ASSESSMENT OF SUNFLOWER GERMPLASM SELECTED UNDER AUTUMN PLANTING CONDITIONS

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ABSTRACT

Agronomic potential of traditional sunflower spring varieties is low because its flowering and grain filling are often exposed to mid and end-season drought. To overcome this, new breeding strategy consisted of selecting varieties tolerant to winter cold in order to shift to autumn or early winter planting. Nowadays, 'Ichraq' is the only one registered autumn variety. The objective of this research is to evaluate various genotypes having been selected in different environments under autumn planting conditions. This germplasm was planted early at winter during two years (2013 and 2014) at 'Annoceur', a mountain site known for its pronounced winter cold. Morphological, physiological, agronomic and technological parameters were considered for the germplasm assessment. Analysis of variance showed significant differences between genotypes for most of these parameters. Plantlet initial vigor average was 3.5 varying from 1 for genotype M32 to 5 for AN8. Leaf area average was 162 cm² varying from 25 to 375 cm² for genotypes M17 and AN34, respectively. Total chlorophyll content average was 43 mg/g, varying from 28 to 79 mg/g for genotypes K7 and M29, respectively. Number of days from sowing to flowering varied from 162 for genotype AN21 to 180 for genotypes M27 and M29. Mean seed yield per plant was 49 g, with a large variation from 8 to 110 g for M18 and K8, respectively. Mean seed oil content was 36%, ranging from 22% for M8 to 47% for K4. Genotypes having exhibited more performance than 'Ichraq' were selected to develop new sunflower germplasm suitable for autumn or early winter sowing.

INTRODUCTION

Agricultural sector continues to dominate Morocco's economic activity. The rural population accounts for 40% of the total population. The Agriculture thus proved an effective engine of economic growth and guaranteed food security. To upgrade and boost domestic agriculture, different strategies have been implemented during the Moroccan contemporary history. The latter being the Green Morocco Plan implemented since 2008. Owing to its importance in the cropping system and the food security challenge in vegetable oils, the oilseed is considered among the priority sectors. Since 2001, the year of oilseed sector reform, and until 2013, the year of signature of a sector program contract, sunflower was the major annual oilseed crop grown in Morocco, with an average area about 50000 ha and an average seed yield below 1 t/ha. Indeed, national seed oil production covers barely 2-3% of the overall needs of the edible oil in the country estimated at over 410.000 t. The gap is covered by importation which has negative repercussion on the economy and food security of our country (Nabloussi et al., 2015).

In Morocco, sunflower traditionally sown in spring has limited productivity as it does not benefit the fall and/or winter precipitation, and it is often exposed to drought and high temperatures of mid and late cropping cycle. Such constraints coincide with periods of flowering and seed filling that are critical for determining seed productivity and seed oil content (Ouattar et al., 1992). Its cultivation is often secondary and is considered as catch crop, following early droughts or floods that affect growing of autumn crops, mainly cereals. However, several studies have shown the benefits of early planting (autumn or early winter) in improving the seed yield and oil content in Morocco (Boujghagh, 1993 ; Gosset and Vear, 1995 ; Aboudrare et al., 2000), Spain (Gimeno et al., 1989) and France (Allinne et al., 2009). Early

sowing, two to three months earlier than conventional sowing, induced a significant drop in temperature at planting and during early stages of vegetative growth (Allinne et al., 2009). The characterization and evaluation of sunflower genotypes adapted to low temperature conditions, during early vegetative growth stages, requires analyzing the impact of such conditions on the physiological processes associated with initial seedling vigor and plant cold tolerance. Agronomic, morphological, physiological, technological and biochemical attributes could be taken as valuable criteria to identify and select genotypes adapted to winter cold conditions. Nowadays, "Ichraq" is the only autumn variety registered in the Moroccan Official Catalogue (Nabloussi et al. 2008). It is a late maturing, winter cold tolerant and combines good seed yield and high seed oil content. Current research continues to develop new sunflower populations, resistant (or tolerant) to winter cold and agronomically performant, which would be the basis for selection of new improved varieties better than the variety "Ichraq". Thus, the present work aimed to evaluate new sunflower genetic materials for agro-morphological, physiological and technological traits under early winter planting conditions.

