Genotype and environmental effects on hullability in sunflower. Results of three years of experimentation.

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Abstract

The aim of this work was to evaluate the effects of different crop management practices during three years of experimentation on sunflower cultivars characterised by different hull anatomy, in order to investigate the 'environment x genotype' interaction on hullability. The experiments were conducted from 1991 to 1993 at the Pisa University Experimental farm, adopting three different combinations of water level and nitrogen availability and three different cultivars, Oscar, Viki and Euroflor of *Helianthus annus* L. Euroflor, characterised by unusual anatomical characteristics, had a positive relationship between seed oil content and hullability, thus offering breeders the possibility of overcoming the barrier of simultaneously improving these two characteristics. However, at the same time, considerable attention must be given to the pedo-climatic environmental conditions. In fact, this cultivar significantly improved its hullability with crop management practices which favoured low water and nitrogen deficits, while in contrast the other two earlier cultivars had better hullability characteristics with good water and nitrogen supplies, especially during the second phase of their vegetative cycle.

Introduction

Theoretically, with the modern sunflower genotypes, if part (60%) of the hull is removed by hulling before oil extraction, the protein content of the seed meal should increase to 40%, which appears to be a reasonable objective (Burghart, 1992). For these reasons, hullability characteristic should be considered as the factor capable of making the greatest contribution to an improvement in the qualitative-nutritional characteristics of sunflower meal. Previous studies have demonstrated that of the numerous factors involved in the expression of hullability, the genetic influence seems to be one of the most important, as confirmed by the high values of narrow heritability (up to 0.96) obtained in many studies (Denis *et al.*, 1994a; Baldini *et al.*, 1994; Denis *et al.*, 1994b, Denis *et al.*, 1994c). This genetic influence is due to the hull anatomy of the achene, which varies with genotype, and is particularly dependent on the organisation of the alternation between two types of sclerenchymatic bulks of cells and their relative width, itself strongly related to the level of splitting of the sclerenchyma by the lines of non-lignified cells which seem to favour achene hulling (Beauguillame and Cadeac, 1992a). Many studies on the hull anatomy of a wide range of cultivated sunflower hybrids have shown that different achene pericarpic characteristics are related to hullability (Beauguillame and Cadeac, 1992b); in contrast, Denis *et al.*, (1994c), with experimental genetic material in different environments, found that the correlations between genotype differences in the pericarpic characteristics and hullability were too weak to be used alone in hullability breeding programmes, demonstrating a possible 'genotype x environment' interaction on the expression of hullability.

The main hurdle to improving hullability is its negative linkage with seed-oil content, which still represents the main sunflower product today (Baldini *et al.*, 1994). This suggests that genetic studies on seed characteristics are not sufficient to give useful indications for improving hullability and seed oil content at the same time. In fact, it is necessary to consider the influence of the environment, i.e. both climatic conditions and crop management practices, on seed characteristics such as density and the degree of lignification of the sclerenchima cell layer which have considerable effects on hullability (Merrien at al., 1992; Beauguillame and Cadeac, 1992a; Denis, 1994c). The present work aimed to evaluate the effect of different pedo-climatic conditions during three years of experimentation on three sunflower hybrids with different hull anatomy, in order to investigate the 'environment x genotype' interaction on hullability.

Materials and Methods

The experiment was perfomed in 1991, 1992 and 1993 at the Pisa University Experimental Farm at S. Piero a Grado (altitude 2 m), 15 km from Pisa, Italy. This region has a Mediterranean climate with mild, rainy winters and hot, dry summers with high evaporation rates. Soil type at the site was a deep alluvial sandy-clay texture.

Three treatments were adopted for the experiment, involving different levels of water and nitrogen availability and in particular: E1N1, high water and nitrogen availability; E1N0, high water availability and a low level of nitrogen; E0N0, low water and nitrogen availability.

The high water availability treatment was assured by overhead sprinkler irrigation maintaining the available soil water approximately 80% above field capacity. The low water availability treatment was obtained by not irrigating. The total amount of water used was determined by the water balance method : water use = soil water at sowing time + rainfall + irrigation - soil water remaining at the end of the experiment.

