheid[1] | **Markus Dier**[2]

[1]Thünen Institute of Biodiversity, Braunschweig, Germany

[2]Institute of Crop Science, Quality of Plant Products, University of Hohenheim, Stuttgart, Germany

Correspondence
Remy Manderscheid, Thünen Institute of Biodiversity, Bundesallee 65, Braunschweig D-38116, Germany.
Email: manderscheid.remy@gmail.com

Funding information
Deutsche Forschungsgemeinschaft, Grant/Award Number: MA-1736/3-1 and MA-1736/5-1

Abstract

Rising atmospheric CO_2 concentration ([eCO_2]) increases the yield of wheat mainly by increasing grain number, but effects on single grain weight are variable. It is discussed whether single grain growth is limited by the sink or the source size under a non-stress environment. This study explores the effect of e[CO_2] combined with varying N supply on the source and sink size during grain filling. Source size was defined as the amount of stem reserves per grain (SRG) and the proportion of incident radiation intercepted by the green canopy per grain ($f_{IR}G$) at anthesis. Data from a 2-year free-air CO_2 enrichment experiment with wheat with three N levels (on average 38 [Nd], 190 [Nad] and 320 kg N ha^{-1} [Nex]) and two CO_2 levels (393 and 600 ppm) on SRG, $f_{IR}G$ and grain filling rate (GFR) and duration (GFD) were evaluated. SRG ranged from 2.5 to 12.9 mg and $f_{IR}G$ from 4.0 × 0.001% to 6.8 x 0.001%. Rising N supply or e[CO_2] decreased SRG and $f_{IR}G$ via their increases in grain number. Accordingly, there was a negative linear relationship between grain number and SRG ($r^2 \geq 0.84$) or $f_{IR}G$ ($r^2 \geq 0.97$). Increasing N supply decreased GFR, but increased GFD, and GFR was increased by e[CO_2] under Nad and Nex. For GFR and final grain weight, there was a strong positive ($r^2 \geq 0.85$), and for GFD, a strong negative linear relationship ($r^2 \geq 0.76$) with $f_{IR}G$ under Nad and Nex. Under these N levels, $f_{IR}G$ supplied the largest share (>86%) for grain growth and thus single grain growth was possibly source limited under Nad and Nex. Under high grain number such as under Nex and e[CO_2], there might be a risk for low final grain weight due to the low SRG that is insufficient for buffering assimilate shortage under unfavourable environmental conditions in early grain filling.

KEYWORDS
climate change, grain growth, nitrogen, source–sink relationship, wheat

Key Points

- The effect of N and CO_2 supply on source size per grain at anthesis and grain growth was studied in a biennial wheat trial.
- Source size was measured as stem reserves (SRG) and percentage radiation intercepted by canopy per grain ($f_{ir}G$).
- Rising N or CO_2 supply decreased SRG and $f_{ir}G$ via the effect on grain number.

- Treatments affected grain filling rate, duration and final weight, and these variables were related to $f_{ir}G$.
- Grain growth becomes source limited at high N and sensitive to favourable (CO_2) and possibly unfavourable conditions.

1 | INTRODUCTION

Wheat is the third most important food crop in the world in terms of production and the most important considering its range of cultivation (FAOSTAT, 2017), and therefore plays a key role in global food security. For wheat and other cereals, the most economically important factor is grain yield, which can be divided into two main components: the number of grains per unit area and single grain weight. Changes in grain yield by environmental factors, such as nitrogen (N) supply and elevated atmospheric CO_2 concentration (e[CO_2]), and increased grain yield by breeding progress have been strongly associated with changes in grain number (Dier et al., 2018; Shearman et al., 2005). In contrast, single grain weight is relatively stable but can vary markedly in high-yielding environments that result in high grain numbers, and this variation can significantly influence grain yield (Lynch et al., 2017).

Single grain weight is considered the product of grain filling rate (GFR) and duration (GFD). Both variables are influenced by temperature due to its effect on crop development (Sofield et al., 1977; Wiegand & Cuellar, 1981). Moreover, grain weight is determined by the interaction between the grain sink capacity and the assimilate supply per grain during grain filling (Fischer, 2011). The grain sink capacity is cultivar specific and is determined by carpel growth occurring between booting and anthesis (Calderini et al., 2001; Calderini & Reynolds, 2000) and by endosperm cell division occurring in the week after anthesis (Fischer, 2011). Both processes, in turn, depend on the assimilate supply in their periods of occurrence (Calderini & Reynolds, 2000). The main assimilate sources for grain growth are assimilates generated by photosynthesis during grain filling and stem reserves of water-soluble carbohydrates (SR). The SR usually peak in the middle of grain filling under an adequate water supply, but then decrease to provide assimilates needed for grain development (Wardlaw & Willenbrink, 1994). Important factors determining the size of the assimilate sources for grain growth are the fraction of global radiation intercepted by the canopy (f_{IR}), which depends on the evolution and size of the green area index (GAI) and radiation use efficiency (RUE).

Grain number is limited by assimilate supply and depends strongly on the biomass accumulation in the ears occurring from the flag leaf stage up to anthesis (Fischer, 2011). In contrast, it is still unclear whether single grain weight is limited by the grain sink capacity or the assimilate source. Several studies (e.g. Borrás et al., 2004; Miralles & Slafer, 2007; Serrago et al., 2013) indicate that grain growth is essentially sink limited. For instance, no correlation between assimilate availability per grain during grain filling and single grain weight was found based on studies with manipulations of the source-to-sink ratio (Miralles & Slafer, 2007). Moreover, in a study where wheat was grown under a low and high N level and the sink-to-source ratio was differently manipulated, single grain weight was relatively stable among the treatments and there were only slight differences in SR at maturity compared with anthesis (Serrago et al., 2013). However, a study conducted in a high-yielding environment indicated source limitation of grain growth as single grain weight did not reach its potential weight (Alonso et al., 2018). A third possibility is that grain growth is co-limited by the sink and source, meaning interaction between sink and source, in which the assimilate demand from growing grains influences assimilate supply and vice versa.

Increased N supply and e[CO_2] have variable effects on single grain growth in wheat. For instance, an increase in N supply from a low-to-high level increased single grain weight (Serrago et al., 2013). However, in another study, single grain weight was the same in plants grown under N deficiency and adequate N supply but decreased under excess N supply (Dier et al., 2018). e[CO_2] was found to increase single grain weight (Fernando et al., 2012, 2014; Tausz-Posch et al., 2015) and GFR (Li et al., 2000, 2001) under high N supply in free-air CO_2 enrichment (FACE) studies, suggesting source limitation of single grain growth. However, other FACE studies showed a reduced grain weight under e[CO_2] (Blandino et al., 2020; Högy et al., 2009), which might indicate sink limitation of single grain growth and more grains with a smaller potential weight or a reduced assimilate availability per grain under e[CO_2]. Therefore, it is unclear how different N supply and e[CO_2] affect the source size per grain and to what degree these influences affect single grain growth.

Increased N supply and e[CO_2] increase grain number primarily through an increased number of tillers (Dier et al., 2018). This effect, however, might result in a decrease in the assimilate source per grain through a decrease in f_{IR} per grain. A possible reason for this is that high N supply and e[CO_2] increase self-shading within the canopy due to increased peak GAI values (Dier et al., 2018), implying a fewer radiation interception by the leaves in lower positions within the canopy compared with plants grown under N deficiency or ambient atmospheric CO_2 concentration (a[CO_2]). On the other hand, e[CO_2] could increase the SR at anthesis due to increased RUE, which was found in barley (Manderscheid et al., 2009) and wheat (Dier et al., 2018). These effects could in turn increase the source size per grain.

The aim of the present study was to investigate interactions between two CO_2 (393 and 600 ppm) and three N levels (N deficiency and adequate and excess N supply) on single grain growth of winter wheat. The present study was conducted as a part of a 2-year FACE experiment with well-watered conditions. The focus was on the analysis of (i) traits that determine the source size per grain that are the stem reserves per grain (SRG) and the f_{IR} per grain ($f_{IR}G$) at anthesis, (ii) the CO_2 × N effects on GFR and GFD and (iii) the degree to which single grain growth is influenced by changes in the source size per grain.

