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Reduced inter-row distance improves yield and competition against weeds in a semi-dwarf durum wheat variety

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ABSTRACT

Weed and nutrient management in cropping systems of semi-arid areas is a major constraint to cereal yield. Where the use of herbicides is banned or discouraged, the competitive ability of a crop is crucial to reduce weed growth and diffusion. Genotypic differences in the competitive abilities of crops are an important trait to reduce weeds, especially for plant height. However, there is contrasting information about the interactions of other management practices and genotypic traits on wheat yield and competitive ability against weeds and weed growth. The present study investigated yield and quality of durum wheat (*Triticum durum* Desf.) and weed growth and composition for two wheat cultivars with contrasting competitive abilities against weeds. Wheat was grown under three spatial arrangements (5-cm, 15-cm, 25-cm inter-row distance) and three sowing densities, and broadleaf weeds were either removed or not. The sowing rate did not affect the yield of these wheat cultivars or the weed growth. Reduced inter-row distance dramatically reduced weed biomass for both wheat cultivars, and increased wheat yield and nitrogen uptake in the low-competitive, high-yielding, semi-dwarf cv. 'PR22D89', when both weed free and with weeds. These results have direct implications for weed and nutrient management in low-input and organic cropping systems.

1. Introduction

Weed control is a major concern for both organic and conventional farming systems, particularly where the use of herbicides is banned or can have high impact on production costs, food safety, and surface and groundwater pollution. For these reasons, sustainable cropping systems call for alternative strategies to reduce competition from weeds, including agronomic and genetic strategies to minimise weed growth and spread, and to decrease yield losses.

In organic weed management, the competitive ability of the cultivated genotype was shown to be important to reduce weed pressure on the crop and weed diffusion through the cropping system (Bastiaans et al., 2008). In particular, plant height, early growth, and tillering capacity were indicated as the most important genotypic traits that confer such competitive abilities to wheat and other cereals (Andrew et al., 2015; Mason and Spaner, 2006). However, the negative relationships between plant height on the one hand, and yield components and potential on the other (Austin et al., 1980; Rebetzke and Richards, 2000) raises the question of the choice of a tall, highly competitive, low-yielding genotype rather than a semi-dwarf, low-competitive, high-yielding genotype when growing wheat under low-nutrient or high-weed conditions. Indeed, the studies to date have suggested that reduced-height cultivars have yields that are more than or similarly to those obtained for tall genotypes under low N availability (Kowalski et al., 2016; Ortiz-Monasterio et al., 1997). In addition, the genotype variability for the yield is not related to the management system (i.e., organic or conventional) (Kitchen et al., 2003; Reid et al., 2011).

Agronomic efforts to reduce weed competition rely on increasing the asymmetric competition between the crop and the weeds (Weiner, 1990; Zimdahl, 2004), either through manipulation of the crop and weed sizes at the early stages of crop growth, or by directly reducing the weeds. The management strategies used to maximise the rate at which a crop occupies space early in the growing season usually minimise the competitive pressure of the weeds (Mohler, 2001). In particular, an increase in crop density and sowing density, use of an efficient seeding method, anticipation of the planting date, and choice of the correct crop rotation and a highly competitive crop genotype are the most important practices.

Nonetheless, the application of these management practices can also have drawbacks in terms of the crop growth and yields. For

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example, higher seeding rates will be accompanied by extra seed costs for planting potentially without increasing the yield or the net return (Kolb et al., 2012). Accordingly, a sowing date earlier than the optimum date for a certain cultivar in a given location can reduce the yield potential, and thus counterbalance the benefit of increased competitiveness against weeds.

Sowing systems for winter cereals in the Mediterranean basin generally use a mechanical or pneumatic seed drill that can distribute the seeds at a fixed inter-row distance (usually >12 cm and ≤25 cm). However, such inter-row distances allow for early growth of weeds in the inter-row spaces considering that the wheat root colonisation ability rarely exceeds 5 cm in the early stage of growth (White and Kirkegaard, 2010). An alternative weed-management strategy relies on an increase in the spatial uniformity of the seed distribution (Fischer and Miles, 1973; Kristensen et al., 2008; Mohler, 2001); i.e., a reduction in the rectangularity of the seed distribution (as the ratio of inter-row to intra-row distance). This should provide better uptake of the soil nutrients and better ground cover by the crop, which can reduce weed germination and growth. In addition, increased spatial uniformity should also reduce the intraspecific competition for light, and thus increase both transpiration of the crop and resource (especially photons) use efficiency (Furbank et al., 2015). However, information regarding the effects of a reduction in the inter-row distance (and an increase in the seeding distance within the rows) on wheat yield and competitive ability against weeds has been contrasting (Boström et al., 2012; Chen et al., 2008; Hu et al., 2015; Rasmussen, 2004; Roberts et al., 2001). This appears to depend on a range of factors, which include: wheat genotype; weed pressure, composition and germination pattern; weed and nutrient management strategies; water availability during the late season; and availability of the correct seed drill. It has been suggested that an increase in the seeding rate can partly compensate for an increase in the inter-row distance (and thus compensate for a reduction in the spatial uniformity). This should increase the crop competition against weeds (Hiltbrunner et al., 2007; Korres and Froud-Williams, 2002; Lafond and Gan, 1999; Mertens and Jansen, 2002; Peltzer et al., 2009). However, it has been shown that this can also increase the intraspecific competition, which can counterbalance the benefits of the reduction in the weeds, and can finally result in a reduction in the yield potential (Hakansson, 2003; Yao et al., 2015). An increase in the spatial uniformity of the seed distribution can be achieved, in theory, by distribution of the seeds with a fertiliser spreader. However, this usually results in an uneven sowing depth, which can negatively affect the outcome and homogeneity of the crop.

