



Mapping of QTLs for source and sink associated traits under elevated CO₂ in rice (*Oryza sativa* L.)

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Abstract

Rice source- and sink-associated traits are important for grain yield and are sensitive to environmental conditions. The continuing increase of CO₂ concentrations in the atmosphere will become a major challenge for rice growth and development in the future due to changes in our climate such as extremes in temperature. To guarantee food safety, novel genetic loci need to be identified for source- and sink-associated traits that are specifically expressed under elevated CO₂ conditions. Eighty chromosome segment substitution lines carrying *japonica* (Nipponbare) chromosome segments in the *indica* (9311) background were used in this study. QTL analysis was conducted for source- and sink-related traits, including flag leaf length, flag leaf width, flag leaf fresh weight, flag leaf dry weight, primary branch number, secondary branch number, grain number per panicle, panicle weight per plant, chlorophyll a, chlorophyll b, and carotenoid contents, under ambient CO₂ concentrations and free-air CO₂ enrichment. A total of 49 QTLs for these traits were detected on chromosomes 1, 3, 5, 6, 7, 9, and 12 under the two conditions; the variance explained by these QTLs varied from 6.22 to 38.15%. Among these QTLs, 19 of them were detected under the natural field conditions and 30 were detected in the elevated CO₂ conditions. In addition, 2 and 13 QTLs were specifically expressed in the natural and CO₂-enriched conditions, respectively. Our findings have important implications on the utilization of germplasm resources for ensuring food security under elevated CO₂ levels, especially for QTLs that were specifically detected under the elevated CO₂ condition.

Keywords CO₂ concentration · Chromosome segment substitution line · QTL · Rice

Introduction

Rice (*Oryza sativa* L.) is a major cereal crop consumed worldwide and increase of its grain yield is extremely important to meet the challenge of increasing population (Gross and Zhao 2014). Grain yield is sensitive to climate change, such as atmospheric CO₂ concentration, temperature, and precipitation (Shi et al. 2017; Wang et al. 2018a; Sharma et al. 2018). Since the industrial revolution, the concentration of greenhouse gases has been increasing annually, especially the CO₂ concentration. The global CO₂ concentration is estimated to have risen from 280 μmol mol⁻¹ before the industrial revolution to 406 μmol mol⁻¹ in 2017, and the annual growth rate was 0.5% (Cheng et al. 2018; Li et al. 2019). It is anticipated that the global CO₂ concentration will reach 421 to 936 μmol mol⁻¹ by the end of the 21st century (Cheng et al. 2018; Li et al. 2019). The increase of CO₂ and other greenhouse gases leads to global warming

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and changes in global precipitation patterns. To ensure food security, we need to develop elite varieties that can adapt to, or even benefit from, these predicted environmental conditions (Ainsworth 2008).

Source and sink traits are the key factors that determine rice grain yield. Source capacity is primarily associated with leaf characteristics, such as the amount of photosynthetic pigments and the size of the leaves. Sink function is mainly associated with panicle traits, such as spike number and grain filling ability. Many studies have reported that chlorophyll content and flag leaf size are positively correlated with grain yield under natural field conditions (Jiang et al. 2012; Zhang et al. 2014; Liu et al. 2014). One study showed that the fast decline of total chlorophyll content, photosynthetic rate, and transpiration rate of the flag leaf in the late growth period led to a decrease of the seed setting rate, which greatly influences grain yield potential (Fu et al. 2012). Chlorophyll content is positively correlated with spike length, spikelet number per panicle, seed setting rate, and 1000-grain weight from heading to maturity in rice, which indicates that chlorophyll content in the flag leaf is closely related to yield after heading, and within a certain range, improvement of the chlorophyll content at the medium-late growth stage contributes to an increase of yield (Liu et al. 2018).