2 | MATERIAL AND METHODS

2.1 | Design of the experiment and crop management

The FACE experiment was conducted with one winter wheat cultivar ('Batis') on a field site (52°18′N, 10°26′E, 79 m.a.s.l.) at the Thünen-Institute in Braunschweig, Germany, in 2014 and 2015. The experiment included two CO_2 treatments with ambient (a[CO_2], 393 ppm) and elevated concentration of CO_2 (e[CO_2], 600 ppm), and three N treatments with deficient, adequate and excess N levels. The CO_2 treatments were conducted in circular plots with a diameter of 20 m that were replicated three times. CO_2 enrichment was carried out with a FACE system constructed according to the Brookhaven National Laboratory design (Lewin et al., 1992). CO_2 enrichment started at the four- and three-leaf stage on 31 March 2014 and 12 March 2015 respectively. CO_2 enrichment took place during the daytime hours and was interrupted when air temperature fell below 5 °C or wind speed exceeded 6 $m\,s^{-1}$. The N treatments were conducted in rectangular subplots with 3 m × 5 m that were randomly established within the CO_2 plots. The quantity of N fertilizer was 40 and 35 $kg\,N\,ha^{-1}$ for the deficient N treatment (Nd), 180 and 200 $kg\,N\,ha^{-1}$ for the adequate N treatment (Nad) and 320 and 320 $kg\,N\,ha^{-1}$ for the excess N treatment (Nex) in 2014 and 2015 respectively. N levels were slightly changed in the second year to strengthen the lack of nitrogen under Nd (Dier et al., 2019) and improve the grain crude protein concentration under Nad (Dier et al., 2020). In general, wheat cultivation was conducted under optimal water availability and according to local farm practice with adequate crop management and pesticide application. Detailed descriptions of the experiment can be found elsewhere (Dier et al., 2018, 2019; Manderscheid et al., 2018). Results of the treatment effects of CO_2 and N on the seasonal course of plant growth (e.g. GAI and biomass) and yield variables (grain number and single grain weight) have already been published (Dier et al., 2018). Readings of daily means of radiation and air temperature were provided from a nearby measuring station of the German Weather Service and used for the calculation of the photothermal quotient, that is the ratio between radiation and temperature.

2.2 | Determination of radiation interception by the whole canopy and the share per single grain

The fraction of global radiation intercepted by the canopy was estimated with the following equation:

$$f_{IR} = 1 - e^{-kGAI} \tag{1}$$

where f_{IR} is the fraction of radiation intercepted by the canopy, *k* is the extinction coefficient and GAI is the green area index. The extinction coefficient was assumed to be 0.58 according to Shearman et al. (2005). f_{IR} per single grain (f_{IR}G) was calculated by dividing f_{IR} by grain number per m^2 ground area (GN):

$$f_{IRG} = \frac{1 - e^{-kGAI}}{GN} \tag{2}$$

where data on GAI and grain number were taken from Dier et al. (2018).

2.3 | Determination of stem reserves per ground area and single grain

Four destructive harvests were taken over the season and total plant biomass was separated into different fractions including the stem fraction, which also contains leaf sheath (Dier et al., 2018). Stem reserves per ground area (SR) ($g\,m^{-2}$) were calculated as the product of stem biomass (SBM) ($g\,m^{-2}$) and the concentration of water-soluble carbohydrates in stem (WSC) ($mg\,g^{-1}$). Stem reserves per single grain (SRG) (mg) were calculated using grain number (GN) (m^{-2}):

$$SRG = \frac{SBM * WSC}{GN} \tag{3}$$

where WSC was determined according to Manderscheid et al. (2009).

2.4 | Analysis of single grain growth

From each N subplot, 10 and 15 main stem ears were harvested every week from anthesis to maturity in 2014 and 2015 respectively. The ears were dried to constant weight at 105°C and threshed by hand. Individual grain weight was then determined from the grain number and the grain weight of the sample. Single grain growth was described by logistic regression using the following equation:

$$y = \frac{a}{1 + be^{-cx}} \tag{4}$$

where *y* is the individual grain weight, *x* is the thermal time from anthesis up to maturity (base temperature 0°C), *a* is the asymptotic parameter for the final grain weight and *b* and *c* are parameters related to the rate and duration of grain filling. The parameters *a*, *b* and *c* were estimated using the SAS (version 9.4) procedure NLIN to obtain the least sum-of-squares estimates. According to the calculated logistic growth curve, the grain filling duration was calculated as the thermal time from the anthesis up to the 95% value of the parameter a. The rate of grain filling was calculated by dividing the 95% value of the parameter a by the duration of grain filling.

2.5 | Statistical analysis

F-tests were implemented with SAS (version 9.4) proc mixed and subsequent mean comparisons were implemented with SAS proc glimmix as already described (Dier et al., 2019).

Analysis of covariance of the effect of e[CO_2] and grain number on f_{IR}G and SRG and the effect of e[CO_2] and f_{IR}G and SRG, respectively, on grain growth parameters was implemented by sequential *F*-tests with SAS proc mixed. The interaction effect was analysed by comparing the full model with the model without the interaction effect. The main effects were analysed by comparing the model without the interaction effect with the model further reduced by the main effect to be tested.

3 | RESULTS

3.1 | Environmental conditions

Figure 1 shows the daily means of the photothermal quotient (PQ) during the grain filling period. The PQ ranged from 0.1 to 1.0 and the average PQ over the whole period (0–45 days after anthesis) was increased in 2014 (0.56) compared with 2015 (0.53). Similarly, the slope of the lines of the relationship between accumulated photosynthetically active radiation and temperature was slightly increased in the second half of the grain filling in 2014 compared with 2015.

3.2 | Effects of CO_2 and N treatments on source sizes for grain growth

3.2.1 | Source sizes referred to ground area

Stem reserves of water-soluble carbohydrates per m^2 ground area (SR) at anthesis were generally similar under N deficiency (Nd) with 128 $g\,m^{-2}$ and under adequate N supply (Nad) with 122 $g\,m^{-2}$, but further rises in excess N supply (Nex) significantly ($p < .001$) decreased SR at anthesis to 64 $g\,m^{-2}$ (Tables 1 and 2). However, in 2014, the SR were significantly ($p < .001$) larger under Nd compared with Nad, but in 2015, it was the opposite. e[CO_2] tended to increase SR at anthesis under Nd and Nad by 13% and 14%, respectively, and tended to decrease the SR under Nex by 14%, but none of the effects were significant.

Figure 2 shows SR at the flag leaf stage, anthesis, milk-ripe stage and maturity. SR were highest at the milk-ripe stage with on average 143 $g\,m^{-2}$ and were almost completely used at maturity so that on average only 7 $g\,m^{-2}$ remained at this stage. N supply influenced the temporal variation of SR and the increase from anthesis to milk-ripe stage was much larger under Nex than under Nd. SR were significantly ($p < .001$) larger in 2015 compared with 2014.

The fraction of global radiation intercepted by the canopy (f_{IR}) at anthesis amounted to 63% up to 97%. Rising N supply significantly ($p < .001$) increased f_{IR} at anthesis from a mean of 67% under Nd to 92% under Nad and 96% under Nex (Tables 1 and 2). e[CO_2] significantly ($p < .05$) increased the f_{IR} in all N levels by on average 3% to 4%.

f_{IR} was very similar at the flag leaf stage, anthesis and milk-ripe stage, even if in the latter stage f_{IR} was somewhat reduced under Nd in 2014 (Figure 3).

3.2.2 | Source sizes referred to single grain

Grain number ranged from 10,000 to 21,000 m^{-2} in 2014 and from 9000 to 24,000 m^{-2} in 2015 and was strongly increased by rising N supply and by e[CO_2] by 8% to 12% (Dier et al., 2018).

SR per single grain (SRG) at anthesis was highest under Nd with on average 12.6 mg (≥11.8 mg) and decreased significantly ($p < .001$) to 6.1 mg (≥4.9 mg) and 3.0 mg (>2.5 mg) under Nad and Nex respectively (Tables 1 and 3). e[CO_2] did not significantly influence SRG at anthesis, but tended to decrease the SRG under Nex. Especially in 2015, SRG was decreased by 34% under e[CO_2].

f_{IR} per single grain (f_{IR}G) at anthesis was highest under Nd (6.7% × 0.001) and decreased significantly ($p < .001$) to 4.7% × 0.001 under Nad and 4.5% × 0.001 under Nex. Moreover, f_{IR}G was significantly ($p < .05$) decreased (by 8%) under Nex compared with Nad in 2015. e[CO_2] resulted in a significant decrease ($p < .05$) of f_{IR}G at anthesis on average by 4% to 9%.

There was a strong ($p < .001$) negative linear relationship between SRG or f_{IR}G at anthesis and grain number (Figure 4), indicating that the treatment effects on grain number determine the assimilate source size per grain. Moreover, the e[CO_2] line of the relationship between SRG and grain number ran significantly ($p < .05$) above the a[CO_2] line so that plants had about 1 mg more stem reserves per grain under e[CO_2].