Durum wheat (*Triticum durum* Desf.) is the main crop in many countries of the Mediterranean basin, and it is the preferred raw material for the worldwide production of pasta, couscous and burghul. Over the last 15 years, the rapidly growing market for organic pasta has created favourable conditions for specialised arable farming systems. However, organic wheat-based farming systems face various agronomic drawbacks, including excessive weed pressure and diffusion. This hampers the use of semi-dwarf, low-competitive varieties. The aim of the present study was to determine the role of the spatial arrangements of the wheat plants in terms of both the grain yields of two contrasting durum wheat genotypes and the weed biomass and composition, at varying sowing densities and weed control. In particular, the spatial arrangements of the wheat plants were modulated by reduced inter-row distance from 25 cm to 15 cm and 5 cm (while maintaining a constant seed rate), which nearly compared to the 5.7-cm spread of the root plate that is theoretically needed by an ideal wheat genotype to achieve the highest yield (Reynolds et al., 2009). This spatial arrangement was achieved using a new seed drill prototype (SEMINBIO project, RM2013A000332 and/or 202015000006429) that was equipped with interchangeable working parts and could be used to sow seeds every 5 cm.

2. Materials and methods

2.1. Experimental site

This study was performed at the Cereal Research Centre (CREA-CER, Foggia, Italy; 41° 28′N, 15° 32′E; 75 m a.s.l.) in the 2012–13 and 2013–14 growing seasons, on a clay-loam soil (Typic Chromoxerert). Soil traits were: 36% clay, 47% sand; pH 7.8; 17.3 g kg^{-1} total C; 1.5 g kg^{-1} total N. The mean long-term rainfall of the experimental site is 479 mm, and the mean air temperatures are 12.2 °C in autumn, 8.2 °C in winter, and 17.6 °C in spring. The mean minimum and maximum annual temperatures are 9.9 °C and 21.0 °C, respectively.

2.2. Experimental design and crop management

The experimental design was a split-split-plot design (six replicates) with the following treatments: main plots were cultivar: cv. 'PR22D89' (a medium flowering, semi-dwarf cultivar, released in 2005) and cv. 'Cappelli' (a late flowering tall cultivar, released in 1915), and inter-row distance (*RD*; 5 cm, 15 cm, 25 cm). The split-plot was sowing density (*SD*; 190, 380, 570 viable seeds m^{-2}); the split–split plot was weed control (*WC*; weed free, with weeds). The size of the plot was 4.5 m × 20 m. The preceding crop for both of the growing seasons was durum wheat. Before sowing, the soil was ploughed in late August and harrowed twice in September and October, to prepare a suitable seedbed and to control the summer weeds. In both of the growing seasons, the pre-sowing fertilisation consisted of 36.0 kg N ha^{-1} and 40.1 kg P ha^{-1} (as diammonium phosphate), top-dressing fertilisation consisted of 54 kg N ha^{-1} (as ammonium nitrate). The top-dressing fertilisation was applied at wheat growth stage 31 (Zadoks et al., 1974). Sowing was performed using a seed drill prototype (SEMINBIO project, RM2013A000332 and/or 202015000006429) that was equipped with a series of delivery elements spaced at 5 cm. This allowed more uniform spatial distribution of the wheat plants compared with conventional sowing using wide (15 or 25 cm) inter-row distances (see Supplemental material Table 1 for the sowing pattern and rectangularity of the seed distribution in the various combinations of inter-row distances × sowing densities). In particular, the seed falling from the hopper through the seed tubes reached the furrow randomly, so a 5-cm inter-row distance was not visible. Previous studies have suggested that a high degree of seed-spacing uniformity is not necessary to achieve major improvements in weed suppression, while sufficient improvement in crop-sowing uniformity can be achieved through the combination of a reduction in the inter-row distance and increased uniformity within the rows (Olsen et al., 2005). The sowing of the 15-cm and 25-cm inter-row distances was performed with the same seed drill, by allowing seeds to pass from every three or five seed bins, respectively, while maintaining the corresponding furrow opener. Durum wheat was sown on 16 December, 2012, and 22 December, 2013.

2.3. Weeds and crop analysis

During growth, each plot was characterised in terms of weed biomass and composition, plant density, grain yield and its protein content. Germinating weeds were removed from the plots for the weed-free treatment, until the end of tillering. At the end of tillering (growth stage 29; Zadoks et al., 1974), plant density (*PD*) was measured in a 1-m^2 microplot inside each subplot. In addition, the weeds were sampled, counted and identified, with the analysis based on the main weed species of *Fumaria*, *Veronica* and *Lamium*.