CO₂ is a major greenhouse gas and the basic substrate for photosynthates (Xu et al. 2014; Jia 2015; Wang et al. 2018b). Plant growth and development are influenced by climate change, such as CO₂ concentrations in the air (Chang et al. 2014). The increase of CO₂ in the atmosphere not only promotes photosynthesis and the water utilization rate of plants, but also contributes to plant growth and crop yield (Song et al. 2008; Wang et al. 2018b). Furthermore, grain yield, grain number, panicle number, and spikelet fertility were significantly increased under elevated CO₂ concentration (Fan et al. 2008a, b, c), which suggests that genes controlling yield-related traits may have different expression patterns under different CO₂ concentrations. Thus, understanding the genetic basis for agronomically important traits of rice under elevated CO₂ is important. However, most of the quantitative trait loci (QTLs) discovered in rice were detected under natural conditions, such as the yield-related and heading date QTLs (Bai et al. 2012; Hori et al. 1972), while only a few QTLs were detected under elevated CO₂ conditions (Fan et al. 2005, 2006, 2008a, b; Zhou et al. 2017). Some QTLs for yield-related traits were detected under both elevated CO₂ and natural field conditions. Interestingly, more yield-related QTLs were detected under elevated CO₂ than under natural field conditions (Fan et al. 2008c). In addition, the number of minor QTLs and the phenotypic variance of major QTLs were also increased under elevated CO₂ (Fan et al. 2008b). Furthermore, alleles of *GNI* (*Grain number 1*) and *APO1* (*Aberrant Panicle Organization 1*) that increase

sink capacity can enhance grain yield under elevated CO₂ conditions (Nakano et al. 2017).

Previously, chromosomal segment substitution lines (CSSLs) derived from Nipponbare (NPB) were developed in the 9311 genetic background; 9311 is the restorer line of the two-line super hybrid rice of Liangyoupeijiu and is widely used in China due to its excellent source and sink capacity and good quality (Gao et al. 2013). NPB is a *japonica* cultivar with relatively poor source and sink capacity. In this study, we performed QTL mapping for source- and sink-associated traits using the 9311/NPB CSSL population under natural and elevated CO₂ conditions. The aims of this study were to dissect the genetic basis of source and sink capacity in rice and to detect novel loci that are specifically expressed under elevated CO₂ conditions. Our results will be useful for breeding rice varieties that are better adapted to high levels of CO₂.

Materials and methods

Free-air CO₂ enrichment

The free-air CO₂ enrichment (FACE) facilities were located in Xiaoji Village (119°42'0" E, 32°35'5" N), Yangzhou City, Jiangsu Province. Three uniform and similarly sized rectangular fields were selected at the FACE experiment site. The spacing between FACE circles was more than 90 m. To reduce the influence of CO₂ release between circles, the FACE circle was designed as a regular octagon with a diameter of 12.5 m. Pure CO₂ gas was injected into the center of the FACE ring through the pipes surrounding the FACE ring. The average CO₂ concentration was controlled at 570 μmol mol⁻¹ with 10% error during the entire growth period (Fan et al. 2008a; Yang et al. 2007).

Rice materials and field trials

Eighty rice (*Oryza sativa* L.) CSSLs with a *japonica* cultivar, Nipponbare (NPB), as the donor parent and an *indica* cultivar, 9311, as the recurrent parent were developed previously and used in this study for QTL mapping. The CSSLs and the parents were planted in Yangzhou from May to October in 2018. Uniform seeds of all CSSLs and their parents were soaked in distilled water at 30 °C for 2 days and then germinated, covered by a damp washcloth at 30 °C for 12 h. The germinated seeds were sown in a paddy field at Hangzhou and transferred to Yangzhou after 30 days. The 30-day-old seedlings were transplanted into a 20-by-20-cm grid pattern in the flooded paddy in Yangzhou (119° 42' 0" E, 32° 35' 5" N). A randomized complete block design was used with three replications. In each replication, 10

plants per line were planted in one row with a spacing of 20 cm between plants and 20 cm between rows.

Trait evaluation

Five plants in the middle of each row were randomly selected for measurement of chlorophyll a (Chl *a*), chlorophyll b (Chl *b*), carotenoids (Car), flag leaf length (LL), flag leaf width (LW), flag leaf fresh weight (FLFW), and flag leaf dry weight (FLDW) at 20 days after heading. Chl *a*, Chl *b*, and Car were measured according to Yang et al. (2016). At maturity, five plants in the middle were harvested, air-dried, and used for measurement of primary branch number (PBN), secondary branch number (SBN), grain number per panicle (GN), and panicle weight per plant (PWPP: grain weight with branch stem weight). The mean values of the five plants within each row were used as the trait value of each line, and the trait values of three replications of each line were averaged and used for data analysis.