3.3 | Effects of CO_2 and N treatments on single grain growth

Single grain growth was influenced by year (Tables 1 and 4), and GFR was significantly increased by 10% and GFD was significantly reduced by 7% in 2014 compared with 2015. N supply affected grain growth with significantly ($p < .001$) larger GFR under Nd (70 µg °Cd^{-1}) and Nad (73 µg °Cd^{-1}) than under Nex (64 µg °Cd^{-1}). Moreover, GFD increased with rising N supply from 635 °Cd under Nd to 674 °Cd under Nad and 711 °Cd under Nex, being significant for the increase from Nd to Nad ($p < .05$). The 95% value of the 'a' parameter of Equation (2) was significantly larger under Nad than under Nex (44.7 mg vs. 42.7 mg).

e[CO_2] increased GFR on average by 5% under Nad and 9% under Nex. This was not significant in the statistical analysis with all N treatments, but if Nd level was excluded, then it became significant at the $p < .1$ level.

3.4 | Relationship between the source size per grain and single grain growth

The average number of SRG was much higher at Nd (12.6 mg) than at Nex (2.9 mg). Values of single grain weight already published by Dier et al. (2018) differed only slightly (Nd: 45.7 mg and Nex: 41.4 mg). Consequently, under high N supply, the role of SRG was minor, and most assimilates for grain growth were provided by photosynthesis

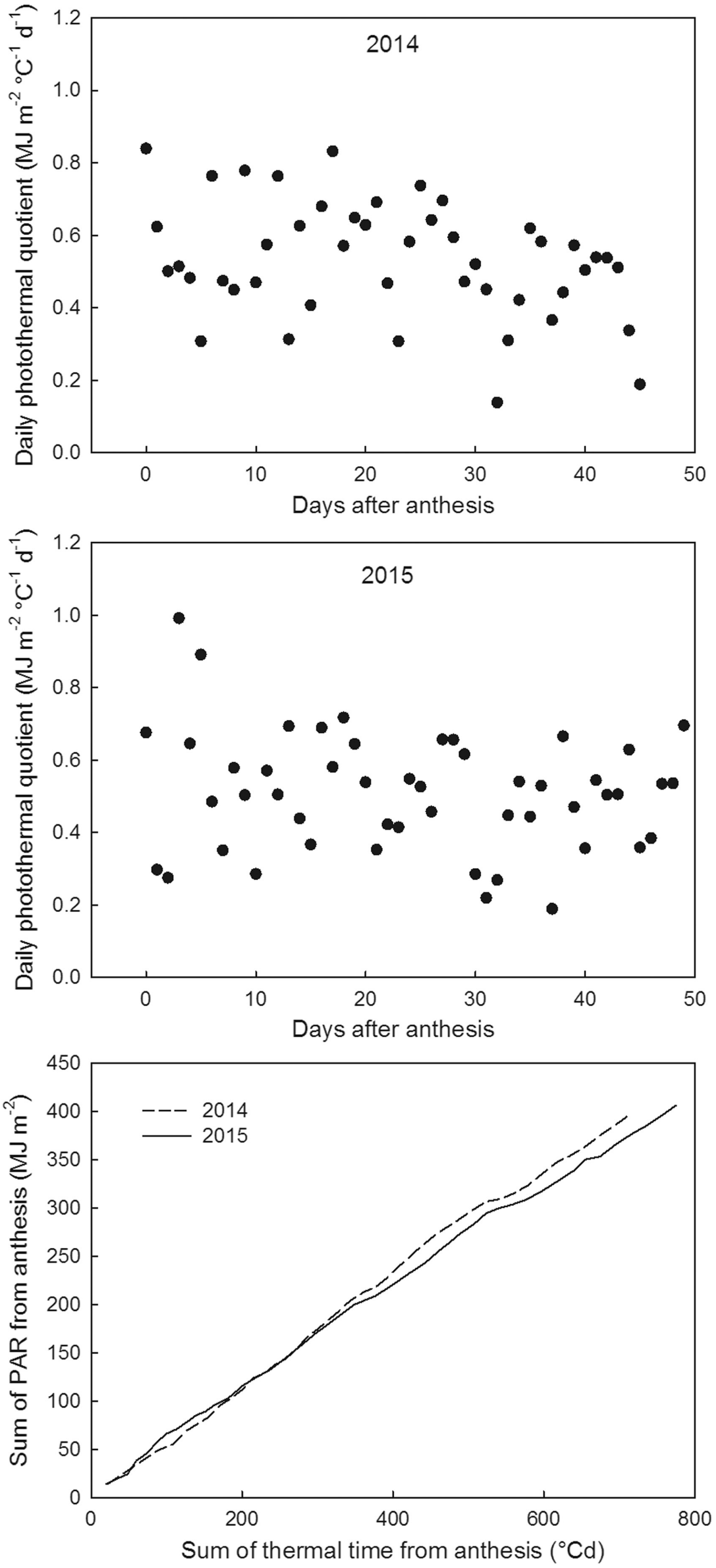


FIGURE 1 Daily variation in photothermal quotient during grain filling in 2014 and 2015 and sum of photosynthetically active radiation (PAR) plotted against the sum of thermal time during grain filling.

TABLE 1 *F* -test result of the effect of the two CO$_2$ and three N levels on stem reserves per unit ground area (SR) and single grain (SRG) at anthesis, on a fraction of intercepted global radiation by the canopy, referred to the unit ground area (f_{IR}) and single grain (f_{IR}G) at anthesis, on grain filling rate (GFR) and duration (GFD) and on the 95% value of the parameter 'a' of Equation (2)

Variable	CO$_2$	N	Year (Y)	CO$_2$ × N	CO$_2$ × Y	N × Y	CO$_2$ × N × Y
SR (g m^{-2})	ns	***	**	*	ns	***	ns
f_{IR} (%)	*	***	*	ns	ns	*	ns
SRG (mg)	ns	***	*	ns	ns	(*)	ns
f_{IR}G (% × 0.001)	*^{1}	***^{1}	ns^1	ns^1	ns^1	**^{1}	ns^1
GFR (μg °Cd^{-1})	ns	***	**	ns	ns	ns	ns
GFD (°Cd)	ns	**	*	ns	ns	ns	ns
95% of a (mg)	ns	*	ns	ns	ns	ns	ns
GFR (μg °Cd^{-1})$^{\#}$	(*)	*	**	ns	ns	ns	ns
GFD (°Cd)$^{\#}$	ns	(*)	*	ns	ns	ns	ns
95% of a* (mg)$^{\#}$	ns	*	ns	ns	ns	ns	ns

Note: (*)$p < .1$, *$p < .05$, **$p < .01$ and ***$p < .001$.

^{1}Data were log-transformed to ensure variance homogeneity and normal distribution of the residual error.

$^{\#}$The N deficiency level was excluded from the analysis.

TABLE 2 Effect of the two CO$_2$ and three N treatments on stem reserves (SR) and on fraction of intercepted global radiation by the canopy (f_{IR}) referred to the unit ground area at anthesis in 2 years (2014 and 2015)

	2014			2015		
N level	a[CO$_2$]	e[CO$_2$]	Δ (%)	a[CO$_2$]	e[CO$_2$]	Δ (%)
SR (g m^{-2})						
Nd	119 a	146 a	22	121 b	124 b	3
Nad	83.8 b	98.1 b	17	144 a	160 a	11
Nex	54.1 c	54.0 c	0	84.6 c	61.5 c	−27
f_{IR} (%)						
Nd	68.9 b	72.6 b	5	63.2 c	64.7 c	2
Nad	90.3 a	94.6 a	5	91.5 b	92.4 b	1
Nex	94.9 a	96.5 a	2	94.7 a	96.7 a	2

Note: Data shown are the mean values ($n = 3$) and the percentage effect of e[CO$_2$] (Δ (%)). Small letters indicate significant differences ($p < .05$) among the marginal N means (mean over the two CO$_2$ treatments) within each year.

during grain filling. In the Nd treatment, there was no positive relationship between the SRG and GFR (*data not shown*) and between the f_{IR}G and GFR (Figure 5a), indicating that grain growth was sink limited under this N level. However, significant positive relationships between the f_{IR}G and GFR ($p < .01$) or TGW ($p < .001$) (Figure 5a,c) were found based on the Nad and Nex treatment. Moreover, in both relationships, the line of the e[CO$_2$] treatment ran significantly (GFR, $p < .01$; TGW, $p < .001$) above the line of the a[CO$_2$] treatment so that GFR was about 10 μg °Cd^{-1} and TGW about 5 g larger at a certain f_{IR}G under e[CO$_2$]. GFD decreased significantly ($p < .01$) with increasing f_{IR}G, while the base level was increased under e[CO$_2$] (Figure 5b).