MATERIALS AND METHODS

Plant materials

The plant material used in this study consisted of 46 sunflower genotypes including 'Ichraq', the first and only one autumn variety, considered as check, and 45 individual selected plants derived from 'Ichraq'. As this latter is a population variety (Nabloussi et al., 2008), there was opportunity to select individual plants (PS) in order to release new germplasm that will be more performant than 'Ichraq'. The 45 PS were selected in various environments for their vigor, habit and agro-morphological performances.

Methods

The 46 genotypes were planted on 2 January 2014 at the INRA experimental station located at 'Annoceur', mountainous area known for its rough winter cold. It is located 50 km from Fez city, in the north of Morocco, at an altitude of 1350 meters. During the cropping cycle, the minimum temperature was -5°C, registered in January and February whilst the maximum temperature was 37°C, recorded in May.

Trial was conducted under rainfed conditions following a randomized complete blocks with two replications. Each genotype was sown in two 5 m rows spaced by 60 cm. In each row, plants were spaced by 30 cm. Initial N-P-K fertilization was 80-80-30 units, respectively, followed by cover N fertilization with two inputs of 40 units, one at stem elongation stage and the other at flowering stage. No phytosanitary treatment was applied.

Morphological, phenological, physiological, agronomic and technological parameters were studied. During plant vegetative growth, plant height (cm), growth rate (cm/d), collar diameter (mm), initial vigor of young seedlings (following a grading scale of 1 to 5), number of leaves per plant, leaf area (cm^2) and number of branches per plant were measured. Flowering time of each genotype was determined by counting the number of days between planting date and the date when 50% of plants of this genotype have flowered. Chlorophyll content (mg/g) was calculated according to the method of Billore and Mall (1975). The optical density (OD) of all the supernatant obtained was measured in a spectrophotometer at 645 nm and 663 nm. The concentrations of chlorophyll pigments are given by the following formulas:

CHL A = 12.7 (OD 663) – 2.69 (DO645)

CHL B = 22.9 (OD 645) – 4.56 (DO663)

At maturity, head diameter and head aborted diameter were measured (cm). After harvest, total seed yield (q/ha), seed yield per plant (g) and its components (number of seeds per propeller and 1000 seeds weight) are determined. Also, seed oil content was determined using RMN method (Oxford 4000).

Descriptive analysis of gathered data, analysis of variance and analysis of correlation were performed using different procedures of SAS program. Duncan's new multiple range test was applied to compare genotypes means.

RESULTS AND DISCUSSION

Morphological parameters

Analysis of variance showed there were significant differences ($P < 0.001$) between the 46 genotypes for all studied parameters (Table 1). Initial vigor of young seedlings varied from 1 for genotype M32 to 5 for genotype AN8, with an average of about 3.5, higher than the check vigor (3). In many studies, seedling and plantlet initial vigor was found as a good selection criterion correlated with the adaptation and the performance of genotypes under environmental abiotic stresses (Foolad and Lin, 2001). In the present work, all genotypes having an initial vigor of 4 or 5 will be selected for further evaluation and germplasm improvement. For growth rate, the overall mean was 2.31 cm/d, with a minimum of 0.53 cm/d, registered for genotype AN21 and a maximum of 3.92 cm/d for genotype M34, slightly higher than that of the check, which was 3.15 cm/d (Table 1). Genotypes having growth rate higher than that of the check will be selected. The average plant height was 147 cm, with a variation from 75 to 200 cm for M18 and K20, respectively. Plant height of the check was about 167 cm. Higher is a plant more it is susceptible to lodging and late drought (Sposaro et al., 2008). Plants with a height less than the observed average (< 145 cm) could be interesting for selection. Number of leaves per plant varied from 17 for M4 to 38 for K3, with an average of 27.5 leaves per plant. The check had 25 leaves per plant. The average leaf area was 162 cm², which is equal to the check value. The genotypes M17 and AN34 exhibited the extreme values: 24.5 and 374.85 cm², respectively. Elevated number of leaves per plant and high leaf area are correlated with high plant transpiration (Romero-Aranda et al., 2001). Thus we aimed to select those plants having less than 25 leaves and a leaf area less than 162 cm². Regarding collar diameter, genotype M7 exhibited the strongest value which was about 31 mm, whilst genotype M18 showed the lowest value which was 11 mm. The overall mean value was 20 mm and the check value was 23 mm. Like initial vigor, collar diameter in an indicator of good adaptation under stressed environments (Liua et al., 2012). Therefore, all the genotypes exhibiting a collar diameter more than the observed average (20 mm) could be selected for further evaluation. Among the 46 studied genotypes, 27 ones, including 'Ichraq' the check variety, had no branching, whilst 19 ones were branched, with a number of branches per plant varying from one to six. Genotype M30 was the most branched, having six branches per plant. The overall average was 0.93. Sunflower branching is an indicator of plants susceptibility to cold conditions (Alba et al., 2010). The plants selected for further evaluation and new germplasm constitution should have no branching.