Data and QTL Analysis

Using the NPB/9311 CSSLs, the effects of putative QTLs were tested according to variance analysis and *t* test with a significance level of $P < 0.001$. When a significant phenotypic difference between a CSSL and the recurrent parent was observed, it was considered the presence of a QTL. The marker interval of a corresponding QTL on a chromosomal location was determined according to the different phenotypic performance between the two contiguous CSSLs with overlapping segments.

Results

Phenotypic performance of CSSLs and parents

The phenotypic performance of CSSLs and parents is summarized in Fig. 1. All measured traits exhibited obvious differences between the parents (NPB and 9311) under both natural field and elevated CO₂ conditions. The contents of Chl *a*, Chl *b*, and Car in NPB were higher than those of 9311. By contrast, the other traits measured were lower in NPB than in 9311. This indicates that NPB may have more alleles that enhance Chl *a*, Chl *b*, and Car but fewer alleles that enhance the remaining traits. With the exception of Chl *a*, Chl *b*, and Car, the mean phenotypic values of the other eight yield-related traits in the CSSL population were closer to those 9311, which is in accordance with the fact that the CSSLs were in the 9311 genetic background.

Correlation analysis of traits

The correlation coefficients among the agronomic traits under natural and CO₂-enriched conditions are presented in Table 1. For the photosynthetic pigments, Chl *a*, Chl *b*, and Car content showed significant positive correlations between each other under both conditions. For the flag leaf size traits, LL, FLFW, and FLDW displayed significant positive correlations between each other under both conditions, whereas LW only showed significant positive correlations with other traits under the natural field condition. For the panicle traits, PBN, SBN, GN, and PWPP exhibited significant positive correlations between each other under both conditions, except for PWPP and PBN, and PWPP and SBN under CO₂ enrichment. These results indicate the existence of an interdependent relationship within the photosynthetic pigments, flag leaf size, and panicle traits.

Significant negative correlations were detected between most of the photosynthetic pigments and flag leaf size as well as panicle traits under the natural field condition, whereas nonsignificant negative correlations were detected between most of the photosynthetic pigments and flag leaf size and panicle traits under CO₂ enrichment. These results suggest that there is probably a negative association between photosynthetic pigment traits and flag leaf size and panicle traits under natural field conditions, whereas their association is diminished under high CO₂ conditions.

QTL analysis under the natural field condition

Under the natural field condition, 19 main-effect QTLs were detected, including one each for Chl *a*, Chl *b*, and Car content, and two for each of the remaining traits investigated (Fig. 2; Table 2). The contributions of a single QTL to the phenotypic variance ranged from 7.96 to 27.95%. There were six regions that had effects on two or more traits, among which three regions showed pleiotropic effects on both source- and sink-associated traits, while the remaining three regions exhibited pleiotropic effects on either source- or sink-associated traits.

For the photosynthetic pigments, three QTLs, *qChla-1N*, *qChlb-3N*, and *qCar-1N*, were detected for Chl *a*, Chl *b*, and Car content, respectively. Two of the enhancing alleles were from NPB, and one was from 9311. This result is in accordance with the content of Chl *a*, Chl *b*, and Car being higher in NPB than in 9311.

For the flag leaf-associated traits, eight QTLs were detected; seven of the enhancing alleles were from 9311 and one was from NPB. For the panicle-associated traits, we detected eight QTLs; five of the enhancing alleles were from 9311 and three were from NPB. This result is in accordance with 9311 showing higher phenotypic values for flag leaf- and panicle-associated traits than NPB.

