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Quantitative trait loci for leaf chlorophyll fluorescence traits in wheat

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Abstract

Chlorophyll fluorescence is closely related to photosystem II (PS II), and chlorophyll fluorescence analysis has become one of the most powerful and widely used techniques that are available to plant physiologists and ecophysiologists. In this article we report the genetic study of leaf chlorophyll fluorescence traits in wheat. 114 wheat recombinant inbred lines (RILs) derived from W-7984×Opata85 were used as samples and QTLs of chlorophyll relative content (C), minimum fluorescence yield of PS II (F_o), maximum fluorescence yield of PS II (F_m), variable chlorophyll fluorescence yield (F_v), maximum quantum yield of PS II (F_v/F_m), and time of achieving maximum fluorescence yield (T_m) were obtained and analyzed. Five QTLs associated with C are detected on 4B, 4D, 6D and 7A, and the relative additive contribution of locus C6D and C7A is 15.60% and 11.04%, respectively; two pairs of interaction loci affecting C are found. Three QTLs are identified for F_o on 1A, 1B and 1D. One QTL for F_v is mapped on 7D, and it is found that five pairs of interaction QTLs influencing F_v. One loci significantly influencing F_m is on chromosome 5A, which acts as a major effect gene and can explain 20.69% of total phenotypic variation for F_m, and closely links with the RFLP marker Xcdo749. There are six QTLs controlling T_m mapped on 2D, 3D, 4A, 4D, 5D and 7D; one pair of interaction QTLs influencing T_m exists. Three pairs of interaction QTLs influence F_v/F_m, showing a general epistatic contribution of 94.62%. The identified QTL markers for chlorophyll fluorescence traits will be useful for understanding the genetics background and marker-assisted selection in wheat photosynthetic traits improving.

Keywords: QTLs; Leaf Chlorophyll Fluorescence Trait; Wheat

Introduction

Light energy absorbed by chlorophyll molecules in a leaf can undergo one of three fates: it can be used to drive photosynthesis (photochemistry), providing the chemical (in the form of ATP and NADPH) for CO_2 fixation in the Calvin cycle; excess energy can be dissipated as heat or it can be re-emitted as light-chlorophyll fluorescence. These three processes occur in competition, such that any increase in the efficiency of one will result in a decrease in the yield of the other two. Hence, by measuring the yield of chlorophyll fluoresce, information about changes in the efficiency of photochemistry and heat dissipation can be gained. Chlorophyll fluorescence closely associated with photosystem II (PS II), which can reflect the photosynthesis efficiency in different plants (Genty *et al.* 1989; Oxborough and Baker 1997; Kornyeyev *et al.* 2003). Chlorophyll fluorescence analysis has become one of the most powerful and widely used techniques available to plant physiologists and ecophysiologists (Biehler and Fock 1995; Govindjee, 1995; Maxwell *et al.* 2000; Barbagallo *et al.* 2003; Proctor *et al.* 2003). This technique has been used in the study of wheat stress response under different unfavorable conditions, such as water stress (Moffat *et al.* 1990; Van Kooten and Snell 1990; Giardi *et al.* 1996; Lu and Zhang 1998; 1999b; Eduardo *et al.* 2002), heat stress (Klinkovsky and Naus 1994; Epron 1997; Lu *et al.* 1999, 2001; Charles *et al.* 2003), cold stress (Fracheboud *et al.* 1999; Haldimann, 1999; Rizza *et al.* 2001; Ying *et al.* 2002), salt stress (Lu *et al.* 1999a, 2001, 2003), nitrogen deficient (Lu *et al.* 2000, 2001; Shangguan *et al.* 2000) and other abiostresses and biostresses (Sampol *et al.* 2003). Up to now, many researches have been reported on the aspect of chlorophyll florescence variance in plant physiology study, however chlorophyll florescence genetic studies are only scarcely reported. Major early genetic study was focused on plant morphological traits and agronomy traits, because they can be easily observed and classified. However, the genetic background of physiology traits such as photosynthesis is difficult to be understood. Using molecular linkage genetic maps and quantitative trait loci (QTLs) mapping technology, it is possible to estimate the number of loci controlling genetic variation in a segregating population and to characterize these loci with regard to their map positions in the genome, gene action, phenotypic effects, pleitropic effects, and epistatic interactions with other QTLs (Xiao *et al.* 1996). Once achieved, targeting genomic regions for crop improvement will be possible through marker-assisted selection (Stuber *et al.* 1999). Combining QTL mapping with other biotechnological techniques such as physical mapping and whole genome sequencing opens the opportunity for the identification of the responsible gene(s) by map-based landing (Tanksley *et al.* 1995)

or a candidate gene approach (Pfieger *et al.* 2001). Based on QTL analysis of photosynthesis traits in plant, three loci controlling chlorophyll content in rice were mapped on the chromosomes 2, 4 and 7 (Wu and Lou 1996); one locus controlling F_m (maximum fluorescence yield) in maize was identified on chromosome 7 under water stress condition (Lebreton *et al.* 1995). Three QTLs for maximum fluorescence ratio (F_v/F_m) were detected on chromosome 1 and 8, and one was detected on chromosomes 8 for F_o in maize (Fracheboud *et al.* 2002). Under chilling stress in tomato, two QTLs affecting non-photochemical quenching (qNP) were detected on chromosomes 1 and 6, and one QTL associated with both photochemical (qP) and F_v/F_m was on chromosome 5 (Oyanedel *et al.* 2000). Because the genetic basis of chlorophyll fluoresce traits in wheat has not yet been fully investigated using molecular markers, the objective of this work was to conduct a QTL analysis on a set of 114 recombinant lines (RILs) to further understand the chlorophyll fluoresce traits at the genetic level. Results about the number and the characteristics of genomic regions that are responsible for chlorophyll fluoresce traits will be useful for future germplasm improvement and high photosynthetic efficiency breeding.

