Protein and Lysine Content, Grain Yield, and Other Technological Traits in Durum Wheat under Mediterranean Conditions

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A major problem for durum wheat production in the Mediterranean region is yield fluctuation. This fluctuation is a result of year-to-year variation in precipitation and heat stress during grain growth, which is typical of the Mediterranean climate. Both yield stability and good quality are needed in adapted durum wheat ideotypes. Ten durum wheat cultivars differing in drought resistance were grown during 1998, under both rainfed and irrigated conditions, at three sites in southern Spain. The main traits studied were protein and lysine content, grain yield, test weight, SDS sedimentation, semolina color, and grain vitreousness. Results show a high influence of site on all traits. Only test weight (TW), SDS sedimentation, grain vitreousness, and protein per kernel appeared to be determined also by cultivar effect. Vitreousness was positively correlated with TW ($r = 0.48$**) and semolina color ($r = 0.46$**). An inverse relationship was found between grain yield and protein content. Regression of cultivar mean values of protein content and grain yield showed a negative correlation ($r = -0.72$***), probably due to dilution of protein by non-nitrogen compounds and reduced starch accumulation in the grain under drought conditions. Lysine content was negatively associated with protein content ($r = -0.86$***), indicating the difficulty of a simultaneous breeding for both characteristics.

Keywords: *Protein content; lysine content; grain yield; quality; durum wheat*

INTRODUCTION

Durum wheat is one of the cereals most grown in the Mediterranean basin. It is cultivated essentially for pasta making, including couscous and bulgur, although in the Mediterranean countries, a large part of the harvest is devoted to bread preparation (*1*). This makes the quality breeding and evaluation a main trend in the research area of durum wheat.

Different environmental variables such as climate, soil, and agronomic practices exert a strong influence on different technological quality parameters of wheat. This effect is particularly marked in Mediterranean environments, where the climate, characterized by increasing water deficit and thermal stress during grain filling, may cause large fluctuations, not only of grain yield, but also of grain quality traits, mainly protein content (*2*). Wheat quality is influenced by genotype, environmental factors, and the interaction between genotype and environment (*3*), but the relative magnitude of environmental, genetic, and G × E effects on quality is unclear (*4*).

Protein content, which is one of the most important traits in quality evaluation and breeding of durum wheat, is known to be influenced mainly by climatic parameters, cultivar, nitrogen fertilizer rate, time of nitrogen application, residual soil nitrogen, and available moisture during grain filling (*5*). Recently, Daniel and Triboi (*6*) reported that the percentage of proteins in the flour increased with the increase of temperature and nitrogen supply, whereas the quantity of proteins per grain was affected negatively by high temperatures and affected positively by N fertilization. Many studies have focused on the relationship between protein content and grain yield. Campbell et al. (*7*) demonstrated that highest protein was obtained under conditions promoting lowest yields in spring wheat, which leads to the inverse relationship between protein content and grain yield. Similar results had been obtained by García del Moral et al. (*8*) in triticale and Novaro et al. (*9*) in durum wheat. In the same context, Beninati and Busch (*10*) reported that the development of high-protein, high-yielding cultivars is believed to be hindered by the predominantly negative correlation of the two traits, which ranged in their study from relatively low (−0.25) to high (−0.77) associations for spring and winter wheat.

Cereal proteins are low in lysine, one of the essential amino acids that cannot be synthesized by humans and must therefore be obtained entirely from dietary sources. Thus, lysine content constitutes an important feature for defining the nutritive value of flour obtained from cereals. There are no works in the literature dealing with the relationship between lysine and protein content in durum wheat, as far as we are aware. In bread wheat, early works in 1960s demonstrated a negative correlation between these two traits (*11*, *12*).

The objectives of this work were (i) to determine the magnitude of variation present in a set of durum wheat cultivars and the degree of additive and multiplicative effects on the traits studied, (ii) to investigate the relationship between grain yield and some quality parameters under Mediterranean conditions, with emphasis on protein content, and (iii) to study the relationships between lysine and protein content.

