Assessment of the phenotypic diversity and agronomic performance of a Mediterranean lentil collection under rainfed conditions: towards efficient use in breeding programs for adaptation to Mediterranean-type environment

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Assessment of the phenotypic diversity and agronomic performance of a Mediterranean lentil collection under rainfed conditions: towards efficient use in breeding programs for adaptation to Mediterranean-type environment

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Abstract

The improvement of lentil productivity and resilience to climate change requires the deployment of breeding approaches and sustainable agronomic practices. Germplasm from the Mediterranean region could be an important source of useful traits for lentil breeding programs. Additionally, no-tillage could also contribute to maintaining lentil productivity in drought-prone environments. However, there are few studies on breeding for adaptation to no-tillage in lentil, as this practice can create growing conditions that differ from those under conventional tillage. The main objectives of this study were to assess the phenotypic diversity of a lentil collection in different environments and to select promising accessions that can be used in lentil breeding programs. A Mediterranean lentil collection of 119 accessions was evaluated in Morocco (under no-till and conventional tillage) and in Turkey (during two growing seasons) under rainfed conditions. There was significant phenotypic variation among accessions for traits assessed. In addition, significant genotype-by-environment interaction effects were observed for grain yield and time to flowering. Moroccan landraces were the earliest to flower compared to landraces from Italy, Turkey, and Greece. Greek landraces displayed the highest mean values of hundred-seed weight. Landraces outperformed advanced lines in low-yielding environment (Turkey in 2022 season) in which higher yield was recorded in Turkish landraces, followed by Moroccan landraces. The high-yielding accessions identified in different environments could be used as donors in breeding programs.

Keywords: Mediterranean lentil landraces, phenotypic diversity, climate change, no-tillage

Introduction

Lentil (*Lens culinaris* Medik.) is a nutritious food legume and an important source of proteins, carbohydrates, fibers, vitamins, and minerals (Johnson et al. 2020; Choukri et al. 2020; Kaale et al. 2023). It is an essential dietary staple in Africa and south Asia in which it plays a vital role in ensuring food and nutritional security. In addition, lentil can deliver many health benefits, including reduced risk of developing diabetes, cardiovascular diseases, and cancer (Ganesan and Xu 2017; Dhull et al. 2023). With its rich nutritional profile, lentil can also be used to develop fortified food products having improved physicochemical, nutritional and technological properties as well as positives health benefits (Bouhlal et al. 2019). Interestingly, lentil has shorter cooking time compared to most other food legumes; consequently, it would be of significant importance in regions with limited cooking fuel supply. Globally, lentil is considered as the third most important cool-season food legume after chickpea (*Cicer arietinum* L.) and pea (*Pisum sativum* L.) (Khazaei et al. 2016). In 2021, the global production of lentil was about 5.61 million metric tons which indicates 111% increase compared to 2.66 million metric tons produced in 1991 (FAOSTAT 2023; Uebersax et al. 2023). Furthermore, the average yield recorded in 2021 (1004 kg ha^{-1}) highlighted an increase of 23.5% compared to average yield registered in 1991 (813 kg ha^{-1}) (FAOSTAT 2023; Uebersax et al. 2023). Currently, lentil is grown in over 50 countries; Canada is the largest producer (1.6 million tons), followed by India (1.49 million tons), Australia (0.85 million tons), Turkey (0.26 million tons), and Nepal (0.24 million tons) (FAOSTAT, 2023). Moreover, global lentil export is dominated by Canada (50.9%), followed by Australia (22.2%), and Turkey with 7.6% (Uebersax et al. 2023).

Lentil, like other legume crops, plays an important role in the sustainability of cropping system, and its incorporation into crop rotation provides significant benefits for succeeding crops due to its ability to enrich the soil with nitrogen, through the formation of symbiotic relationships with nitrogen-fixing bacteria (Quinn 2009), and to reduce diseases severity for succeeding crops such as wheat (Fernandez et al. 1998). It has been reported in a recent meta-analysis study that inclusion of pluses in rotation can benefit wheat crop, which is reflected by increased grain yield, and nitrogen and water use efficiency (Lasisi and Liu 2023). Interestingly, this study reported that lentil resulted in more yield increase for subsequent wheat crop compared to chickpea and pea (Lasisi and Liu 2023). Therefore, it is of significant importance to exploit the rotational value of lentil crop in different cropping systems.

Lentil is grown in three major agro-ecological zones (i.e., Mediterranean, South Asian, and northern temperate) that are characterized by different photoperiods and temperatures (Tullu et al. 2011; Khazaei et al. 2016). The adaptation of lentil to these diverse agro-ecological regions is mainly driven by phenological response to the variation in photoperiod and temperature (Erskine et al. 1994). The genetic variability is important for the success of a breeding program; in fact, breeders are particularly interested in the diversity present in genetic resources to develop improved varieties with superior traits, such as resistance to abiotic and biotic stresses, increased and stable grain yield, and improved grain quality (Swarup et al. 2021). Lentil crop is facing various biotic and abiotic constraints that limit its yield and productivity (Singh et al. 2022; Zeroual et al. 2022; Roy et al. 2023). Therefore, it is important to harness the potential of lentil genetic resources housed in different gene banks around the world to address these challenges. Efficient use of these genetic resources can lead to the development and release of high-yield climate-ready lentil varieties that can contribute in increasing lentil yield and productivity and gain quality, thereby ensuring global food and nutritional security. The Mediterranean region is known by its rich history of domestication and cultivation of lentil and referred as “diversity hotspot” of different species (Idrissi et al. 2018). Studies have reported substantial genetic variability in lentil germplasm originating from the Mediterranean region (Lombardi et al. 2014; Idrissi et al. 2016, 2018; Khazaei et al. 2016; Sahri et al. 2023). Therefore, lentil germplasm collected from this region could be a possible source of diversity that can be used in lentil breeding program for the introgression of favorable alleles into elite cultivars.

In addition to breeding approaches, best crop management practices can also help address different production constraints by establishing beneficial growing conditions. In this context, no-tillage, as a principal component of the conservation agriculture practice, plays a pivotal role in reducing soil erosion and improving its physicochemical and biological proprieties, contributing, therefore, in improving crop productivity (Mrabet 1993, 2011; Ouhemi et al. 2023). In lentil, it has been reported that no-tillage with early sowing, compared to conventional tillage and late sowing, reduces the effects of drought stress on grain yield by optimizing physiological and biochemical responses and promoting soil microbial activity (Saha et al. 2020). However, it has been reported that crop performance under no-tillage is influenced by other genetic, environmental, and management factors (Devkota et al. 2021). To harness the full potential of no-tillage system, it may be advantageous to develop varieties that are specifically adapted to new growing conditions associated with the adoption of this agricultural practice (Herrera et al. 2013). However, the implementation of a breeding program for adaptation to no-tillage system requires the demonstration of the existence of genotype by tillage system interaction (Serraj and Siddique 2012). In previous studies, significant genotype by tillage system interactions were documented in wheat (Honsdorf et al. 2018; Roohi et al. 2022; Mohammadi et al. 2024) and chickpea (Devkota et al. 2021; Abderemane et al. 2023).

The objectives of this study are to: (1) assess the phenotypic variability of a Mediterranean lentil collection in Morocco under two tillage systems and in Turkey during two growing seasons; (2) investigate the significance of tillage and genotype × tillage interaction for grain yield and other agronomic traits; and (3) select promising accessions in different environments that could be used in lentil breeding programs.

Materials and methods

Plant material, experimental design, and growing conditions

A Mediterranean lentil collection of 119 accessions (Table S1), including landraces (from Morocco, Turkey, Italy, and Greece), advanced lines, local cultivars, and improved cultivars was evaluated under no-till (no soil disturbance) and conventional tillage (full tillage) at the experimental station of Sidi El Aidi, Settat, Morocco (33.17° N, 7.40° W, altitude 230 m) of the National Institute of Agricultural Research, Morocco (INRA-Morocco) during the 2020–2021 cropping season. The no-till field had been under no-tillage for more than 10 years without any soil plowing; it is used for growing cereals and legumes as rotations or left as fallow.

The collection was also evaluated during two growing seasons (2020-2021 and 2021-2022) at the experimental area located in the Application and Research Area of the Department of Field Crops, Faculty of Agriculture, University of Çukurova, Adana/Turkey (37°00'59.2"N, 35°21'22.0"E). All trials were grown under rainfed conditions without irrigation.

Sidi El Aidi experimental station in Morocco is located in a semi-arid region with typically Mediterranean climate characterized by hot and dry summers, cold and wet winters, and highly variable inter and intra-annual rainfall (average annual rainfall of about 300 mm). Adana experimental station has a typical Mediterranean climate with mild and rainy winters and hot and dry summers (average annual rainfall of about 670 mm). The Soil of Sidi El Aidi experimental station is classified as Vertic Calcixeroll, while the soil of Adana experimental station is classified as sandy-loam soil. Further soil characteristics of both experimental stations are presented in Table S2 and S3.

All trails were laid out in an augmented randomized complete block design with six lentil checks (ILL8094, L8PS05-5-13, Bakria, "Nilou" Gara, L24, L56). ILL8094 and L8PS05-5-13 are advanced breeding lines; Bakria is a Moroccan cultivar selected from Precoz, an Argentinian landrace, and released in 1989; L24 and L56 are two cultivars selected from Moroccan landraces and registered in Morocco in 1989; "Nilou" Gara is a local cultivar grown in Morocco. Each trial had a total of 149 plots (number of plots = (6 checks × 6 blocks) + 113 test entries). Each block contains 25 accessions, including the six lentil checks, with exception of the block No. 5 which contains 24 accessions, including the six lentil checks.

Each accession was seeded in 4-rows plot of 6m-length; the inter-row distance was 0.35 m and the seeding rate was 150 seeds per m^2. At Sidi El Aidi, for both tillage systems, sowing date was late November 2020; at Adana the sowing date was late December in the 2020-2021 season and in late November in 2021-2022 season. At Sidi El Aidi, both trials received 50 kg/ha of ammonium sulfate (21%); in addition, 0.5 l/ha of Haloxyfop was applied to control narrow-leaf weeds and manual and surface mechanical weeding were used for broad-leaf weeds. At Adana, an average of 20-40 kg/ha of nitrogen and 50-70 kg/ha of phosphorus were supplied; for weeds controls, manual and surface mechanical weeding were adopted;

The agronomics traits namely, time to 50% flowering (days), plant height (cm), hundred-seed weight (g), and grain yield (kg ha^{-1}) were recorded; days to 50% flowering and grain yield were recorded on a plot basis. Hundred-seed weight was estimated from randomly samples (100 seeds) from the plot harvest, while plant height was recorded based on 10 randomly selected plants from each plot.

Statistical analysis

All statistical analyses were performed in R software ver.4.2.3 (R Core Team 2013). Before analysis, 16 accessions from Adana 2022 trial were excluded as they were damaged by cold stress. In each environment (a combination of location-growing season), analysis of variance (ANOVA) of the augmented complete block design, descriptive statistics (mean and range), and heritability estimation were carried out using the R package "augmentedRCBD" (Aravind et al. 2020); the adjusted means obtained by "augmentedRCBD" package were used for further analyses. The analysis of the effect of origin of accessions on different traits and Duncan test were performed using "agricolae" package (Mendiburu 2019). Boxplots and density plots were generated using "ggplot2" package (Wickham et al. 2016). Pearson's correlation between traits studied was performed by "psych" package (Revelle 2017). Principal Component Analysis (PCA) was performed using "Factoextra" (Kassambara 2016) and "FactoMineR" (Husson et al. 2013) packages. Furthermore, for grouping lentil accessions into similar clusters a hierarchical clustering analysis was computed in each environment, based on adjusted means of traits, through Euclidean distances and Ward.D2 method using "cluster" package (Maechler 2018); data were standardized before computing pairwise Euclidean distances between lentil accessions and the result was graphically represented in a Dendrogram.

The combined ANOVA across environments (combination of location-growing season) was performed to evaluate the effects of genotype, environment, and their interaction on different traits. For this, a linear mixed effect model was fit using "lme4" package (Bates et al. 2015). The following model was used:

$$Y_{ijk} = \mu + B_{jk} + G_i + E_j + GE_{ij} + e_{ijk}$$

where Y_{ijk} denotes the observations of the genotype i, in the environment j, and block k; μ is the overall mean; B_{jk} is the random effect of the block k within environment j; $G_i = A_{i'} + A_{ki}$ with G_i denotes the effect of the genotype, $A_{i'}$ is the fixed effect of check i' and A_{ki} is the random effect of test entry i within the block k; E_j is the random effect of the environment j; GE_{ij} is the random effect of the interaction of genotype i with environment j; e_{ijk} is the random residual error associated with observation Y_{ijk}. The variance components were estimated using restricted maximum likelihood (REML) methodology.

Heritability estimates across environments were obtained using the method suggested by Cullis et al. (2006)

$$H^2 = 1 - \frac{\bar{\upsilon}_{\mathrm{BLUP}}}{2\sigma_{\mathrm{g}}^2}$$

where σ_{g}^2 is the genotypic variance and $\bar{\upsilon}_{\mathrm{BLUP}}$ is the mean variance of a difference of two best linear unbiased predictions (BLUP).

