



# Article Agronomic Performances and Seed Yield Components of Lentil (Lens culinaris Medikus) Germplasm in a Semi-Arid Environment

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Abstract: Lentil (Lens culinaris Medik.) is widely known among grain legumes for its high nutritional quality, playing an important role in enhancing Mediterranean farming systems as a sustainable crop. Field experiments comparing 121 lentil accessions (microsperma and macrosperma types from different countries) were conducted in a semi-arid environment of south Italy over two growing seasons (2016/2017 and 2017/2018). Their agronomic performance was determined, focusing on phenological, morphological, productive, and qualitative variability. Changes in rainfall and temperatures affected the agronomic traits, especially yield components. In both years, the average grain yield (GY) (2.31 and 2.22 t ha<sup>-1</sup>, respectively) was above the threshold of 2 t ha<sup>-1</sup>. Consistent yield exceeding the field average in both growing seasons revealed the superiority of accessions from Egypt, Cyprus, Algeria, Nepal, and Tunisia. Moreover, *microsperma* yielded more (+0.31 and +0.41 t ha<sup>-1</sup> in the first and second year, respectively) than macrosperma accessions. Flowering (DASF-days after sowing to flowering) and thousand seed weight (TSW) appeared to be the most important traits related to grain yield. Flowering earliness seems to act as a mechanism for overcoming abiotic stresses. The analysis of yield components revealed a different productive determinism within the two subspecies. As also highlighted by the Principal Component Analysis, microsperma accessions presented on average a significantly higher number of pods per plant (PP) and seeds per pod (SP), despite the considerable variability among countries of origin. The results showed phenological and morphological variability among genotypes, which should be taken into account in view of future selection programs focused on obtaining lentil ideotypes suitable for the Mediterranean environment.

Keywords: lentil; agronomic traits; yield components; germplasm collection; ICARDA genotypes

# 1. Introduction

Lentil (*Lens culinaris* Medik., 2n = 2x = 14) is a self-pollinating annual crop and one of the most important grain legume species in the world [1]. It was one of the first crops to initiate agriculture in the Neolithic period in the "Fertile Crescent", which then spread westward in other areas of Asia, the Mediterranean, and temperate lands of Europe and Egypt [2].

Lentil was grown on an estimated global area of 5,585,879 ha in 2021 for an average yield of roughly 1 t ha<sup>-1</sup>. Canada, India, and Australia are among the leading producing countries [3]. The Italian agricultural areas under its cultivation have rapidly risen from 3215 ha in 2016 to 5587 ha in 2023: in detail, from 2006 to 2016, the average area cultivated with lentils was 2354 ha, increasing then to 5442 ha in the period 2017–2023. The total production in 2023 was 6083 t for an average yield of about 1.1 t ha<sup>-1</sup> [4].

According to the classification of Barulina (1930), cultivated lentil can be grouped in two categories based on seed size and 100 seed weight: large-seeded or *macrosperma* type with a diameter of 6–9 mm and a weight higher than 4 g; small-seeded or *microsperma* type



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**Copyright:** © 2024 by the authors. Licensee MDPI, Basel, Switzerland. This article is an open access article distributed under the terms and conditions of the Creative Commons Attribution (CC BY) license (https:// creativecommons.org/licenses/by/ 4.0/). with a diameter ranging from 2 to 4 mm and a weight of less than 4 g [5]. Several market classes are also defined on the basis of seed size and color such as red, green, yellow, and Spanish brown [6]. Consumption of the last two classes is limited to a few countries, while the popular type in Italy is the green one with its multiple sizes, i.e., extra small, small, and medium [7]. Lentil straw may also find useful application as a resource for livestock feed [8].

