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Mapping and analysis of quantitative trait loci for grain oil content and agronomic traits using AFLP and SSR in sunflower (*Helianthus annuus* L.)

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Abstract Crosses were made between two inbred lines of sunflower. Parents and 118 F_3 families were planted in the field in a randomized complete block design in two replications. Genetic control for some agronomical traits: grain weight by plant (GWP), 1,000-grain weight (TGW), percentage of oil in grain (POG) and sowing to flowering date (STF) was investigated in F_3 families and their parents. Genetic variability was observed among the 118 F_3 families for all the traits studied. Genetic gain was obtained when the best F_3 family, or the mean of 10% of the selected families was compared with the best parent for GWP, TWG and POG. Heritability was 0.23 for GWP, 0.55 for TGW, 0.57 for POG and 0.32 for STF. A set of 244 F_3 families from the same cross, including the above 118 mentioned families and their two parents, were screened with 276 AFLP and microsatellite markers and a linkage map was constructed based on 170 markers. Two putative QTLs for the GWP trait (*gmp*), one QTL for TGW (*tgw*), six QTLs for POG (*pog*) and two for STF (*stf*) were detected. The percentage of phenotypic variance explained by each QTL ranged from 2.6% to 70.9%. The percentage of total phenotypic variance explained was 50.7% for GWP, 5.4% for TGW, 90.4% for POG and 89.3% for STF. Although these regions need to be more-precisely mapped, the information obtained should help in marker-assisted selection.

Keywords Sunflower · Genetic variability · AFLP · SSR · QTL · Genetic map

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Introduction

Sunflower (*Helianthus annuus* L.) is grown mostly as a source of vegetable oil and proteins in the world. The main objectives of sunflower breeding programs are the development of productive F_1 hybrid cultivars with high oil yield. Sunflower oil yield is determined by the product of grain yield per unit area and the oil percentage in the grain. Therefore, consideration of both components is important when breeding for high oil yield (Leon et al. 1995). Yield in sunflower, as in all other crops, depends on many characters, especially yield components which are controlled by several genes, their effects being modified with environment (Fick and Miller 1997). Heritability for yield is relatively low compared to other agronomic traits (Fick 1978), whereas oil content in grain heritability is rather high and was estimated between 0.65 to 0.70 (Fick 1975). The number of days from sowing to maturity varies widely among sunflower cultivars with a range of 75 to 150 days (Fick 1978). Therefore, selection of genotypes with the most appropriate cycle length is critical. Polygenic inheritance patterns are reported for days from sowing to flowering in sunflower (Stoenescu 1974; Machacek 1979). Heritability of the sowing to flowering date ranges from 0.62 to 0.95 (Jan 1986). Gene action is usually additive (Alvarez et al. 1992; El-Hity 1992) but dominant effects are also observed for this trait (Jan 1986). The vegetative period in sunflower is controlled by several genes affecting the date of flowering and maturity, and others controlling photoperiodism (Stoenesn 1974).

The importance of molecular markers in sunflower genetic analysis was demonstrated by several research works. Relationships among inbred lines of sunflower were studied by Gentzbittel et al. (1994) and by Berry et al. (1994) using low-copy genomic and cDNA probes. Species with a big genome, like sunflower (2n = 34), require techniques which provide a high number of markers. The AFLP (amplified fragment length polymorphism) is considered to be an efficient marker technology due to its high multiple ratio (Pejic et al. 1998).

Hongtrakul et al. (1997) showed that AFLP is a powerful technique for genetic fingerprinting in sunflower. This technique was previously used in the establishment of several genetic maps, like rice (Mackill et al. 1996), maize (Castiglioni et al. 1999), ryegrass (Bert et al. 1999), tomato (Haanstra et al. 1999), melon (Wang et al. 1997), pine (Remington et al. 1999), eucalyptus (Marques et al. 1998) and recently sunflower (Flores Berrios et al. 2000; Rachid Al-Chaarani et al. 2001). Simple sequence repeats (SSRs), called microsatellites, are also used as molecular markers. Their polymorphism has shown high efficiency and are used for genetic mapping, population and evolutionary studies, as well as for fingerprinting and pedigree analysis (Hazan et al. 1992; Plaschke et al. 1995; Rongwen et al. 1995; Guilford et al. 1997). SSR markers are thus now recognized as one of the most-efficient molecular markers. However, in some dicot species, a lack of polymorphism on an uneven repartition on the genome has been reported, as in tomato (Broun and Tanksley 1996). For sunflower, only a few SSR markers are described, thus limiting their use in academic research programs. The first RFLP map of sunflower was reported by Gentzbittel et al. (1995) based on three different F_2 crosses and using RFLP of cDNA probes. Gentzbittel et al. (1999) presented an updated version of this map of sunflower also using RFLP markers. Two other RFLP maps were also published (Berry et al. 1995; Jan et al. 1998). Finally Flores Berrios et al. (2000) established a sunflower map by AFLP markers.