4 | DISCUSSION

The first objective of the present study was to determine the effect of N and CO$_2$ supply on the assimilate source size per grain at anthesis, defined as stem reserves per grain (SRG) and the fraction of incident radiation intercepted by the canopy per grain (f_{IR}G). The second objective was to test to what degree grain growth (i.e. GFR, GFD and TGW) is affected by variation in the source size per grain at anthesis.

4.1 | SRG and f_{IR}G decrease with the increase in grain number controlled by N and CO$_2$ supply

The data of the present study clearly demonstrate that N and CO$_2$ supply affect the source size per grain at anthesis via their effects on grain number. As expected, increased N and CO$_2$ supply increased f_{IR} (Table 2) but decreased f_{IR}G (Table 3). Both effects were associated with an increase in the number of tillers per unit of ground area (Dier et al., 2018). GAI at anthesis varied between 1.7 and 2.3 under Nd, 4.1 and 5.0 under Nad and 5.2 and 5.9 under Nex, and e[CO$_2$] increased GAI on average by about 10%. However, the effects of N and CO$_2$ supply on GAI and the associated f_{IR} were less than that on grain number, resulting in a decrease in f_{IR}G. The decrease in SRG by increasing N supply could be explained by increased assimilate allocation to ear growth relative to SR accumulation (Schnyder, 1993).

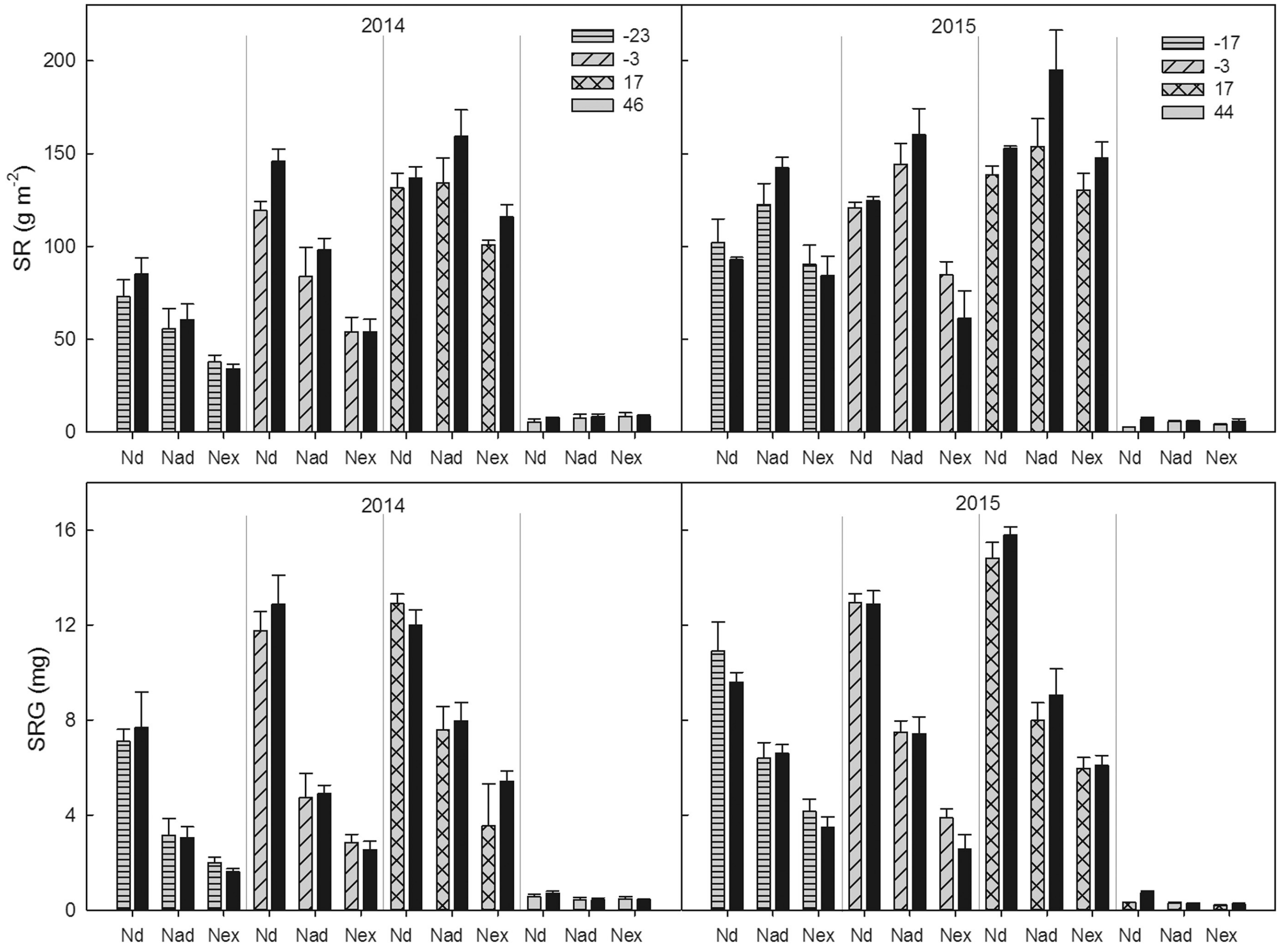


FIGURE 2 Seasonal change in stem reserves referred to the unit ground area (SR) and grain (SRG) in 2 years (2014 and 2015). Each bar represents the mean value (±SEM, $n = 3$). The grey bar represents the a[CO_2] and the black bar the e[CO_2] treatment. The numbers in the legend indicate the days after anthesis.

In this connection, SR were decreased under Nex compared with Nd and Nad, which is in line with Knapp et al. (1987) and partly with Valluru et al. (2011).

There was a strong negative linear relationship between grain number and $f_{IR}G$ or SRG (Figure 4). The negative relationship of $f_{IR}G$ indicates that single grain growth becomes more and more source limited in terms of photosynthesis when grain number increases. SR can supply the grain with assimilates under adverse conditions for photosynthesis (Asseng et al., 2017). However, SRG at anthesis decreased strongly with grain number from 13 to 3 mg, indicating that the plant has less potential to stabilize early grain filling by SR at high grain numbers. Under e[CO_2], the plant had about 1 mg more SRG at a given grain number, but under Nex, SRG was not increased but decreased by e[CO_2] due to the grain number effect on SRG.

The contribution of SRG to final single grain weight ranged from ≥3 mg at Nex to ≥5 mg at Nad and ≥ 12 mg at Nd in the present study. This is in agreement with other wheat field studies, in which the contribution of stem reserves to final grain weight ranged from 2 to 16 mg (Dreccer et al., 2009; Ehdaie et al., 2008; Lynch et al., 2017).

$f_{IR}G$ is a measure of the fraction of photosynthetically active area per grain present at anthesis and this variable was strongly influenced by N fertilization. Besides $f_{IR}G$, the amount of assimilates per grain, depends on the environmental conditions (irradiance, temperature and atmospheric CO_2 concentration) and photosynthetic capacity of green tissue, which determine the photosynthetic activity of this area and is usually not strongly influenced by N supply.

Stem reserves are used very efficiently in grain filling and losses associated with metabolism are low (Gebbing et al., 1999). Thus, the relationship of SRG measured in this study to single grain weight published in Dier et al. (2018) can be taken as a measure of the relative contribution of SRG to single grain weight. SRG amounted on average to 12.6 mg at Nd, 6.1 mg at Nad and 2.9 mg at Nex. The corresponding values of single grain weight amounted to 45.7 mg, 45.4 and 41.4 mg. Consequently, the relative contribution of SRG to final grain weight ranged from 7% at Nex to 28% at Nd. Besides C from stem reserves, protein-C in vegetative plant parts and especially in leaf blades is also an important source for grain filling (Gebbing et al., 1999). However, the amount of remobilized C from proteins was found to be smaller than C from water-soluble carbohydrates.