MATERIALS AND METHODS

Three experimental field trials were conducted during the 1997−1998 season in a representative area of the cereal-growing region of southern Spain, a Mediterranean-type

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Table 1. Mean Squares from the Combined Analysis of Variance Across Sites for the Studied Characteristics of Durum Wheat Grown in the 1997−1998 Season in Southern Spain[a]

source of variance	df	grain yield (× 10^6)	TKW	TW	semolina color	SDS	vitreous-ness	grain protein %	grain protein mg kernel^{-1}	df	lysine content
site	2	55.8***	583.6***	84.9***	8.59***	10.1***	84.2***	82.3***	1.21	2	1.60***
cultivar	9	1.5***	83.1***	21.5***	3.00***	9.1***	88.2***	7.9***	1.91**	9	0.14
site × cultivar	18	0.8***	50.3***	1.8	0.31***	0.5**	53.7***	4.1***	1.36***	18	0.10
blocks (site)	9	1.2***	18.0	2.8*	0.07*	0.8***	6.8*	2.8**	1.13*	3	0.25
error	81	0.2	12.4	1.3	0.03	0.2	3.0	1.0	0.56	27	0.11

[a] TKW = thousand kernel weight; TW = test weight; SDS = sedimentation volume. *, **, and *** indicate significant at 0.05, 0.01, and 0.001 levels of probability, respectively.

environment. The sites were the following: (i) CIFA, Granada (37° 11′ N, 3° 40′ W; Typic Xerofluvent soil) under irrigated conditions; (ii) Ochíchar, Granada (37° 3′ N, 3° 52′ W, loamy Calcixerolic Xerochrept soil) under rainfed conditions, and (iii) La Merced Experimental Station, Jerez (36° 70′ N, 6° 20′ W; Vertisol Uderts soil) under rainfed conditions. Total amounts of rainfall were 549.8 mm, 458.2 mm, and 609.7 mm at CIFA, Ochíchar, and Jerez, respectively.

Ten durum wheat (*Triticum durum* Desf.) cultivars were studied: three commercial varieties (Jabato, Mexa, and Vitrón), one advanced line (ID-1049) from IRTA (Institut de Recerca i Tecnologia Agroalimentaries) of Lerida (Spain), and six advanced lines from the durum wheat core collection of ICARDA (Awalbit, Korifla, Lagost-3, Omrabi-3, Sebah, and Waha).

Cultivars were sown at an adjusted seed rate of 170 kg ha^{-1} (350 viable seeds m^{-2}) in plots 10 m long by 1.2 m wide, with six rows placed 20 cm apart. Each plot received fertilizer according to the standard recommendation as follows (kg ha^{-1}): 60−60−60 N−P−K at CIFA-Granada, 45−45−45 at Ochíchar-Granada, and 32−49−32 at La Merced-Jerez. The experimental design at each site was a randomized complete block with four replications.

Different commercial and technological characteristics other than grain yield were considered: thousand kernel weight (TKW), test weight (TW), protein content (expressed in % and in mg kernel^{-1}), lysine content (in percentage of protein), SDS (sodium dodecyl sulfate) sedimentation test, grain vitreousness, and semolina color (designated also as pigment content). TKW was calculated as mean weight of three sets of 100 kernels. TW, a widespread commercial parameter, was measured on a 250-g sample and expressed as kg hL^{-1}. Protein content was determined using the standard Kjeldhal method. Grain protein percentage was calculated after multiplying Kjeldhal nitrogen by 5.7, and was expressed on a dry basis. The SDS test was performed according to Axford et al. (*13*) using stoppered 25-mL graduated cylinders. Vitreousness was evaluated by visual inspection of 200 grains. A grain was considered defective if it was completely or partially starchy or affected by fungic or viral infections. Semolina color was assessed by the "tristimulus" measure, using a color quality control system and retaining the parameter b as an indicator of pigment content (*14*). Lysine was analyzed quantitatively with high-performance liquid chromatography (HPLC) using the Waters Pico-Tag Method, which involves precolumn derivatization with phenylisothiocyanate (*15*). Protein was hydrolyzed in 6 N hydrochloric acid + 1% phenol in sealed evacuated tubes at 110 °C for 24 h.

Statistical Analysis System (*16*) procedures and programs were used for data analyses. Analyses of variance were calculated using the general linear model procedure, and cultivars and sites were treated as fixed effects. Mean squares were used to compare the relative magnitude of main effects and interaction. Site means of all characters were compared using LSD estimates ($\alpha = 0.05$). Cultivar means for protein content at all sites were regressed on grain yield to determine the relationship between these two traits, and a matrix of correlation was calculated for all characters. Similarly, the relationship between lysine and protein content was graphically represented. Principal component analysis (PCA) were performed on the correlation matrix, calculated on the mean data of all replicates.