Identification of chromosome regions with effects on grain yield, oil percentage in grain and other agronomic traits would increase our understanding of the genetic control of the characters. Starting a program of marker-assisted selection becomes possible after the identification of QTLs for traits of interests. QTLs controlling important traits such as resistance to downy mildew and black stem (Rachid Al-Chaarani et al. 2001), percentage of oil in grain, 1,000-grain weight and Sclerotinia tolerance (Mestries et al. 1998), days from sowing to flowering with the effect of the environment (Leon et al. 2001) and photosynthesis parameters (Hervé et al. 2001), are detected in sunflower. In this paper, we present a new genetic map of sunflower, constructed with both AFLP and microsatellite markers, based on a population of 244 F_3 families, obtained by crossing two inbred lines of sunflower. QTLs for some agronomical characters like seed yield, the percentage of oil in grains and the time of sowing to flowering, were identified.

Materials and methods

Field experiment

Two inbred lines, L1 (restorer line) and L2 (maintainer line) of sunflower, were selected from the Syngenta Seeds company sunflower collection. Crosses were made between them, and 118 F_3 families of this cross were used in this study. Parents and the 118 F_3 families were planted at 2000 in the experimental field of INRA (Toulouse France), in a randomised block design with two replications. Each replication per parent or F_3 family consisted of a plot with three rows 4.6-m long. Spacing was 30 cm between plants in rows 50-cm apart, which makes a total number of about 48 plant per plot. Plants were harvested at maturity and grain weight per plant (GWP), 1,000-grain weight (TGW) and the percentage of oil in grains (POG) were measured for each parent or F_3 family in each replication. Days from sowing to flowering (STF) were also recorded when 50% of the plants of a plot were at anthesis.

Statistical analysis was carried out in order to determine the main effect of F_3 families for the studied traits and means, separated using a Newman-Keuls-test ($P = 0.05$). Heritability was estimated according to the following formula: $h^2 = 100\ \sigma^2g/\sigma^2p$, where σ^2g = genetic variance and σ^2p = phenotypic variance. The mean of the 118 F_3 families and that of their parents were compared for all traits. Genetic gain expressed as the difference between the mean of the best F_3 family, or as the mean of 10% selected F_3 families and the best parents, was also determined for the studied traits.

DNA extraction

Two hundred and forty four F_3 families of the cross 'L1×L2', including the 118 F_3 families used in the field experiment and their two parents, were grown in pots in the greenhouse and used for AFLP and SSR analysis. Leaf tissue from 16 10 day-old plants of each F_3 family or parent was collected for the extraction of total DNA. Genomic DNA was isolated according to the Nucléon Phytopure Kit extraction and purification protocol (Amersham Life Science). Briefly, leaf samples (50 mg) were ordered in deep-well plates in order to be frozen at –20 °C. After breaking the cell wall, the cells were lysed in a reagent containing potassium SDS which is known to form complexes with proteins and polysaccharides. Chloroform was then added along with a modified resin. After incubation and centrifugation, DNA pellets were washed once with 70% ethanol and re-suspended in 100 µl of TE.