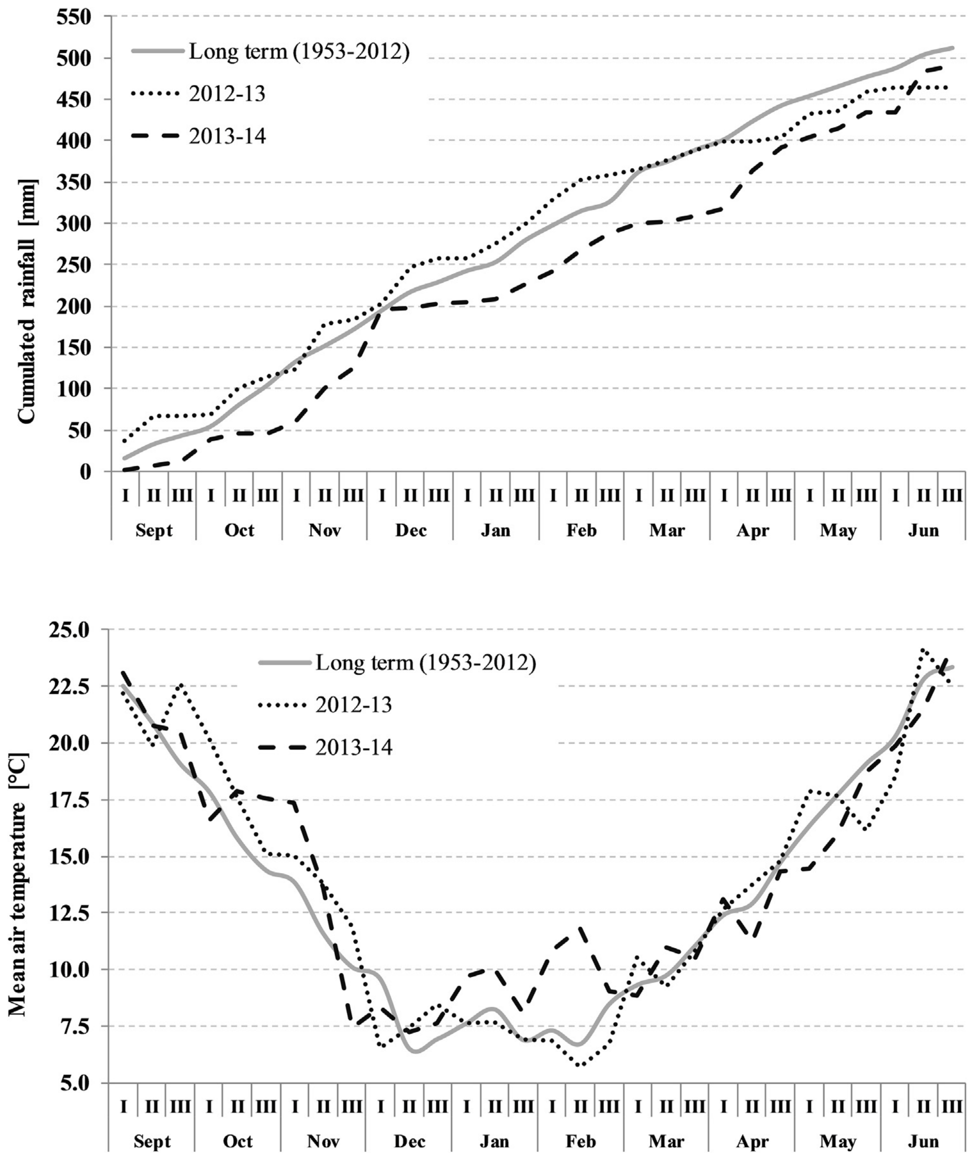

Fig. 1. Cumulated rainfall (upper panel) and mean air temperatures (lower panel) at the CREA-CER experimental station during the two growing seasons (2012-13, 2013-14, comparing to the long-term mean over 1953–2012) using a 10-day period.

Weed biomass was determined with drying in an oven at 65 °C, until constant weight. Heading date (*HD*; days after 1 April) was recorded when about half of the culms showed emerging spikes (growth stage 55; Zadoks et al., 1974). Plant height (*PH*) was measured at the milk-waxy maturation stage. In the second half of June, the whole plots were mechanically harvested and the grain yield (*GY*) was determined and recorded at 13% moisture content. Thousand kernel weight (*TKW*) was measured. Grain N content was determined using the Kjeldahl method, and grain protein concentration (*PC*) calculated by multiplying the Kjeldahl N by 5.7, and is expressed on a dry weight basis (AACC International, 2000). Grain N uptake was calculated as $GY \times PC \times 10^{-2}$.

2.4. Calculations and statistical analysis

The contribution of each major weed to the total weed biomass was computed and the percentage data were arcsin square-root transformed before running the statistical analyses. Data were subjected to analysis of variance (Glimmix procedure in SAS/STAT, by using the random statement with 'rep*main factor' and 'rep*main factor*first-order split factor' as random factors), according to the experimental design, and differences among means compared by applying t-grouping at the 5% probability level to the LSMEANS. The following were computed separately per wheat cultivar: standard correlation among total weed biomass at tillering, and percentage variation (computed as with-weeds compared to weed-free plots) of plant density, grain yield, grain protein content, 1000-kernel weight, plant height, and grain N uptake. In addition, the correlation coefficient between plant density and date of heading, grain yield, grain protein content, 1000-kernel weight, plant height, and grain N uptake were also computed. The CORR procedure in SAS/STAT was used to test the statistical significance of the correlation coefficients.