RESULTS AND DISCUSSION

Analyses of Variance. The analyses of variance (Table 1) showed a large effect of sites and cultivars and a relative low magnitude of the interaction. Within the main factors, sites and cultivars had a considerable effect on all the characters with the exceptions of protein per kernel in the case of sites, and lysine content in the case of cultivars, but these effects were not significant. The most important interaction effects were found on TKW, vitreousness, and protein content per kernel.

The results clearly show the high influence of site on all characters. In this regard, it is useful to underline that in previous works Peterson et al. (4), Pecetti and Annicchiarico (*17*), and Novaro et al. (*9*) found similar results in durum wheat. In fact, Peterson et al. (*4*) encountered a large effect of environment on many traits but effect of genotype was more determinant for SDS test. For protein content, the environmental effect overcomes genotype and genotype by environment (G × E) interaction effects. These results are confirmed in our study and were also found in other works (*18*, *19*, *20*). A small G × E interaction reveals the lack of multiplicative effects in determining the considered character that appeared essentially influenced by additive effects of the environment and genotype. Novarro et al., (*9*), and as also shown in our results (Table 1), revealed that for characteristics such as grain yield and protein content, highly influenced by additive environmental effects, a large variability over environments must be expected. On the contrary, for traits such as SDS test and TW, for which the genotype effect is important, it is easier to identify the interest of a cultivar over sites.

Lysine content (expressed in % of protein) was mainly influenced by environmental growing conditions, as shown in the analysis of variance, and this might be due to its dependency on protein level in the grain, which is also under environmental control.

Mean Comparison Among Sites. The site mean values of the recorded traits are shown in Table 2. The irrigated site (CIFA) favored a good grain yield, higher TKW, and better lysine content. Technological traits such as TW, semolina color, SDS test, and vitreousness were better expressed in Jerez (rainfed site) probably, because of soil fertility and adequate distribution of rains.

Protein content showed a high percentage in samples from the lowest-yielding site (Ochíchar), whereas in samples from a site with favorable conditions (CIFA), where high grain yield was obtained, low protein contents were recorded. Our results are in accordance with those obtained by Campbell et al. (*7*). Differences

Table 2. Mean Site Values of Different Traits Studied in Durum Wheat Grown at Three Sites of Southern Spain during the 1997–1998 Growing Season[a]

	grain yield	TKW	TW	semolina color	SDS	vitreousness	grain protein		lysine content
	(kg ha^{-1})	(g)	(kg hL^{-1})	b values	(ml)	(%)	(%)	(mg kernel^{-1})	(g/100 g of protein)
CIFA	4523 a	49.9 a	81.1 c	14.7 c	5.4 b	91.0 c	12.8 c	6.4 a	2.5 a
Ochíchar	2162 c	42.4 c	81.8 b	15.1 b	4.8 c	92.2 b	15.7 a	6.7 a	1.9 c
Jerez	3292 b	44.7 b	83.9 a	15.7 a	5.8 a	93.9 a	14.3 b	6.4 a	2.2 b
LSD	184	1.6	0.5	0.1	0.2	0.8	0.5	0.3	0.1

[a] TKW = thousand kernel weight; TW = test weight; SDS = sedimentation volume. Values with the same letter do not differ significantly at the 0.05 probability level using the least significant difference test (LSD).

Table 3. Coefficients of Correlation among Traits of Durum Wheat Grown in Three Sites of Southern Spain during the 1997–1998 Growing Season (n = 30)[a]

	TKW	TW	semolina color	SDS	vitreous ness	grain protein %	grain protein mg kernel^{-1}	lysine content % protein
grain yield	0.50**	0.01	−0.14	0.16	0.35	−0.72***	−0.27	0.60***
TKW		−0.33	−0.38*	−0.10	−0.20	−0.65***	0.28	0.59***
TW			0.66***	−0.05	0.48**	0.15	−0.10	−0.16
semolina color				0.09	0.46**	0.07	−0.27	−0.13
SDS					0.13	−0.20	−0.41*	0.24
vitreousness						−0.10	−0.24	−0.09
grain protein (%)							0.53**	−0.86***
grain protein (mg kernel^{-1})								−0.46*

[a] TKW = thousand kernel weight; TW = test weight; SDS = sedimentation volume. *, **, and *** indicate significant at 0.05, 0.01, and 0.001 levels of probability, respectively.

between sites in the quantity of protein accumulated per kernel were statistically nonsignificant and appear to be mainly under genotypic dependency as has been shown in Table 1. Lysine content exhibited higher values under the irrigated conditions of CIFA, and the lowest percentages were obtained in the rainfed site of Ochíchar.