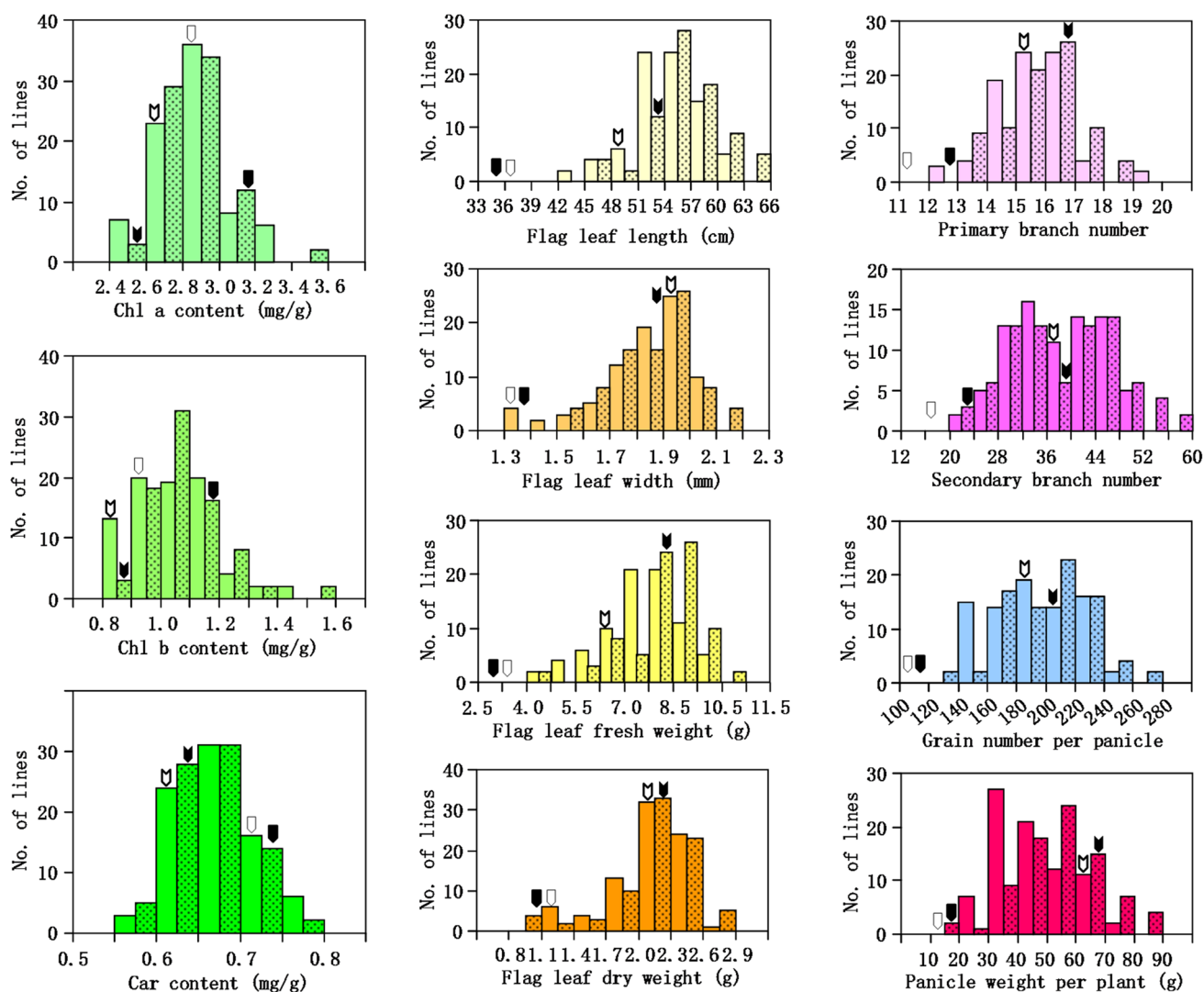


Fig. 1 Performance of source- and sink-associated traits in the CSSL population and parents. Histograms with dots represent traits measured under FACE and without dots represent traits measured under natural field conditions. Black and white arrows represent parental

phenotypic values measured under FACE and natural field conditions, respectively. Arrows with blunt end represent Nipponbare, and arrows with notched end represent 9311

QTL analysis under elevated CO₂

Under the elevated CO₂ condition, 30 main-effect QTLs were detected and summarized in Fig. 2 and Table 2. The contributions of a single QTL to the phenotypic variance ranged from 6.22 to 38.15%. There were nine regions that had effects on two or more traits, among which only three regions showed pleiotropic effects on both source- and sink-associated traits, while the remaining six regions only exhibited pleiotropic effects on either source- or sink-associated traits.

For the photosynthetic pigments, five QTLs were detected: only one each for Chl *a* and Car, and three for Chl *b*. Four of the enhancing alleles were from NPB, and one was from 9311. This result is in accordance with the

content of Chl *a*, Chl *b*, and Car being higher in NPB than in 9311.