The evaluation of environments was carried out using Genotype plus genotype by environment (GGE) analyses (Yan and Tinker 2006) in “metan” package (Olivoto and Lúcio 2020). The data were environment-centered (centering = 2) and scaled by the standard deviation of environments (scaling = 1) in order to reduce heterogeneity among environments. The biplots were drawn using environment-focused singular value partitioning (SVP = 2) (Yan and Tinker 2006; Yan 2015; Hoyos-Villegas et al. 2016).

To evaluate the effect of tillage system and genotype × tillage system interaction, the following model was used:

$$Y_{ijk} = \mu + B_{jk} + G_i + T_j + GT_{ij} + e_{ijk}$$

where Y_{ijk} denotes the observations of the genotype i, in tillage system j, and block k; μ is the overall mean; B_{jk} is the random effect of the block k within tillage system j; $G_i = A_{i'} + A_{ki}$ with G_i denotes the effect of the genotype, $A_{i'}$ is the fixed effect of check i' and A_{ki} is the fixed effect of test entry i within the block k; T_j is the fixed effect of the tillage system j; GT_{ij} is the fixed effect of the interaction of genotype i with tillage system j; e_{ijk} is the random residual error associated with observation Y_{ijk}.

To evaluate the effect of growing season and genotype × growing season interaction, at Adana, the following model was used:

$$Y_{ijk} = \mu + B_{jk} + G_i + S_j + GS_{ij} + e_{ijk}$$

where Y_{ijk} denotes the observations of the genotype i, in growing season j, and block k; μ is the overall mean; B_{jk} is the random effect of the block k within growing season j; $G_i = A_{i'} + A_{ki}$ with G_i denotes the effect of the genotype, $A_{i'}$ is the fixed effect of check i' and A_{ki} is the fixed effect of test entry i within the block k; S_j is the fixed effect of the growing season j; GS_{ij} is the fixed effect of the interaction of genotype i with growing season j; e_{ijk} is the random residual error associated with observation Y_{ijk}.

Results

Climatic conditions

Monthly rainfall and average temperature registered in each trial are presented in Fig. 1. In 2021-2022 growing season the total rainfall recorded at Sidi El Aidi was 347 mm, while at Adana it was 336 mm; in 2021-2022 season the total rainfall registered at Adana was 489 mm. In all trials, rainfalls were consistently distributed throughout the crop growing seasons, but higher amounts of rainfall were received in vegetative growth stage compared to reproductive stage. The average temperature ranged from 10.27 to 20 °C at Adana during the 2021 season, from 6.46 to 25.7 °C at Adana during the 2022 growing season, and from 10.34 to 18.74 °C at Sidi El Aidi during 2021 season. The lowest minimum temperature values were recorded at Adana during the 2022 season in January (2.06 °C) and March (3.94 °C).

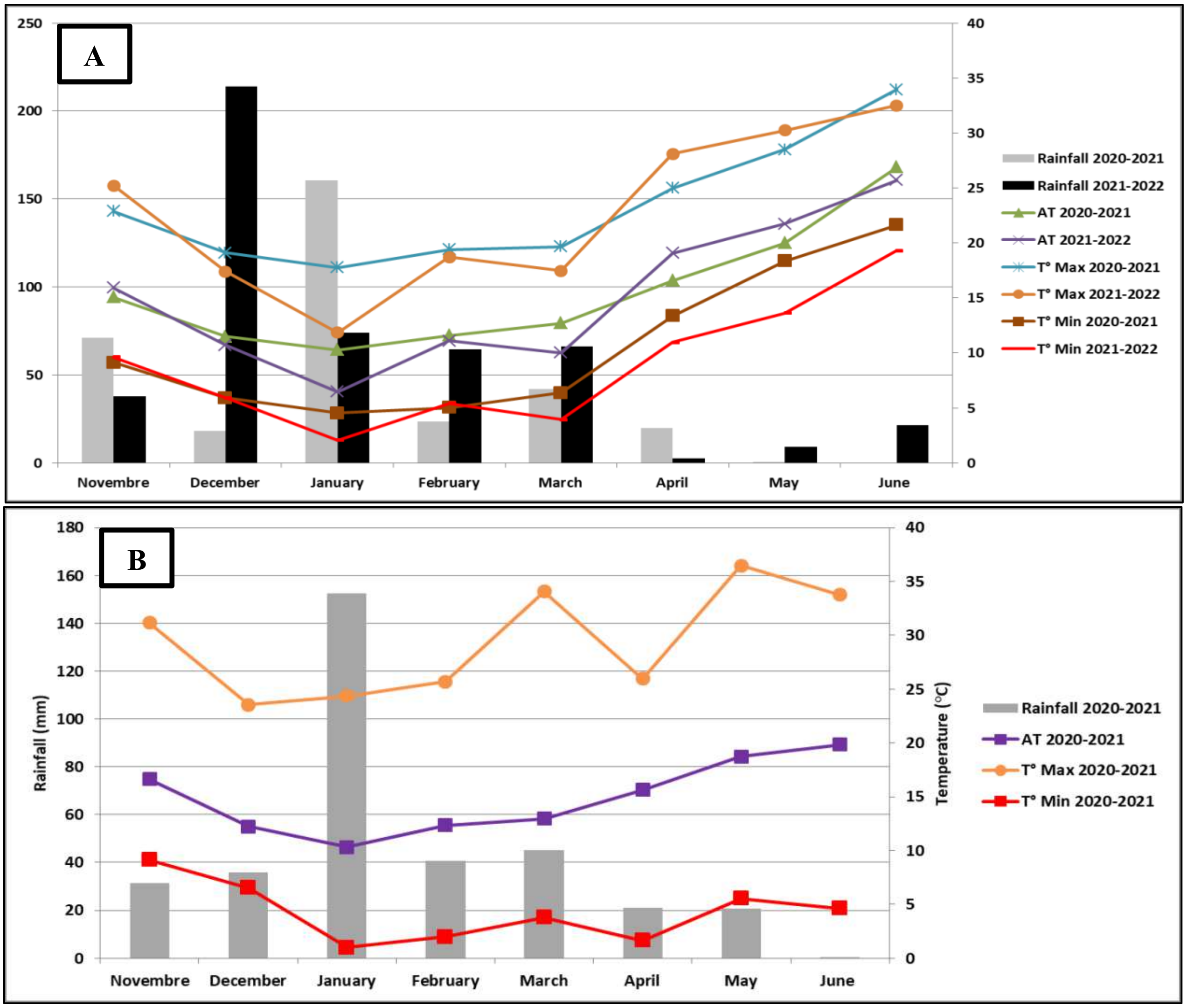


Fig. 1 Rainfall distribution (mm) and average temperature (°C) registered at Adana during 2020-2021 and 2021-2022 cropping seasons (A) and at Sidi El Aidi (B) during 2020-2021 cropping season

Phenotypic variation of the Mediterranean lentil collection in different environments

At Sidi El Aidi, the results showed no significant difference between tillage systems (No-till and conventional tillage) for all traits (Table S4); hence, the data from both tillage systems were combined. At Adana, the results showed significant differences between years (2021 and 2022) for all studied traits, except plant height (Table S5); therefore, each year was analyzed separately.

Analysis of variance indicated significant difference among tested genotypes for the number of days to 50% flowering in all experiments, except at Adana in 2022. The results revealed significant difference for plant height at Adana in 2021 and Sidi El Aidi. For hundred-seed weight, significant variations among genotypes were observed in all tested environments, except at Adana in 2022. In addition, significant genotypic effects on grain yield were recorded at Sidi El Aidi, whereas no significant effects were observed at Adana (Table S6).

Mean, range, and heritability of traits in different environments are listed in Table S7. The number of days to 50% flowering ranged from 82 to 142 days with an average of 104 days (Sidi El Aidi), from 67 to 114 days with an average of 87 days (Adana 2021), and from 61 to 151 days with an average of 111 days (Adana 2022). Plant height ranged from 26.85 to 41.65 cm with an average of 33.4 cm (Sidi El Aidi), from 33.87 cm to 52.7 cm with an average of 41.65 cm (Adana 2021), and from 26.92 to 55.84 cm with an average of 43.28 cm (Adana 2022). Hundred-seed weight ranged from 2.63 to 5.44 g with an average of 3.98 g (Sidi El Aidi), from 2.14 to 6.47 g with an average of 3.51 g (Adana in 2021 trial), and from 1.56 to 4.31 g with an average of 2.76 g (Adana in 2022 trial). For grain yield, the obtained values ranged from 41.59 to 1768.62 kg ha^{-1} with an average of 943.85 kg ha^{-1} (Sidi El Aidi), from 256.61 to 2173.28 kg ha^{-1} with an average of 1252.35 kg ha^{-1} (Adana in 2021 trial), and from 65.92 to 1207.35 kg ha^{-1} with an average of 408.78 kg ha^{-1} (Adana in 2022 trial).

The number of days to 50% flowering showed high heritability at Sidi El Aidi and at Adana during the 2021 season (85.70% and 83.62%, respectively), while moderate heritability was recorded at Adana during the 2022 season (31.51%). Plant height exhibited high heritability at Adana during the 2021 season (60.77%) and Sidi El Aidi (64.92). High heritability estimates were obtained for hundred-seed weight at Sidi El Aidi (87.91%) and at Adana during 2021 season (89.34%); while medium heritability was recorded at Adana during 2022 season (35.87%). For grain yield, the results revealed medium heritability at Sidi El Aidi and Adana during the 2022 season (51.48% and 31.76%, respectively); however, low estimate was observed at Adana during the 2021 season (25.84%).

Phenotypic variability according to geographical origin of the Mediterranean lentil collection accessions

The variation of traits, assessed in the lentil Mediterranean collection, according to geographical origin is illustrated in Fig. 2 and 3. The effect of origin was significant for days to 50% flowering in all trials (Table S8). The hundred-seed weight exhibited significant variation according to geographical origin of accessions at Sidi El Aidi and Adana in 2021. The effect of origin on grain yield was statistically significant at Sidi El Aidi only. On the other hand, no significant effect of the geographical origin on plant height was observed in all trials.

On average, the Moroccan landraces exhibited the shortest time to flowering compared to other landraces in all trials, while the Italian landraces exhibited the longest time to flowering. Greek landraces displayed the highest mean hundred-seed weight as compared to other landraces at Sidi El Aidi and Adana in 2021(Table S8 and S9).

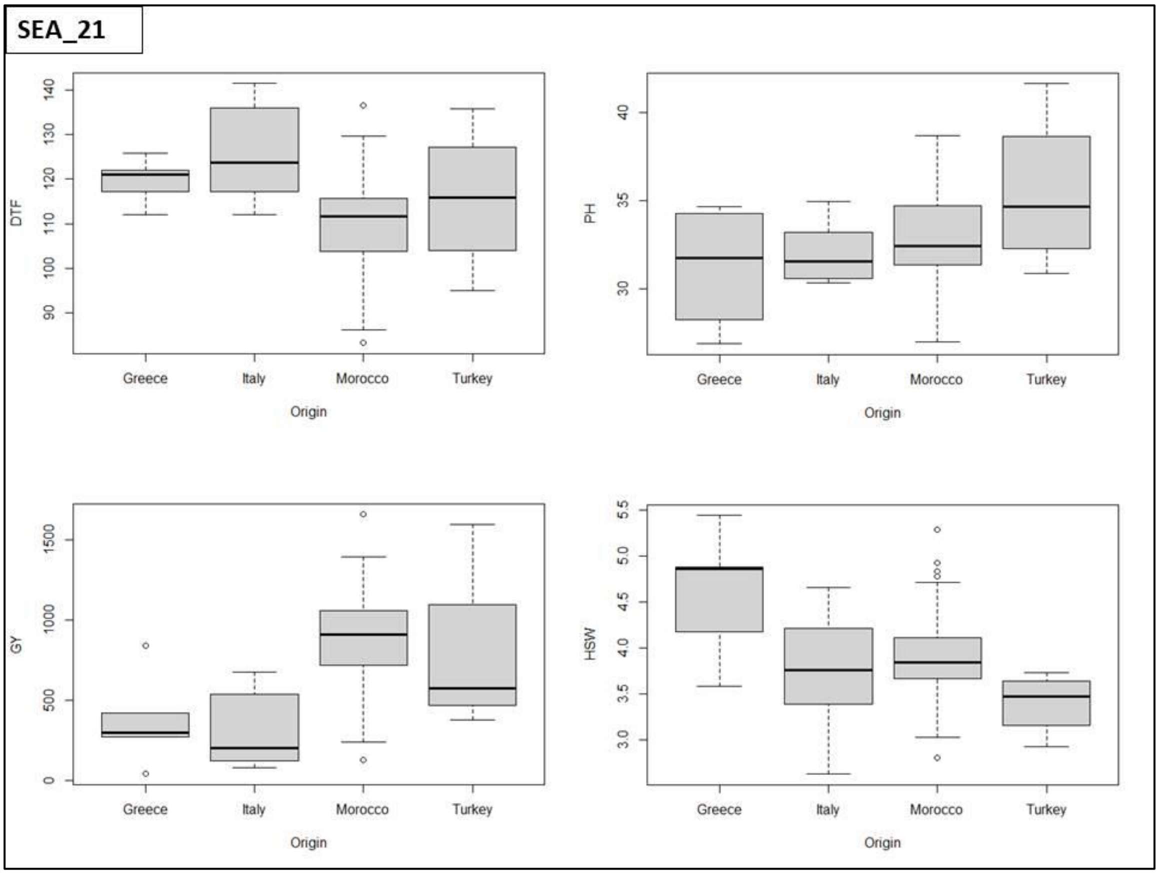
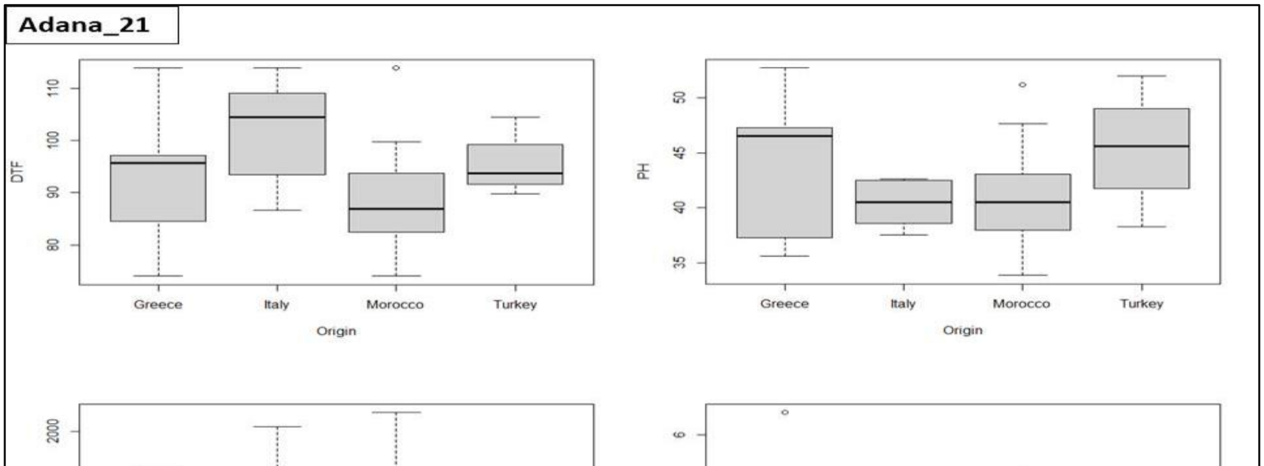