Correlations Among Characteristics. Table 3 shows the correlation matrix calculated over cultivar means in each site (i.e., $n = 30$). Protein content was negatively correlated with grain yield ($r = -0.72$***), TKW($r = -0.65$***), and lysine content ($r = -0.86$***). The inverse relationship between protein content and grain yield was found in numerous works in bread wheat (*7, 21*), triticale (*8*), and barley (*22*). Similar works on bread wheat underlined the negative correlation between protein and lysine content (*11, 12*).

Vitreousness was positively correlated with TW ($r = 0.48$**) and semolina color ($r = 0.46$**). Novaro et al. (*9*) stated that the increase in yellow-berry percentage (= 100 − vitreousness) determines a significant decrease of test weight (TW) and then negatively influences the semolina yield. Porceddu (*1*) noted that vitreousness is correlated to semolina color, and industries continue to prefer durum wheat low in nonvitreous grain. The positive correlation between grain protein percentage and protein per kernel was probably due to a decrease in kernel weight demonstrated with the inverse relationship between TKW and grain protein percentage (Table 3). These results contradict those of Motzo et al. (*23*) who encountered no correlation between kernel weight and grain protein and thus concluded that the positive relation between grain protein percentage and protein content per kernel was not a consequence of decrease in kernel weight.

Environment Characterization Based on the Studied Traits. Analyses of variance showed that most of the variability present in our results could be attributed to differences between sites. To detect the combination of characters that best explained the existing variability, principal component analysis (PCA) was carried out on the correlation matrix based on the 9 traits shown in Figure 1. The first three PCA axes accounted for 77.3% of total variance: 36.2, 27.2, and 13.9 for axes 1, 2, and 3, respectively.

Toward the positive direction of axis 1 (Figure 1a), there was a joint increase of grain yield, TKW, and lysine content. Toward its negative direction, there was an increase in protein percentage. On axis 2, a positive relationship appeared between TW, pigment content, and vitreousness in the negative side, whereas on its positive direction, only protein per kernel appeared. On axis 3 (Figure 1b), there was a clear separation of SDS volume, which interacted negatively with a group of traits in the other side of the axis formed by protein per kernel, TW, and vitreousness.

The points plotted on the plane determined by the first two axes (Figure 1a) are grouped in clusters related to sites. Axis 1 discriminates between sites according to its moisture regimen whereas axis 2 separated, at a lesser extent and in its negative direction, between Granada (CIFA and Ochíchar) and Jerez trials. Grain yield, TKW, and lysine content were higher in the irrigated site CIFA. Ochíchar favored better protein percentages. In addition to acceptable values of grain yield and protein percentage, Jerez favored a good expression of other quality parameters such as TW, pigment content, and vitreousness. In general, rainfed sites of our study better stimulated the genetic potential of cultivars in terms of producing grains with good quality. This finding agrees with similar work by Borghi et al. (*2*) who concluded that the Mediterranean climate causes a large fluctuation in grain yield, but offers the opportunity for production of high-quality wheats.

Similarly to Figure 1a, points plotted on the plane determined by axes 1 and 3 are grouped in clusters related to sites (Figure 1b). Cultivars in each site appear to be distributed along axis 3, which lead to the conclusion that this axis mainly discriminates between cultivars. The variation between cultivars was mostly explained by traits that were under genetic determination, such as protein per kernel, TW, vitreousness, and