Materials and methods

Plant material

The mapping population consists of 114 recombinant inbred lines (RIL) developed from a cross between the spring wheat variety 'Opata85' and a synthetic hexaploid wheat 'W-7984'. W-7984 was derived from the cross between the durum wheat (*Triticum turgidum L.* 2n=4x=28, AABB) cultivar 'Altar 84' and *Aegilops tauschii Coss. [syn.Aegilops squarrosa L.,syn.T.tauschii (Coss) Schmal*. 2n=2x=14, DD], as described by Nelson *et al.* (1995c). The seeds of this RILs population were kindly provided by Dr. Patrick McGuire and Shaoke Wang at University of California.

Experimental design and physiology traits measurement

The experiments were conducted in a growth chamber at the Key Laboratory of Crop Germplasm and Biotechnology of Agriculture Ministry, Institute of Crop Germplasm, Chinese Academy of Agriculture Sciences, Beijing. In the growth chamber, temperature was controlled at 20 ^oC and light was on (400 $\mu mol \cdot m^{-2} \cdot s^{-1}$ PAR) at daytime, and temperature was kept at 18 ^oC and light was off at night; the relative humidity is 60% at daytime and 65% at night. Seeds were sterilized and germinated in petri dishes. After growing to about 10 cm in length, three healthy seedlings of each RIL were aquicultivated in three Erlenmeyer flask (50 ml) with Hoagland's nutrient solution. The experimental units (one plant per accession per flask) were randomized in complete blocks with three replications. When the wheat has 5 leaves, chlorophyll fluorescence of the third leaf was measured by FIM 1500 apparatus (Analytical Development Company, UK). The transients were induced by red light of 3000 $\mu mol \cdot m^{-2} \cdot s^{-1}$, provided by an array of six light-emitting diodes (peak 650 nm), which focused on the exposed area of the sample (4 mm in diameter). All samples were dark-adapted for 30 minutes prior to the fluorescence measurement. Five chlorophyll fluorescence traits were output by FIM 1500 apparatus as follows: F_o, minimum fluorescence yield of PS II; F_m, maximum fluorescence yield of PS II; F_v, variable fluorescence yield, calculated as F_m-F_o; F_v/F_m, maximum quantum yield of PS II, calculated as $(F_m-F_o)/F_m$; T_m, time of achieving maximum fluorescence yield. Chlorophyll relative content (C) was measured on the third leaf of each plant by a hand held meter (SPAD 502, Minolt, Spectrum Technologies In., Plainfied, IL, Japan) as flag leaf greenness in arbitrary absorbency or SPAD units. These units are linearly related to chlorophyll concentration (Yadava, 1986; Fischer *et al.* 1998).

Data and map analysis

The SAS system for windows release 6.12 (Copyright 1989-1999, SAS Institute Inc., USA) was employed for data analysis and cartography. The mapping population has been the subject of an extensive genome mapping effort by investigators of the International Triticeae Mapping Initiative (ITMI) (Marino *et al.* 1996; Nelson *et al.* 1995a,b,c; Van Deynze *et al.* 1995). The QTL analysis was performed using QTLMapper 1.0 (Wang *et al.* 1999), software for mapping quantitative trait loci (QTLs) with main effects, epistatic effects and QTL× environment interactions. A LOD score of 2.0 was used as the threshold for detecting QTL locations in the QTL Mapper 1.0 program.

Results

Analysis of the phenotypic values

The phenotypic values and statistical data of all traits are shown in Table 1 and Fig 1. Between the two parental varieties, C, F_o, F_m, F_v and T_m of Opata 85 are higher than those of W-7984. But no significant difference exhibits between the two parents for C, T_m and F_v/F_m. Significant difference exhibits in 114 RILs for six traits examined by T test method. The order of coefficient of variation (CV) is T_m>C>F_o>F_v>F_m>F_v/F_m. Compared with the mean values of the two parents and minimum and maximum of all traits, phenotypic values of 114 RILs are greater than that of the higher parent and less than that of the lower parent, which indicate that transgressive segregation have occurred. All traits are fitted normal distribution.

Traits correlations

The simple correlation coefficients among the traits are presented in Table 2. C is significantly positive correlated with F_o, F_m and F_v. F_o is significantly positive correlated with F_m, and significantly negative correlated with F_v/F_m. F_m is significantly positive correlated with F_v, T_m and F_v/F_m. F_v is significantly positive correlated with T_m and F_v/F_m.