3. Results

3.1. Weather conditions

Total rainfall in 2012–13 and 2013–14 were 464 mm and 484 mm (Fig. 1), respectively. These were very close to the long-

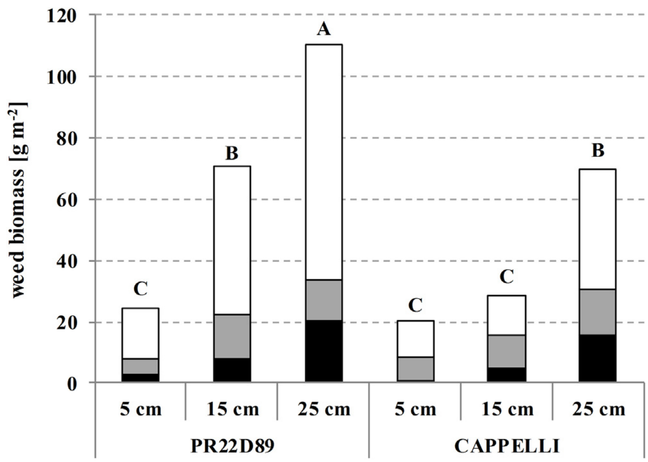

Fig. 2. Weed biomass [g m^{-2}] for 'PR22D89' and 'Cappelli' at varying inter-row distance (5, 15, 25 cm) at the end of tillering in with-weeds plots. Black bars, *Fumaria* weeeds; grey bars, *Veronica* weeds; white bars, *Lamium* weeds. Treatments with a letter in common are not different at p < 0.05 (according to the t-grouping of least square means differences). See Supplemental Material Table 2 for data on sowing densities and analysis of variance.

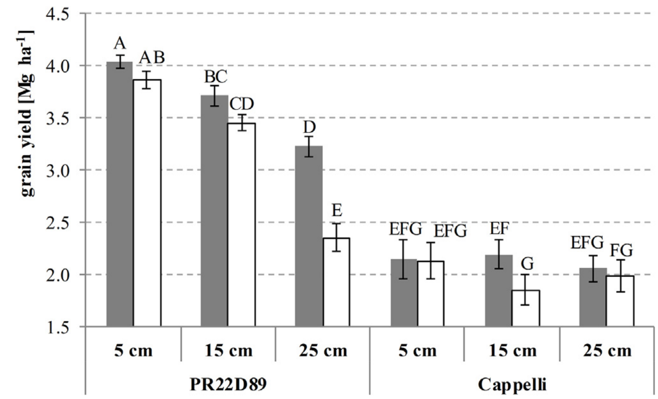

Fig. 3. Grain yield [Mg ha$^{-2,}$ data are means ± S.E., n = 36] for 'PR22D89' and 'Cappelli' at varying inter-row distances (5, 15, 25 cm) and weed control (black bars, weed-free plots; white bars, with-weeds plots). Treatments with a letter in common are not different at p < 0.05 (according to the t-grouping of the least square means differences). See Supplemental Material Table 3 for raw data.

term annual mean (479 mm year^{-1}; Fig. 1). The rainfall during both of the growing seasons was well distributed. Mean temperature was slightly higher than the long-term mean during autumn, and slightly lower during spring. In 2013–14, the winter temperature was also slightly higher than the long-term mean.

3.2. Weed biomass and composition

At the time of wheat tillering, there were no weeds in the weed-free plots. In general, the biomass of weeds in the with-weeds treatments increased with increased inter-row distance (Fig. 2), with significant differences according to cultivar. In particular, at the lowest inter-row distance (i.e., 5 cm), there were similar biomass of weeds for cvs. 'PR22D89' and 'Cappelli', whereas at 15-cm and 25-cm inter-row distances there were higher biomass of weeds in plots sown with 'PR22D89', compared to 'Cappelli'. Increased sowing density showed slightly, but not significantly, reduced biomass of total weeds in the with-weeds 'Cappelli', whereas it did not affect the total weed biomass in the with-weeds 'PR22D89' (data not shown).

The incidence of each weed on the total weeds varied according to the wheat genotype and row distance: for both genotypes, at increased inter-row distances, *Fumaria* increased and *Veronica* decreased, whereas the incidence of *Lamium*, which was the most abundant weed, was similar among row distances and genotypes (Supplemental material Table 2). However, the incidence of *Veronica* on the total weeds was higher in 'Cappelli' than 'PR22D89'. The opposite was seen for *Lamium* and *Fumaria*, and differences in the $CV \times RD$ interactions were only in the magnitude of the effects.

3.3. Wheat plant density at tillering, time of heading, and plant height

When the sowing density was reduced by 50% (i.e., from 380 to 190 seeds m^{-2}), plant density reduced by a mean of 42.7%, between both wheat genotypes (Table 1 and Supplemental material Table 3), whereas when sowing density was increased by 50% (from 380 to 570 seeds m^{-2}), plant density was increased by 36.9%, with a greater increase for 'Cappelli' (+40.7%) than 'PR22D89' (+33.0%). In addition, plant density was reduced with increases in both inter-row distance and sowing density. As expected, with the weeds removed (i.e., weed free), there was higher plant density at tillering compared to the with-weeds plots (+17.4%), which occurred irrespective of sowing density, cultivar or inter-row distance.