For the flag leaf-associated traits, 13 QTLs were detected; nine of the enhancing alleles were from 9311 and four were from NPB. For the panicle-associated traits, 12 QTLs were detected; nine of the enhancing alleles were from 9311 and three were from NPB. This result is in accordance with 9311 having higher phenotypic values for flag leaf- and panicle-associated traits than those of NPB.

Similarities and differences of QTL identification under the two conditions

A total of 49 main-effect QTLs distributed on chromosomes 1, 3, 5, 6, 7, 9, and 12 were detected among the two

Table 1 Correlations between source- and sink-associated traits in the CSSL population

NF	FACE										
	<i>Chla</i> content	<i>Chlb</i> content	Car content	LL	LW	FLFW	FLDW	PBN	SBN	GN	PWPP
Chl <i>a</i> content		0.703**	0.688**	-0.115	0.098	-0.046	-0.222	0.043	-0.052	-0.029	-0.232*
Chl <i>b</i> content	0.774**		0.601**	-0.209	0.034	-0.095	-0.179	0.065	-0.214	-0.229*	-0.223*
Car content	0.859**	0.622**		-0.210	0.005	-0.156	-0.223*	0.069	-0.194	-0.129	-0.054
LL	-0.244*	-0.349**	-0.194		-0.080	0.816**	0.330**	-0.236*	-0.172	-0.116	-0.218
LW	-0.231*	-0.212	-0.232*	0.298**		0.218	0.080	-0.144	-0.020	0.231*	-0.166
FLFW	-0.328**	-0.357**	-0.314**	0.847**	0.545**		0.444**	-0.250*	-0.106	-0.034	-0.163
FLDW	-0.425**	-0.439**	-0.405**	0.818**	0.506**	0.912**		-0.139	0.101	-0.019	0.052
PBN	-0.084	0.020	-0.171	0.125	-0.072	0.111	0.206		0.506**	0.601**	0.070
SBN	-0.368**	-0.315**	-0.261*	0.152	0.147	0.264*	0.416**	0.593**		0.837**	0.171
GN	-0.356**	-0.281*	-0.287**	0.115	0.113	0.132	0.350**	0.647**	0.885**		0.317**
PWPP	-0.189	0.067	-0.197	0.026	0.085	0.090	0.164	0.211	0.419**	0.420**	

LL flag leaf length, LW flag leaf width, FLFW flag leaf fresh weight, FLDW flag leaf dry weight, PBN primary branch number, SBN secondary branch number, GN grain number per panicle, PWPP panicle weight per plant, NF natural field, FACE free air CO₂ enrichment