Fig. 2 Variation of agronomic traits assessed in the Mediterranean lentil collection at Sidi El Aidi during 2021 season according to geographic origin. DTF, days to 50% flowering (days); PH, plant height (cm); HSW, hundred-seed weight (g); GY, grain yield (kg ha^{-1})



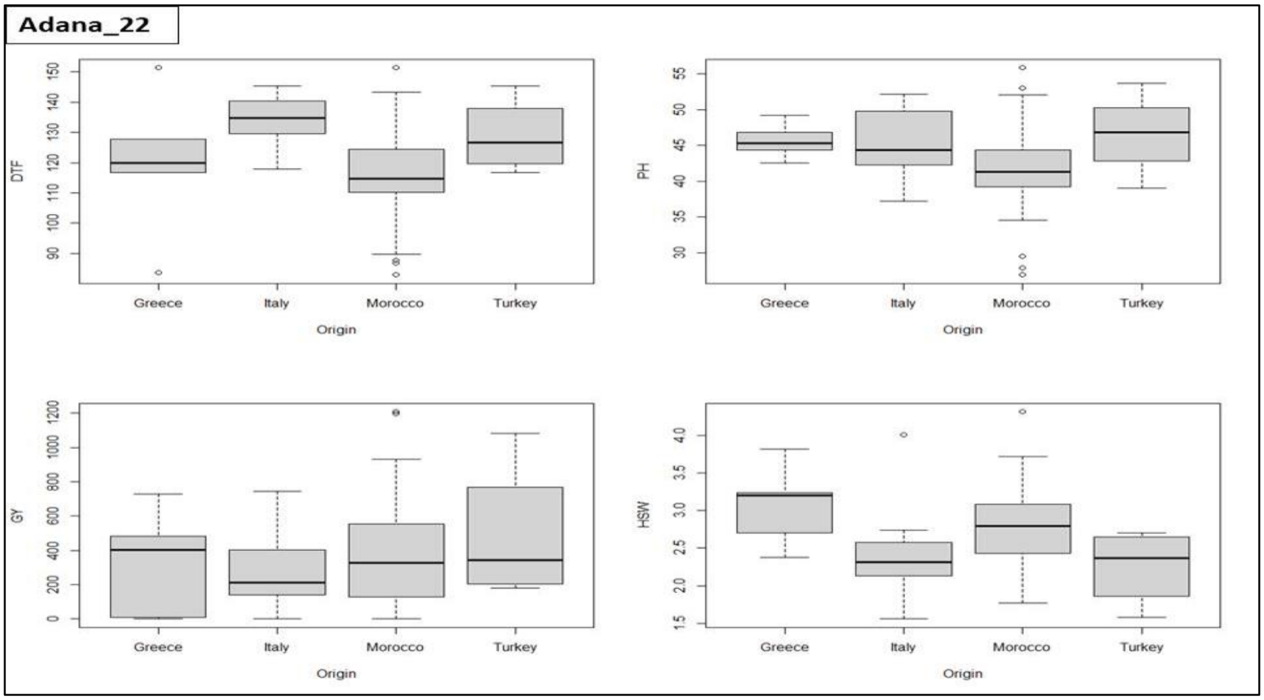


Fig. 3 Variation of agronomic traits assessed in the Mediterranean lentil collection at Adana during 2021 (Adana_21) and 2022 (Adana_22) seasons according to geographic origin. DTF, Days to 50% flowering (days); PH, plant height (cm); HSW, hundred-seed weight (g); GY, grain yield (kg ha^{-1})

Genotype × environment interaction

Results across all environments showed significant effects of genotype on all traits. In addition, significant effects of genotype × environment interaction on days to 50% flowering and grain yield were observed. High heritability estimates were obtained for all traits across all environments which ranged from 64.01% for grain yield to 90.83 for hundred-seed weight (Table S10).

The environments were evaluated for their representative and discriminating ability for grain yield using GGE biplot (Fig. 5). A detailed analysis and interpretation of different GGE biplots can be found in (Yan and Tinker 2006;

Hoyos-Villegas et al. 2016). In Fig. 5a environments with longer vectors are more discriminative; therefore, all environments exhibited consistent discriminating ability, with Adana 2022 representing the most discriminative environment. Furthermore, In Fig. 5a the representativeness of environments can be determined according to their angles with Average-Environment Axis (AEA), the line passing through the average environment and the biplot origin. Accordingly, Adana 2021 is the highest representative (Fig. 5a), followed by Sidi El Aidi 2021. In Fig. 5b, the “ideal” test environment is represented by the arrow at the center of the concentric circles on AEA. Adana 2021 is the closest to this “ideal” environment, highlighting that this environment could be considered ideal; it is followed by Sidi El Aidi 2021.

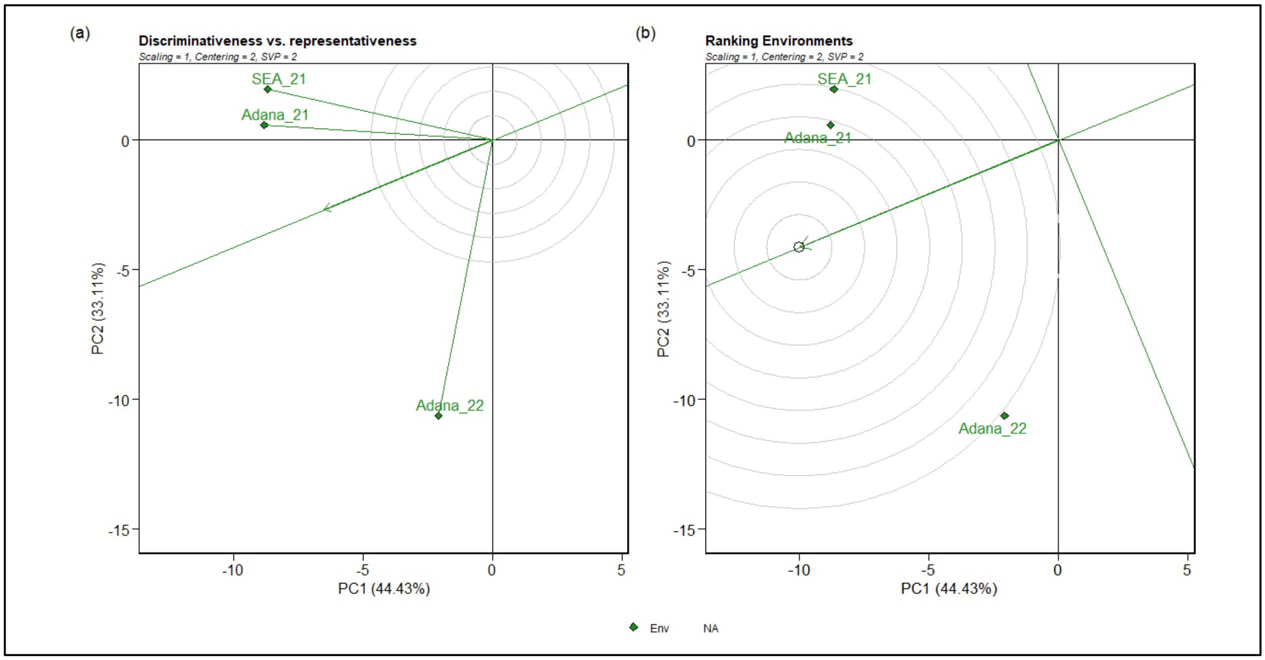


Fig. 5 View of discriminating ability and representativeness of test environments (a) and ranking of environments relative to the “ideal” environment (b) for grain yield. SEA_21, Sidi El Aidi 2021 season; Adana_21, Adana 2021 season; Adana_22, Adana 2022 season

Tillage and genotype × tillage interaction effects

The effect of tillage system on different traits was not significant. Genotype × tillage interaction was only significant for hundred-seed weight (Table S4).

On average conventional tillage outperformed no-till in terms of grain yield (10.23%), while no-till recorded the highest average hundred-seed weight (9.73%). Similar mean values of days to 50% flowering and plant height were noted in both tillage systems (Fig. 6)

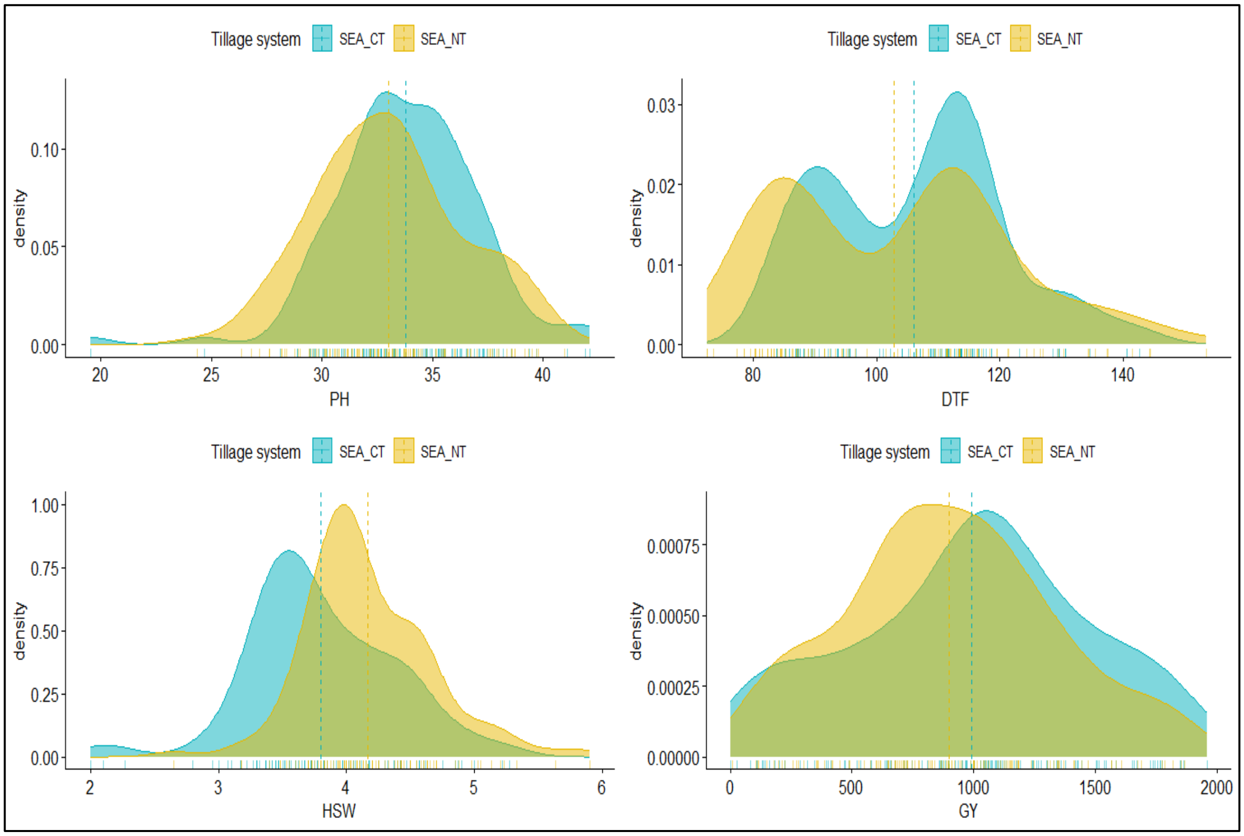


Fig. 6 Density plots of traits assessed in the Mediterranean lentil collection under conventional tillage (SEA_CT) and no-till (SEA_NT) at Sidi El Aidi. DTF, days to 50% flowering (days); PH, plant height (cm); HSW, hundred-seed weight (g); GY, grain yield (kg ha^{-1})

Correlations between traits

Pearson correlation matrix was used to identify correlations among studied traits in each experiment (Fig. 7). Grain yield showed significant negative correlations with days to flowering at Sidi El Aidi (r = -0.76), and at Adana 2021 (r = -0.37), while negative and non-significant correlation was recorded at Adana 2022. The correlation between grain yield and plant height was positive and significant at Sidi El Aidi (r = 0.30), and in Adana during the 2021 season (r = 0.33). Plant height showed a significant negative correlation with days to flowering at Sidi El Aidi (r = -0.23) and at Adana during the 2021 season (r = -0.21). Furthermore, the results indicated significant negative correlations between hundred-seed weight and days to flowering at Sidi El Aidi (r = -0.21) and at Adana in the 2022 season (r = -0.39).