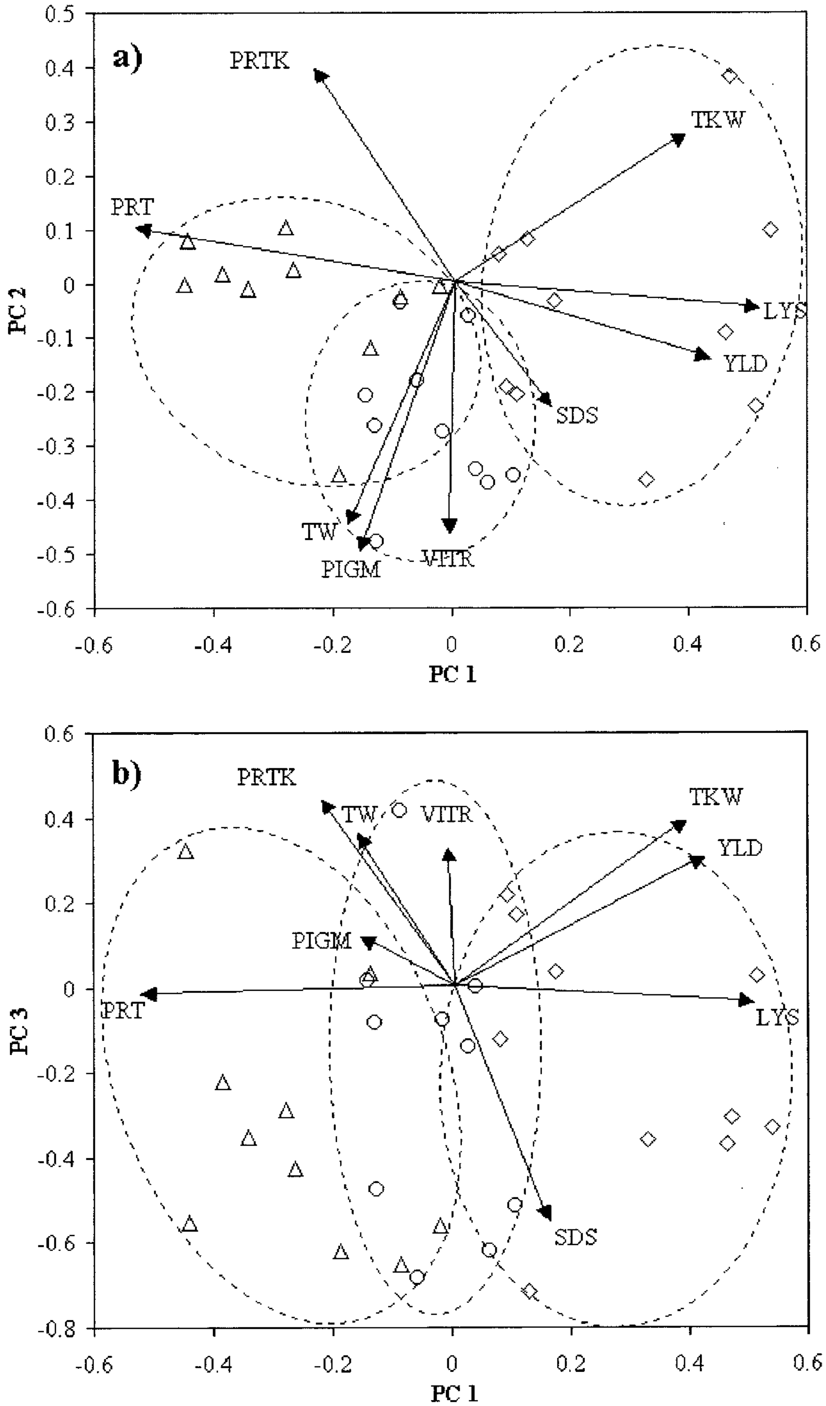

Figure 1. Principal component analysis (PCA) projections on axes 1 and 2 (Figure 1a), and axes 1 and 3 (Figure 1b), accounting for 63.4 and 50.1 of total variance, respectively. In each figure, the eigenvalues of the correlation matrix are symbolized as vectors representing traits that most influence each axis. YLD = grain yield, PRT = protein content, PRTK = protein per kernel, LYS = lysine content, TKW = thousand kernel weight, TW = test weight, PIGM = pigment content, VITR= vitreousness, and SDS = sedimentation volume. The 30 points representing cultivar mean for each site (◇, CIFA; △, Ochíchar; and ○, Jerez) are plotted on the plane determined by axes 1 and 2 (Figure 1a) and 1 and 3 (Figure 1b).

SDS volume. These results confirmed those obtained by the analyses of variance that were previously mentioned.

Relationship between Protein Content and Grain Yield. Regression of cultivar mean values in each site for protein content on grain yield (Figure 2) shows a high negative correlation between these two traits. This inverse relationship between grain yield and grain protein concentration is reported widely in the literature and is frequently confirmed in studies of grain yield and protein. As discussed by Simmonds (*24*), this relationship is curiously consistent, but no simple physiological interpretation is available.