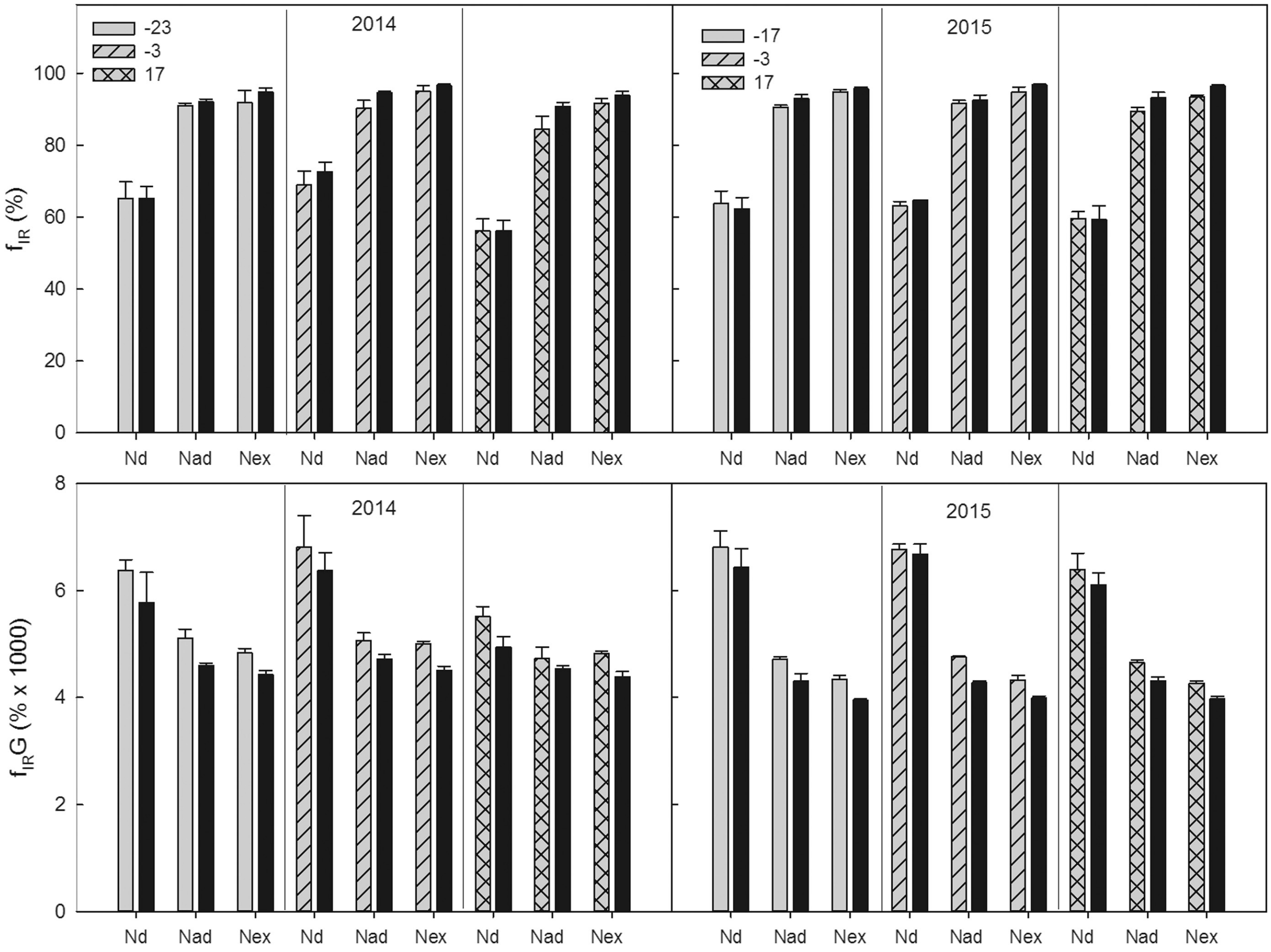


FIGURE 3 Seasonal change in the fraction of intercepted radiation by the canopy referred to the unit ground area (f_{IR}) and grain (f_{IR}G) in 2 years (2014 and 2015). Each bar represents the mean value (±SEM, *n* = 3). The grey bar represents the a[CO_2] and the black bar the e[CO_2] treatment. The numbers in the legend indicate the day after anthesis.

For a rough estimation of total C remobilization, the contribution of SRG-C and protein-C can be assumed to be the same size. With this assumption, remobilization of C from water-soluble carbohydrates and proteins contributed 56% and 14% of the assimilates for grain filling under Nd and Nex respectively. Consequently, 44% and 86% of the assimilates for grain filling came from f_{ir}G under Nd and Nex respectively. This contrast with the finding that f_{IR}G was 70% larger under Nd than Nex.

4.2 | Grain growth seems to depend on source size below a specific limit

A clear effect of N supply on GFR, GFD and TGW was observed in the present study (Table 4) and in previous studies with wheat (Elia et al., 2018; Yan et al., 2019) and barley (Manderscheid et al., 2009). e[CO_2] influenced GFR only under high N supply which corresponds to other studies (Li et al., 2000; Manderscheid et al., 2009). The decrease in GFR and increase in GFD in 2015 compared with 2014 can be attributed to differences in grain number (Dier et al., 2018) and thus differences in the source size per grain as far as the Nad and Nex treatment is concerned. In addition, the slight difference in the mean photothermal quotient during grain filling between 2014 and 2015 might also have contributed to the year effect on GFR and GFD.

Our data show that single grain growth was controlled by source size if f_{IR}G was <5.5 ×0.001% and <4.7 ×0.001% under a[CO_2] and e[CO_2] respectively (Figure 5). Above these values, grain growth was sink limited. The difference between the CO_2 treatments reflects the effect of e[CO_2] on the activity of photosynthesis. In addition to the decrease in GFR, there was an increase in GFD with decreasing f_{IR}G. This result suggests that grain sink demand results in increased GFD when grain growth is limited by the source size and this causes stabilization of TGW. It has been assumed that GFD is controlled almost exclusively by temperature (Miralles & Slafer, 1999; Otero et al., 2021), but in the present study air temperature did not differ among the N treatments and the effect of e[CO_2] on temperature is negligible (Burkart et al., 2011). Therefore, GFD seems to depend on source size and to compensate for the decrease in GFR. For example, values of TGW observed in 2014 under Nd (46.1 g) and Nad (45.9 g) at a[CO_2] (Dier et al., 2018)

TABLE 3 Effect of the two CO_2 and three N treatments on SR per grain (SRG) and f_{IR} per grain ($f_{IR}G$) at anthesis in 2 years (2014 and 2015)

	2014			2015		
N level	**a[CO_2]**	**e[CO_2]**	**Δ (%)**	**a[CO_2]**	**e[CO_2]**	**Δ (%)**
SRG (mg)						
Nd	11.8 a	12.9 a	9	12.9 a	12.9 a	−1
Nad	4.7 b	4.9 b	3	7.5 b	7.4 b	−1
Nex	2.8 c	2.5 c	−11	3.9 c	2.5 c	−34
$f_{IR}G$ (% × 0.001)						
Nd	6.8 a	6.4 a	−6	6.8 a	6.7 a	−1
Nad	5.1 b	4.7 b	−7	4.8 b	4.3 b	−10
Nex	5.0 b	4.5 b	−10	4.3 c	4.0 c	−8

Note: Data shown are the mean values ($n = 3$) and the percentage effect of e[CO_2] (Δ (%)). Small letters indicate significant differences ($p < .05$) among the marginal N means (mean over the two CO_2 treatments) within each year.