Reduced inter-row distance slightly delayed time of heading, especially at the lower sowing density. As expected, 'PR22D89' headed before 'Cappelli' (6.3 days, across all treatments). 'Cappelli' showed a mean height that was 38.3 cm greater than 'PR22D89'. At increased sowing density, plant height increased similarly for both cultivars. Weed control slightly reduced plant height for 'PR22D89', but not for 'Cappelli'.

3.4. Grain yield and quality

The effects of inter-row distance (*RD*) on grain yield and N uptake varied according to the cultivar (*CV*) and weed control (*WC*) (significant $CV \times RD \times WC$ interaction; P = 0.017), whereas no effects were seen for sowing density (Table 1; *SD*). In particular, grain yield of 'Cappelli' was not affected by inter-row distance and weed control, whereas for 'PR22D89', at increased inter-row distance, grain yield differences between weed-free and with-weeds plots increased (Fig. 3). Similar differences among treatments were found for total grain N uptake (Supplemental material Table 3). Row distance, sowing density and weed control did not affect grain protein content, which showed a mean of 12.2% ±0.08% (±S.E.) for 'PR22D89', and 14.6% ±0.10% for 'Cappelli' (Supplemental material Table 3). Few differences among treatments were found for 1000-kernels weight, which decreased at increased sowing density and decreased inter-row distance. When the wheat was sown at the 5-cm inter-row distance, the weed control achieved resulted in 3.7% reduction in 1000-kernel weight for 'PR22D89′ and 2.9% increase for 'Cappelli'.

3.5. Correlations between total weeds and wheat density and other traits

The linear correlation between total weeds at tillering and percentage variation of grain yield for with-weeds comparing to weed-free plots (Fig. 4) was significant for 'PR22D89' (P = 0.004), but not for 'Cappelli' (P = 0.553). Similar correlations were found between total weeds at tillering and percentage variation of grain N uptake (Table 2). Total weeds also correlated with variations in 1000-kernel weight for 'PR22D89'. For both cultivars, significant and negative correlations were seen between plant density and date of heading and 1000-kernel weight (Table 2), and between date of heading and 1000-kernel weight (r = +0.60, p = 0.008 for 'PR22D89' and r = +0.63, p = 0.005 for 'Cappelli'), whereas no corre-

Table 1
Analysis of variance of effects of cultivar (*CV*), inter-row distance (*RD*), sowing density (*SD*) and weed control (*WC*) on grain yield, protein content and N uptake, and plant density at tillering and height at maturity.

Variable	Grain yield ($Mg\,ha^{-1}$)	Grain protein content (%)	Grain N uptake ($kg\,N\,ha^{-1}$)	1000-kernel weight (g)	Plant density ($n\,m^{-2}$)	Plant height (cm)
Cultivar (*CV*)	**<0.001***	**<0.001**	**<0.001**	0.160	**<0.001**	**<0.001**
Inter-row distance (*RD*)	**<0.001**	0.076	**<0.001**	**0.005**	**<0.001**	**<0.001**
CV × *RD*	**<0.001**	0.301	**<0.001**	0.932	**<0.001**	**<0.001**
Sowing density (*SD*)	0.290	0.882	0.460	**<.001**	**<0.001**	**<0.001**
CV × *SD*	0.054	0.989	**0.010**	0.524	**<0.001**	**0.039**
RD × *SD*	0.886	0.783	0.837	0.611	**<0.001**	0.182
CV × *RD* × *SD*	0.996	0.976	0.975	0.462	0.118	0.359
Weed control (*WC*)	**<0.001**	0.069	**<0.001**	0.556	**<0.001**	**<0.001**
CV × *WC*	**0.024**	0.354	**0.005**	0.221	0.837	**<0.001**
RD × *WC*	0.057	0.266	0.089	0.863	0.897	**0.020**
CV × *RD* × *WC*	**0.017**	0.163	**0.002**	**0.023**	0.941	0.205
SD × *WC*	0.862	0.757	0.759	0.353	0.980	0.093
CV × *SD* × *WC*	0.860	0.553	0.914	0.123	0.986	0.391
RD × *SD* × *WC*	0.532	0.996	0.331	0.887	0.919	0.284
CV × *RD* × *SD* × *WC*	0.865	0.962	0.879	0.593	0.950	0.667

* **values in bold**, significant at p < 0.05.

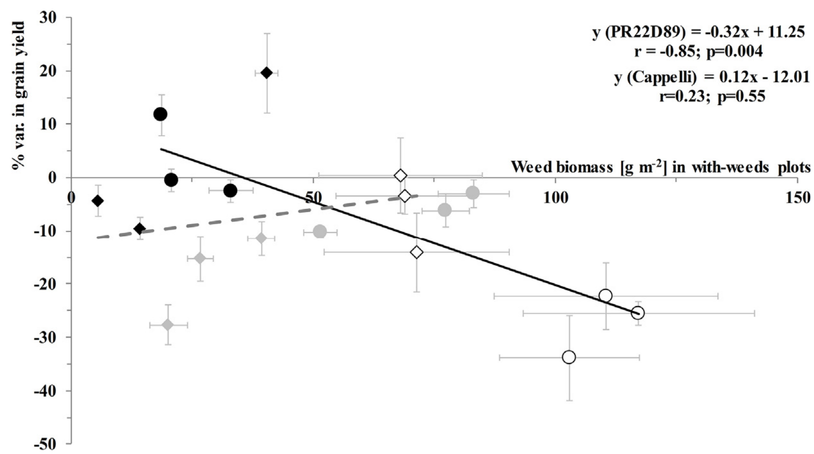

Fig. 4. Correlations between weed biomass [$g\,m^{-2}$] in with-weeds plots and percentage variations in grain yield for with-weeds compared to weed-free plots. Data are means ±standard errors, n = 6. Circles and straight lines are for 'PR22D89'; diamonds and dashed lines are for 'Cappelli'. Black symbols are for plots sown at 5 cm; grey symbols at 15 cm; and white symbols at 25 cm inter-row distances.