AFLP procedure

Amplified fragment length polymorphism was resolved according to the AFLP Analysis System-I and the AFLP Starter Primer Kit protocol from Life Technologies, which is based on a two-step amplification strategy using *Eco*RI and *Mse*I primers (Vos et al. 1995). Two hundred and fifteen nanograms of total DNA for each F_3 family or parent sample was used in the restriction-ligation of *Eco*RI and *Mse*I adapters. All reactions were made in the Gene Amp PCR System 9700 Thermocycler (Perkin Elmer Applied Biosystems). The bands were first amplified with primers each having one selective nucleotide. Then, the diluted PCR products were employed as a template for the second amplification using primers containing three selective nucleotides (Table 2). The procedure was performed as described in the kit, using *Taq* DNA Polymerase (Life Technologies) and γ^{33}P[ATP] from Amersham Pharmacia Biotech. Following the amplification, reactions products were mixed with an equal volume of formamide dye (98% formamide, 10 mM EDTA, bromophenol blue and xylene cyanol). The gel was pre-equilibrated by passing an electric current through it (at a constant 100 W) for 30-min denaturation for 3 min at 90 °C, and 5 µl of each sample was loaded on a 6% denaturing polyacrylamide gel in a 38 × 50 cm Sequi-Gen GT Nucleic Acid Electrophoresis Cell (BIO-RAD). The reactions products were then separated under these conditions, for 2 h 30 min. Following the separation, the gel was vacuum-dried and exposed to Hyperfilm MP (Amersham Pharmacia Biotech) for 2 days. Mapping data were obtained by visual scoring of autoradiograms. AFLP markers were named using the universal code of primers, followed by the molecular weight of the band.

SSRs procedure

A total of 465 pairs of proprietary SSR primers was screened for polymorphism, among which 61 pairs presenting polymorphism between the parents were used in this study. SSR markers employed in the present study are the property of the Syngenta Seeds Company (B.P.27 Saint Sauveur France). SSR primers could be made available for collaborative research purposes under written request. PCR amplifications were performed in a solution of 10 µl containing 27 ng of each primer, 0.1875 U of Platinum *Taq* DNA polymerase (Life Technologies), 125 µM of each dNTP, 1 × reaction buffer (20 mM Tris-HCl; pH = 8.4, 50 mM KCl), 1.65 mM of $MgCl_2$, ddH_2O and 10 ng of template DNA. All SSR amplifications were performed in a Gene Amp PCR System 9700 Thermocycler (Perkin Elmer Applied Biosystems). Two different PCR programs were used at two different annealing temperatures depending on the SSR primers. Program 1: denaturation at 94 °C for 2 min followed by 32 cycles of: 1 s at 94 °C, 1 s at 55 °C and 5 s at 72 °C, then held at 15 °C. Progam 2: denaturation at 94 °C for 2 min followed by 40 cycles of: 15 s at 94 °C, 45 s at 60 °C, 2 min at 72 °C, then held at 15 °C. Products were separated by electrophoresis in a cooled horizontal gel system (Sigma-Aldrich SARL). Gels were prepared using 0.5 × TBE (Eurobio) and Superfine Resolution Agarose (Interchim) at 30 g/l. Eight microliters of the PCR mix were loaded into the wells of a 1-mm wide comb. The gels were run at 400 V for 1.5 h and stained using a solution of 1 µ g/ml of ethidium bromide for 20 min.

Linkage analysis

AFLP and microsatellite polymorphic bands were scored as present (1) or absent (0) on autoradiograms, whereas unreliable ambiguous bands were scored missing (–). Data coded (0) and (1) was transformed to A,B,C,D genotype codes, according to the presence of the band for the parents L1 or L2, following the Mapmaker convention. A molecular linkage map based on a data set consisting of 276 marker loci (61 SSRs and 215 AFLPs) was constructed using MAPMAKER 3.0 (Lander et al. 1987). Linkage groups were obtained with a LOD score of 3.5. The Kosambi mapping function was used to transform the recombination frequency to genetic distances (Kosambi 1944). The distance between two markers differs from 5 cM to 33.1 cM.

QTL analysis

The chromosomal location of QTLs for the studied traits was resolved by composite Interval mapping (CIM) using QTL Cartographer Version 1.13 model 6 (Basten et al. 1999). This QTL cartographer integrated two parameters for CIM: the number of markers which control the genetic background (n_m = 15), and a window size (w = 20) that will block out a region of the genome on either side of the markers flanking the test site. The inclusion of background markers makes the analysis more-sensitive to the presence of a QTL in the target interval. At each interval, the significance of the association is tested by the likelihood-ratio statistic (LRS) (Haley and Knott 1992). The LRS test is statistically convenient because it is asymptotically a χ_2 distribution (Manly and Olson 1999).