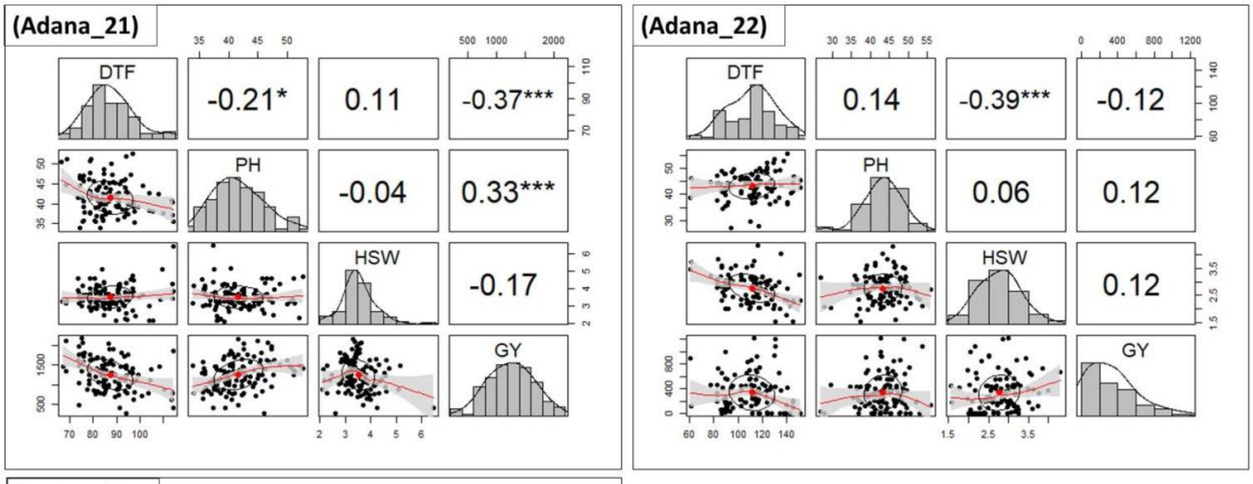


Fig. 7 Correlations between traits screened in the Mediterranean lentil collection grown at Sidi El Aidi during the 2021 season (SEA_21), Adana during the 2021 season (Adana_21), and at Adana during the 2022 season (Adana_22). Upper triangle shows Pearson correlation coefficients between traits; lower triangle shows the scatter plots for correlations between traits, while histograms of traits are presented on the diagonal. DTF, days to 50% flowering (days); PH, plant height (cm); HSW, hundred-seed weight (g); GY, grain yield (kg ha^{-1}). "*", and "*" represent significance at the 0.05, and 0.001 probability levels, respectively**

Principal component analysis (PCA)

Principal component analysis (PCA) was performed using traits assessed in different environments (Fig. 8 and 9). At Sidi El Aidi, the results of PCA showed that the first two principal components (PC1 and PC2) captured 74.5% of the total variation. PC1 (49.51% of the total variation) was highly and negatively correlated with days to flowering and positively correlated with grain yield and plant height, while PC2 (24.99% of the total variability) was mainly associated with hundred-seed weight. At Adana during the 2021 season, the first two principal components explained 66% of the total variability. PC1 (41.8% of the total variation) showed negative correlation with days to 50% flowering and positive correlation with plant height and grain yield, while PC2 (24.2% of the total variation) indicated positive association with hundred-seed weight and plant height. At Adana during the 2022 season, the first two principal components explained 67.7% of the total variability. In fact, PC1 (36.6% of the total variability) was negatively correlated with days to 50% flowering and positively associated with grain yield and hundred-seed weight, while PC2 (28.1% of the total variability) was positively associated with plant height and grain yield.

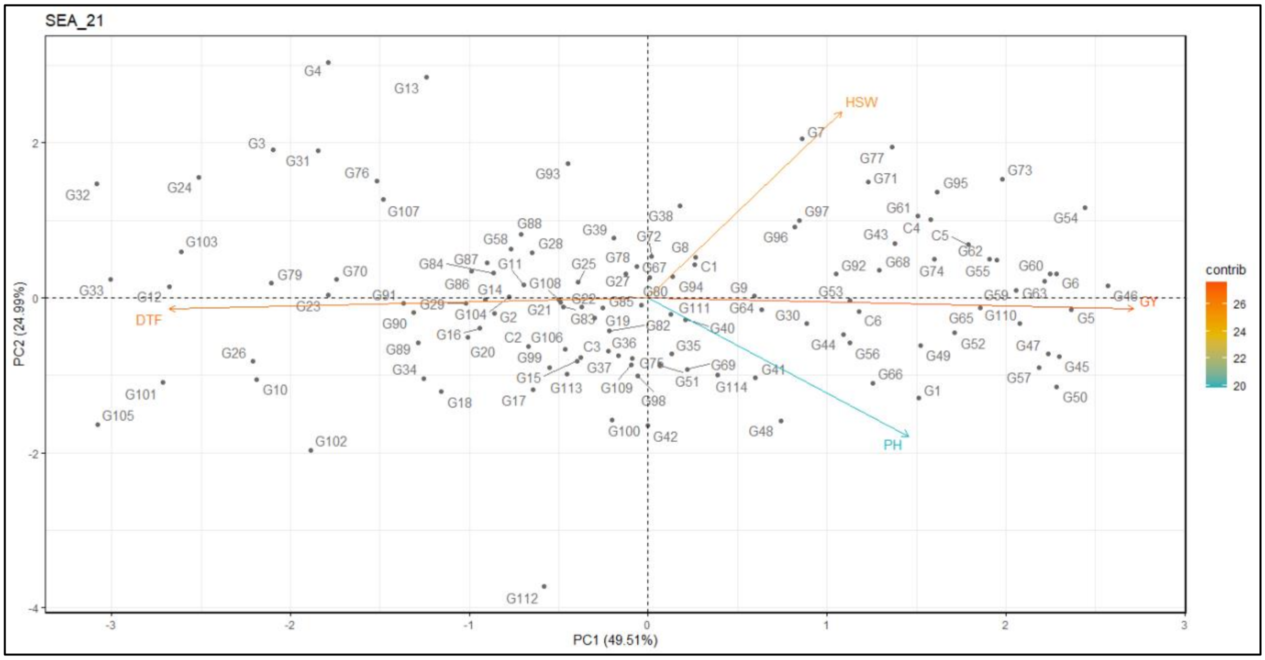


Fig. 8 Principal component analysis (PCA) of four agronomic traits assessed in the Mediterranean lentil collection at Sidi El Aidi in 2021 season. DTF, days to 50% flowering (days); PH, plant height (cm); HSW, hundred-seed weight (g); GY, grain yield (kg ha^{-1})

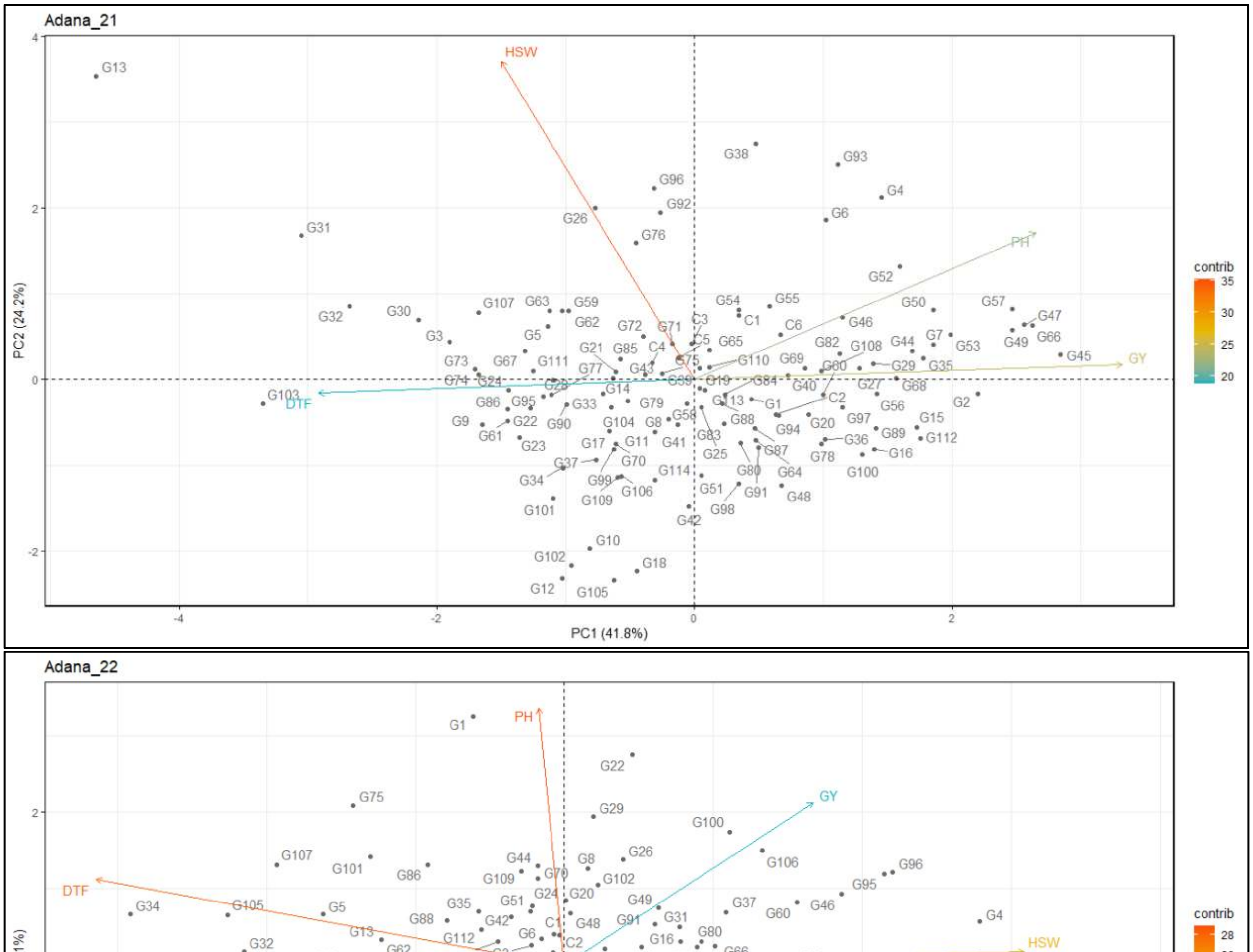


Fig. 9 Principal component analysis (PCA) of four agronomic traits assessed in the Mediterranean lentil collection at Adana in the 2021 season (Adana_21), and at Adana during the 2022 season (Adana_22). DTF, days to 50% flowering (days); PH, plant height (cm); HSW, hundred-seed weight (g); GY, grain yield (kg ha^{-1})

Cluster analysis among Mediterranean lentil collection accessions based on agronomic traits

Hierarchical cluster analysis was used to assess the relationships among accessions of the Mediterranean lentil collection. The accessions were grouped into four different clusters in all trials according to their phenotypic traits (Fig. 10 and 11; Table S11).

At Sidi El Aidi, cluster 1 consisted of accessions with high number of days to 50% flowering (127 days) and hundred-seed weight (4.12 g), low plant height (31.47 cm) and grain yield (187.61 kg ha^{-1}). Cluster 2 resembled late flowering genotypes (112 days) with moderate hundred-seed weight (3.84 g) and plant height (31.52 cm) and low grain yield (682.29 kg ha^{-1}). Genotypes grouped in cluster 3 had intermediate number of days to 50% flowering (102 days), hundred-seed weight (3.91 g) and plant height (33.16 cm), and high grain yield (1044.11 kg ha^{-1}). Genotype grouped in cluster 4 revealed low number of days to 50% flowering (89 days), high plant height (34.32 cm) and hundred-seed weight (4.2 g), and high grain yield (1459.39 kg ha^{-1}). At Adana in the 2021 season, the genotypes from cluster 1 exhibited high number of days to 50% flowering (92 days), moderate plant height (39.7 cm) and hundred-seed weight (3.74 g), and low grain yield (760.65 kg ha^{-1}). Genotypes from cluster 2 had low number of days to 50% flowering (82 days), moderate hundred-seed weight (3.38 g), and high plant height (43.42 cm) and grain yield (1779.17 kg ha^{-1}). Cluster 3 grouped accessions with moderate number of days to 50% flowering (88 days), plant height (40.27 cm), hundred-seed weight (3.35 g) and low grain yield (1080.75 kg ha^{-1}). Accessions grouped in cluster 4 revealed moderate value of number of days to 50% flowering (86 days), plant height (42.7 cm), hundred-seed weight (3.52 g), and high grain yield (1367.89 kg ha^{-1}).