lations were seen between date of heading and grain yield (r = 0.07, p = 0.601 for 'PR22D89' and r = 0.08, p = 0.266 for 'Cappelli').

4. Discussion

Weed management represents one of the main constraints to diffusion of organic wheat-based farming systems. However, the usual sowing system for cereals (i.e., rows >15 cm apart) results in little ground cover, and thus allows weeds to rapidly colonise the soil and compete with the crop for nutrients, light and space. In the present study, we evaluated the yield and competition against weeds of two contrasting durum wheat genotypes (a semi-dwarf, early heading genotype, 'PR22D89', and a tall, late-heading genotype, 'Cappelli',) sown at three sowing densities and three inter-row distances. Both growing seasons were characterised by high homogeneity of rainfall distribution, which appears to have favoured complete expression of the yield potential of the wheat and growth of the weeds. There was consistent reduction in weeds with increasingly homogenous plant arrangements that were achieved by reduced inter-row distance (from 25 cm to 5 cm) while keeping the sowing density constant, and thus increased in-row plant-to-plant distance. This appears to depend on both higher early competition of the crop due to a higher early soil cover and depletion of soil water and nutrients. Indeed, reduction of inter-row distance also led to stronger decrease in the contribution of the later-growing weeds *Fumaria* and *Lamium* to the total weeds biomass, compared to the earlier growing *Veronica* (Berti et al., 2008; Viggiani and Montemurro, 1998), for both wheat cultivars. Boström et al. (2012) showed that increased inter-row distance from 12 cm to 24 cm did not promote increased weed density, but reduced crop yield, whereas Rasmussen (2004) showed that weed control was efficient to reduce weeds only at the highest inter-row distance and when weed pressure was high. Similar results were obtained by Evers and Bastiaans (2016), which showed that a uniform planting pattern can strongly reduce weed biomass with minor effect on crop biomass, especially when time of emergence of weed is early. This implies that the effects of inter-row distance and seeding rate on weed density and biomass depend on both weed pressure and resources availability for both weeds and crop. The increased competitive ability that resulted from reduced inter-row distance in the present study also depended on the wheat cultivars under study. In particular, the tall cultivar 'Cappelli' depressed weed growth more than 'PR22D89' at the 15-cm and 25-cm inter-row distances, whereas similar competition against weeds between cultivars was seen with the plants arranged in a more uniform grid pattern; i.e., at the 5-cm inter-row distance. This was expected for the highly competitive tall cultivar 'Cappelli', as observed in bread wheat (Drews et al., 2009), but not for the modern semi-

Table 2
Linear correlation coefficients between total weeds [g dry mass m^{-2}] at end of tillering and percentage variations of traits measured at maturity (n = 9); and between plant density and date of heading, grain and grain N yield, grain quality and plant height (n = 18). Each correlation coefficient was calculated separately per genotype.

	PR22D89		Cappelli	
	r	*p*	r	*p*
	Total weeds			
date of heading	−0.221	*0.568*	0.134	*0.730*
% variation of plant density*	−0.218	*0.574*	−0.311	*0.416*
% variation of grain yield	**−0.849**	***0.004***	0.229	*0.553*
% variation of grain protein content	−0.075	*0.849*	0.588	*0.096*
% variation of 1000-kernels weight	**−0.840**	***0.005***	0.453	*0.220*
% variation of plant height	−0.478	*0.194*	0.082	*0.833*
% variation of grain N uptake	**−0.694**	***0.038***	−0.545	*0.129*
	Plant density			
Date of heading	**−0.822**	***0.000***	**−0.918**	***0.000***
Grain yield	0.418	*0.084*	−0.107	*0.671*
Grain protein content	0.068	*0.789*	0.035	*0.891*
1000-kernels weight	**−0.833**	***0.000***	**−0.704**	***0.001***
Plant height	**0.653**	***0.003***	**0.844**	***0.000***
Grain N uptake	0.415	*0.087*	−0.248	*0.321*

*calculated as percentage variation for with-weeds comparing to weed-free plots.