QTLs for traits

Chlorophyll content (C)

Five QTLs significantly affecting chlorophyll content (C) are detected on chromosomes 4B, 4D, 6D and 7A (Table 3 and Fig 1). The general contribution of additive effect of the five loci is 46.72%, which means that 46.72% of the phenotypic variation is accounted for by these five loci. The relative additive contribution of locus C (chlorophyll content) 6D (chromosome

Table 1. Statistics of each trait of the RILs

Trait	Parental variety			RILs								
	Opata85	W7984	Prob	Mean	Std Dev	Minimum	Maximum	CV	Skewness	Kurtosis	T value	Prob
C	40.16	30.8	0.0835	34.971	2.555	28.7	42.37	7.306	-0.01	-0.049	146.138	0.0001
Fo	645	596	0.0251	601.289	28.577	529	690	4.753	0.298	0.846	224.656	0.0001
Fm	3968	3667	0.0251	3525.772	137.235	3070	3796	3.892	0.556	0.556	274.31	0.0001
Fv	3323	3071	0.0252	2925.035	135.077	2415	3211	4.618	-0.689	1.365	231.207	0.0001
Tm	306.3	209	0.1185	269.851	43.539	172	404	16.134	0.291	0.116	66.176	0.0001
Fv/Fm	0.837	0.837		0.829	0.009	0.784	0.842	1.083	-1.689	5.486	986.196	0.0001

Table 2. Correlation Matrix

	C	Fo	Fm	Fv	Tm	Fv/Fm
C	1.0000					
Fo	0.2719**	1.0000				
Fm	0.4625**	0.2484**	1.0000			
Fv	0.4155**	0.0576	0.9387**	1.0000		
Tm	0.1521	0.1199	0.3553**	0.3046**	1.0000	
Fv/Fm	0.0834	-0.6325**	0.5103**	0.6451**	0.1043	1.0000

**Correlation was significant at the 0.01 probability level

6D) and C7A is 15.60% and 11.04%, respectively, which are higher than that of other three loci (5.59%-8.08%). Opata 85 contributes just only C6D and the other four loci are contributed by W-7984. It is interesting that loci C7Aa and C7Ab are mapped on the same chromosome 7A, and these two loci together can explain 17.45% of the total phenotypic variation for chlorophyll content. It is notable that loci Ci2Da and Ci2Db (i, interaction) are on the same chromosome 2D, which significantly interact with two loci on the other two different chromosomes 5B and 3A.

The general espistasis contribution of the two pairs of interaction loci (Ci2Da-Ci5B, Ci2Db-Ci3A) is 47.98%, which means 47.98% of the phenotypic variation for chlorophyll content can be explained by the two pairs interaction loci. Moreover, the espistasis contribution of Ci2Db-Ci3A (31.86 %) is almost double of that of Ci2Da-Ci5B (16.12%) (Table 4 and Fig 2).

F_o (Minimum fluorescence yield of PS II)

Three QTLs are identified for F_o, and 23.67% of the total phenotypic variation of F_o can be explained by the three loci. Opata 85 alleles increase this trait at chromosome 1A and 1B on two loci (F_o1A, F_o1B), which together explain 17.36% of the total phenotypic variation for F_o. W7984 alleles increase this trait at chromosome 1D that can only explain 6.31% of the total phenotypic variation (Table 3 and Fig 2). No interaction QTLs significantly influencing F_o has been found on different chromosomes having epistasis effect.

F_m (Maximum fluorescence yield of PS II)

Just one locus for F_m is mapped on chromosome 5A, and Opata 85 contributes alleles increasing F_m at F_m5A locus, which can explain 20.69% of the total phenotypic variation. That means locus F_m5A is a major gene controlling F_m, setting exactly on the left marker Xcdo749 of the markers interval Xcdo749-Xfba131 (Table 3 and Fig 2). No interaction QTLs significantly influencing F_m is examined out on different chromosomes having epistasis effect.

F_v (Variable chlorophyll fluorescence yield)

Also one locus on chromosome 7D significantly affects F_v contributed by W7984, which can explain 10.40% of the total phenotypic variation (Table 3 and Fig 2). F_v7D are closely linked with RFLP markers XksuE3. There are five pairs of interaction QTLs influencing F_v (Table 4 and Fig 2). The general epistatic effect contribution can explain 70.78% of the total phenotypic variation for F_v. The espistasis contribution of F_v5A- F_v6Ab (28.17%) is the highest of these five pairs of interaction QTLs.

T_m (Time of achieving maximum fluorescence yield)

There are six QTLs for T_m, and their general additive contribution is 53.72%. The sum of additive contribution of Opata 85 alleles (T_m2D, T_m4A, T_m4D and T_m7D) is 38.44%, which is higher than that of W7984 alleles (T_m3D and 5D, 15.28%). Two QTLs (T_m2D and T_m4A) have higher additive contribution (>10%) from Opata 85 (Table 3). There is one pair interaction QTLs (T_mi2B-T_mi7B) influencing T_m, and the contribution of the epistasis effect is 30.99% (Table 4 and Fig 2).

F_v/F_m (maximum quantum yield of PS II)

No one putative QTLs has been identified for F_v/F_m, but three pairs of interaction QTLs influence F_v/F_m and the general epistatic contribution is 94.62%. There are one pair of interaction QTL$_S$ from 1 group genome on 1A and 1D chromosomes and two pairs of interaction QTLs on chromosome 2B and 5B. Results show that the episitasis effect is from the interaction QTLs on the same chromosome (Table 4 and Fig 2).