Physiological parameters

Analysis of variance revealed significant effect of genotype on flowering earliness, chlorophyll a content and chlorophyll b content ($P < 0.001$), and non-significant effect on total chlorophyll content (Table 1). However, a large variation was observed, ranging from 28 mg/g for genotype K7 to 79 mg/g for genotype M29. The average total chlorophyll content was 43.21 mg/g, while the content concerning the check variety was 54.6 mg/g (Table 1). Genotypes maintaining high chlorophyll content under abiotic stresses, like as drought or cold, exhibit tolerance to such stresses (Yang et al 2015). All genotypes having total chlorophyll content higher than that of the check will be selected. Regarding chlorophyll a and chlorophyll b content, the genotype K4 exhibited the highest values for both types, 11.3 and 19.8 mg/g, respectively. The lowest contents were 0.86 mg/g, registered in genotype K9, and 1.74 mg/g, registered in genotype K8, for chlorophyll a and chlorophyll b, respectively. The check variety had 1.86 and 2.77 mg/g for these parameters, respectively. Vegetative period before flowering was too long, with an average duration exceeding 170 days from sowing date to flowering date. It ranged from 162 days for genotype AN21 to 180 days for genotypes M27 and M29. The check variety has bloomed in 170 days after sowing.

Flowering earliness is a desired character in environments under terminal drought stress (Ribot et al., 2012). Thus, genotypes having a sowing-flowering period shorter than that of the check will be selected.

Table 1. Analysis of variance (Mean square and significance level of differences) for agromorphological, physiological and technological traits of 45 sunflower accessions evaluated under early winter planting conditions in Annoceur 2014.

Parameter	Genotype	Average	Minimum		Maximum		value from Control Ichraq	Threshold for selection
			Value	Genotype	Value	Genotype		
IV(1)	*** (2)	3.49	1	M32	5	AN8	3	4-5
GR	***	2.31	0.53	AN21	3.92	M34	3.15	>3.15
PH	***	146.77	75	M18	200	K20	166.66	<145
NLP	***	27.46	17	M4	38	K3	25.33	<25
LA	**	162.23	24.5	M17	374.85	AN34	161.7	<162
CD	***	20.08	11.11	M18	30.82	M7	23.11	>20
CHLT	ns	43.21	28.2	K7	79.1	M29	54.6	>54
CHLA	***	4.04	0.86	K9	11.31	K4	1.86	>1.86
CHLB	***	6.77	1.74	K8	19.89	K4	2.77	>2.77
DSF	***	170.48	162	AN21	180	M29 M27	170	<170
NBP	***	0.93	0		6	M30	0	0
THD	***	12.79	6	M22 M18	23	K9	16.33	>16
AHD	ns	2.48	0		7	K3 K9	2	<2
PAD	ns	21.2	0		62.5	M32	13.96	<13.96
NSP	***	18.39	8	M22	27	K20 K8	20	>20
HSY	***	49.36	8.184	M18	110.3	K8	62.14	>62
TSW	***	47.52	12.4	M18	83.6	K8	56.86	>56
SOC	***	36.43	21.83	M8	46.85	K4	38.52	>38
TSY	***	27.42	4.54	M18	61.3	K8	34.52	>34

(1) IV: initial vigor, GR: growth rate, PH: plant height, NLP: number of leaves per plant, LA: leaf area, CD: collar diameter, CHLT: total chlorophyll content, CHLA: chlorophyll a content, CHLB: chlorophyll b content, DSF: days from sowing to flowering, NBP: Number of branches per plant, THD: total head diameter, AHD: aborted head diameter, PAD: percentage of aborted diameter, NSP: number of seeds per propeller, HSY: head seed yield, TSW: 1000 seeds weight, SOC: seed oil content, TSY: total seed yield (per hectare).