Concerning the results recorded at Adana in the 2022 season, accessions in the cluster 1 are characterized by low number of days to 50% flowering (107 days), intermediate plant height (44.55 cm), and high hundred seed weight (3.12 g) and grain yield (679.09 kg ha^{-1}). Cluster 2 grouped accessions with intermediate number of days to 50% flowering (109) and hundred-seed weight (2.66 g), and high plant height (45.16 cm) and grain yield (992.56 kg ha^{-1}). The accessions of cluster 3 exhibited intermediate number of days to 50% flowering (110 days), plant height (42.8 cm) and hundred-seed weight (2.85 g), and high grain yield (422.98 kg ha^{-1}). The accessions grouped in cluster 4 had moderate number of days to 50% flowering (113 days), plant height (43.07 cm), hundred-seed weight (2.67), and low grain yield (115.12 kg ha^{-1}).

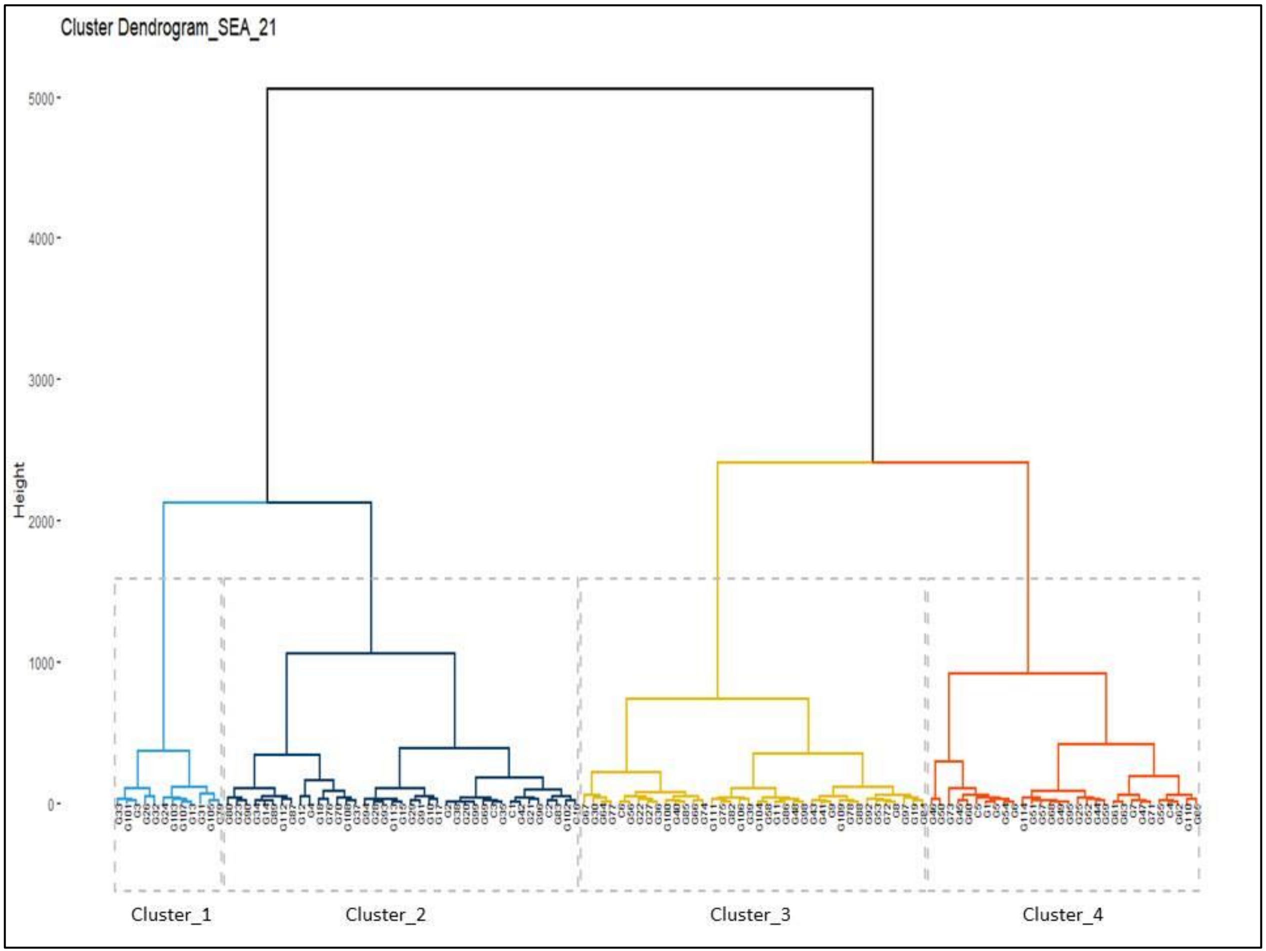
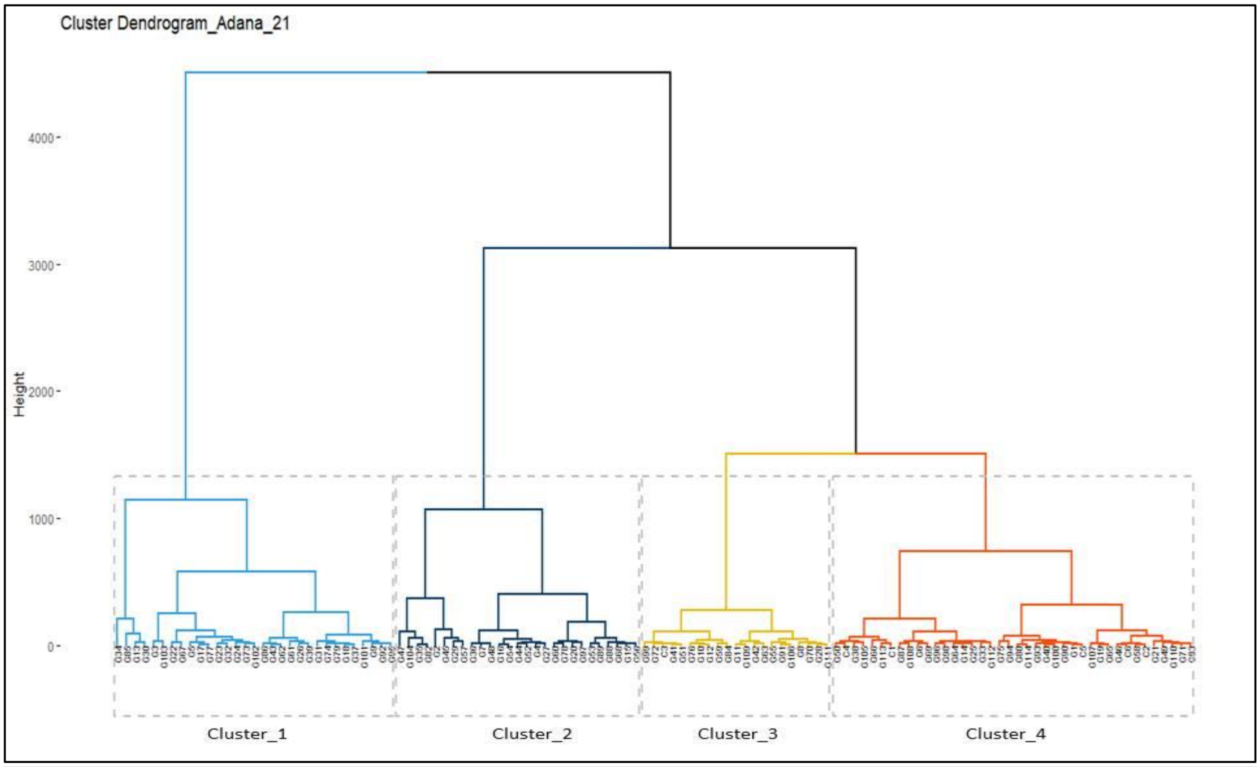


Fig. 10 Hierarchical clustering of Mediterranean lentil collection accessions at Sidi El Aidi


Cluster Dendrogram_Adana_21
Height
4000
3000
2000
1000
0
Cluster_1
Cluster_2
Cluster_3
Cluster_4

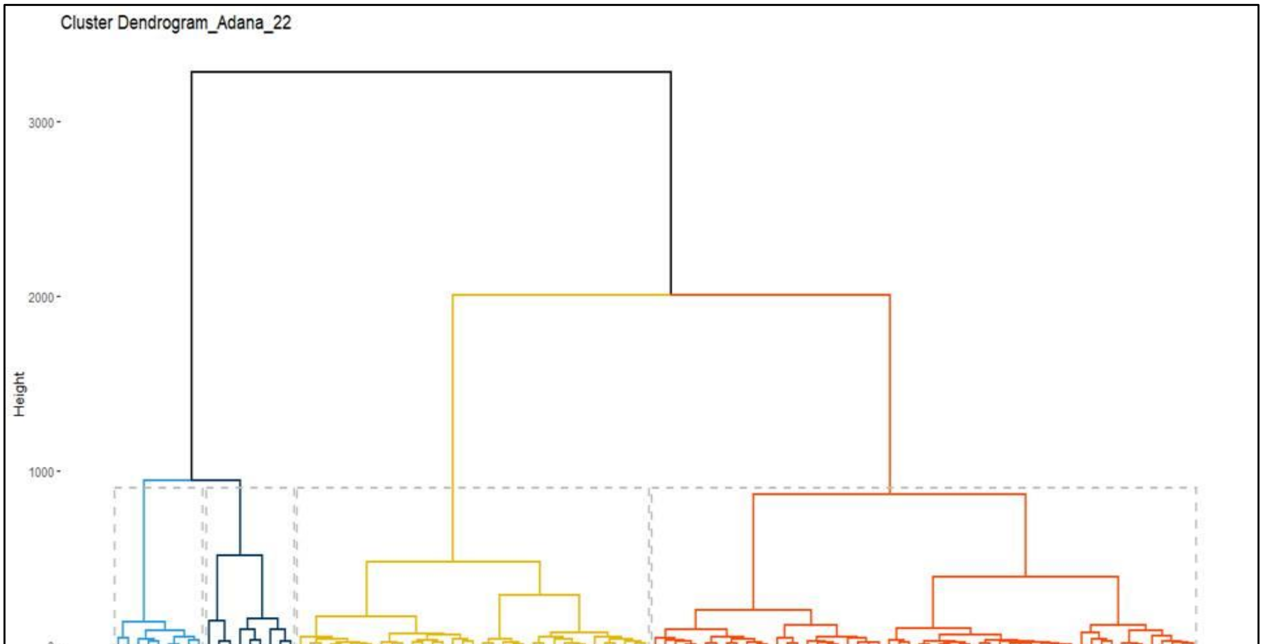

Cluster Dendrogram_Adana_22
Height
3000
2000
1000
0

Fig. 11 Hierarchical clustering of Mediterranean lentil collection accessions at Adana during the 2021 (Adana_21) and 2022 (Adana_22) seasons

Selection of promising accessions

The top 10 lentil accessions were selected based on the grain yield in different environments (Table 1). At Sidi El Aidi, the accessions selected belong mainly to the cluster 4. Generally, the accessions selected in Sidi El Aidi consisted of advanced lines characterized by the early flowering which is an important trait for adaptation to terminal drought and heat stress in Moroccan dry areas; nevertheless, it should be noted that advanced line LL543 and LL599 were derived from landraces.

In Adana during the 2021 season, six landraces against four advanced lines were selected for grain yield production; all the selected accessions belong to cluster 2. At Adana during the 2022 season, the highest performing accessions were generally landraces which highlights their adaptation to stress conditions; further, in this experiment only three landraces produced more than 1000 kg ha^{-1} viz., MGB7391 and MGB1034 from Morocco, and ILL171 from Turkey.

Across environments, the advanced line F04-52 was selected at Adana in both season and could be of interest for further studies of adaptation and stability; the landrace ILL171 was selected twice (Sidi El Aidi and Adana in the 2022 season); the advanced line LL599 was selected at Sidi El Aidi and in Adana during the 2021 season, while the Italian landrace MG111863 was selected at Adana in both seasons.