Table 3. Additive effect (A) and relative contribution [H^2 (Ai)] of QTLs affecting chlorophyll fluorescence traits

Trait	Chrom	Interval	Site (cM)	LOD	A	Prob	H^2 (Ai)%
C	4B	Xfba147-Xbcd1250	0.5	2.9	0.8105	0.0003	5.59
	4D	Xcdo669-Xfbb13	14.5	2.37	0.9747	0.0012	8.08
	6D	XksuD27-Xfbb59	2.5	4.14	-1.3545	0	15.6
	7Aa	Xcdo1395-Xabc158	6	2.42	0.8684	0.0009	6.41
	7Ab	Xfba354-Xfba69	2.5	2.53	1.1393	0.0007	11.04
Fo	1A	Xbcd1889-Xbcd8082	2	2.51	-8.587	0.0008	7.77
	1B	Xrz166-Xbcd1796	7.5	2.9	-9.537	0.0003	9.59
	1D	XGli1-XksuD141	8.5	2.24	7.7346	0.0015	6.31
Fm	5A	Xcdo749-Xfba131	0	2.83	-75.5235	0.0005	20.69
Fv	7D	XksuE3-Xfbb189	0	2.05	40.2379	0.0026	10.4
Tm	2D	Xfba88-Xfbb274	22.5	2.6	-18.5306	0.0007	11.1
	3D	Xbcd907-Xbcd1802	2.5	2.1	16.1145	0.0021	8.4
	4A	Xbcd1670-Xfba4	0.5	3.83	-18.2639	0	10.78
	4D	Xcdo669-Xfbb13	0	3.22	-15.9105	0.0002	8.18
	5D	Xfbb238-Xfba137	4.5	2.51	14.5903	0.0009	6.88
	7D	Xrz2-Xfba377	7.5	2.02	-16.0966	0.0027	8.38

Site (cM) column is the genetic distance (in cM) of the testing points from the left marker on the interval on which the testing points are set. A column shows the additive genetic effects estimated at the test points. Positive value implies that the P_1 parent (W7984) takes positive value for the additive effect, while the P_2 parent (Opata85) takes the negative.
LOD column is the LOD scores >2.0

Table 4. Analysis of epistatic effect (Aij) and relative contribution [H^2(Aij)] of QTLs affecting chlorophyll fluorescence traits

Trait	Ch-Ini	Int. Namei	Sitei (cM)	Ch-Inj	Int. Namej	Sitej (cM)	LOD	Ch-Ini-Inj	AAij	Probij	H^2 (AAij)%
C	2Da	Xbcd718-Xbcd102	2	5B	Xabg705-Xcdo749	1	5.01	2Da-5B	-1.3065	0	16.12
	2Db	Xfba3411-Xbcd260	8	3A	Xcdo1345-Xabg471	0	5.47	2Db-3A	1.8366	0	31.86
Fv	1A	Xbcd1930-XksuH9	0	2B	Xcdo388-Xbcd1119	0	4.49	1A-2B	49.8511	0	6.23
	1D	Xbcd1261-XksuE11	0	3D	Xbcd372-Xbcd1555	0	5.03	1D-3D	49.8828	0	6.24
	3A	Xmwg961-Xfbb277	0	6Aa	Xfbb95-Xfbb192	0	3.77	3A-6Aa	63.8314	0.0001	10.22
	5A	Xfbb249-Xcdo20	0	6Ab	Xcdo772-Xpsr463	0	4.74	5A-6Ab	-105.965	0	28.17
	7A	Xcdo347-Xfba134	1	7D	Xrz2-Xfba377	19	7.67	7A-7D	89.1159	0	19.92
Tm	2B	Xfbb284-Xfba622	0	7B	Xbcd385-Xbcd1338	0	5.81	2B-7B	23.0829	0	30.99
Fv/Fm	1A	Xmwg67-Xcdo580	0	1D	XksuD141-Xwhs1791	0	6.57	1A-1D	-0.0046	0	22.92
	2Ba	Xfba280-Xbcd1184	8	2Bb	Xfbb121-Xfba004	0	5.97	2Ba-2Bb	0.0073	0	37.73
	5Ba	Xabg473-Xbcd508	0	5Bb	Xcdo504-Xfbb328	7	4.54	5Ba-5Bb	0.0056	0.0001	33.97

Sitei (cM) and Sitej (cM) columns show the genetic distances of the two putative QTLs from the left markers on their intervals.
A positive AAij value implies that the two –locus genotypes being the same as those in P_1 parent or P_2 parent take the positive effects, while the two –locus genotypes of recombination between the P_1 parent and P_2 parent take the negative effects. The case of negative AAij values is just opposite.