* and **significant at 0.05 and 0.01

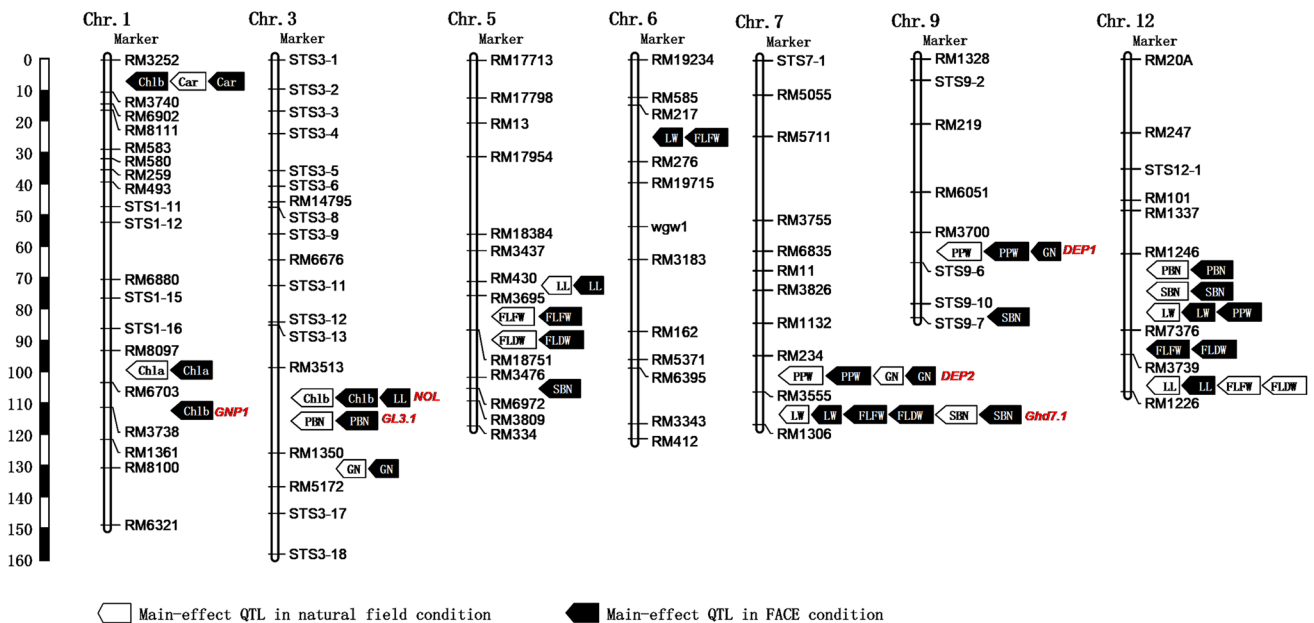


Fig. 2 Main-effect QTLs detected in the natural field and elevated CO₂ conditions. Chl *a* chlorophyll a, Chl *b* chlorophyll b, Car carotenoids, LL flag leaf length, LW flag leaf width, FLFW flag leaf fresh

weight, FLDW flag leaf dry weight, PBN primary branch number, SBN secondary branch number, GN grain number per panicle, PWPP panicle weight per plant

conditions, with more of the QTLs detected in the elevated CO₂ condition than in the natural field condition (Fig. 2; Table 2). In addition, 2 and 13 QTLs were specifically detected under the natural and CO₂ enriched conditions, respectively. These results suggest that some QTLs were detected depending on the CO₂ concentration and that a considerable number of QTLs were specifically expressed under the elevated CO₂ condition.

Discussion

Rice grain yield is largely determined by the source–sink relationship. Flag leaf area and its photosynthetic pigment content are key factors that influence source ability, while panicle traits greatly affect the sink capacity. The genetic basis underlying these traits remains poorly

Table 2 Main-effect QTLs detected under natural field and elevated CO₂ conditions