The treatment with high nitrogen availability was obtained by the application at planting of 80 Kg N/ha and later a further 120 Kg N/ha, giving a total of 200 Kg/ha, while nitrogen fertiliser (urea) was not applied in correspondence with the treatments with the low level of nitrogen. Each year, nitrogen soil tests were performed before sowing time. The main pedo-climatic conditions and the level of the adopted crop management practices are reported in Tab.1.

Three commercial sunflower (*Helianthus annuus* L.) hybrids, Viki, Euroflor and Oscar were used, differing in their origin and most of their characteristics, mainly in terms of the anatomical characteristics of the transversal architecture of the hull of the achenes (Beauguillaume and Cadeac, 1992b).

On 26 May 1991, 3 May 1992 and 26 April 1993, seeds were sown following a split-plots experimental design, with water and nitrogen levels as main treatments and genotypes as sub-treatments in 4 replicates. At harvest, ten plants were manually taken at random from the central two rows of each plot. Seed yield (Y) was estimated per hectare and the following measurements were performed on the seeds :

- seed moisture (SM) at the physiological maturity of each plot;
- 1000 seed weight, (SW), estimated from the weight of 250 seeds per sample;
- seed density, (SD);
- hull content (HC), mechanical hull extraction (MH) and hullability (H) were measured as previously described by Baldini *et al.*,(1994):
- oil (OIL) seed content, measured by nuclear magnetic resonance (N.M.R.);
- nitrogen (NT) seed content, measured with the Kjeldahl method;

Flowering (DAE) and physiological maturity (DAF) for each plot were recorded and growing degree-days (GDD) were calculated by the formula reported by MacMaster (1993), with a temperature base of 7°C, as indicated by Merrien (1986) for sunflower.
All the data acquired were submitted to the analysis of variance which tested the effects of year, treatment, genotype and their interactions. Simple correlations (and regression in some cases) were calculated for some variable means.

Results and Discussion

The statistical analysis of the treatments and the interactions during three years of experimentation on sunflower characteristics is reported in Table 2. All characteristics were significantly influenced by the treatments adopted and particularly the 'cultivar x year' and 'cultivar x treatment' interactions seemed to be critical in influencing hullability, confirming the importance of the 'genotype x environmental conditions' interaction on the expression of the above characteristics (Beaguillaume and Cadeac, 1992b; Denis *et al.*, 1994; Baldini *et al.*, 1994).

Fig. 1 reports the '*cv* x treatment' interaction, with Oscar always having higher hullability than *cv* Viki in each treatment and both *cv* decreased their hullability with treatments E1N1 and E0N0.
The *cv* Euroflor clearly reacted to different crop management practices; in fact, while with treatment E1N1 its hullability values were the same as *cv* Viki, in treatment E1N0 and particularly E0N0, its values were the same as *cv* Oscar.
In Fig. 2, where the '*cv* x year' interaction is reported, it could be argued that the three cultivars always had their hullability differences in the first two years of trials; *cv* Oscar showed significantly higher values than Viki and Euroflor and this latter *cv* had intermediate values. In 1993, *cv* Euroflor had a significant increase in its hullability, reaching the same values as *cv* Oscar.
The correlation between achene oil content and hullability was negative for the pooled data (Tab. 3), in agreement with results from many other studies (Roath *et al*:, 1987; Dedio, 1989; Baldini *et al.*, 1994; Denis *et al.*, 1994a; Denis *et al.*, 1994b).
Even if the data are considered for each of the 3 years of trials, the correlation between these two characteristics was always highly significant and negative (data not shown), underlining the fact that different climatic conditions are not sufficient to invert such a trend. However, when the same data were evaluated for each of the three cultivars (Fig. 3), while the relationship remained almost unvaried for *cv* Oscar and Viki, for *cv* Euroflor the achene oil content and hullability significantly and positively co-varied, confirming that the negative correlation between oil content and hullability, although present in sunflowers, is not absolute, as previously reported by Denis *et al.*, (1994a) and Baldini *et al.*, (1994). In addition, of the seed characteristics analysed, seed density also affected hullability, with a negative relationship in this environment (Tab. 3) as reported by Dedio and Dorrell (1989) in other studies.
Of the environmental parameters considered, thermal time, expressed as growing degree-days (GDD), required to complete the seed maturity phase, negatively influenced hullability (Tab. 3). All this confirms that the environment and the crop management practices, during the grain filling period and maturation when the lignification of the ovary wall occurs, strongly influence hullability (Denis *et al.*, 1994).
The negative correlation between hullability and achene moisture at harvest (Tab. 3) could have been caused by the particular environmental conditions during the seed filling up to maturity found in *cv* Euroflor. In fact, treatments E1N1 and E1N0, with optimal water and nitrogen supply, contribute to the determination of an excessive prolongation of the final