Table 1 Days to 50% flowering (DTF), plant height (PH), hundred-seed weight (HSW), and grain yield (GY) of the top 10 high-performing lentil accessions selected in different environments based on grain yield

Code	Accession name	DTF (days)	PH (cm)	HSW (g)	GY (kg ha^{-1})	Cluster	Biological status
				SEA_21			
G50	L904-6-15	86	36.15	3.71	1768.62	Cluster_4	Advanced line
G46	LL543	82	34.95	4.31	1746.08	Cluster_4	Advanced line
G73	LR6	86	31.46	4.64	1657.1	Cluster_4	Landrace
C5	L8PS05-5-13	92	33.23	4.39	1617.46	Cluster_4	Advanced line
G1	ILL171	95	35.65	3.56	1596.16	Cluster_4	Landrace
G5	L905-4-1	87	36.46	4.34	1595.26	Cluster_4	Advanced line
G54	F00-24	91	35.75	5.03	1583.02	Cluster_4	Advanced line
G6	F05-15	82	34.54	4.33	1579.09	Cluster_4	Advanced line

G45	LL599	84	36.59	3.99	1568.13	Cluster_4	Advanced line
G60	L805-2-7	89	35.85	4.53	1554.2	Cluster_4	Advanced line
Adana_21							
G2	MGB996	76	41.59	3.3	2173.28	Cluster_2	Landrace
G45	LL599	69	44.93	3.37	2101.85	Cluster_2	Advanced line
G57	LC 960254	76	46.59	3.65	2066.14	Cluster_2	Advanced line
G29	MG111863	87	42.59	3.52	2042.33	Cluster_2	Landrace
G47	F04-52	74	47.2	3.48	1974.87	Cluster_2	Advanced line
G35	MGB1045	88	47.37	3.24	1905.42	Cluster_2	Landrace
G82	ZR-5	87	42.59	3.61	1899.47	Cluster_2	Landrace
G104	MGB1032	114	40.31	3.41	1863.76	Cluster_2	Landrace
G53	ILL81S-15-19	76	45.59	3.53	1804.23	Cluster_2	Advanced line
G89	ZR-12	92	45.65	2.8	1800.26	Cluster_2	Landrace
Adana_22							
G106	MGB7391	113	42.61	2.99	1207.35	Cluster_2	Landrace
G22	MGB1034	131	48.89	2.84	1198.43	Cluster_2	Landrace
G1	ILL171	131	53.65	2.14	1083.45	Cluster_2	Landrace
G49	F05-8	90	42.79	2.02	997.78	Cluster_2	Advanced line
G109	MGB1051	131	42.65	2.48	932.4	Cluster_2	Landrace
G102	MGB9305	116	43.02	2.49	917.39	Cluster_2	Landrace
G47	F04-52	51	38.85	3.47	895.85	Cluster_2	Advanced line
G100	MGB1026	112	47.93	3.16	862.82	Cluster_2	Landrace
G8	ZR-2	108	46.02	2.31	837.54	Cluster_2	Landrace
G29	MG111863	118	50.16	2.74	743.04	Cluster_1	Landrace

SEA_21, Sidi El Aidi in 2021 season; *Adana_21*, Adana in 2021 season; *Adana_22*, Adana in 2022 season

Discussion

The assessment of the phenotypic diversity for traits of interest is a critical step in the breeding process. National and international gene banks harbor diverse germplasm collections that are inexpensive source of beneficial alleles of agronomic and economic significances. These genetic resources are used in breeding programs to develop new varieties with superior traits such as high-productivity and nutritional quality, and tolerance or resistance to abiotic/biotic stresses. In lentil, up to 58,000 germplasm accessions are stored in different gene banks around the globe (Khazaei et al. 2016). The largest collection of lentil germplasm (14,597 accessions) is stored in the International Center for Agricultural Research in the Dry Areas (ICARDA).

In this study, we assessed the phenotypic diversity of a Mediterranean lentil collection of 119 accessions including landraces (from Morocco, Turkey, Italy, and Greece), advanced breeding lines, local cultivars, and improved cultivars. The results unveiled significant variation among Mediterranean lentil collection accessions for traits examined, highlighting the possibility for the selection of promising genotypes that may be exploited in lentil breeding program. Similarly, previous studies reported considerable genetic variation for agronomic and grain quality traits as well as adaptation to biotic and abiotic stresses in different lentil germplasm (Erskine 1983; Tullu et al. 2001; Fernández-Aparicio et al. 2009; Choukri et al. 2020, 2022; El Haddad et al. 2021). The Mediterranean region holds an important lentil genetic diversity thanks to the history of domestication and cultivation and the frequency of abiotic and biotic stress that act as selection pressure (Idrissi et al., 2016). Hence, it can be expected that genetic resources from this region may offer useful adaptive traits for lentil breeding programs. Several studies reported considerable genetic diversity among lentil accessions from Mediterranean region using agro-morphological and phenological traits (Toklu et al. 2009; Bacchi et al. 2010; Torricelli et al. 2012; Zaccardelli et al. 2012; Baggar et al. 2023a), and an arsenal of molecular markers (Lombardi et al. 2014; Idrissi et al. 2015, 2016, 2018; Khazaei et al. 2016; Mbasani-Mansi et al. 2019).

Phenotypic variability for relevant agronomic traits in different environments

Flowering time is an important trait for lentil adaptation to different agro-ecological conditions. In this study, the results showed significant effect of genotype on time to 50% flowering in all trials except at Adana during the 2021 season (Table S6). In addition, significant effects of genotype, environment, and genotype × environment interaction on this trait were obtained (Table S10). These results are consistent with those reported in previous studies (Bermejo et al. 2020; Hossain et al. 2023). In lentil, temperature and photoperiod are the major factors determining flowering time (Summerfield et al. 1985; Erskine et al. 1990). In addition, light quality can also influence the time to flowering with a specie- and genotype-dependent manner (Yuan et al. 2017); in fact, flowering time of some lentil wild accessions was less affected by the variation in light quality as compared to cultivated genotypes (Yuan et al. 2017).

Concerning the plant height, the analysis of variance in each environment indicated significant differences between accessions at Sidi El Aidi and Adana during 2021 season (Table S6). Across environments, there were significant differences between accessions and environments, but the genotype × environment interaction was not significant (Table S10). Another study found significant effect of genotype, season, and their interaction on plant height in lentil (Sharma et al. 2022). Furthermore, it was noted that plant height is influenced by genotype, environment and their interaction (Bermejo et al. 2020). In another investigation, the plant height in lentil crop was significantly varied according to genotype and environment, while the genotype × environment interaction was not significant (Balech et al. 2023).

Hundred-seed weight has great economic significance because it influences the market value of the lentil. In the present study, the effect of genotype on hundred-seed weight was significant in all environments with the exception of Adana in 2022 (Table S6). Combined analysis of variance across environments demonstrated highly significant effects of genotype and environment, whereas the genotype × environment interaction was not significant (Table S10). In accordance with these results, previous studies reported the variation of hundred-seed weight according to the genotype and environmental conditions (El haddad et al. 2020; Choukri et al. 2020). In addition, other investigations, as in the present study, indicated that hundred-seed weight in lentil is not significantly affected by genotype × environment interaction (Abo-Hegazy et al. 2013); however, other researchers reported that this effect was statistically significant (Bermejo et al. 2020; Baggar et al. 2023a). Additionally, significant variation has been reported for hundred-seed weight between and within yellow and red-cotyledon types, with ranges of 1.7-7.4 g for yellow-cotyledon types and 1.3-5.2 g for red-cotyledon types (Tullu et al. 2001).

In the present study, grain yield was significantly influenced by the genotype at Sidi El Aidi, while this effect was not significant at Adana in both seasons (Table S6). Combined analysis of variance across environments revealed that grain yield was significantly affected by genotype, environment, and their interaction (Table S10); in the same sense similar results have been reported previously (Mohebodini et al. 2006; Sabaghnia et al. 2008; Mohammed et al. 2016; Abbas et al. 2019; Idrissi et al. 2019; Chen et al. 2022; Baggar et al. 2023b, a; Ghaffar et al. 2023; Hossain et al. 2023).

The above-discussed results highlight that the Mediterranean lentil collection could be a useful genetic resources for improving traits namely time to flowering, plant height, hundred-seed weight and grain yield. In addition, these results also indicate the importance of the genotype-by-environment interaction for grain yield and time to flowering compared to plant height and hundred-seed weight which should be considered in the selection process.

On average Adana during the 2021 season produced the highest yield followed by Sidi El Aidi, while the lowest average yield was obtained at Adana during the 2022 season and this can be explained by the variability in weather conditions between environments. At Adana during the 2022 season the trial experienced cold stress which negatively impacted the performance of the accessions. Low temperatures observed in January 2022 (2.06 °C) and March 2022 (3.94 °C) may have significantly disturbed the normal growth and development of plants, leading to significant yield reduction. In lentil, the optimum average temperature during reproductive stage is around 15–25/8–10 °C day/night (Kumar et al. 2020). In March 2022, the flowering stage of the majority of accessions was coincided with cold stress which hampered the normal development of flower and pod formation. In another study, cold stress (4 °C for 48 h) induced cellular damages in lentil that were reflected by increased lipid peroxidation and expression of transcripts related to reactive oxygen species (ROS) scavenging (Sohrabi et al. 2022).

Cold tolerance is an important trait to increase lentil production under winter sowing and in highland regions. In highlands of Central and West Asia and North Africa (CWANA) region, lentil crop experiences cold stress at seedling stage and its productivity in such regions could be increased by the development of cold/frost tolerant, winter-hardy cultivars (Kumar et al. 2013). Several cultivars with improved winter hardiness such as 'Kafkas' in Turkey, 'Morton' in USA, 'Gachsaran' in Iran, 'Shiraz-96' in Pakistan, and 'Bichette' in Morocco have been released (Sakr et al. 2004; Sarker and Erskine 2006; Aydogan et al. 2007; Muehlbauer and McPhee 2007; Sabaghpour et al. 2007; Sarker et al. 2009; Kumar et al. 2013). In a recent study conducted in Morocco across nine contrasted environments (combination of three locations and three growing years), the cultivar 'Bichette' showed adaptation to low temperature conditions (Benbrahim et al. 2021). 'Bichette' is a single plant selection from a Jordanian landrace '76TA 66005' (Sakr et al. 2004; Idrissi et al. 2019); therefore, landraces may constitute an important source of useful traits enabling the development of improved cultivars with high productivity that can prosper in winter conditions. These genetic resources might provide a starting point for breeding programs targeting winter hardiness as well as other traits of interest.

Phenotypic diversity of accessions according to geographical origin

Considering the performance of accessions as function of their country of origin, the results indicated that the effect of origin varied depending on the environment and the trait concerned (Fig. 2 and 3; Table S8 and S9). The highest effect of origin was observed in the time to 50% flowering across all environments (Fig. 2 and 3; Table S8 and S9). Hundred-seed weight was affected by origin in Sidi El Aidi and Adana in the 2021 season, while the effect of origin on grain yield was significant only at Sidi El Aidi. Similarly, in another study, highly significant effect of country of origin on different agro-morphological traits was observed in 615 lentil accessions from 13 lentil-producing countries (Erskine et al. 1989). Furthermore, significant effect of geographical origin on different phenological and morphological traits was observed in a lentil core collection of 287 accessions across two years under no-till and conventional tillage conditions (Tullu et al. 2001).

On average, Moroccan landraces were the earliest to flower in all environments (Fig.2 and Fig. 3) as compared to landraces from Italy, Greece and Turkey. The same pattern was reported previously (Idrissi et al. 2018). It has been well established that phenology has played a pivotal role in the adaption of lentil to different agro-ecological regions after the spread from its center of origin (Erskine et al. 1989). The dissemination of lentil from its center of origin to new agro-ecological environments has been accompanied by selection for an appropriate phenological response to regionally-specific balance between photoperiod and temperature (Erskine et al. 1994; Neupane et al. 2023). The response of lentil flowering time to photoperiod has proven to be related to latitude of origin (Erskine et al. 1990). The spread of lentil from its center of origin has resulted in reduced photoperiod sensitivity but increased temperature sensitivity (Erskine et al. 1994; Erskine 1997). The differences between the Mediterranean lentil landraces for the time to flowering could also be attributed to the time of sowing adopted in each region (Idrissi et al. 2018). Additionally, both early and later-flowering were found among genotype from different Mediterranean countries (Neupane et al. 2021). Early-flowering observed in Moroccan landraces is the result of successive selection for avoidance of terminal drought and heat which are considered as important limiting factors for lentil production in Morocco. In fact, it is well known that early phenology is an important trait to escape terminal drought and heat stress (Erskine et al. 1993; Sarker et al. 2005; El haddad et al. 2020).

Considering the performance of accessions regardless of their geographic origins, Moroccan landraces were ranked among the top performing at Adana during 2022 growing season (Table 1), which clearly illustrates that they have also acquired a certain level of tolerance to cold stress in their growing areas. This is consistent with an earlier study indicating the differentiation of Moroccan landraces according to their agro-environmental origins with distinction of landraces from highland regions of Morocco, medium Atlas mountains, which are characterized by the occurrence of cold stress (Idrissi et al. 2015). Furthermore, there is evidence that Moroccan lentil landraces from highland zones and favorable areas may potentially share proportions of the genome with landraces from northern Mediterranean (i.e. Turkey, Italy and Greece) (Idrissi et al. 2018).

Greek landraces displayed the highest mean values of hundred-seed weight at Sidi El Aidi and Adana in 2021. In fact, hundred-seed weight is an important trait of significant market value in lentil crop; it is mostly affected by human-mediated selection processes and with low adaptive value (Erskine 1997).