Discussion

The order of significant putative QTLs per trait is T_m (6) > C (5) > F_o (3) > F_m, F_v (1) > F_v/F_m (0), which is of the same order of CV of the six traits (Table 1). That means if one trait has more phenotype variation, more additive effect genes will control this trait. The relative contribution of additive effect ranges from 5.59% to 20.69%. In most cases, QTLs are found at different genomes and chromosomes, but two additive QTLs affected C (C7Aa, C7Ab) are identified from the same chromosome 7A. Opata 85 contributes more additive loci for F_o, F_m and T_m, whereas W-7984 contributes more additive loci for C and F_v (Table 3 and Fig 2). The number of pairs of significant interaction effect QTLs of each trait is in the following order: F_v (5) > F_v/F_m (3) > C (2) > T_m (1) > F_o (0). This sequence is opposite to that of the additive effect genes controlling the traits. That means if one trait has lesser phenotype variation, more pairs of interaction effect genes will affect the trait. This result is in consistence with actual situation in wheat genetics and breeding. The relative contribution of epistasic effect ranges from 6.23 % to 37.73%. There are four types of interaction QTLs among the eleven pairs of QTLs detected from the above-mentioned epistasis analysis results. Two pairs of interaction QTLs affect F_v/F_m on the same chromosome 2B (F_v/F_mi2Ba–F_v/F_mi2Bb) and 5B (F_v/F_mi5Ba-F_v/F_mi5Bb); two pairs of interaction QTLs are found on the homologues chromosomes (F_vi7A-F_vi7D, F_v/F_mi1A-F_v/F_mi1D); four pairs interaction QTLs are mapped on the same genomes (F_vi1D-F_vi3D, F_vi3A-F_vi6Aa, F_vi5A-F_vi6Ab, T_mi2B-T_mi7B); and three pairs of interaction QTLs are identified on different genomes and different chromosomes (Ci2Da-Ci5B, Ci2Db-Ci3A, F_vi1A-F_vi2B) (Table 4 and Fig 2). The results of epsitasis QTLs analysis offer more information about genes interaction among different chromosomes and loci, which also

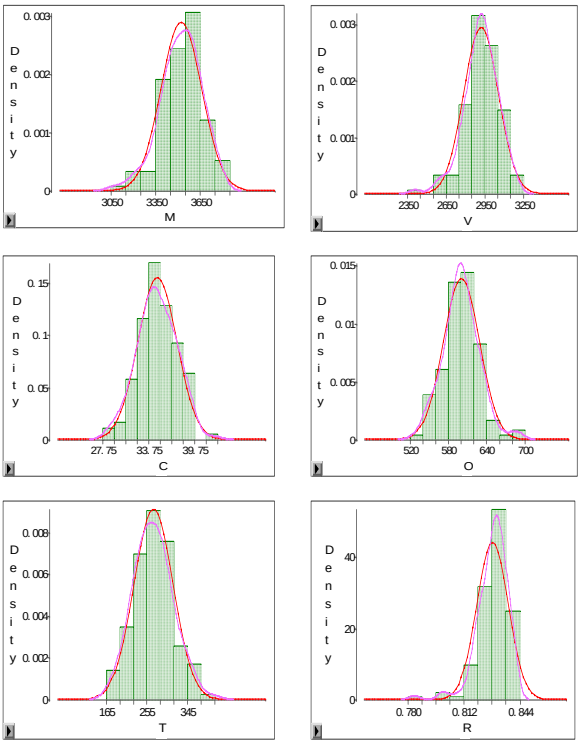
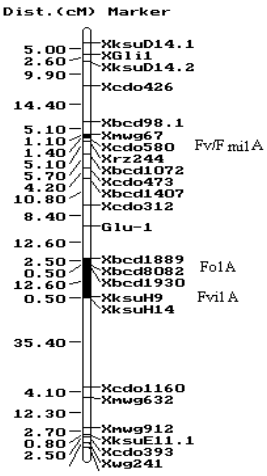
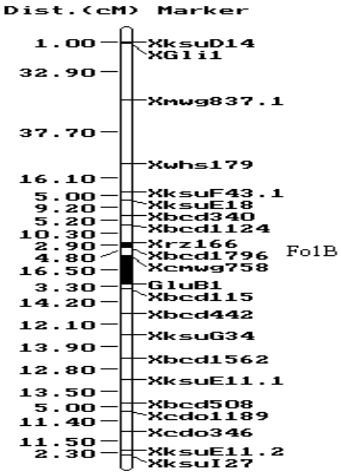


Fig 1. Parametric density estimation for distribution of phenotypes for each trait of the 114 RILs derived from W-7984×Opata85. C: Chlorophyll content; O: F_o; M: F_m; V: F_v; T: T_m; R: F_v/F_m.