dwarf cultivar 'PR22D89'. Indeed, tall genotypes are normally more competitive against weeds than semi-dwarf genotypes, and this has been mainly attributed to differences in early vigour, plant stature, tillering ability, leaf display (Lemerle et al., 2001, 1996; Ruisi et al., 2015; Murphy et al., 2008; Mason et al., 2007), and allelopathic potential (Bertholdsson, 2005; Fragasso et al., 2013; Oueslati, 2003). Sanguineti et al. (2007) reported that plant height was positively associated with shoot length and shoot dry matter accumulation recorded during the juvenile phase of wheat, and that a similar feature was observed for all durum wheat cultivars lacking of the *Rht-B1* dwarfing gene (such as 'Cappelli'), which was introduced in the modern durum wheat germplasm in the early 1970s. In a study carried out to characterise the variability in the duration of various pre-anthesis phases, Sanna et al. (2014) showed that the substantial vernalisation requirement of 'Cappelli' was higher than for cv. 'Ofanto' (a modern semi-dwarf durum wheat cultivar), and that this resulted in higher leaf numbers, longer vegetative phase, and more rapidly cover of the soil surface. Other studies have also reported higher water use efficiency in 'Cappelli' than 'Ofanto', and that this difference was likely to be associated with lower stomatal conductance over the range of relative soil water contents tested (Panio et al., 2013; Rizza et al., 2012). This might explain the greater ability of 'Cappelli' to suppress weeds comparing to 'PR22D89' at the 15-cm and 25-cm inter-row distances. In the present study, reduced inter-row distance also resulted in increased wheat plant density, especially at the highest sowing density, irrespective of the weed control. Moreover, this increase was higher for 'PR22D89' than 'Cappelli'. The differences between 'PR22D89' and 'Cappelli' in terms of plant-to-plant intraspecies competition at the 15-cm and 25-cm inter-row distances might be due to the lower metabolic cost of formation of the root system in 'Cappelli' than 'PR22D89' (Nazemi et al., 2015), and thus to the growth of deeper roots (Sanguineti et al., 2007; Siddique et al., 1990). When inter-row distance was reduced to 5 cm, the differences in plant development between cultivars decreased. In this plant arrangement pattern, the modern cultivar 'PR22D89' was as competitive as 'Cappelli' against weeds, and this appeared to depend on several mechanisms, which included earlier onset of competition for light, nutrients, water and space.

Thus, we hypothesise that reduced inter-row distance also resulted in reduced in-row-plant-to-plant wheat competition, which would lead to greater early vigour of the wheat, especially for the semi-dwarf genotype. Accordingly, reduced or increased sowing density by 50% (from 380 to 190 or 570 seeds m^{-2}) reduced or increased plant density by only 42.7% and 36.9%, respectively, with little difference between cultivars. Increased sowing density, but not significantly so, reduced the amount of total weeds for with-weeds 'Cappelli', whereas it did not affect total weeds for with-weeds 'PR22D89' (data not shown). Similar data were reported by Chen et al. (2008), who showed that reduced inter-row distance of wheat resulted in more rapid crop biomass accumulation and resource depletion from the soil, and that these effects were not achieved by an increase in seeding rate. In contrast to Chen et al. (2008), and the present study, increased sowing density generally resulted in higher competitive ability of wheat (Champion et al., 1998; Korres and Froud-Williams, 2002; Olsen et al., 2005). However, this effect might depend on the high accumulation of day-degrees during the early season and the low water availability during the late season, which would favour emergence of early weeds (Grundy, 2003), and which resulted in most of the crop–weed competition in the early phases of the present study. This calls for further investigations into the role of the plant arrangements on the critical weed-free period in cereals, with small-seeded, and slow growing species, which in Mediterranean countries and under organic cropping systems can depend on the time needed by the canopy to completely cover the inter-row space (Frenda et al., 2013; Welsh et al., 1999), and thus to rapidly remove the early competition from weeds (Fahad et al., 2015).

4.1. Grain yield and quality

As expected, the modern cultivar 'PR22D89' provided higher yield than 'Cappelli', and this confirms the genetic gain in the yield associated with a few key genes that affect the morpho-phenological traits, mainly *Rht* (De Vita et al., 2007). This also suggests that the choice of a tall genotype, such as 'Cappelli', to increase the crop-to-weed biomass ratio should deal with the drawback related to its intrinsically low yield potential (De Vita et al., 2010). For the semi-dwarf 'PR22D89' cultivar, the increase in weed biomass resulted in decreased grain yield (compared to weed-free treatment). In addition, reduced inter-row distance from 25 cm to 15 cm, and from 15 cm to 5 cm, increased grain yield of 'PR22D89' but not that of 'Cappelli', and this occurred for both weed-free and with-weeds treatments. Similarly, reduced inter-row distance resulted in increased total grain N uptake of 'PR22D89', irrespective of weed control. On the contrary, wider inter-row distance limited expression of the yield potential of 'PR22D89', and this especially occurred with the with-weeds plots, as the expression of yield potential strongly relies on nutrient availability and lack of competition from weeds. A similar result was reported by Schillinger and Wuest (2014), when they increased winter-wheat inter-row distance from 40 cm to 80 cm. This suggests that for the present study, the wider inter-row distances limited expression of the yield potential of 'PR22D89′, and confirms the hypothesis of Griepentrog (1999), who stated that increased uniformity of plant distribution leads to enhanced nutrient uptake. Our data also agree with those from Solie et al. (1991), showing that reduced inter-row distance significantly increased modern wheat yields in weed-free and weed-infested fields. The lack of any effects of the treatment applied on the grain yield and N uptake of 'Cappelli' probably depended on several factors, which includes the low yield potential of this genotype (De Vita et al., 2007), its higher water use efficiency (WUE) compared to the dwarf genotypes (Panio et al., 2013; Rizza et al., 2012) and its competitive ability against weeds (Ruisi et al., 2015). In addition, this effect might also depend on differences between 'PR22D89' and 'Cappelli' in terms of the accumulation of dry matter during grain filling, as has also been observed between other old and modern cultivars (Arduini et al., 2006), where it has been shown that most of the grain dry matter depends on post-anthesis photosynthesis (Masoni et al., 2007), whereas most of the grain N content depends on uptake before grain filling (Barraclough et al., 2014). This suggests that the old durum wheat genotype was characterised by a minimal responsiveness to improved environmental conditions (e.g., from with weeds to weed free), as it showed an almost stable grain yield potential regardless of the agronomical conditions, in agreement with the concept of 'biological' or 'static' stability (De Vita and Maggio, 2006). The present findings also show that reduced inter-row distance and increased sowing density (from 190 to 570 seeds m^{-2}) slightly, but significantly, brought forward the heading date and increased plant height for both cultivars and weed control treatments. This suggests that intraspecies competition occurred. This delay in heading date and increased plant height did not affect grain yield, but negatively correlated with 1000-kernel weight of both cultivars (Table 2).