period of the cycle for *cv* Euroflor, already considered late in our environment. This fact probably led to the achene maturity period progressing under non-optimal conditions of temperature, which, linked to excessive soil water and nitrogen availability, caused not only high seed humidity but also low lignification of the sclerenchima cells and a reduction of hull content, with an increase in specific weight and a better filling up of empty spaces between amond and pericarp, which negatively influenced hullability (Laprince *et al.*, 1988; Dedio and Dorrel, 1989; Beauguillaume and Cadeac, 1992).
In fact, from Table 4 it is evident that in Euroflor the values between hullability and nitrogen availability and water use during the flowering-maturity period were negative, above all in water use, while for Oscar and Viki, genotypes with a middle cycle and earlier than Euroflor in our environment, the correlation was not significant or quite positive, confirming the hypothesis of Merrien *et al.*, (1992) and Denis *et al.*, (1994a).

Conclusions

In this experiment, the results obtained with the hybrid Euroflor (Fig. 4) allow, for the first time, the description of hullability as a character not exclusively linked to low achene oil content or high hull content of the achene, thus allowing breeders to overcome the barrier of simultaneously improving seed oil content and hullability.
Thus, the strategy for improving this characteristic could be, on the one hand, to try to introduce, by an adequate breeding programme, the same or better anatomical characteristics than Euroflor in a hypothetical new genotype, but on the other, at the same time, to specify the pedo-climatic and cultural practice conditions, for the selected genotype and environment examined, to favour hullability.
In fact, it is necessary to consider that the environment influences both cells sclerenchimatic lignification, a process generally completed about 30 days after the end of flowering (Perestova, 1976), and the hull thickness. Thus, when such anatomical characteristics of the hull are improved or incorporated *ex novo* in a genotype, the crop management practices will favour a small water and nitrogen deficit, especially during the grain-filling period, if its crop cycle is late for the environment of cultivation, otherwise this fact could produce a lignification defect of the pericarpic cells, too high a humidity content at physiological maturity and a decrease of oil content in the achene in favour of a larger protein accumulation. In contrast, with early or middle-early genotypes, with different anatomical characteristics of the hull, it seems that hullability is favoured by good water and nitrogen supply during the second phase of the crop cycle.
This suggests that breeders must aim to produce cultivars adapted to specific regions and the level of hullability will depend on the 'genotype x crop management practice' interaction and considerable attention must be given to the stability of the characteristics observed.

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Tab. 1 - Soil reserves and amount of available water and nitrogen for plants during three years of experimentation.

	1991	1992	1993
ETM sunflower (mm)	550	496	701
Rainfall during crop cycle (mm)	238	285	89
Water available in the soil (mm)[a]	100	90	130
Irrigation (mm)	200	100	400
Total water available (mm):			
-E0 treatment (without irrigation)	348	375	219
-E1 treatment (with irrigation)	538	475	619
N fertilization (kg/ha)	180	180	180
N mineral in the soil (kg/ha)[a]: N0	62	60	68
N fertilization + N mineral soil: N1	242	240	248

[a] in the first 1 m of soil deep at sowing

Tab.2 - Analysis of variance for sunflower characters analyzed during 1991-1993 period.