supply more about complex traits controlled by polygenes. The sequence of significant additive and interaction QTLs mapped on each chromosome is 2B (4) >1A, 1D, 2D, 5B, 7A and 7D(3) > 3A, 3D, 4D, 5A and 6A(2) > 1B, 4A, 4B, 5D, 6D and 7B (1). No QTL has been found on 2A, 3B and 6B for chlorophyll fluorescence traits in this work (Fig 2). Chlorophyll fluorescence traits are related to the light capture, transmission, consumption and distribution in the photosystem, and thus it can directly reflect the intrinsic PS II efficiency. It provides the route to estimate photosynthetic performance. PS II is also accepted as the most vulnerable part of the photosynthetic apparatus to light–induced damage. The damage to PS II will often be the first manifestation of stress in a leaf (Maxwell *et al.* 2000). It is generally recognized that different plants or different genotypes may show different photosynthesis efficiencies. To the best of our knowledge, photosynthesis efficiency and chlorophyll fluorescence of wheat are quantitative traits that controlled by multiple genes. The mechanism of plant photosynthesis is very complex and has attracted many researchers in physiology, biochemical, physics, genetic, and other fields. Researches crossing different disciplines such as chlorophyll fluorescence and QTLs can supply more information for discovering the genetic background of photosynthesis. For example, it is useful in exploring the relationship between the high photosynthesis ability and stress resistance. The recombinant inbred lines (RILs) from W-7984×Opata 85 was constructed by the International Triticeae Mapping Initiative (ITMI), and widely used in QTL for different traits and genomic comparative study. A microsatellite map of the RILs population has been set up by Röder *et al.* (1998). Plastid ACCase genes were located on maize chromosomes 2 and 10 (Caffrey *et al.* 1995). Comparative mapping experiments have demonstrated that regions of conserved synteny exist between the short arms of wheat group 2 chromosomes and maize chromosomes 2, 7, and 10 (Ahn *et al.* 1993; Van *et al.* 1995; Gornicki *et al.* 1997). Using this RILs population, plastid-localized acetyl-CoA carboxylase (ACC) gene was identified on three ancestral chromosome sets of wheat, and the gene maps close to the telomere on the short arm of chromosomes 2A, 2B, and 2D (Piotr *et al.* 1997). Hypersensitive response (HR) genes such as peroxidase (Per2) and superoixde dismutase (Sod) were mapped on the up location of short arm of chromosome 2D (Li *et al.* 1999) (Fig 2). Two interaction QTLs for F_v/F_m, one QTL for C and one for T_m were also mapped on near telomere or up location of the short arm of chromosomes 2B and 2D in this work (Fig 2). The results show that chlorophyll fluorescence traits have close relationship with acetyl-CoA carboxylase and oxidases activity at the gene level. As we know, acetyl-CoA carboxylase catalyzes the first committed step in *de novo* fatty acid biosynthesis in plastids. It also provides malonyl-CoA for the synthesis of very long chain fatty acids and a variety of important secondary metabolites, and for alonylation. The activity of oxidase closely associates with stress resistance and photosynthesis maintenance. Therefore, the corresponding genes play an important role in plant energy transfer system, and have close relationship with photosynthetic dynamical system and chlorophyll fluorescence traits. Using this RILs population, fifty-eight marker loci representing defense response genes were detected on 21 linkage maps (Li *et al.* 1999). The QTL marker *Xfba314*, located down on long arm of chromosome 2D, shows significant association with the activity of polyphenol oxidase (PPO) (Tigst *et al.* 2001) (Fig 2). Sh2 (shrunken2) codes for the large subunit of ADP-glucose pyrophosphorylase (Fig 2). Probe Aga 7 for Sh2 was mapped on the distal region on the long arm of chromosome 1D (Li and Bikram 2002) (Fig 2). Conidial genes that resistant to tan spot (*Pyrenophora tritici-repentis*) was identified and mapped: one gene with a major effect on 1AS, one gene with a minor effect on 4AL, and an interaction between the 1AS gene and a gene on 2DL. Together, these loci explain 49.0% of the variation in this population for the resistance to tan spot. Two regions, one on 1BL and the other on 3BL, are significantly associated with the resistance to extensive chlorosis (Faris *et al.* 1997). Marker *XGli1* that located on the short arm of chromosome 1A is linked to the tan spot insensitivity locus within 5.7 cM (Effertz *et al.* 2002). Stripe (yellow) rust (YR) gene *Yr28* is located on chromosome arm 4DS. Opata 85 marker alleles near the adult-plant resistance (APR) gene *Yr18* on chromosome arm 7DS. Gene *Yr18* is tightly linked with leaf-rust resistance gene *Lr34*. Three other regions from Opata 85 on chromosome arms 3BS, 3DS, and 5DS are also associated with APR (Singh *et al.* 2000). It is well known that many diseases can cause the chlorosis of wheat, which grievously decreases the photosynthetic ability and greatly influences chlorophyll fluorescence traits. Therefore, if some QTLs that are resistant to these diseases and QTLs for chlorophyll fluorescence traits were mapped on the same chromosome or closely linked in gene map, they are useful for disease and other stress resistance and chlorophyll fluorescence traits genes. For example, by comparing the maps for defense response genes from the GrainGenes database (http//:wheat.pw.usda.gov/graingenes.htm/) (Li *et al.* 1999) and the maps for chlorophyll fluorescence traits (Fig 2), we can



1A



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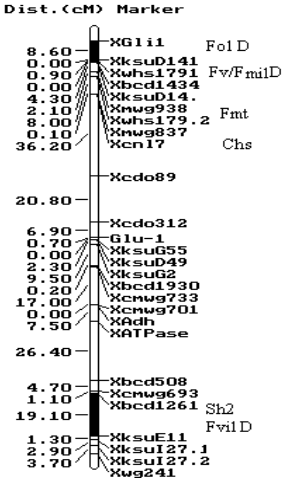

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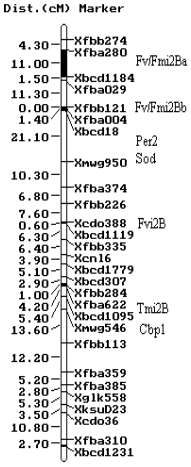