Dilution of protein by non-nitrogen compounds in the grain seemed to be the primary cause for the negative association between grain yield and protein content (*21*). Penning de Vries et al. (*25*) have estimated that, in theory, 1 g of glucose produced by photosynthesis can be used by the crop to produce 0.83 g of carbohydrates or 0.40 g of protein when NO_3 is the N source. This implies that an increase in protein content uses more photosynthate, thus decreasing available photosynthate

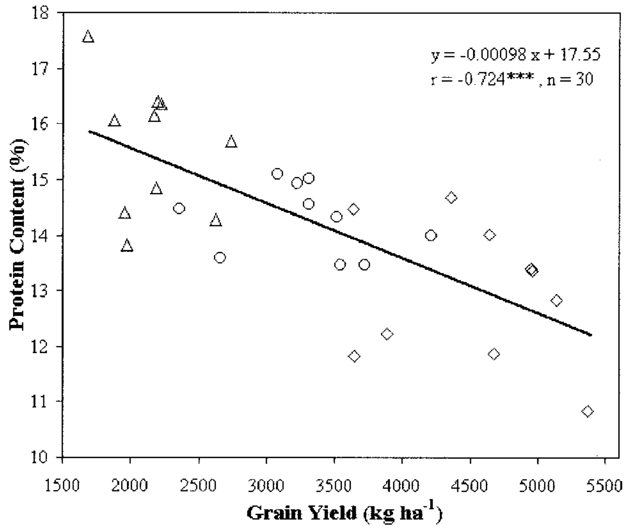

Figure 2. Regression of protein content on grain yield in 10 durum wheat genotypes grown in CIFA (◇), Ochíchar (△), and Jerez (○) during 1997−1998 season. ***, $P < 0.001$.

for carbohydrate synthesis and leading to relative decreases in grain yield. Because nitrogen is the most predominant constituent of proteins, nitrogen accumulation and redistribution during the plant life cycle is of great interest in order to understand the protein−grain yield relationship. Cox et al. (*26*) demonstrated that total grain nitrogen was influenced both by translocation and current assimilation of nitrogen, whereas its concentration was influenced by grain yield and the dilution effect of carbohydrate deposition through photosynthesis.

Conditions that promote leaf senescence during grain growth, such as drought or higher temperatures, favor protein deposition over starch accumulation in the grain because the production and/or translocation of carbohydrates to the grain is more sensitive to adverse growing conditions that is protein production (*7, 27*). In the same way, under conditions that shorten the duration of grain filling period, starch deposition appears to be more affected than protein accumulation (*8, 28*). This is also confirmed by the climatic conditions of our experiments (data not shown), so that the rainfed trial (Ochíchar) was the warmest and driest site during grain filling period, followed by Jerez trial, and finally the irrigated trial (CIFA).

Although the grain protein percentage was negatively related to grain yield and varied with growing conditions, the protein per kernel appeared unchangeable over the sites (Table 2), thus confirming the suggestions made by several authors (*7, 8, 27, 28, 29*) that protein synthesis in the grain is less affected by environmental factors than is starch synthesis, which could determine variation in grain protein percentage.

Relationship between Lysine and Protein Content. Under the conditions of our study, a highly negative correlation ($r = -0.864$***) was encountered between lysine content (as expressed in % of protein) and protein content (Figure 3), suggesting that the percentage of lysine increases as the protein level decreases. There are no literature references on the nature of this relationship in durum wheat as far as we are aware. Nevertheless, in bread wheat an inverse relationship was found in the early works by Lawrence et al. (*11*) and McDermott and Pace (*12*). As discussed by Lawrence et al. (*11*), this negative correlation might

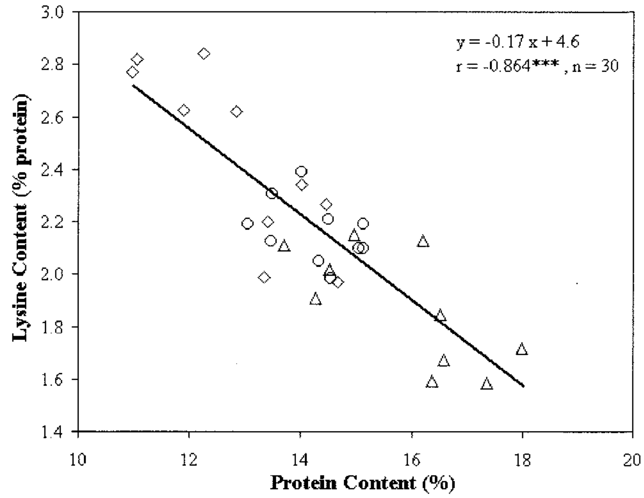

Figure 3. Relationship between lysine and protein content of 10 durum wheat genotypes grown in CIFA (◇), Ochíchar (△), and Jerez (○) during 1997−1998 season. ***, $P < 0.001$.