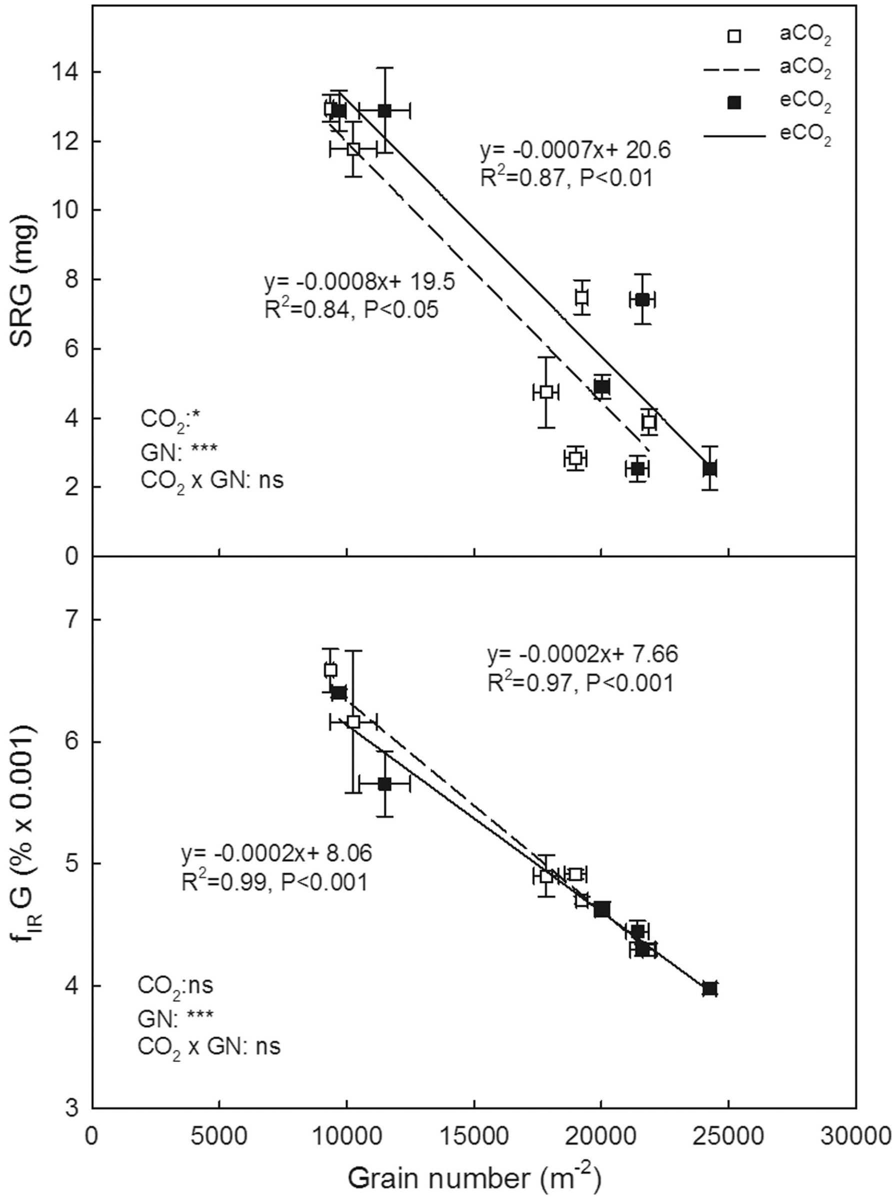


FIGURE 4 Linear regression of grain number on stem reserves per grain (SRG) and on fraction of intercepted global radiation by the canopy referred to single grain ($f_{IR}G$) at anthesis. Each data point represents the mean value (±SEM, $n = 3$) for each CO_2 × N × Year combination. Each diagram includes the result of the analysis of covariance where the CO_2 and grain number (GN) effect was tested. *$p < .05$ and ***$p < .001$.

TABLE 4 Effect of the two CO_2 and three N treatments on individual grain growth in 2 years (2014 and 2015)

	2014			2015		
N level	a[CO_2]	e[CO_2]	Δ (%)	a[CO_2]	e[CO_2]	Δ (%)
GFR (µg °Cd^{-1})						
Nd	77.3 a	75.6 a	−2.3	70.5 a	68.5 a	−2.8
Nad	70.8 a	74.4 a	5.2	65.5 a	69.1 a	5.3
Nex	65.9 b	71.1 b	7.9	55.9 b	62.3 b	11.3
GFD (°Cd)						
Nd	610 b	630 b	3.2	641 b	660 b	3.0
Nad	668 a	636 a	−4.8	703 a	691 a	−1.7
Nex	698 a	662 a	−5.1	732 a	749 a	2.3
95% of a (mg)						
Nd	44.7 ab	45.0 ab	0.6	42.9 ab	42.8 ab	−0.3
Nad	44.9 a	45.0 a	0.1	43.8 a	45.3 a	3.3
Nex	43.5 b	44.7 b	2.7	38.9 b	43.6 b	12.3

Note: Data shown are the mean values ($n = 3$) and the percentage effect of e[CO_2] (Δ (%)) of grain filling rate (GFR) and duration (GFD) and on 95% of the parameter a of Equation (2). Small letters indicate significant differences ($p < .05$) among the marginal N means (mean over the two CO_2 treatments) within each year.

were similar, although $f_{IR}G$ was 40% less in Nad than Nd. Longer GFD in Nad compared with Nd was coupled with a longer green area duration (Dier et al., 2019) and thus a longer period of photosynthetic activity. However, there is probably an upper limit on GFD and thus an upper limit on the compensation of a small source size as suggested by the lower 95% value of the parameter a and the lower TGW under Nex (38.5 g) compared with Nad (44.1 g) and Nd (45.4 g) in 2015 (Dier et al., 2018).

4.3 | Reflections on the variation in the CO_2 effect on TGW

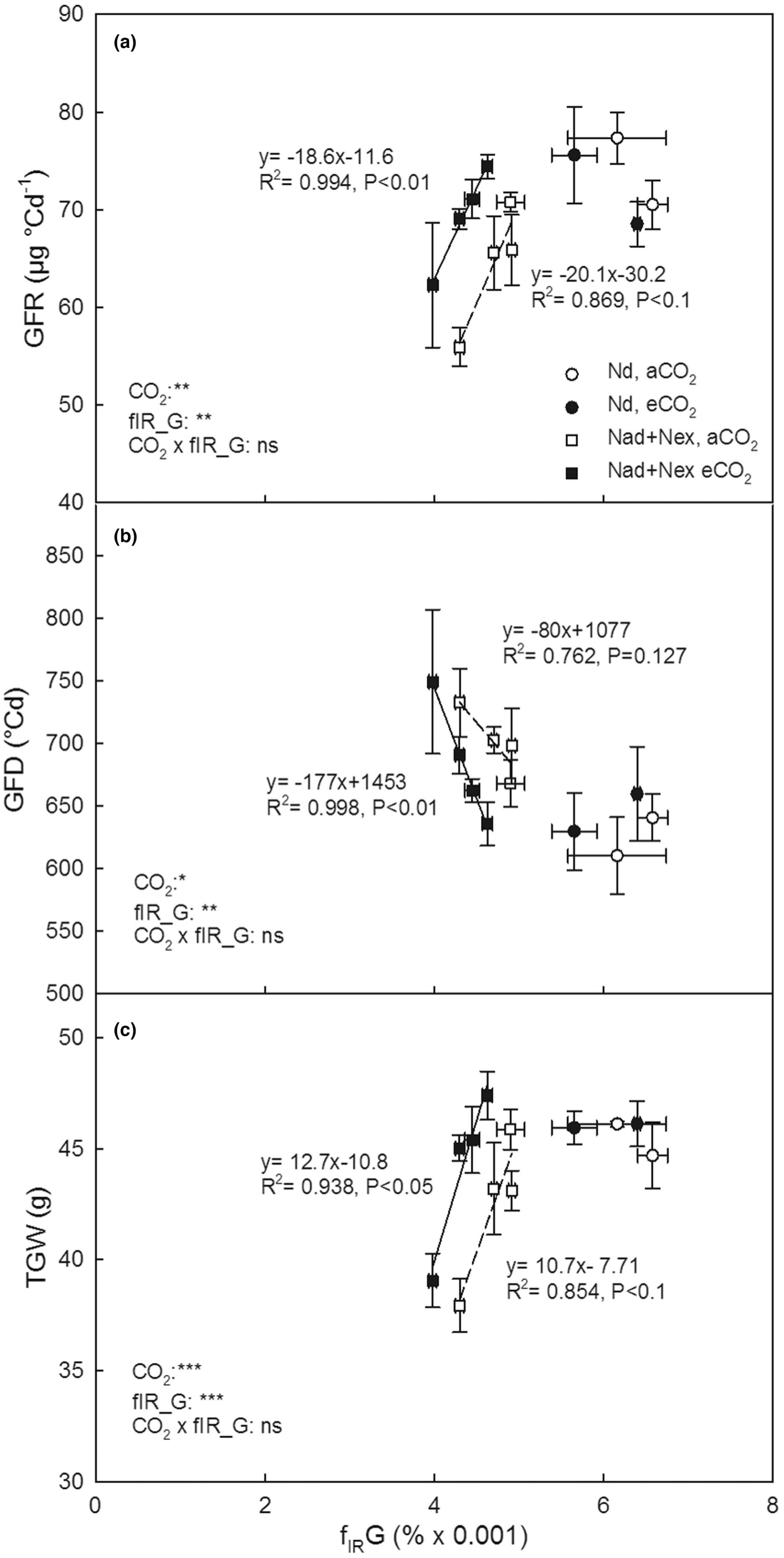
Under low N supply, grain growth is sink limited and therefore almost unaffected by e[CO_2] as shown herein for wheat and in another study for barley (Manderscheid et al., 2009). With rising N supply, grain growth becomes increasingly source limited and increased by e[CO_2]. However, a large grain number is associated with a larger proportion of grains with low potential weight (Acreche & Slafer, 2006). In the present study (Dier et al., 2018), grain number per m^2 varied between 9342 and 11,488 under Nd, 17,827 and 21,607 under Nad and 18,977 and 24,254 under Nex. Moreover, e[CO_2] increased grain number less under Nd (+8%) than under Nad or Nex (+12%). This might be an explanation why e[CO_2] increased TGW less under Nex (3%) than Nad (4.2%) in 2015, although grain growth was more source limited under Nex due to very high grain number (Dier et al., 2018). Another reason for that could be a short-term occurrence of assimilates shortage at early grain filling. $f_{IR}G$ at the start of grain filling was lowest under Nex in 2015 and SRG was lowest under Nex and e[CO_2] with 2.5 mg. Additionally, the daily photothermal quotient representing the weather effect on the source in relation to sink activity was extremely low over the 2 days just after anthesis in 2015 (Figure 1). However, at early grain filling, the maximum grain volume that determines final grain weight is set, and thus assimilate shortage at early grain filling has a higher impact on TGW compared with later stages (Borrás et al., 2004; Harcha & Calderini, 2014). Consequently, assimilate shortage just after anthesis might explain why e[CO2] increased TGW less under Nex (from 37.9 g to 39.1 g) than under Nad (from 43.2 g to 45.0 g) and the very low TGW under Nex in 2015 (38.5 g averaged over both CO_2 treatments) than in 2014 (44.3 g) (Dier et al., 2018). In a 2-year field study, large differences in TGW were also discussed to result from variation in SRG, which amounted to 6 and 2 mg when TGW was high and low respectively (Lynch et al., 2017). Moreover, an assimilate shortage directly after anthesis might be associated with a decrease in grain number under high N supply by 10 days warming treatment from 10 days after anthesis (Elia et al., 2018).