(2) *, ** and *** significant at 0.05, 0.01 and 0.001 levels, respectively. ns not significant.

Agronomic and technological parameters

Analysis of variance showed there were significant differences ($p < 0.001$) between the studied genotypes for all agronomic and technological parameters, excepted aborted head diameter (AHD) and percentage of aborted diameter (PAD) (Table 1). However, one could observe some variation between genotypes for AHD and PAD (Table 1). Most of the evaluated genotypes had no AHD, and among the few ones having

AHD, genotypes K3 and K9 exhibited the largest AHD, 7 cm. The overall average AHD was about 2.5 cm. The overall average PAD was about 21%, ranging from 0%, for most of the genotypes, to more than 62%, for genotype M32. All genotypes exhibiting some AHD should be discarded from the selected population as aborted sunflower head is an indicator of plant susceptibility to cold (Hladni et al., 2010). Average total head dimeter (THD) was 12.8 cm, ranging from 6 cm, for genotypes M22 and M18, to 23 cm, for genotype K9. The check variety 'Ichraq' had a THD of 16 cm, an AHD of 2 cm and a PAD of 14%. A large range was observed for number of seeds per propeller, from 8 in genotype M22 to 27 in genotypes K8 and K20. The check variety had a number of 20 seeds per propeller. Regarding seed yield per head, the overall mean was slightly higher than 49 g, and a large range was found, from 8 g in genotype M18 to 110 g in genotype K8, which is much higher than head seed yield of the check (62 g). Thousand seed weight (TSW) ranged from 12.4 g in genotype M18 to 83.6 g in genotype K8, and the average was 47.52 g. TSW of the check was about 57 g. The average total seed yield (TSY) was around 27 q/ha and there was a large variation from 4.54 q/ha in genotype M18 to 61.30 q/ha in K8. TSY of the check was slightly higher than 34 q/ha. Total head diameter, number of seeds per propeller, single head seed yield and TSW are components of TSY which are correlated with this latter, and thus could be considered as selection criteria for seed yield breeding (Yasin and Singh, 2010). In our study, we will select all those genotypes showing values higher than those of the check. Finally, seed oil content (SOC) fluctuated from 21.80% in genotype M8 to 46.85% in genotype K4, and had a mean value of 36.43%. The check 'Ichraq' had a SOC of 38.52%, which was slightly higher than the overall average. Genotypes with SOC exceeding that of the check will be selected. Table 2 shows the pools of genotypes selected, according to described threshold, for each of the studied parameters.

Pearce 1999 subdivided the plants into three categories according to their tolerance to cold and ability to adapt to low temperatures. Susceptible plants to low temperatures that suffer damage as early as 12 °C, tolerant plants to low positive temperatures and plants capable to acclimate to survive under temperatures below zero degree. Xin and Browse 2000 showed that they are a large number of physiological mechanisms that allow plants to better withstand severe stress (temperatures below zero) after a long time at low temperature (acclimatization). Many studies have shown low temperature direct effects on cells (Pearce, 1999), on seed germination (Durr et al., 2001), on photochemical reactions of photosynthesis and carbon fixation (Liua et al., 2012). Likewise, cold causes reduction of cell water content (Kacperska, 2004).

Our findings have shown there was a genetic diversity among the sunflower genotypes evaluated for most of the studied parameters. In all cases, these genotypes were compared with the check variety 'Ichraq'. This study allowed us to identify and select genotypes more interesting than the check for morphological, physiological, agronomic and technological parameters under winter early planting conditions. Globally, taking into account all these parameters, the genotypes AN8, AN3, AN34, AN33, AN27, AN23, AN21, AN24, K30, K20, K10, K3, K8, K7, K4 seemed to be performant and promising. After confirming their performance in further seasons, they could be useful for intercrossing to develop a new variety more performant and more tolerant to winter cold than 'Ichraq', the only one autumn variety registered to day in Morocco.