Trait	QTL	Chr	Marker interval	R ² (%)	Additive effect	P value
Chl <i>a</i> content	<i>qChla-1N</i>	1	RM8097–RM6703	26.12	−0.1169	<1 × 10 ^{−6}
	<i>qChla-1C</i>	1	RM8097–RM6703	25.93	−0.0999	<1 × 10 ^{−6}
Chl <i>b</i> content	<i>qChlb-3N</i>	3	RM3513–RM1350	7.96	0.0484	2.5 × 10 ^{−4}
	<i>qChlb-1.1C</i>	1	RM3252–RM3740	17.5	−0.0833	<1 × 10 ^{−6}
	<i>qChlb-1.2C</i>	1	RM6703–RM3738	20.59	−0.084	<1 × 10 ^{−6}
	<i>qChlb-3C</i>	3	RM3513–RM1350	13.39	0.0551	4.8 × 10 ^{−4}
Car content	<i>qCar-1N</i>	1	RM3252–RM3740	27.95	−0.0264	1.4 × 10 ^{−6}
	<i>qCar-1C</i>	1	RM3252–RM3740	25.26	−0.0167	2.0 × 10 ^{−6}
LL	<i>qLL-5N</i>	5	RM430–RM3695	11.76	2.2635	6.5 × 10 ^{−5}
	<i>qLL-12N</i>	12	RM3739–RM1226	23.44	5.663	<1 × 10 ^{−6}
	<i>qLL-3C</i>	3	RM3513–RM1350	9.47	−2.8965	<1 × 10 ^{−6}
	<i>qLL-5C</i>	5	RM430–RM3695	12.04	2.7442	3.7 × 10 ^{−5}
	<i>qLL-12C</i>	12	RM3739–RM1226	24.95	4.5562	<1 × 10 ^{−6}
LW	<i>qLW-7N</i>	7	RM3555–RM1306	11.99	−0.0813	8.5 × 10 ^{−5}
	<i>qLW-12N</i>	12	RM1246–RM7376	16.13	0.0858	3.0 × 10 ^{−5}
	<i>qLW-6C</i>	6	RM217–RM276	17.36	0.0727	6.4 × 10 ^{−5}
	<i>qLW-7C</i>	7	RM3555–RM1306	10.73	−0.0701	3.5 × 10 ^{−4}
	<i>qLW-12C</i>	12	RM1246–RM7376	19.05	0.0923	<1 × 10 ^{−6}
FLFW	<i>qLFW-5N</i>	5	RM3695–RM18751	15.61	0.8226	<1 × 10 ^{−6}
	<i>qLFW-12N</i>	12	RM3739–RM1226	12.12	0.7291	<1 × 10 ^{−6}
	<i>qLFW-5C</i>	5	RM3695–RM18751	14.42	0.797	<1 × 10 ^{−6}
	<i>qLFW-6C</i>	6	RM217–RM276	22.28	0.9707	<1 × 10 ^{−6}
	<i>qLFW-7C</i>	7	RM3555–RM1306	7.83	−0.4372	1.3 × 10 ^{−5}
	<i>qLFW-12C</i>	12	RM7376–RM3739	11.24	0.6896	<1 × 10 ^{−6}
FLDW	<i>qLDW-5N</i>	5	RM3695–RM18751	11.97	0.3203	3.4 × 10 ^{−5}
	<i>qLDW-12N</i>	12	RM3739–RM1226	12.82	0.315	1.2 × 10 ^{−5}
	<i>qLDW-5C</i>	5	RM3695–RM18751	10.35	0.2662	7.8 × 10 ^{−5}
	<i>qLDW-7C</i>	7	RM3555–RM1306	27.07	−0.3693	<1 × 10 ^{−6}
	<i>qLDW-12C</i>	12	RM7376–RM3739	11.35	0.2759	4.6 × 10 ^{−5}
PBN	<i>qPBN-3N</i>	3	RM3513–RM1350	14.84	0.6476	5.2 × 10 ^{−4}
	<i>qPBN-12N</i>	12	RM1246–RM7376	16.42	0.5945	3.1 × 10 ^{−4}
	<i>qPBN-3C</i>	3	RM3513–RM1350	15.19	0.6891	1.0 × 10 ^{−3}
	<i>qPBN-12C</i>	12	RM1246–RM7376	19.65	0.7409	1.3 × 10 ^{−5}
SBN	<i>qSBN-7N</i>	7	RM3555–RM1306	15.4	−3.4601	9.1 × 10 ^{−5}
	<i>qSBN-12N</i>	12	RM1246–RM7376	22.68	4.1695	<1 × 10 ^{−6}
	<i>qSBN-5C</i>	5	RM3476–RM6972	6.22	2.5048	9.4 × 10 ^{−3}
	<i>qSBN-7C</i>	7	RM3555–RM1306	12.97	−3.3466	1.8 × 10 ^{−4}
	<i>qSBN-9C</i>	9	STS9-10–STS9-7	38.15	7.2768	<1 × 10 ^{−6}
	<i>qSBN-12C</i>	12	RM1246–RM7376	16.47	2.8501	<1 × 10 ^{−6}
GNPP	<i>qGN-3N</i>	3	RM1350–RM5172	18.01	16.4719	2.7 × 10 ^{−5}
	<i>qGN-7N</i>	7	RM234–RM3555	23.35	−22.7838	<1 × 10 ^{−6}
	<i>qGN-3C</i>	3	RM1350–RM5172	14.52	10.9267	2.1 × 10 ^{−4}
	<i>qGN-7C</i>	7	RM234–RM3555	18.01	−16.4251	5.6 × 10 ^{−5}
	<i>qGN-9C</i>	9	RM3700–STS9-6	30.08	25.7084	<1 × 10 ^{−6}
PWPP	<i>qPPW-7N</i>	7	RM234–RM3555	14.44	−5.5252	1.1 × 10 ^{−4}
	<i>qPPW-9N</i>	9	RM3700–STS9-6	19.38	5.9554	8.8 × 10 ^{−5}
	<i>qPPW-7C</i>	7	RM234–RM3555	12.73	−4.6724	7.1 × 10 ^{−4}
	<i>qPPW-9C</i>	9	RM3700–STS9-6	20.16	7.2295	4.7 × 10 ^{−4}
	<i>qPPW-12C</i>	12	RM1246–RM7376	18.03	8.5246	2.1 × 10 ^{−5}