Old crop cultivars, when grown under ample N supply, possibly have a larger source size per grain at anthesis than modern ones due to differences in grain number and stem biomass (Manderscheid & Weigel, 1997). Thus, these are probably less vulnerable to assimilate shortage around anthesis. This might be an explanation for higher yield response to e[CO_2] of old compared to modern wheat cultivars (Manderscheid & Weigel, 1997; Ziska et al., 2004), which was possibly associated with less abortion of grains in old cultivars due to more SRG around anthesis (Manderscheid & Weigel, 1997).

4.4 | N status influences the response of grain growth to unfavourable conditions

A model that the N status of the plant determines the source size per grain for grain filling and that a low source size per grain is partly compensated by an increased GFD is supported by several studies

FIGURE 5 Linear regression of fIRG on (a) grain filling rate (GFR), (b) grain filling duration (GFD) and (c) thousand-grain weight (TGW). Each data point represents the mean value (±SEM, *n* = 3) for each $CO_2 \times N \times$ Year combination. Each diagram includes the result of the analysis of covariance where the effect of CO_2 and $f_{IR}G$ was tested. The analysis of covariance bases on the Nad and Nex treatment. **p* <.05, ***p* <.01 and ****p* <.001.



in which the source size has been manipulated. Those manipulations were (i) a decrease in interception of incident radiation by shading (Asseng et al., 2017), (ii) a decrease in the number of days for grain filling by warming (Elia et al., 2018) or (iii) a decrease in GAI during grain filling by fungal infection (Parker et al., 2004). In the study of Asseng et al. (2017), a decrease in grain yield by shading at grain filling was less than predicted by the N wheat model, and this might result from an increase in GFD that has partially compensated for the decrease in GFR. In the study of Elia et al. (2018), TGW was strongly decreased by warming under high than under low N supply and this might be due to an N×Warming interaction on GFD. Under high N, GFD might be already close to the limit, and warming hardly changed the thermal time for grain filling but decreased duration in days. However, under low N, GFD was possibly below the limit and a decreased source size by warming was possibly compensated by increased GFD in thermal time. Another recent study showed that GFD measured in thermal time increased under warming and thus mitigated the adverse effect of high temperature on TGW (Luig et al., 2016). In the same study, GFR and GFD were found to be related to the mean photothermal quotient of the grain filling period. Finally, such a model in which grain number defines the source size per grain would explain why wheat cultivars with a low-yield potential are less affected by the fungal disease during grain filling than high-yield cultivars (Parker et al., 2004). In addition to a lower potential of GFD to compensate for a low GFR, the risk of assimilate shortage at early grain filling with a negative impact on TGW (Borrás et al., 2004) and possibly on grain number (Elia et al., 2018) seems higher under high than low N supply. In this context, it can be speculated whether heat stress at anthesis is also compounded by high nitrogen fertilization if assimilate shortage is involved in grain number loss (Farooq et al., 2011).

5 | CONCLUSION

Based on the effect of N supply on source size per grain at anthesis found in this study, plants grown under high N might be more affected by favourable (e[CO_2]) and unfavourable conditions (shading, warming or fungal infection) during grain filling than plants grown under low N. Thus, N management can be used as a tool to adjust wheat growth to either favourable or unfavourable conditions during grain filling as already suggested by Slafer and Savin (2018). A slight increase in stem reserves at anthesis by plant breeding might reduce the risk of assimilate shortage during early grain filling and improve plant performance under high N fertilization when combined with e[CO_2] or heat waves as demanded in a recent paper (Senapati & Semenov, 2020).

AUTHOR CONTRIBUTIONS

Remy Manderscheid: Conceptualization; funding acquisition; investigation; project administration; supervision; writing – original draft; writing – review and editing. **Markus Dier:** Investigation; methodology; writing – original draft; writing – review and editing.

ACKNOWLEDGEMENT

This work was partly funded by the Deutsche Forschungsgemeinschaft (DFG) (grant nos. MA-1736/3-1 and MA-1736/5-1). Open Access funding enabled and organized by Projekt DEAL.

DATA AVAILABILITY STATEMENT

The data that support the findings of this study are openly available at http://doi.org/10.18174/odjar.v6i0.16397.

ORCID

Remy Manderscheid https://orcid.org/0000-0001-6975-0238

REFERENCES

Acreche, M. M., & Slafer, G. A. (2006). Grain weight response to increases in number of grains in wheat in a Mediterranean area. *Field Crops Research*, *110*, 98–105.

Alonso, M. P., Abbate, P. E., Mirabella, N. E., Merlos, F. A., Panelo, J. S., & Pontaroli, A. C. (2018). Analysis of sink/source relations in bread wheat recombinant inbred lines and commercial cultivars under a high yield potential environment. *European Journal of Agronomy*, *93*, 82–87.

Asseng, S., Kassie, B. T., Labra, M. H., Amador, C., & Calderini, F. (2017). Simulating the impact of source-sink manipulations in wheat. *Field Crops Research*, *202*, 47–56.

Blandino, M., Badeck, F. W., Giordano, D., Marti, A., Rizza, F., Scarpino, V., & Vaccino, P. (2020). Elevated CO_2 Impact on Common Wheat (Triticum aestivum L.) Yield, Wholemeal Quality, and Sanitary Risk. *Journal of Agricultural and Food Chemistry*, *68*(39), 10574–10585.

Borrás, L., Slafer, G. A., & Otegui, M. E. (2004). Seed dry weight response to source–sink manipulations in wheat, maize and soybean: A quantitative reappraisal. *Field Crops Research*, *86*(2–3), 131–146.

Burkart, S., Manderscheid, R., Wittich, K. P., Löpmeier, F. J., & Weigel, H. J. (2011). Elevated CO_2 effects on canopy and soil water flux parameters measured using a large chamber in crops grown with free-air CO_2 enrichment. *Plant Biology*, *13*, 258–269.

Calderini, D. F., & Reynolds, M. P. (2000). Changes in grain weight as a consequence of de-graining treatments at pre-and post-anthesis in synthetic hexaploid lines of wheat (Triticum durum x T. tauschii). *Functional Plant Biology*, *27*(3), 183–191.

Calderini, D. F., Savin, R., Abeledo, L. G., Reynolds, M. P., & Slafer, G. A. (2001). The importance of the period immediately preceding anthesis for grain weight determination in wheat. *Euphytica*, *119*(1), 199–204.

Dier, M., Hüther, L., Schulze, W. X., Erbs, M., Köhler, P., Weigel, H. J., Manderscheid, R., & Zörb, C. (2020). Elevated atmospheric CO_2 concentration has limited effect on wheat grain quality regardless of nitrogen supply. *Journal of Agricultural and Food Chemistry*, *68*(12), 3711–3721.

Dier, M., Sickora, J., Erbs, M., Weigel, H. J., Zörb, C., & Manderscheid, R. (2018). Decreased wheat grain yield stimulation by free air CO_2 enrichment under N deficiency is strongly related to decreased radiation use efficiency enhancement. *European Journal of Agronomy*, *101*, 38–48.

Dier, M., Sickora, J., Erbs, M., Weigel, H. J., Zörb, C., & Manderscheid, R. (2019). Positive effects of free air CO_2 enrichment on N remobilization and post-anthesis N uptake in winter wheat. *Field Crops Research*, *234*, 107–118.

Dreccer, M. F., van Herwaarden, A. F., & Chapman, S. C. (2009). Grain number and grain weight in wheat lines contrasting for stem water soluble carbohydrate concentration. *Field Crops Research*, *112*, 43–54.