Relationships between traits

The type and magnitude of correlation between traits of interest can inform on the selection strategy in a breeding program. In the present study, the type and magnitude of correlation varied according to environmental conditions (Fig. 7). Grain yield showed negative correlation with days to 50% flowering, highly significant in most environments, except from Adana in 2022. In agreement with these results, a previous study conducted in two locations in Morocco reported correlation with different type and magnitude in different conditions (El haddad et al. 2020). Early flowering is an important trait that enables the adaptation to Mediterranean-type environment which is characterized by terminal drought and heat stress; this was confirmed by strong association of early flowering and yield in stressful environments (Lake and Sadras 2021). Plant height exhibited significant positive correlation with grain yield at Sidi El Aidi and at Adana 2021 and positive, but not significant, correlation in Adana 2022 (Fig. 7). In agreement with these results significant positive (El haddad et al. 2020; Choukri et al. 2020; Naik et al. 2024) as well as non-significant correlations (El haddad et al. 2020; Choukri et al. 2020) between grain yield and plant height were reported. Overall, the results of the present study indicate that environmental conditions affect the relationships between traits. Hence, for using a trait as selection criteria to improve grain yield, it is important to characterize the target environments as such trait cloud have positive, negative or no effect on grain yield according to the target environment.

Principal Component Analysis and clustering patterns of accessions based on agronomic traits

Results of Principal Component Analysis (PCA) (Fig. 8 and 9) and cluster analysis (Fig. 10 and 11) confirmed the wide variability between accessions phenotyped in the present study in different environments. In addition, the results of PCA and cluster analysis highlight that environmental conditions have significant effect on the performance of accessions for traits studied. The accessions were grouped into four clusters in different environments. Generally, there was no clear relationship between clustering patterns and geographical origin of landraces as landraces from different countries were grouped in the same cluster in different environments. This is in agreement with other reports in common bean (Rana et al. 2015), faba bean (Karaköy et al. 2014), and wheat (Mohammadi and Amri 2022). Furthermore, it has been observed that drought tolerance was not related to landraces origin, indicating that the selection for improved drought tolerance could be focused on the performance of individual accessions rather than their geographical origin (Idrissi et al. 2016). Additionally, other reports indicated differentiation of Moroccan landraces (southern Mediterranean) from landraces from Italy, Turkey and Greece (northern Mediterranean regions) (Idrissi et al. 2016, 2018).

Tillage and genotype × tillage interaction effects

In the present study, effects of tillage and genotype × tillage interaction were evaluated based on experiments conducted at Sidi El Aidi under no-till and conventional tillage system. No-tillage is an important component of conservation agriculture practices that contributes to improving soil physical, chemical and biological properties, thus increasing crop yield and productivity (Mrabet 1993, 2011). No-tillage can maintain yield stability in dry seasons through the conservation of soil moisture that helps crops cope with adverse effects of drought and heat stress. In lentil, it has been reported that no-till could be a buffer system for adaptation to drought stress (Saha et al. 2020). However, to exploit its full benefits, it is essential to tailor no-tillage technology to specific contexts, which may differ according to different factors such soil type, crop and climatic conditions. In addition, the genotypes developed for conventional tillage system may not exhibit the same level of performance when cultivated under conservation agriculture, making it necessary to set up a breeding program for each system; nevertheless, the implementation of such specialized breeding programs require justifying the presence of significant genotype × tillage system interaction (Serraj and Siddique 2012; Roohi et al. 2022). In the present study, there was no-significant effect of tillage system on all studied traits, suggesting that to achieve more sustainability and resource use efficiency no-tillage can be applied without any detrimental effects on lentil productivity; however, these results are derived from single growing season which should be taken in consideration. However, in agreement with these results, another study reported similar yield between no-till and conventional tillage in 13 lentil genotypes under Moroccan rainfed conditions (Devkota et al. 2021), suggesting that lentil can produce acceptable yield under conservation agriculture compared to conventional tillage without yield penalty. In addition, the result of the present study showed no significant effect of tillage system on time to flowering, plant height, and hundred-seed weight. Similarly, in another study, traits such as days to flowering and days to maturity were not influenced by tillage system; however, plant height was influenced by tillage system in wet year compared to dry one (Devkota et al. 2021). In another investigation, in lentil, tillage system effect was not significant on seed yield and seed weight (Das et al. 2019).

In the present study, there was no genotype × tillage system interaction effect observed for all traits, except hundred-seed weight. Previous studies have documented genotype × tillage interaction for grain yield in wheat (Roohi et al. 2022; Mohammadi et al. 2024) and chickpea (Devkota et al. 2021; Abderemane et al. 2023). Although frequent genotype × tillage system interaction was observed in chickpea and wheat compared to lentil and barley, the major variation in grain yield is attributed principally to the variation in rainfall in terms of amount and distribution (Devkota et al. 2021). Overall, no significant effect of genotype × tillage system interaction on grain yield highlights that the establishment of a specialized breeding program could not be justified, which implicates that varieties developed for conventional tillage system could be adopted in conservation agriculture system. However, the efficiency of the lentil selection under conservation agriculture condition was not yet addressed and warrant additional investigations, particularly for grain yield that indicated high heritability under no-tillage as compared to conventional tillage.

Conclusions

The Mediterranean lentil collection assessed in this study showed useful phenotypic variability that could be exploited in lentil breeding programs. This also indicates that the Mediterranean region may contain an important lentil genetic diversity that warrants collection, conservation and utilization in pre-breeding program. Lentil landraces outperformed advanced breeding lines and improved cultivars in low-yielding environment, highlighting the importance of integrating them in lentil breeding programs in order to make available to farmer improved cultivars that can thrive in hash environments.

The geographical origin showed high significant effect on the time to flowering compared to other traits, indicating the importance of phenology in the adaptation of lentil to different ago-ecological regions. Moroccan landraces were the earliest to flowers in all environments compared to landraces from Turkey, Italy and Greece and could harbors interesting alleles associated with phenological adaptation to terminal drought and heat stress. However, it is important to add further accessions from countries considered in the present study and other Mediterranean regions in order to draw tangible conclusions and leverage the genetic diversity present in lentil Mediterranean landraces.

On the other hand, the evaluation of the performance of accessions under no-till and conventional tillage showed that the effects of tillage system and genotype × tillage interaction on grain yield were mostly not significant; thus, considering pedo-climatic conditions and the time frame of the present study, the development of separate lentil breeding programs for no-till and conventional tillage may not be efficient. Nevertheless, additional field trials in different years and locations are needed in order to extrapolate these findings as the current results are based on single growing season. In addition, it is important to evaluate, and consider in lentil breeding programs, the production constraints (soil borne disease, physical constraint for seedling establishment and roots growth) that may arise after shifting from conventional tillage to a no-tillage system.

The accessions selected in different environments are valuable genetic resources that could be incorporated in cross-breeding blocks in order to introgress useful traits/genes/alleles into elite cultivars, obtain transgressive segregants, and develop mapping populations. Furthermore, phenotypic data resulted from this study could be integrated with genotypic data to identify marker-trait associations thought genome wide association studies (GWAS).

Author contributions

Abdelmonim Zeroual: Data curation; methodology; software; formal analysis; investigation; validation; writing—original draft; writing—review and editing; **Mohammed Mitache:** writing—review and editing; **Aziz Baidani**: Supervision; project administration; funding acquisition; Validation; writing—review and editing; **Bacar Abdallah Abderemane**: Methodology; writing—review and editing; **Nadia Benbrahim**: Writing—review and editing; **Hanane Ouhemi:** Writing—review and editing; **Esra Çakır:** Writing—review and editing; **Valerio Hoyos-Villegas**: Writing—review and editing; **Agata Gadaleta:** Resources; project administration; Writing—review and editing; **Elisabetta Mazzucotelli:** Resources; project administration; Writing—review and editing; **Hakan Özkan:** Data curation; investigation; resources; project administration; Writing—review and editing; **Omar Idrissi**: Conceptualization; data curation; methodology; project administration; resources; funding acquisition; supervision; validation; writing—review and editing. All authors read and approved the final manuscript.

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Declarations

Conflict of interest The authors declare no confict of interests.

References

Abbas G, Asghar MJ, Shahid M, et al (2019) Yield performance of some lentil genotypes over different environments. Agrosystems, Geosciences & Environment 2:1–3

Abderemane BA, Fakiri M, Idrissi O, et al (2023) Evaluation of the Productive Potential of a World Collection of Chickpeas (Cicer arietinum L.) for the Initiation of Breeding Programs for Adaptation to Conservation Agriculture. Sustainability 15:11927

Abo-Hegazy SRE, Selim T, Ashrie AAM (2013) Genotype\times environment interaction and stability analysis for yield and its components in lentil. J Plant Breed Crop Sci 5:85–90

Aravind J, Mukesh Sankar S, Wankhede DP, Kaur V (2020) Augmented RCBD: Analysis of augmented randomised complete block designs. R package version 01 2:

Aydogan A, Sarker A, Aydin N, et al (2007) Registration of 'Kafkas' lentil. J Plant Regist 1:44–45

Bacchi M, Leone M, Mercati F, et al (2010) Agronomic evaluation and genetic characterization of different accessions in lentil (Lens culinaris Medik.). Italian Journal of Agronomy 5:303–314

Baggar A, Safi A, Gaboun F, et al (2023a) Identification of stable lentil genotypes through genotype by environment interactions on yield potential in Morocco. Plant Science Today 10:57–66

Baggar A, Safi A, Rakhila Y, et al (2023b) Targeting resilient lentil genotypes with an adding value of nutritional quality by using AMMI and GGE biplots analysis. Plant Science Today 10:56–65

Balech R, Maalouf F, Patil SB, et al (2023) Assessing the Stability of Herbicide-Tolerant Lentil Accessions (Lens culinaris Medik.) under Diverse Environments. Plants 12:854

Bates D, Maechler M, Bolker B (2015) Walker., S. Fitting linear mixed-effects models using lme4. J Stat Softw 67:1–48

Benbrahim N, MONA T, ASMAE B, et al (2021) GRAIN YIELD STABILITY OF LENTIL RELEASED VARIETIES IN MOROCCAN CLIMATIC CONTRASTED ENVIRONMENTS USING AMMI-GGE-BIPLOT ANALYSIS. environment 4:6

Bermejo C, Cazzola F, Maglia F, Cointry E (2020) Selection of parents and estimation of genetic parameters using BLUP and molecular methods for lentil (Lens culinaris Medik.) breeding program in Argentina. Experimental Agriculture 56:12–25

Bouhlal O, Taghouti M, Benbrahim N, et al (2019) Wheat-lentil fortified flours: health benefits, physicochemical, nutritional and technological properties. J Mater Environ Sci 10:1098–1106

Chen C, Etemadi F, Franck W, et al (2022) Evaluation of environment and cultivar impact on lentil protein, starch, mineral nutrients, and yield. Crop Science 62:893–905

Choukri H, El Haddad N, Aloui K, et al (2022) Effect of High Temperature Stress During the Reproductive Stage on Grain Yield and Nutritional Quality of Lentil (Lens culinaris Medikus). Frontiers in nutrition 9:

Choukri H, Hejjaoui K, El-Baouchi A, et al (2020) Heat and Drought Stress Impact on Phenology, Grain Yield, and Nutritional Quality of Lentil (Lens culinaris Medikus). Front Nutr 7:596307. https://doi.org/10.3389/fnut.2020.596307

Cullis BR, Smith AB, Coombes NE (2006) On the design of early generation variety trials with correlated data. Journal of agricultural, biological, and environmental statistics 11:381–393

Das A, Layek J, Ramkrushna GI, et al (2019) Effects of tillage and rice residue management practices on lentil root architecture, productivity and soil properties in India's Lower Himalayas. Soil and Tillage Research 194:104313

Devkota M, Patil SB, Kumar S, et al (2021) Performance of elite genotypes of barley, chickpea, lentil, and wheat under conservation agriculture in Mediterranean rainfed conditions. Experimental Agriculture 57:. https://doi.org/10.1017/S0014479721000107

Dhull SB, Kinabo J, Uebersax MA (2023) Nutrient profile and effect of processing methods on the composition and functional properties of lentils (Lens culinaris Medik): A review. Legume Science 5:e156

El Haddad N, Choukri H, Ghanem ME, et al (2021) High-temperature and drought stress effects on growth, yield and nutritional quality with transpiration response to vapor pressure deficit in lentil. Plants 11:95

El haddad N, Rajendran K, Smouni A, et al (2020) Screening the FIGS Set of Lentil (Lens culinaris Medikus) Germplasm for Tolerance to Terminal Heat and Combined Drought-Heat Stress. Agronomy 10:1036. https://doi.org/10.3390/agronomy10071036

Erskine W (1983) Relationship between the yield of seed and straw in lentil. Field Crops Research 7:115–121

Erskine W (1997) Lessons for breeders from land races of lentil. Euphytica 93:107–112

Erskine W, Adham Y, Holly L (1989) Geographic distribution of variation in quantitative traits in a world lentil collection. Euphytica 43:97–103. https://doi.org/10.1007/BF00037901

Erskine W, Ellis RH, Summerfield RJ, et al (1990) Characterization of responses to temperature and photoperiod for time to flowering in a world lentil collection. Theoretical and Applied Genetics 80:193–199

Erskine W, Hussain A, Tahir M, et al (1994) Field evaluation of a model of photothermal flowering responses in a world lentil collection. Theoretical and Applied Genetics 88:423–428

Erskine W, Tufail M, Russell A, et al (1993) Current and future strategies in breeding lentil for resistance to biotic and abiotic stresses. Euphytica 73:127–135

FAOSTAT (2023) FAOSTAT. https://www.fao.org/faostat/en/#data/QCL. Accessed 18 Jun 2023