Source	d.f.	EF	FM	SW	SD	SM	OIL	P	Y	GDD	HC	MH	H
		Mean squares											
years	2	465.1**	114.2**	174.0**	9.0**	78.7**	25.4*	11.4**	1064.5**	28933.6**	0.37	99.8**	1414.5**
treatments	2	62.2**	98.5**	725.5**	17.1**	16.1**	55.4**	82.3**	1925.9**	4007.5**	14.6**	6.5*	233.2**
cultivars	2	480.5**	111.2**	575.6**	96.9**	124.0**	274.3**	5.7**	22.8*	69259.5**	277.2**	632.7**	4146.6**
tr x years	4	3.8**	1.8	240.2**	1.5	2.0	1.3	3.9**	36.1*	1642.8**	2.4**	2.1	20.6
cv x years	4	22.8**	9.8**	61.9**	2.0*	15.2**	1.0	0.7**	58.3**	12283.2**	2.0*	5.9**	179.3**
cv x tr	4	1.7*	0.2	37.2**	2.2*	2.2	2.2	1.1**	41.1**	1346.3**	2.2*	22.8**	448.2**
cv x tr x years	8	1.4*	0.6	11.4	1.2	0.6	3.0	0.5**	5.6	321.5	0.6	2.8	48.6**
error	54	0.6	0.4	7.1	0.7	1.2	1.7	0.1	6.8	185.1	0.7	1.0	15.1

*,** Significant at 0.05 and 0.01 levels, respectively

EF=emergence-flowering period in days; FM=flowering-maturity period in days; SW=seed weight in g; SD=seed density in g/l; SM=seed moisture at harvest in %; OIL=seed oil content in %; P=seed protein content in %; Y=seed yield in q/ha; GDD= growing degree-days in °C; HC=seed hull content %; MH=hull extracted %; H=hullability %.

Tab.3 - Correlation coefficients of hullability and other yield characters and components across hybrids and years of experimentation.

Characteristics	Correlation
Emergence-flowering period (EF)	-0.33**
Flowering-maturity period (FM)	-0.36**
Seed weight (SW)	0.27**
Seed density (SD)	-0.49**
Hull content (HC)	0.43**
Seed yield (Y)	0.07 n.s.
Oil seed content (OIL)	-0.56**
Seed protein content (P)	0.24*
Seed moisture at harvest (SM)	-0.43**
Flowering-maturity - Growing Degree Days (GDD)	-0.44**
Nitrogen availability (N)	0.07 n.s.
Water availability (W)	0.05 n.s.
Mechanical hull extraction (MH)	0.93**

*,** signficant for $P \leq 0.05$ and $P \leq 0.01$ respectively (n=106)

Tab.4 - Correlation of hullability (over treatments and years) and water use and soil N availability during the flowering- maturity period in each of the three cultivars

Cultivars	water use	soil N availability
Viki	0.39*	0.32 n.s.
Euroflor	-0.53**	-0.45**
Oscar	0.34*	0.46**

*,** signficant for $P \leq 0.05$ and $P \leq 0.01$ respectively (n=34).

Fig.1- Treatment x cv interaction on hullability character (over years)