Results and discussion

Genetic variability

Analysis of variance of 118 F_3 Families and their parents L1 and L2 showed that the effect of the genotype was highly significant for all the characters studied (data not presented). The genetic variability of parental genotypes and the 118 F_3 Families, together with the genetic gain for each character, are presented in Table 1. The difference between parents is significant for the percentage of oil in grains (POG) and sowing to flowering (STF). However, the difference between parents is not significant for the grain weight per plant (GWP) and the 1,000-grain weight (TGW). The difference between all F_3 families ($\bar{x}$ F_3) and their parents ($\bar{x}$ P) for the studied traits was not significant (Table 1) indicating that the 118 F_3 families in this experiment are representative of the total possible recombinations from the cross 'L1×L2'.

Table 1 Genetic gain and heritability for grain weight by plant (GWP), 1,000-grain weight (TGW), percentage of oil in grain (POG) and sowing to flowering date (STF) in a population of 118 F_3 families of sunflower and their two parents

Item	GWP(g)	TGW(g)	POG	STF(d)
L_1 (P1)	44.03	54.45	49.23	102.00
L_2 (P2)	41.97	62.00	46.67	98.00
P1 – P2	2.06^{ns}	-7.55^{ns}	2.56*	4.00*
$\bar{X}_P = (L_1 + L_2)/2$	43.00	58.23	47.95	100.00
$\bar{X}_{F3}$ [a]	48.92	60.12	48.95	100.89
$\bar{X}_{F3} - \bar{X}_P$	5.92^{ns}	1.89^{ns}	1.00^{ns}	0.89^{ns}
Best family (BF_3)	71.67	95.50	53.26	96.50
BP	44.03	62.00	49.23	98.00
GG[c]=BF_3 – BP[b]	27.1*	33.50*	4.03*	-1.50^{ns}
10% SF_3[d]	62.22	74.02	52.26	97.71
GG[e]=10% SF_3 – BP	18.19*	12.02*	3.03*	-0.29^{ns}
h^2	0.23	0.55	0.57	0.32

*Significant at $P = 0.05$; ns, non significant at $P = 0.05$
[a] $\bar{X}_{F3}$, mean of all the F_3 families
[b] BP, best parent
[c,e] GG genetic gain when the best F_3 family or 10% of the selected F_3 families are compared with the best parent
[d] 10%SF_3: 10% of the best F_3 families in the population

From a cross between two lines, breeders are always expecting to obtain genetic gain, which is expressed by the superiority of one or several progenies compared to the parents. This phenotypic superiority is genetically translated by the polygenic nature of the trait and the existence of positive alleles coming from both parents in some progenies of a cross. A significant genetic gain was obtained by comparing the best parent (BP) with the best family (BF_3) for grain weight per plant (GWP), 1,000-grain weight (TGW) and the percentage of oil in grains (POG). We have noticed that the best F_3 family (BF_3) has the highest values for grain weight per plant (GWP) and 1,000-grain weight (TGW). By comparing the best parent (BP) with 10% of the selected F_3 families having the best values, all the traits presented a genetic gain. Heritabilities were rather high for 1,000-grain weight (TGW) and the percentage of oil in grains (POG) (0.55 and 0.57 respectively). Whereas, low values for heritability (0.23 and 0.33) were observed for grain weight per plant (GWP) and sowing to flowering (STF) respectively. Fick (1978) reported also a high heritability for the percentage of oil in the grain (POG) and low values for grain weight per plant (GWP). Our results concerning the low value (0.33) of heritability for sowing to flower-