Fernández-Aparicio M, Sillero JC, Rubiales D (2009) Resistance to broomrape in wild lentils (Lens spp.). Plant Breeding 128:266–270

Fernandez MR, Zentner RP, McConkey BG, Campbell CA (1998) Effects of crop rotations and fertilizer management on leaf spotting diseases of spring wheat in southwestern Saskatchewan. Can J Plant Sci 78:489–496. https://doi.org/10.4141/P97-082

Ganesan K, Xu B (2017) Polyphenol-rich lentils and their health promoting effects. International journal of molecular sciences 18:2390

Ghaffar M, Asghar MJ, Shahid M, Hussain J (2023) Estimation of G× E Interaction of Lentil Genotypes for Yield using AMMI and GGE Biplot in Pakistan. Journal of Soil Science and Plant Nutrition 1–15

Herrera JM, Verhulst N, Trethowan RM, et al (2013) Insights into genotype× tillage interaction effects on the grain yield of wheat and maize. Crop Science 53:1845–1859

Honsdorf N, Mulvaney MJ, Singh RP, et al (2018) Genotype by tillage interaction and performance progress for bread and durum wheat genotypes on irrigated raised beds. Field Crops Research 216:42–52

Hossain MA, Sarker U, Azam MG, et al (2023) Integrating BLUP, AMMI, and GGE Models to Explore GE Interactions for Adaptability and Stability of Winter Lentils (Lens culinaris Medik.). Plants 12:2079

Hoyos-Villegas V, Wright EM, Kelly JD (2016) GGE Biplot Analysis of Yield Associations with Root Traits in a Mesoamerican Bean Diversity Panel. Crop Science 56:1081–1094. https://doi.org/10.2135/cropsci2015.10.0609

Husson F, Josse J, Le S, Mazet J (2013) FactoMineR: multivariate exploratory data analysis and data mining with R. R package version 1:

Idrissi O, Piergiovanni AR, Toklu F, et al (2018) Molecular variance and population structure of lentil (*Lens culinaris* Medik.) landraces from Mediterranean countries as revealed by simple sequence repeat DNA markers: implications for conservation and use. Plant Genet Resour 16:249–259. https://doi.org/10.1017/S1479262117000260

Idrissi O, Sahri A, Houasli C, Nsarellah N (2019) Breeding Progress, Adaptation, and Stability for Grain Yield in Moroccan Lentil Improved Varieties. Crop Science 59:925–936. https://doi.org/10.2135/cropsci2018.07.0431

Idrissi O, Udupa MS, De Keyser E, et al (2016) Functional Genetic Diversity Analysis and Identification of Associated Simple Sequence Repeats and Amplified Fragment Length Polymorphism Markers to Drought Tolerance in Lentil (Lens culinaris ssp. culinaris Medicus) Landraces. Plant Mol Biol Rep 34:659–680. https://doi.org/10.1007/s11105-015-0940-4

Idrissi O, Udupa SM, Houasli C, et al (2015) Genetic diversity analysis of Moroccan lentil (*Lens culinaris* Medik.) landraces using Simple Sequence Repeat and Amplified Fragment Length Polymorphisms reveals functional adaptation towards agro-environmental origins. Plant Breed 134:322–332. https://doi.org/10.1111/pbr.12261

Johnson N, Johnson CR, Thavarajah P, et al (2020) The roles and potential of lentil prebiotic carbohydrates in human and plant health. Plants, People, Planet 2:310–319

Kaale LD, Siddiq M, Hooper S (2023) Lentil (Lens culinaris Medik) as nutrient-rich and versatile food legume: A review. Legume Science 5:e169

Karaköy T, Baloch FS, Toklu F, Özkan H (2014) Variation for selected morphological and quality-related traits among 178 faba bean landraces collected from Turkey. Plant Genetic Resources 12:5–13

Kassambara A (2016) Factoextra: extract and visualize the results of multivariate data analyses. R package version 1:

Khazaei H, Caron CT, Fedoruk M, et al (2016) Genetic Diversity of Cultivated Lentil (Lens culinaris Medik.) and Its Relation to the World's Agro-ecological Zones. Front Plant Sci 7:. https://doi.org/10.3389/fpls.2016.01093

Kumar J, Sen Gupta D, Djalovic I (2020) Breeding, genetics, and genomics for tolerance against terminal heat in lentil: Current status and future directions. Legume Science 2:https://doi.org/10.1002/leg3.38

Kumar S, Barpete S, Kumar J, et al (2013) Global lentil production: constraints and strategies. SATSA Mukhapatra-Annual Technical 17:1–13

Lake L, Sadras VO (2021) Lentil yield and crop growth rate are coupled under stress but uncoupled under favourable conditions. European Journal of Agronomy 126:126266. https://doi.org/10.1016/j.eja.2021.126266

Lasisi A, Liu K (2023) A global meta-analysis of pulse crop effect on yield, resource use, and soil organic carbon in cereal-and oilseed-based cropping systems. Field Crops Research 294:108857

Lombardi M, Materne M, Cogan NOI, et al (2014) Assessment of genetic variation within a global collection of lentil (Lens culinarisMedik.) cultivars and landraces using SNP markers. BMC Genet 15:150. https://doi.org/10.1186/s12863-014-0150-3

Maechler M (2018) Cluster: cluster analysis basics and extensions. R package version 20 7–1

Mbasani-Mansi J, Ennami M, Briache FZ, et al (2019) Characterization of genetic diversity and population structure of Moroccan lentil cultivars and landraces using molecular markers. Physiology and Molecular Biology of Plants 25:965–974

Mendiburu F de (2019) Agricolae: statistical procedures for agricultural research. (No Title)

Mohammadi R, Amri A (2022) Assessment of the suitability of Triticum turgidum accessions for incorporation into a durum wheat breeding program. Euphytica 218:70

Mohammadi R, Rajabi R, Haghparast R (2024) On-farm assessment of agronomic performance of rainfed wheat cultivars under different tillage systems. Soil and Tillage Research 235:105902

Mohammed YA, Chen C, McPhee K, et al (2016) Yield performance and stability of dry pea and lentil genotypes in semi-arid cereal dominated cropping systems. Field Crops Research 188:31–40

Mohebodini M, Dehghani H, Sabaghpour SH (2006) Stability of performance in lentil (Lens culinaris Medik) genotypes in Iran. Euphytica 149:343–352

Mrabet R (1993) Revue bibliographique sur les systèmes de labour, de conservation de l'eau et leurs effets sur le système sol-plante. 3–38

Mrabet R (2011) No-Tillage Agriculture in West Asia and North Africa. In: Rainfed Farming Systems

Muehlbauer FJ, McPhee KE (2007) Registration of'Morton'winter-hardy lentil

Naik YD, Sharma VK, Aski MS, et al (2024) Phenotypic profiling of lentil (Lens culinaris Medikus) accessions enabled identification of promising lines for use in breeding for high yield, early flowering and desirable traits. Plant Genetic Resources 1–9

Neupane S, Dhakal R, Wright DM, et al (2021) Strategic Identification of New Genetic Diversity to Expand Lentil (Lens culinaris Medik.) Production (Using Nepal as an Example). Agronomy 11:1933

Neupane S, Wright DM, Martinez RO, et al (2023) Focusing the GWAS Lens on days to flower using latent variable phenotypes derived from global multienvironment trials. The Plant Genome 16:e20269

Olivoto T, Lúcio AD (2020) metan: An R package for multi-environment trial analysis. Methods in Ecology and Evolution 11:783–789

Ouhemi H, Aboutayeb R, El Aissaoui A, et al (2023) Effet du système de travail du sol sur les propriétés chimiques du sol sous différents génotypes de lentille et de pois chiche. African and Mediterranean Agricultural Journal-Al Awamia 99–115

Quinn MA (2009) Biological nitrogen fixation and soil health improvement. In: Erskine W, Muehlbauer FJ, Sarker A, Sharma B (eds) The lentil: botany, production and uses. CABI, Wallingford, pp 229–247

R Core Team R (2013) R: A language and environment for statistical computing

Rana JC, Sharma TR, Tyagi RK, et al (2015) Characterisation of 4274 accessions of common bean (Phaseolus vulgaris L.) germplasm conserved in the Indian gene bank for phenological, morphological and agricultural traits. Euphytica 205:441–457. https://doi.org/10.1007/s10681-015-1406-3

Revelle WR (2017) psych: Procedures for personality and psychological research

Roohi E, Mohammadi R, Niane AA, et al (2022) Genotype× tillage interaction and the performance of winter bread wheat genotypes in temperate and cold dryland conditions. Journal of Integrative Agriculture 21:3199–3215

Roy A, Sahu PK, Das C, et al (2023) Conventional and new-breeding technologies for improving disease resistance in lentil (Lens culinaris Medik). Frontiers in Plant Science 13:1001682

Sabaghnia N, Dehghani H, Sabaghpour SH (2008) Graphic analysis of genotype by environment interaction for lentil yield in Iran. Agronomy Journal 100:760–764

Sabaghpour SH, Sarker A, Safikhani M (2007) Registration of 'Gachsaran'Lentil. Journal of plant registrations 1:39–39

Saha M, Bandyopadhyay PK, Sarkar A, et al (2020) Understanding the impacts of sowing time and tillage in optimizing the micro-environment for rainfed lentil (Lens culinaris Medik) production in the lower Indo-Gangetic Plain. Journal of Soil Science and Plant Nutrition 20:2536–2551

Sahri A, Chegdali Y, Qariouh N, Ouabbou H (2023) Diversité phénotypique et structure des populations de lentilles conservées à la banque de gènes de l'INRA. African and Mediterranean Agricultural Journal-Al Awamia 77–98

Sakr B, Sarker A, El Hassan H, et al (2004) Registration of'Bichette'lentil. Crop science 44:686–688

Sarker A, Aydogan A, Chandra S, et al (2009) Genetic enhancement for yield and yield stability. In: The lentil: botany, production and uses. CABI Wallingford UK, pp 102–120

Sarker A, Erskine W (2006) Recent progress in the ancient lentil. The Journal of Agricultural Science 144:19–29

Sarker A, Erskine W, Singh M (2005) Variation in shoot and root characteristics and their association with drought tolerance in lentil landraces. Genet Resour Crop Evol 52:89–97. https://doi.org/10.1007/s10722-005-0289-x

Serraj R, Siddique KH (2012) Conservation agriculture in dry areas. Elsevier

Sharma R, Chaudhary L, Kumar M, et al (2022) Phenotypic diversity analysis of Lens culinaris Medik. accessions for selection of superior genotypes. Sustainability 14:5982

Singh B, Padhy AK, Ambreen H, et al (2022) Understanding Abiotic Stress Responses in Lentil Under Changing Climate Regimes. In: Developing Climate Resilient Grain and Forage Legumes. Springer, pp 179–204

Sohrabi SS, Ismaili A, Nazarian-Firouzabadi F, et al (2022) Identification of key genes and molecular mechanisms associated with temperature stress in lentil. Gene 807:145952

Summerfield RJ, Roberts EH, Erskine W, Ellis RH (1985) Effects of temperature and photoperiod on flowering in lentils (Lens culinaris Medic.). Annals of Botany 56:659–671

Swarup S, Cargill EJ, Crosby K, et al (2021) Genetic diversity is indispensable for plant breeding to improve crops. Crop Science 61:839–852

Toklu F, Biçer B, Karaköy T (2009) Agro-morphological characterization of the Turkish lentil landraces. African Journal of Biotechnology 8:

Torricelli R, Silveri DD, Ferradini N, et al (2012) Characterization of the lentil landrace Santo Stefano di Sessanio from Abruzzo, Italy. Genetic resources and crop evolution 59:261–276

Tullu A, Diederichsen A, Suvorova G, Vandenberg A (2011) Genetic and genomic resources of lentil: status, use and prospects. Plant Genetic Resources 9:19–29

Tullu A, Kusmenoglu I, McPhee KE, Muehlbauer FJ (2001) Characterization of core collection of lentil germplasm for phenology, morphology, seed and straw yields. Genet Resour Crop Evol 48:

Uebersax MA, Siddiq M, Kaale LD (2023) An Overview of Lentil Production, Trade, Processing, and Nutrient Profile. Lentils: Production, Processing Technologies, Products, and Nutritional Profile 1–24

Wickham H, Chang W, Wickham MH (2016) Package ‘ggplot2.’ Create elegant data visualisations using the grammar of graphics Version 2:1–189

Yan W (2015) Mega-environment analysis and test location evaluation based on unbalanced multiyear data. Crop Science 55:113–122

Yan W, Tinker NA (2006) Biplot analysis of multi-environment trial data: Principles and applications. Can J Plant Sci 86:623–645. https://doi.org/10.4141/P05-169

Yuan HY, Saha S, Vandenberg A, Bett KE (2017) Flowering and growth responses of cultivated lentil and wild Lens germplasm toward the differences in red to far-red ratio and photosynthetically active radiation. Frontiers in plant science 8:386

Zaccardelli M, Lupo F, Piergiovanni AR, et al (2012) Characterization of Italian lentil (Lens culinaris Medik.) germplasm by agronomic traits, biochemical and molecular markers. Genetic Resources and Crop Evolution 59:727–738

Zeroual A, Baidani A, Idrissi O (2022) Drought Stress in Lentil (Lens culinaris, Medik) and Approaches for Its Management. Horticulturae 9:1

Supplementary Files

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