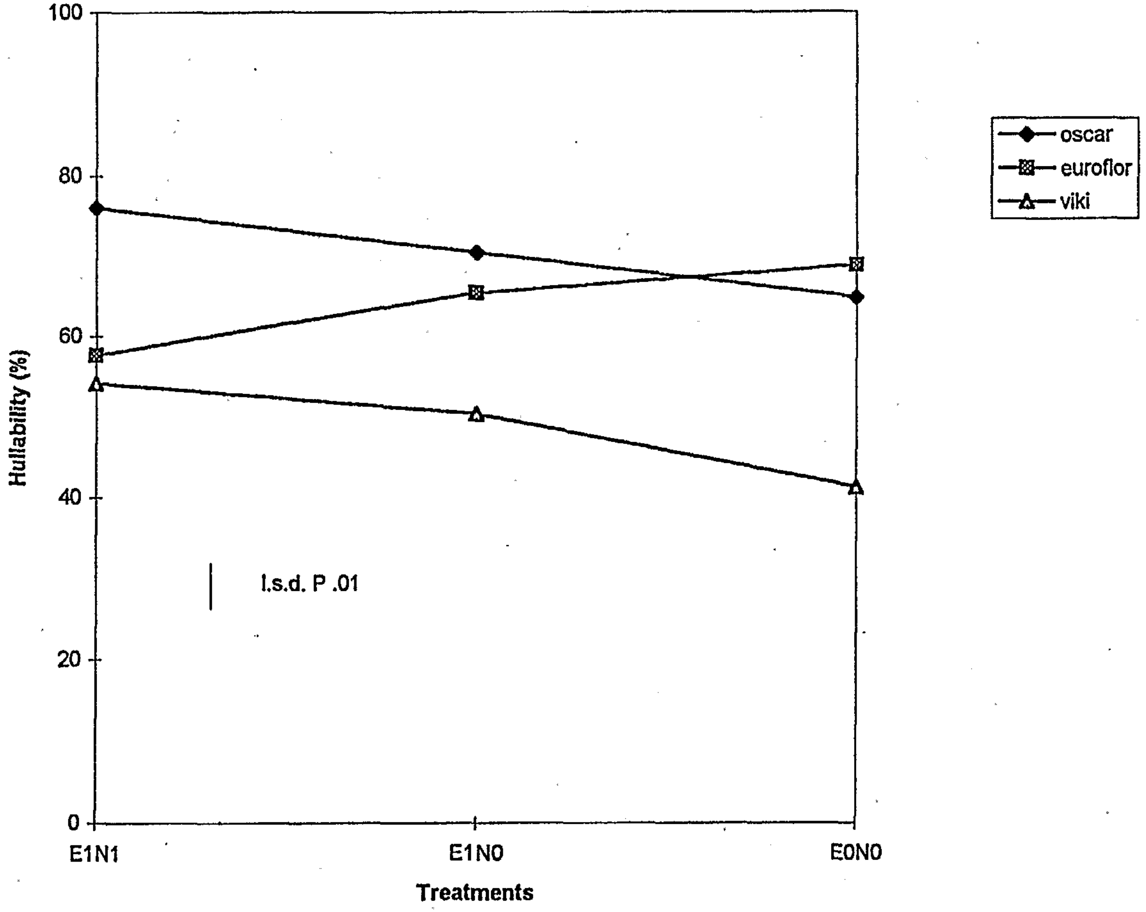

Fig.2 - Years x cv interaction on hullability character (over treatments)

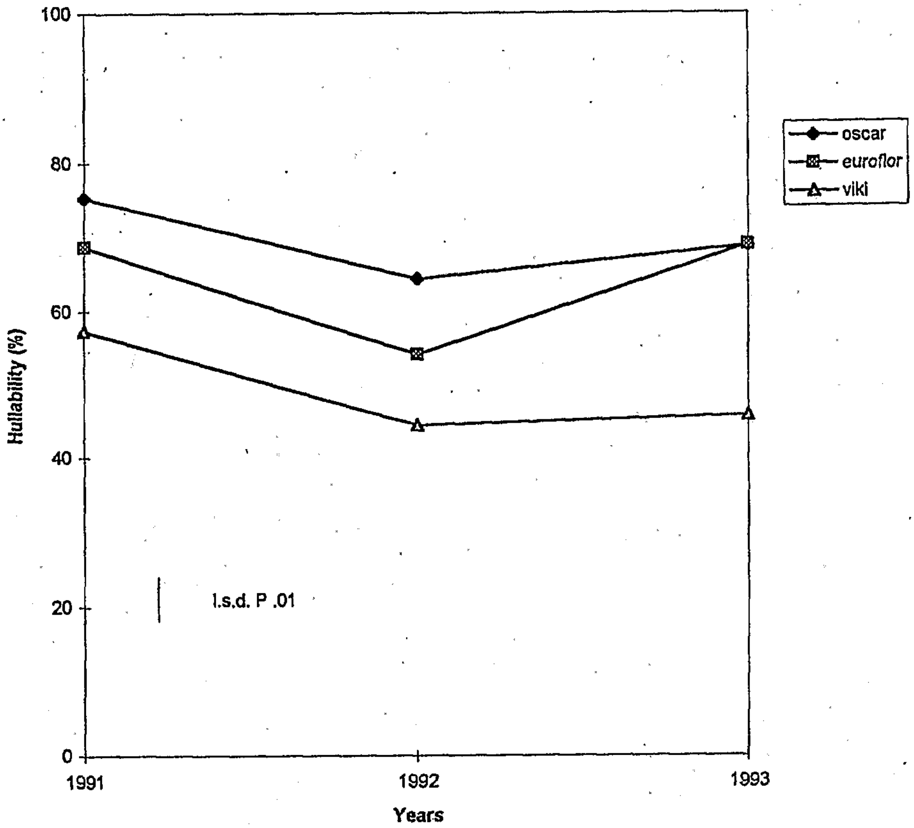

Fig.3 - Relationship of hullability and oil content in each cultivar.
Values are means across years and treatments.