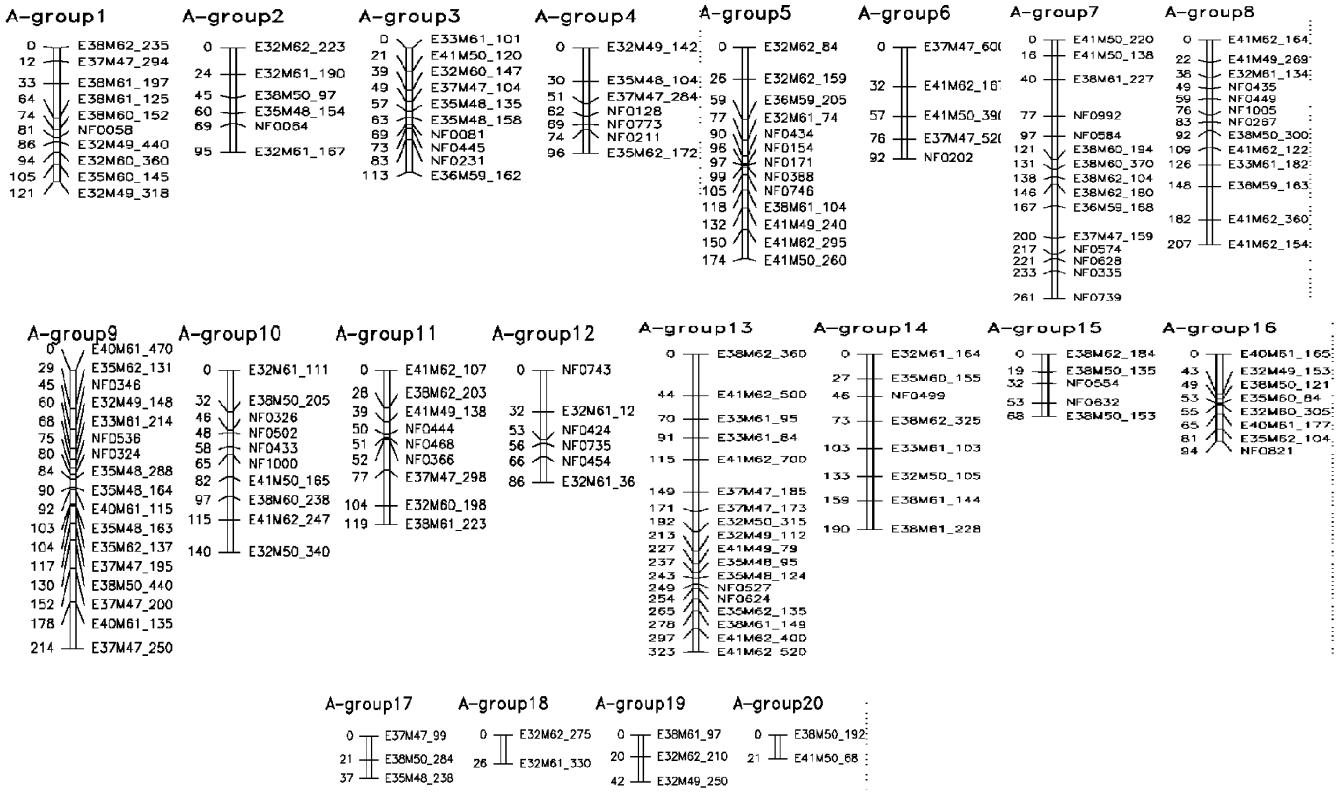


Fig. 1 AFLP and microsatellites linkage map of sunflower based on a population of 244 F3 families. The names of the markers are shown at the right and their map position (cM) at the left. Distances are in Kosambi cM

Table 2 AFLP primer combinations tested and number of mappable markers in each primer pair used for the map construction

Primer combination	Number of markers	Primer combination	Number of markers
E41M62	24	E35M62	9
E41M49	11	E41M50	12
E32M62	16	E32M61	15
E40M61	9	E32M50	12
E32M60	10	E35M48	14
E38M50	15	E38M60	6
E38M61	17	E32M49	22
E36M59	10	E37M47	22
E33M61	14	E35M60	7
E38M62	9		

ing (STF), comparing with those obtained by Alvarez et al. (1992) and El-Hity (1992), might be explained by the low variability for genes controlling this character in the parents of the cross.