Chr chromosome, R² proportion of phenotypic variance explained by the QTL effect, Additive effect additive effect of replacing a 9311 allele by a Nipponbare allele

LL flag leaf length, LW flag leaf width, FLFW flag leaf fresh weight, FLDW flag leaf dry weight, PBN primary branch number, SBN secondary branch number, GN grain number per panicle, PWPP panicle weight per plant

understood, particularly in elevated CO₂ environments. In this study, QTL analysis for source- and sink-associated traits was conducted under natural field and elevated CO₂ conditions. A total of 49 QTLs for Chl *a*, Chl *b*, Car, LL, LW, FLFW, FLDW, PBN, SBN, GN, and PWPP were detected among the two conditions, among which 19 and 30 QTLs were detected under natural field and elevated CO₂ conditions, respectively. They fell into 16 regions of 7 chromosomes, and the phenotypic variation explained by a single QTL ranged from 6.22 to 38.15%. Notably, five of these regions were reported cloned genes that related to source- and sink-associated traits (Fig. 2). The five regions are RM6703–RM3738 that is co-located with *GNPI*, RM3513–RM1350 that covers *GL3.1* and *NOL*, RM234–RM3555 that coincided in position with *DEP2*, RM3555–RM1306 that co-localized with *Ghd7.1*, and RM3700–STS9-6 that contains *DEP1* (Huang et al. 2009; Sato et al. 2009; Li et al. 2010; Qi et al. 2012; Yan et al. 2013; Wu et al. 2016).

Source and sink capacity are both important for rice high-yielding potential. However, both positive and negative correlations were reported between source- and sink-associated traits (Li et al. 1998; Liu et al. 2018). In this study, significant positive correlations were only detected between a few source- and sink-associated traits, while negative correlations were detected between multiple source- and sink-associated traits, such as Chl *a* and PWPP, Chl *b* and GN, and leaf length and PBN (Table 1). However, both the positive and negative correlations between the two types of traits were relatively weak. Furthermore, we detected six and nine QTL clusters under the natural field and enhanced CO₂ conditions, respectively, but only three regions showed pleiotropic effects on both source- and sink-associated traits under each condition. The remaining QTL regions exhibited allelic variation for either source- or sink-associated traits. Thus, the source- and sink-associated traits may have relatively weak relationship genetically. With regard to these results, pyramiding of genes that have superior effects on source- or sink-associated traits is an effective and practive way to develop high-yielding cultivars.

Rice source- and sink-associated traits are controlled by multiple genes and are easily influenced by environmental change, such as the temperature and CO₂ concentration. It is important to identify genes that confer increased yield potential under elevated CO₂ concentrations. In this study, a total of 34 QTLs were detected under both conditions. Among which, 13 QTLs were specifically detected under the elevated CO₂ condition, and only two QTLs were specifically detected under the natural field condition. The QTLs expressed only under the elevated CO₂ condition will be useful in breeding to improve rice yield potential.

The size of the flag leaf is an important indicator for breeding cultivars with high yield potential. QTLs with

pleiotropism on both leaf size and yield traits were also reported. For example, Wang et al. (2011) detected a QTL, *qFL1*, for flag leaf size on the short arm of chromosome 1 and fine-mapped it to a 31-kb region and found that *qFL1* has pleiotropic effects on secondary branch number, number of spikelets per panicle, and panicle weight. Shen et al. (2012) fine-mapped a major QTL, *qFLL6.2*, that simultaneously controlled flag leaf length and yield traits. Zhang et al. (2015) fine-mapped a major QTL, *qFLW7.2*, responsible for flag leaf size and grain yield. In this study, three regions, RM3513–RM1350, RM3555–RM1306, and RM1246–RM7376, showed pleiotropic effects on both source- and sink-associated traits. However, whether the effects of these three regions result from a single QTL or a QTL cluster needs to be validated by further study.

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Data availability The datasets supporting the conclusions of this article are provided within the article.

Compliance with ethical standards

Conflict of interest The authors declare that they have no competing interests.

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