Table 2. Pools of sunflower genotypes selected for their performance on basis of morphological, physiological, agronomic and technological parameters under early winter planting conditions.

Parameters	Sunflower genotype pools
IV	AN8;AN6;AN3;M45;M43;M30;M26;M17;K8;K7;K6;K5;K4; A35;A34;A33; A32;A27;A23;A21;A13;A11
RG	M34;M32;M30;K30;K20;K10;K9;K8;K5;
PH	AN8;AN3;M43;M41;M37;M34;M32;M29;M26;M27;M22;M19;M18;M17;M13;M8;M7;M6; M5;M4;AN21
NLP	AN8;AN3;M41;M37;M34;M32;M29;M27;M22;M19;M18;M17;M8;M6;M4;K8;AN33;AN27;AN21;
LSA	AN6;AN3;M45;M37;M34;M32;M29;M26;M22;M19;M18;M17;M8;M7;M6;M4;K30;K20;K9; K8;K4;K3;AN32;AN31;AN27;AN24;AN23;AN21;AN13;AN11;AN9;
CD	AN8;AN3;M45;M41;M37;M34;M30;M26;M22;M13;M7;M5;K30;K20;K10;K9;K8;K7;K6; K5; K4;K3;AN35;AN34;AN32;AN31;AN27;AN24;AN23;AN21;AN13;AN11;
CHLT	M29;M27;M22;M8;M6;K30;K3;AN31;
CHLA	All genotypes except: M45;M34;M30;K9;K8;AN35;AN31;AN23;AN9
CHLB	All genotypes except: M30;K8;AN35;AN34;AN31;AN23;
DSF	AN8;AN6;M22;M18;M7;M4;K30;K7;K4;AN35;AN34;AN33;AN27;AN31;AN24;AN23; AN21;AN13;AN9
NGB	AN8;AN6;AN3;AN45;M43;M41;M37;M13;M7;K30;K20;K10;K9;K8; K7;K3;K4;K5;K6;AN35;AN32;AN27;AN24;AN9;AN11;AN13
DTC	AN8;M45;M37;K20;K10;K9;K8;K7;K5;K4;K3;AN31;AN24;AN27;
DFA	AN3;M43;M34;M32;M27;M29;M30;M18;M7;K20;K10;K9;K8;K7;K5 ;K3;AN34;AN32;AN27;AN24;AN23;AN13;AN9
PDA	AN3;M43;M37;M34;M32;M27;M29;M30;M18;M7;K20;K10;K9;K8;K7; K5;K3;AN34;AN32;AN31;AN27;AN24;AN23;AN13;AN9
NGP	AN8;AN3;M45;M43;M41;M37;M34;M32;M30;M13;M8;M7;M6;M5;K30;K20; K10;K9;K8;K7;K6;K5;K4;K3;AN34;AN31;AN27;AN24; AN23;AN11;AN9
SYC	AN8;AN6;AN3;M45;M41;M37;M32;M30;M8;M7;K30;K20;K10;K8;K7;K4;K3; AN35;AN34;AN27;AN24;AN23;AN21;AN11;
TSW	AN8;AN6;AN3;M45;M43;M41;M37;M34;M32;M30;M18;M7;K30;K20;K10;K8;K7;K5; K4;K3;AN35;AN34;AN33;AN27;AN24;AN23;AN21;AN13;
SOC	AN8;AN6;AN3;M41;M22;M19;M17;M6;M5;M4;K30;K20;K10;K9;K8;K7; K6;K5;K4;K3; AN34;AN33;AN32;AN31;AN27;AN24;AN23;AN21;AN13; AN11;AN9
SYP	AN8;AN6;AN3;M45;M41;M37;M32;M30;M8;M7;K30;K20;K10;K8;K7;K4;K3 ;AN35;AN34;AN33;AN27;AN24;AN23;AN21;AN11;

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