Ehdaie, B., Alloush, G. A., & Waines, J. G. (2008). Genotypic variation in linear rate of grain growth and contribution of stem reserves to grain yield in wheat. *Field Crops Research*, *106*, 34–43.

Elia, M., Slafer, G. A., & Savin, R. (2018). Yield and grain weight responses to post-anthesis increases in maximum temperature under field grown wheat as modified by nitrogen supply. *Field Crops Research*, *221*, 228–237.

FAOSTAT. (2017). *Suite of food security indicators*. Food and Agriculture Organization of the United Nations http://www.fao.org/faostat/en/#data/FS

Farooq, M., Bramley, H., Palta, J. A., & Siddique, K. H. M. (2011). Heat stress in wheat during reproductive and grain-filling phases. *Critical Reviews in Plant Sciences*, *30*, 491–507.

Fernando, N., Panozzo, J., Tausz, M., Norton, R. M., Fitzgerald, G. J., Myers, S., Walker, C., Stangoulis, J., & Seneweera, S. (2012). Wheat grain quality under increasing atmospheric CO_2 concentrations in a semi-arid cropping system. *Journal of Cereal Science*, *56*(3), 684–690.

Fernando, N., Panozzo, J., Tausz, M., Norton, R. M., Neumann, N., Fitzgerald, G. J., & Seneweera, S. (2014). Elevated CO_2 alters grain quality of two bread wheat cultivars grown under different environmental conditions. *Agriculture, Ecosystems & Environment*, *185*, 24–33.

Fischer, R. A. (2011). Wheat physiology: A review of recent developments. *Crop and Pasture Science*, *62*(2), 95–114.

Gebbing, T., Schnyder, H., & Kühbauch, W. (1999). The utilization of pre-anthesis reserves in grain filling of wheat. Assessment by steady-state $^{13}CO_2/^{12}CO_2$ labelling. *Plant, Cell and Environment*, *22*, 851–858.

Harcha, C. I., & Calderini, D. F. (2014). Dry matter and water dynamics of wheat grains in response to source reduction at different phases of grain filling. *Chilean Journal of Agricultural Research*, *74*, 380–390.

Högy, P., Wieser, H., Köhler, P., Schwadorf, K., Breuer, J., Franzaring, J., & Fangmeier, A. (2009). Effects of elevated CO_2 on grain yield and quality of wheat: Results from a 3-year free-air CO2 enrichment experiment. *Plant Biology*, *11*, 60–69.

Knapp, J. S., Harms, C. L., & Volenec, J. J. (1987). Growth regulator effects on wheat culm nonstructural and structural carbohydrates and lignin. *Crop Science*, *27*(6), 1201–1205.

Lewin, K. F., Hendrey, G. R., & Kolber, Z. (1992). Brookhaven National Laboratory free-air carbon-dioxide enrichment facility. *Critical Reviews in Plant Sciences*, *11*, 135–141.

Li, A., Hou, Y., & Trent, A. (2001). Effects of elevated atmospheric CO_2 and drought stress on individual grain filling rates and durations of the main stem in spring wheat. *Agricultural and Forest Meteorology*, *106*(4), 289–301.

Li, A. G., Hou, Y. S., Wall, G. W., Trent, A., Kimball, B. A., & Pinter, P. J., Jr. (2000). Free-air CO_2 enrichment and drought stress effects on grain filling rate and duration in spring wheat. *Crop Science*, *40*(5), 1263–1270.

Luig, A., Manderscheid, R., Erbs, M., Ratjen, A. M., Weigel, H. J., & Kage, H. (2016). Towards a simple model for winter wheat's grain filling dynamics considering heat effects. In *International Crop Modelling Symposium* 15–17 March 2016, Berlin (pp. 312–313). Book of Abstracts.

Lynch, J. P., Doyle, D., McAuley, S., McHardy, F., Danneels, Q., Black, L. C., & Spink, J. (2017). The impact of variation in grain number and individual grain weight on winter wheat yield in the high yield potential environment of Ireland. *European Journal of Agronomy*, *87*, 40–49.

Manderscheid, R., Dier, M., Erbs, M., Sickora, J., & Weigel, H. J. (2018). Nitrogen supply–A determinant in water use efficiency of winter wheat grown under free air CO_2 enrichment. *Agricultural Water Management*, *210*, 70–77.

Manderscheid, R., Pacholski, A., Frühauf, C., & Weigel, H. J. (2009). Effects of free air carbon dioxide enrichment and nitrogen supply on growth and yield of winter barley cultivated in a crop rotation. *Field Crops Research*, *110*(3), 185–196.

Manderscheid, R., & Weigel, H. J. (1997). Photosynthetic and growth responses of old and modern spring wheat cultivars to atmospheric CO_2 enrichment. *Agriculture, Ecosystems and Environment*, *64*, 65–73.

Miralles, D. J., & Slafer, G. A. (1999). Wheat development. In E. H. Satorre & G. A. Slafer (Eds.), *Wheat: Ecology and physiology of yield determination* (pp. 14–43). Food Product Press.

Miralles, D. J., & Slafer, G. A. (2007). Sink limitations to yield in wheat: How could it be reduced? *Journal of Agricultural Science*, *145*(2), 139–149.

Otero, E. A., Miralles, D. J., & Benech-Arnold, R. L. (2021). Development of a precise thermal time model for grain filling in barley: A critical assessment of base temperature estimation methods from field-collected data. *Field Crops Research*, *260*, 108003.

Parker, S. R., Welham, S., Paveley, N. D., Foulkes, J., & Scott, R. K. (2004). Tolerance of septoria leaf blotch in winter wheat. *Plant Pathology*, *53*, 1–10.

Schnyder, H. (1993). The role of carbohydrate storage and redistribution in the source-sink relations of wheat and barley during grain filling—A review. *New Phytologist*, *123*, 233–245.

Senapati, N., & Semenov, M. A. (2020). Large genetic yield potential and genetic yield gap estimated for wheat in Europe. *Global Food Security*, *24*, 100340.

Serrago, R. A., Alzueta, I., Savin, R., & Slafer, G. A. (2013). Understanding grain yield responses to source–sink ratios during grain filling in wheat and barley under contrasting environments. *Field Crops Research*, *150*, 42–51.

Shearman, V. J., Sylvester-Bradley, R., Scott, R. K., & Foulkes, M. J. (2005). Physiological processes associated with wheat yield progress in the UK. *Crop Science*, *45*(1), 175–185.

Slafer, G. A., & Savin, R. (2018). Can N management affect the magnitude of yield loss due to heat waves in wheat and maize? *Current Opinion in Plant Biology*, *45*, 1–8.

Sofield, I., Evans, L. T., Cook, M. G., & Wardlaw, I. F. (1977). Factors influencing the rate and duration of grain filling in wheat. *Functional Plant Biology*, *4*(5), 785–797.

Tausz-Posch, S., Dempsey, R. W., Seneweera, S., Norton, R. M., Fitzgerald, G., & Tausz, M. (2015). Does a freely tillering wheat cultivar benefit more from elevated CO_2 than a restricted tillering cultivar in a water-limited environment? *European Journal of Agronomy*, *64*, 21–28.

Valluru, R., Link, J., & Claupein, W. (2011). Natural variation and morpho-physiological traits associated with water-soluble carbohydrate concentration in wheat under different nitrogen levels. *Field Crops Research*, *124*, 104–113.

Wardlaw, I. F., & Willenbrink, J. (1994). Carbohydrate storage and mobilisation by the culm of wheat between heading and grain maturity: The relation to sucrose synthase and sucrose-phosphate synthase. *Functional Plant Biology*, *21*(3), 255–271.

Wiegand, C. L., & Cuellar, J. A. (1981). Duration of grain filling and kernel weight of wheat as affected by temparature. *Crop Science*, *21*(1), 95–101.

Yan, S., Wu, Y., Fan, J., Zhang, F., Qiang, S., Zheng, J., Xiang, Y., Guo, J., & Zou, H. (2019). Effects of water and fertilizer management on grain filling characteristics, grain weight and productivity of drip-fertigated winter wheat. *Agricultural Water Management*, *213*, 983–995.

Ziska, L. H., Morris, C. F., & Goins, E. W. (2004). Quantitative and qualitative evaluation of selected wheat varieties released since 1903 to increasing atmospheric carbon dioxide: Can yield sensitivity to carbon dioxide be a factor in wheat performance? *Global Change Biology*, *10*, 1810–1819.

How to cite this article: Manderscheid, R., & Dier, M. (2023). Effect of N and CO_2 supply on source size per grain at anthesis and its relationship with grain growth in wheat. *Journal of Agronomy and Crop Science*, *209*, 273–285. https://doi.org/10.1111/jac.12624