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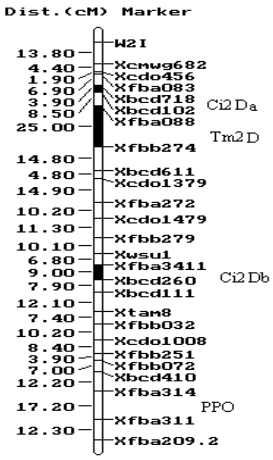

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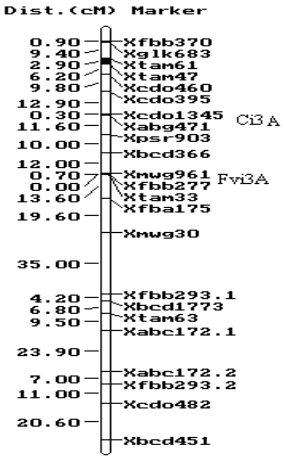

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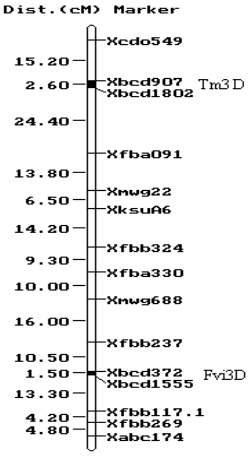

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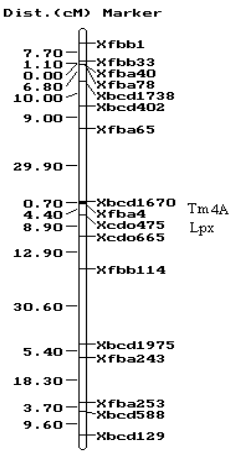

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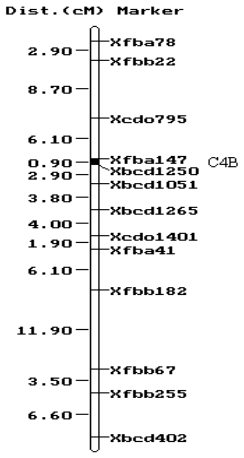

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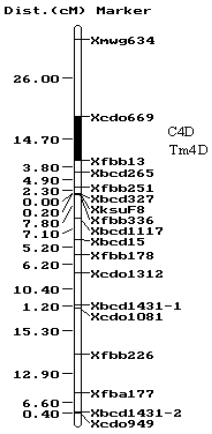

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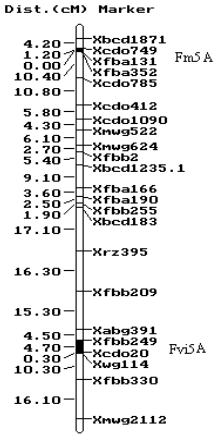

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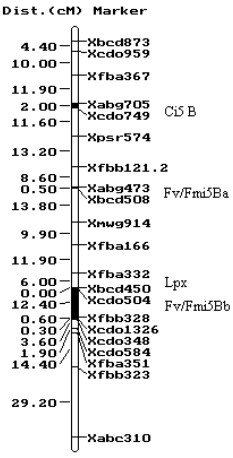