Linkage map

The genetic map presented in this paper (Fig. 1) was constructed using 215 AFLP markers which were identified by the use of 19 primer combinations (Table 2) and 61 microsatellite markers. In order to construct this map, two preliminary maps were realized, based on molecular markers in the same linkage phase. Typically, they concern the data coming from markers presenting the bands for the parent L1, and the other by using the data from markers presenting the bands for the parent L2. Problems in estimating recombination frequency between dominant markers in the repulsion phase are thus minimized. These two maps had the 61 microsatellites markers in common, which enable us to construct the final map (Fig. 1). Out of 276 markers analysed, 170 were placed in 20 groups by the use of a minimum LOD score of 3.5 and a maximum recombination value of 0.29. The groups ranged from 21 to 323 cM in length and carried 2–18 markers. An unexpected clustering of SSR markers was observed. The total length of the map is 2,539 cM which represents at least one marker for every 14.9 cM on average (Table 3). Flores Berrios et al. (2000) using recombinant inbred lines and AFLP markers constructed a genetic map of 2,833.7 cM, updated in Rachid Al-Chaarani et al. (2001). Many factors as to the nature of

Table 3 Marker distribution among the linkage groups

Group	Marker number	Number of markers placed	Length (cM)	Average distance (cM)
1	14	10	121	12.1
2	6	6	95	15.8
3	15	10	113	11.3
4	10	7	96	13.7
5	16	13	174	13.4
6	5	5	92	18.4
7	33	15	261	17.4
8	15	13	207	15.9
9	29	17	214	12.6
10	15	10	140	14
11	20	9	119	13.2
12	6	6	86	14.3
13	27	18	323	17.9
14	8	8	190	23.7
15	5	5	88	17.6
16	10	8	94	11.7
17	3	3	37	12.3
18	2	2	26	13
19	3	3	42	14
20	2	2	21	10.5
Total	276	170	2,539	14.9

Table 4 Map position and effect of QTLs detected in a population of 118 F_3 families for some agronomical traits

QTL	Linkage group	Position[a]	LOD	Additive	Dominance	R2[b]	TR2[c]
Grain weight per plant:							
gwp-9-1	9	83.75	3.11	2.25	–5.09	16.40	43.90
gwp-9-2	9	99.73	4.90	3.07	–5.44	23.30	50.70
1,000-grain weight:							
tgw-16-1	16	54.73	3.38	–3.08	–1.66	5.40	24.70
Oil percentage in grain:							
pog-9-1	9	74.14	3.12	–0.65	0.72	11.40	44.30
pog-9-2	9	91.73	3.83	–1.03	–0.19	8.40	41.30
pog-11-1	11	41.41	4.90	1.13	0.13	12.50	43.20
pog-11-2	11	56.43	3.87	1.03	0.26	9.20	40.00
pog-12-1	12	26.01	5.54	1.46	1.44	3.80	47.10
pog-12-2	12	31.56	7.6	1.48	1.47	6.00	43.10
pog-13-1	13	313.24	3.12	–1.15	1.48	47.10	90.40
Sowing to flowering date:							
stf-9-1	9	95.73	3.62	0.90	0.85	2.60	27.80
stf-10-1	10	20.01	4.31	1.89	–0.80	70.90	89.30

[a] Expressed in Kosambi CM, from north of the linkage group
[b] Percentage of individual phenotypic variance explained, value determined by QTL Cartographer, Version 1.13 (Basten et al. 1999)
[c] Percentage of phenotypic variance considering epistasis effects, value determined by QTL Cartographer, Version 1.13 (Basten et al. 1999)