be accounted for by changes in the proportion of individual proteins with different lysine contents as the total protein content changes; by changes in the lysine content of the proteins themselves; or by changes in the proportions of free lysine present. Moreover, the same authors stated that the proportion of endosperm proteins tends to be lower in wheats of low total protein, and that the endosperm proteins of low-protein wheat tend to have a higher concentration of lysine.

It is worthy to comment here on the positive relationship found in our work between lysine content and grain yield (Table 3), which means that an increase in grain yield will be accompanied with a decrease in protein quantity, but an increase in the nutritional quality of these proteins. However, improvement of lysine content while maintaining or increasing protein content in durum wheat appears to be hampered by the negative correlation between both characters.

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LITERATURE CITED

(1) Porceddu, E. Durum wheat quality in the Mediterranean countries. In *Proceedings of the Seminar on Durum Wheat Quality in the Mediterranean Region*; Di Fonzo, N., Kaan, F., Nachit, M., Eds.; CIHEAM (Centre International de Hautes Etudes Agronomiques Méditerranéennes): Zaragoza, Spain, 1995; pp 11−21.

(2) Borghi, B.; Corbellini, M.; Minoia, C.; Palumbo, M.; Di Fonzo, N.; Perenzin, M. Effects of Mediterranean climate on wheat bread-making quality. *Eur. J. Agron.* **1997**, *6*, 145−154.

(3) Uhlen, K. A.; Hafskjold, R.; Kalhovd, A. H.; Sahlström, S.; Longva, Å.; Magnus, E. M. Effects of cultivars and temperature during grain filling on wheat protein content, composition, and dough mixing properties. *Cereal Chem.* **1998**, *75*, 460−465.

(4) Peterson, C. J.; Graybosch, P. S.; Baenziger, P. S.; Grombacher, A. W. Genotype and environment effects on quality characteristics of hard red winter wheat. *Crop Sci.* **1992**, *32*, 98−103.