5B

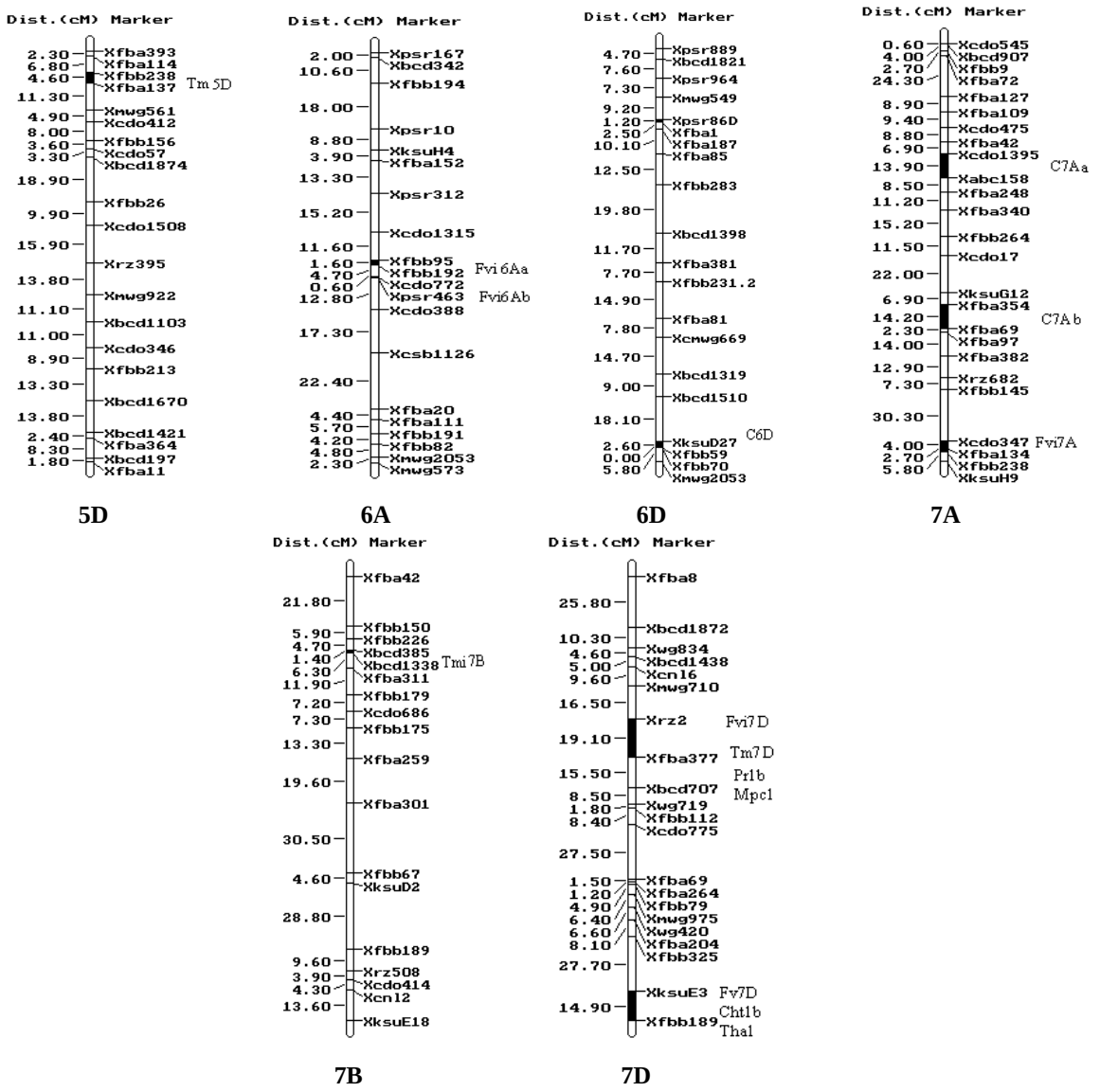

Fig 2. Thirty-eight QTLs of chlorophyll fluorescence physiology traits are placed on the existing genetic linkage map of 18 chromosomes generated in the population of recombinant inbred lines, which are derived from the cross between the synthetic hexaploid wheat W7984 and Opata 85. Fmt: Flavonol 7-o-methyl transferase; Chs: Chalcone synthase; Per: Peroixdase; Sod: Superooxide dismutase; Cbp1: Chitin binding protein; Lpx: Lipoxygenase; Pathogenesis-related gene; Mpc1: myb protein c1; Cht1b: Chitinase; Tha1: Thaumatin (from the GrainGenes database http//:wheat.pw.usda.gov/graingenes.htm/, Li *et al.* 1999); Sh2: shrunken2 (Li and Bikram 2002); PPO: polyphenol oxidase (Tigst *et al.* 2001).

find that, Fmt (Flavonol 7-o-methyl transferase) and Chs (Chalcone synthase) are mapped on the short arm of chromosome and closely linked with F_o1D and F_v/F_mi1D; Per (Peroixdase) and Sod (Superooxide dismutase) are closely linked with F_v/F_mi2Bb on 2BS; Cbp1 (Chitin binding protein) closely linked with Tm2Bb on 2BL; Lpx (Lipoxygenase) closely linked with T_m4A on 4AS and with F_v/F_mi5B on 5BL; Pr1b (Pathogenesis-related gene) and Mpc1 (myb protein c1) linked with T_m7D and F_vi7D on 7DS; F_v7D linked with Cht1b (Chitinase 1b); and Tha1 (Thaumatin) on the distal of 7DL. One locus for F_m was identified on chromosome 7 in maize (Lebreton *et al.* 1995). Maize chromosome 7 was of colinearity with wheat chromosome 5 (Moore *et al.* 1995). One major additive QTL for F_m was also mapped on the telomere of 5AS in this work (Fig 2). Three QTLs were detected for F_v/F_m on chromosome 1 and 8, and one QTL on chromosomes 8 for F_o in maize (Fracheboud *et al.* 2002). Maize chromosome 1 was of colinearity with wheat chromosome 4, 5 and 7, and chromosome 8 was of colinearity with wheat chromosome 1 and 3 (Moore *et al.* 1995). In our work one interaction QTL for F_v/F_m was mapped on 1A and 1D, two interaction QTLs for F_v/F_m were mapped on 3B, and one additive QTL for F_o was mapped on 1A, 1B and 1D (Fig 2). Three loci controlling chlorophyll content was mapped on the chromosomes 2S, 4S

and 7S respectively in rice (Wu *et al.* 1996). Rice chromosome 2 and wheat chromosome 6 showed colinearity; whereas wheat chromosome 2 and rice chromosome 4 and 7 showed colinearity (Moore *et al.* 1995). In our work two interaction QTLs for chlorophyll content were mapped on 2D, and one additive QTL for chlorophyll content was detected on 6D. In summary, our results are valuable for orthologous genes from different plant species, and for closely linked genes for different related traits in the same chromosome map region. It should be feasible to introgress these resistance loci into adapted genetic backgrounds by using a marker-assisted selection scheme, and is important for plant evolution and gene cloning and gene function and transgenic crop improving.

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