Chen et al. (2008) also showed that an increase in seeding rate can be responsible for reduced grain protein content, although such a response was not observed in other studies (Beavers et al., 2008; Ozturk et al., 2006), as well as in the present study. On the contrary, grain protein content was seen to increase for the winter wheat grown in wide inter-row distances, and this was attributed to an augmented N availability, either from the inter-row distances or from the vegetative parts of the plant, as plants grown in wider rows tended to have greater shoot dry matter, and hence more nutrients to translocate per kg grain (Capouchová et al., 2008; Hiltbrunner et al., 2005). In the present study, the modulation of row distance and the increase in sowing density did not have any effects on the grain protein content.

5. Summary and conclusions

(1) Reduced inter-row distance led to reduced weed biomass in both the tall cv. 'Cappelli' and the semi-dwarf cv. 'PR22D89', and this occurred at a similar degree at each sowing density. The contribution of the later-growing weeds *Fumaria* and *Lamium* to the total weed biomass decreased at reduced wheat inter-row distance, whereas the contribution of the early-growing weed *Veronica* increased.
(2) At reduced inter-row distance, yield and N uptake of cv. 'PR22D89' increased, whereas those of cv. 'Cappelli' were constant. In addition, weed control resulted in further increase in 'PR22D89' yield, and this mostly occurred at the widest row distance, whereas weed control did not increase 'Cappelli' yield.
(3) For 'PR22D89', weed biomass directly correlated with reduction in grain yield compared to weed-free treatments ($r = -0.85$, $p = 0.004$), whereas there was no correlation between weed biomass and 'Cappelli' yield for with-weeds compared to weed-free treatments ($r = +0.23$, $p = 0.55$).
(4) Few effects of sowing density were observed. In particular, increased sowing density slightly increased grain N uptake and plant height and pushed forward the date of heading, for both cultivars.

The best weed control strategies should rely on integrated approaches to weed management using chemical, mechanical, and/or preventive weed control methods, including modulation of rotation, seed rates, and others. In the present study, we demonstrated that adopting an increasingly homogenous plant arrangement (by reduced inter-row distance from 25 cm to 15 cm and 5 cm at the same time as increased in-row plant-to-plant distance) while keeping constant the sowing density resulted in reductions of the weeds, in both the tall and semi-dwarf wheat cultivars.

Reduction in inter-row distance also resulted in strong increases in grain yield and N uptake of the semi-dwarf cv. 'PR22D89', for both with-weeds and weed-free treatments. In particular, the reduction in row distance increased 'PR22D89′ grain yield more than the weed control. Thus, the choice of the correct row distance has an importance when aimed at reducing weed pressure and herbicide use in field crops, and for increased yield. Indeed, the minimum inter-row distance used in the present study nearly corresponded to the wheat root colonisation ability, which rarely exceeds 5 cm (White and Kirkegaard, 2010); the 5.7 cm root plate spread theoretically needed by the ideal wheat genotype to achieve the highest yield (Reynolds et al., 2009); and the 6.6-cm optimum row spacing suggested by Solie et al. (1991).

The present data have direct implications for weed management, and can enable farmers operating under low-input and organic cropping systems to adopt semi-dwarf, high-yielding varieties with high qualitative standards, and to expand their choice of cultivars, which is often restricted to old, low-yielding genotypes which are more competitive against weeds. In addition, it has been shown that reduced inter-row distance can contemporarily lead to increased transpiration from the crop and reduced evaporation from the soil, and thus lead to increased water use efficiency and nutrient uptake from the crop (Chen et al., 2008, 2010; Hu et al., 2015; Kleemann and Gill, 2010; Schillinger and Wuest, 2014; Tompkins et al., 1991a,b). However, the slight delay in the heading date, and the rapid soil ground cover at reduced inter-row distance, could also have implications in arid environments, where earlier heading and less soil evaporation are both needed, respectively. Finally, the increase of grain yield while keeping constant grain protein concentration at reduced inter-row distance calls for further investigation on the role of the spatial distribution and seed rate on the efficiency of the use of resources, and especially nitrogen (Dai et al., 2013; Mao et al., 2016).

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Appendix A. Supplementary data

Supplementary data associated with this article can be found, in the online version, at http://dx.doi.org/10.1016/j.eja.2017.02.003.

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