(5) Rao, A. C. S.; Smith, J. L.; Jandhyala, V. K.; Papendick, R. I.; Parr, J. F. Cultivar and climatic effects on protein content of soft white winter wheat. *Agron. J.* **1993**, *85*, 1023−1028.
(6) Daniel, C.; Triboi, E. Effects of temperature and nitrogen nutrition on the grain composition of winter-wheat. Effects on gliadin content and composition. *J. Cereal Sci.* **2000**, *32*, 45−56.
(7) Campbell, C. A.; Davidson, H. R.; Winkelman, G. E. Effect of nitrogen, temperature, growth stage and duration of moisture stress on yield components and protein content of Manitou spring wheat. *Can. J. Plant Sci.* **1981**, *61*, 549−563.
(8) García del Moral, L. F.; Boujenna, A.; Yañez, J. A.; Ramos, J. M. Forage production, grain yield and protein content in dual-purpose triticale grown for both grain and forage. *Agron. J.* **1995**, *87*, 902−908.
(9) Novaro, P.; D'Egidio, M. G.; Bacci, L.; Mariani, B. M. Genotype and environment: their effect on some durum wheat quality characteristics. *J. Genet. Breed.* **1997**, *51*, 247−252.
(10) Beninati, N. F.; Busch, R. H. Grain protein inheritance and nitrogen uptake and redistribution in a spring wheat cross. *Crop Sci.* **1992**, *32*, 1471−1475.
(11) Lawrence, J. M.; Day, K. M.; Huey, E.; Lee, B. Lysine content of wheat varieties, species, and related genera. *Cereal Chem.* **1958**, *35*, 169−178.
(12) McDermott, E. E.; Pace, J. Comparison of the amino acid composition of the protein in flour and endosperm from different types of wheat, with particular reference to variation in lysine content. *J. Sci. Food Agric.* **1960**, *11*, 109−115.
(13) Axford, D. W. E.; McDermott, E. E.; Redman, D. G. Small scale test of bread making quality. *Milling Feed Fert.* **1978**, *161*, 18−20.
(14) Trentesaux, E. Evaluation de la qualité du blé dur. In: *Proceedings of the Seminar on Durum Wheat Quality in the Mediterranean Region*; Di Fonzo, N., Kaan, F., Nachit, M., Eds.; CIHEAM (Centre International de Hautes Etudes Agronomiques Méditerranéennes): Zaragoza, Spain, 1995; pp 53−59.
(15) Cohen, S. A.; Meys, M.; Tarvin, T. L. *The Pico-Tag Methodology. A Method of Advanced Techniques for Amino-Acids Analysis*; Millipore Corporation: Bedford, MA, 1989.
(16) *SAS/STAT Software: Changes and Enhancements through Release 6.12*; SAS Institute Inc.: Cary, NC, 1997.
(17) Pecetti, L.; Annicchiarico, P. Grain yield and quality of durum wheat landraces in a dry Mediterranean region of northern Syria. *Plant Breed.* **1993**, *110*, 243−249.
(18) Mariani, B. M.; D'Egidio, M. G.; Novaro, P. Durum wheat quality evaluation: Influence of genotype and environment. *Cereal Chem.* **1995**, *72*, 194−197.
(19) Boggini, G.; Doust, M. A.; Annicchiarico, P.; Pecetti, L. Yielding ability, yield stability, and quality of exotic durum wheat germplasm in Sicily. *Plant Breed.* **1997**, *116*, 541−545.
(20) Ames, N. P.; Clarke, J. M.; Marchylo, B. A.; Dexter, J. E.; Woods, S. M. Effect of environment and genotype on durum wheat gluten strength and pasta viscoelasticity. *Cereal Chem.* **1999**, *76*, 582−586.
(21) Pleijel, H.; Mortensen, L.; Fuhrer, J.; Ojanpera, K.; Danielsson, H. Grain protein accumulation in relation to grain yield of spring wheat (*Triticum eastivum* L.) grown in open-top chambers with different concentrations of ozone, carbon dioxide and water availability. *Agric. Ecosys. Environ.* **1999**, *72*, 265−270.
(22) García del Moral, L. F.; García del Moral, M. B.; Boujenna, A.; Ramos, J. M. Acumulación de proteínas en el grano de cebada en ambiente mediterráneo. In: *Nutrición mineral de las plantas en la agricultura sostenible*; Sarmiento, R., Leidi Montes, E. O., Troncoso del Arce, A., Eds.; Junta de Andalucía, Conserjería de Agricultura y Pesca: Sevilla, Spain, 1997; pp 275−279.
(23) Motzo, R.; Giunta, F.; Deidda, M. Relationships between grain-filling parameters, fertility, earliness and grain protein of durum wheat in a Mediterranean environment. *Field Crops Res.* **1996**, *47*, 129−142.
(24) Simmonds, N. W. The relation between yield and protein in cereal grain. *J. Sci. Food Agric.* **1995**, *67*, 309−315.
(25) Penning de Vries, F. W. T.; Brunsting, A. H. M.; Van Laar, H. H. Products, requirements and efficiency of biosynthesis: A quantitative approach. *J. Theor. Biol.* **1974**, *45*, 339−377.
(26) Cox, M. C.; Qualset, C. O.; Rains, D. W. Genetic variation for nitrogen assimilation and translocation in wheat. III. Nitrogen translocation in relation to grain yield and protein. *Crop Sci.* **1986**, *26*, 737−740.
(27) Fernandez-Figares, I.; Marinetto, J.; Royo, C.; Ramos, J. M.; García del Moral, L. F. Amino acid composition and protein and carbohydrate accumulation in the grain of triticale grown under terminal water stress simulated by a senescing agent. *J. Cereal. Sci.* **2000**, *32*, 249−258.
(28) Bhullar, S. S.; Jenner, C. F. Differential responses to high temperatures of starch and nitrogen accumulation in the grain of four cultivars of wheat. *Aust. J. Plant Physiol.* **1985**, *12*, 363−375.
(29) Jenner, C. F.; Uglade, T. D.; Aspinall, D. The physiology of starch and protein deposition in the endosperm of wheat. *Aust. J. Plant Physiol.* **1991**, *18*, 211−226.

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