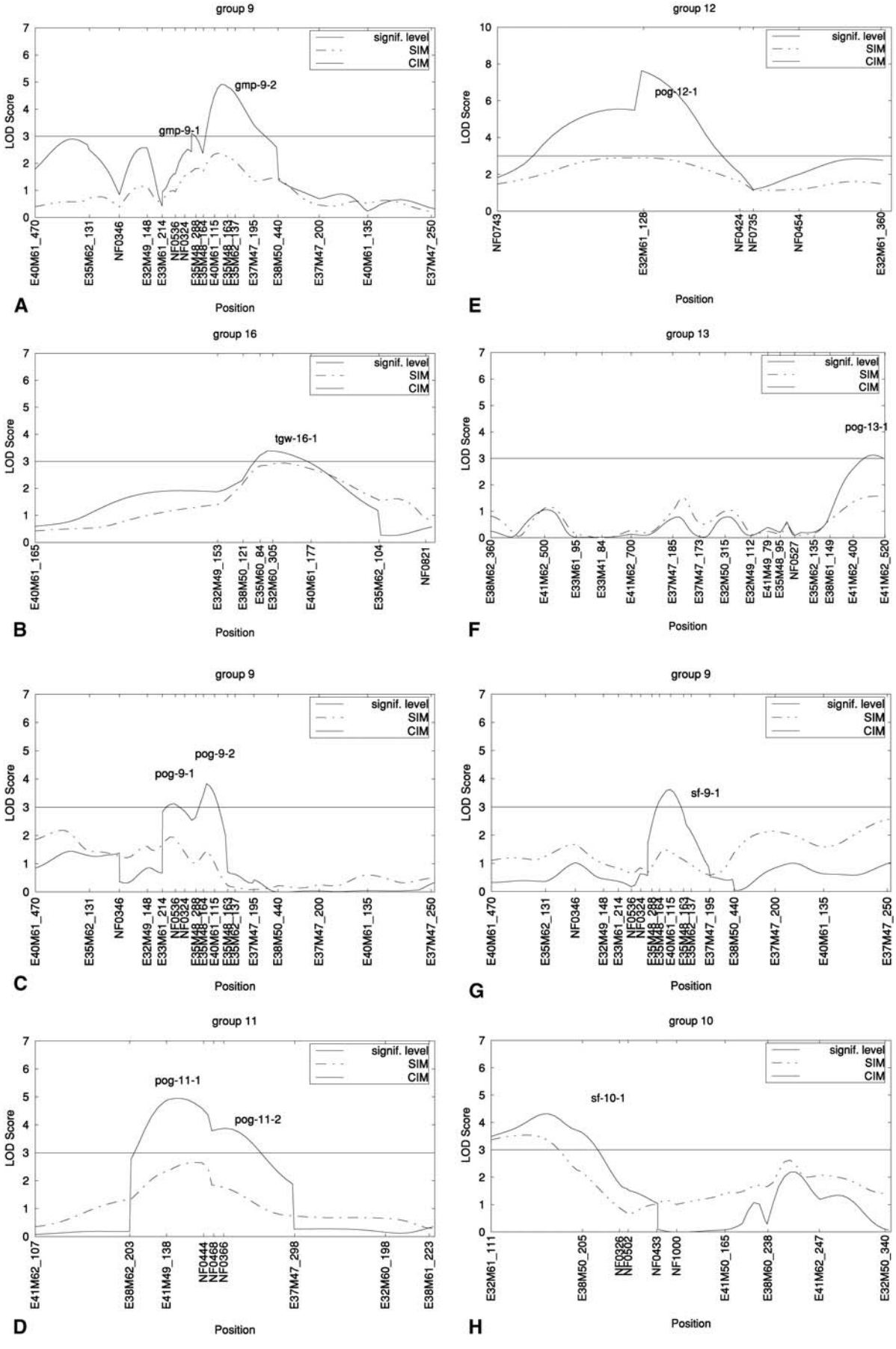
the population studied, the number of individuals and the number of markers, might change the recombination rate and in consequence the distance between two loci. Some other genetic maps of sunflower were also developed using different techniques: RFLP (Berry et al. 1995; Gentzbittel et al. 1995; Jan et al. 1998; Gentzbittel et al. 1999) and RAPD (Reiscberg et al. 1996).

QTL analysis

Fourteen putative loci associated with the four studied traits were identified (Table 4), which are located on six different linkage groups (Fig. 2). The QTLs were designated as follows: *gwp* for grain weight per plant, *tgw* for 1,000-grain weight, *pog* for percentage of oil in grain and *stf* for sowing to flowering, followed by the corresponding number of linkage groups and the corresponding number of QTLs on the group.

The map position and characteristics of QTLs detected are referred to in Table 4. Two QTLs were detected for the grain weight per plant (GWP), at 83.75 and 99.73 cM on group 9, respectively. The LOD score was 3.1 for the first QTL (*gmp-9-1*) and 4.9 for the second one (*gmp-9-2*). The phenotypic variance explained (R2) by *gmp-9-1* and *gmp-9-2* was 16.4% and 23.3%, whereas the total phenotypic variation (TR2) explained in the model was 43.9% and 50.7%, respectively. For this character, the dominant effect is more important than the additive one, and alleles having positive effects come from the L1 parent (Table 4).

As for as 1,000-grain weight (TGW) is concerned only one QTL was observed (*tgw-16-1*) which is situated


group 9
signif. level
SIM
CIM
LOD Score
gmp-9-2
gmp-9-1
E40M61_470
E35M62_131
NF0346
E32M49_148
E33M61_214
NF0536
NF0324
E35M48_288
E35M48_164
E40M61_115
E35M62_137
E37M47_195
E38M50_440
E37M47_200
E40M61_135
E37M47_250
Position
A
group 12
pog-12-1
NF0743
E32M61_128
NF0424
NF0735
NF0454
E32M61_360
E
group 16
tgw-16-1
E40M61_165
E32M49_153
E38M50_121
E35M60_84
E32M60_305
E40M61_177
E35M62_104
NF0821
B
group 13
pog-13-1
E38M62_360
E41M62_500
E33M61_95
E33M41_84
E41M62_700
E37M47_185
E37M47_173
E32M50_315
E32M49_112
E41M49_79
E35M48_95
NF0527
E35M62_135
E38M61_149
E41M62_400
E41M62_520
F
group 9
pog-9-2
pog-9-1
C
group 9
sf-9-1
G
group 11
pog-11-1
pog-11-2
E41M62_107
E38M62_203
E41M49_138
NF0444
NF0468
NF0366
E37M47_298
E32M60_198
E38M61_223
D
group 10
sf-10-1
E32M61_111
E38M50_205
NF0433
NF1000
E41M50_165
E38M60_238
E41M62_247
E32M50_340
H

◀ **Fig. 2A–H** Genetic map and LOD-score plots showing the localisation of putative QTLs associated with: (**A**) grain weight by plant (*gwp*), linkage group 9, (**B**) 1,000-grain weight (*tgw*), linkage group 16, (**C**) percentage of oil in grains (*pog*), linkage group 9, (**D**) percentage of oil in grains (*pog*), linkage group 11, (**E**) percentage of oil in grains (*pog*), linkage group 12, (**F**) percentage of oil in grains (*pog*), linkage group 13, (**G**) sowing to flowering date (*stf*), linkage group 9 and H sowing to flowering date (*stf*), linkage group 10, in sunflower as detected by composite interval mapping (CIM)

on linkage group 16 at 54.73 cM. The LOD score was 3.8. The individual effect of this QTL on the expression of the character was 5.4%. Mestries et al. (1998) have detected three QTLs for 1,000-grain weight (TGW) in the progenies of a cross, but their analysis was based only on Simple Interval Mapping. The percentage of oil in grains coming from F_3 families presents six QTLs. The most important QTL detected is located on linkage group 13 (*pog-13-1*) with a very important effect on phenotypic variance (R^2 = 47%). The epistasis effect is important in the expression of this QTL, and its effect on oil synthesis can reach 90.4% of the phenotypic variance (TR2). This could indicate an epistasis effect between this QTL and those detected in the other groups. The additive effect is more important than dominance, and alleles with a positive effect come from the parent L2 for this QTL. Mestries et al. (1998) have also identified three QTLs for oil content in the sunflower. For sowing to flowering (STF) two QTLs were detected in linkage groups 9 and 10 (*stf-9-1* and *stf-10-1*). The individual effects of these two QTLs are 2.6% and 70.9%. The second QTL (*stf-10-* 1) explains 89.3% of the total phenotypic variance (TR2), which could indicate an epistasis effect. The additive effect is predominant and alleles giving a positive effect come from the parent L1. Recently six QTLs associated with growing-degree-days to flowering and photoperiod response were identified by Leon et al. (2001). The results presented here revealed regions related to different agronomic traits studied, especially the percentage of oil in the grain (POG). Comparison of the present results with previously described data are, however, difficult as different markers and nomenclature were used. Although these regions need to be more precisely mapped, the information obtained should help in marker-assisted selection.

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