Lentil is well known to be an excellent source of protein (circa 25%), slow-digesting carbohydrates, dietary fiber, low fat, a diversity of micronutrients [9], and bioactive healthpromoting compounds, including polyphenols [10]. Different ranges of protein values attributed to genetic variability [11] and growing conditions related to year and location [12] are reported in the literature within the same species. A study comparing 39 landraces and seven cultivars resulted in the former having an average greater protein content (25.60% and 24.43%, respectively), together with a wider range of values (22.72–31.88% and 23.60–26.54%, respectively) [13]. This nutritional profile makes the pulse an important component of a healthy diet in the context of prevention against health conditions such as obesity [14], diabetes [15], and cardiovascular disease risk [16]. It is also worth mentioning that the consumption of improperly processed lentil involves the intake of antinutritional factors and consequential clinical implications, therefore traditional and cutting-edge treatments are recommended to reduce their content [17]. Nonetheless, a recent study conducted in China [18] showed that lectin derived from lentil can act as a strong infection inhibitor against variants of SARS-CoV-2, having also shown stronger antiviral activity than other plant-derived lectins.

The cultivation of this legume plays an important role in enhancing Mediterranean farming systems thanks to its adaptability, especially in marginal areas characterized by rainfall shortage and high temperatures [19]. Introducing lentil into crop rotations helps to enrich nitrogen and water availability of the soil for the benefit of the following crop [20] and limit the potential soil erosion caused by wind and water, also improving soil properties [21]. According to [22], "pulses serves as a mini nitrogen factory", thereby reducing the use of synthetic fertilizers and promoting environmentally friendly agricultural systems. Several agronomic traits of wheat can be positively influenced when crop rotation is preferred to monoculture, leading to various and significant effects depending on the preceding legume [23]. For instance, in an eight-year experiment alternating wheat after pea, lentil, and chickpea, the cultivation of the first two legumes led to significant yield increases for the cereal. Additionally, the positive effect was also observed in the soil, with greater nitrogen and organic carbon content at 30 and 15 cm depth, respectively [24].

Lentil exhibits high heterogeneity in terms of yield and morphological traits due to genetic and environmental variability [25]. Crop productivity and economic return are also affected by seeding rate and row spacing. Specific recommendations have been indicated based on agronomic management and area of cultivation, suggesting to increase seed density in organic systems to values of 375 seeds  $m^{-2}$  [26] or 280–320 seeds  $m^{-2}$  with narrower row spacing in rainfed Mediterranean environments [27]. Several traits related to grain yield and protein content have been indicated as relevant aspects for the selection of genotypes for breeding improvement [28]. The generation of hybrids through crosses using commercial varieties and crosses between accessions could be an alternative practice to conventional breeding methods for boosting seed yields [29] as well as for increasing resistance to abiotic and biotic stress [30,31].

The production of legumes is the result of different plant growth processes, often expressed in the yield components of pods/plant, seeds/pod, and mean seed weight. The highest seed yields are generally obtained when all of them are maximized. Yield components analysis has been used to explain variations in the yield of grain legumes [32].

The International Center for Agricultural Research in the Dry Areas (ICARDA) owns one of the most extensive lentil collection in the world [33]. Studies conducted in Brazil on a range of lentil genotypes provided by ICARDA on the basis of morpho-agronomic characteristics have made it possible to identify promising genotypes that can be exploited in breeding programs [34].

A recent study conducted in the same area of Calabria region focused on salinity tolerance of lentil genotypes. It was clearly shown that some Italian accessions can tolerate soil salinity stress to a greater extent. In contrast, a commercial variety used as tester (Eston) presented generalized decreases of several plant growth parameters in the same condition [35]. Furthermore, previous research had already revealed the considerable yield and production stability of 22 lentil accessions in the same region [36]. This study characterized and evaluated, ex situ, a part of accessions selected within the IBBR-CNR Lentil collection provided by Mediterranean Germplasm Database (MGD). The aim of this work was to identify promising genotypes that can be exploited in breeding programs in semi-arid conditions, based on morpho-agronomic performances already shown in previous trials. Availability of information of part of this collection of lentil germplasm would facilitate the utilization of new sources for developing improved lentil lines with higher yields and height of first pod insertion, well-suited for harvest mechanization. The plant material mostly included Asian and Mediterranean germplasm (including the Nile valley area from Egypt to Ethiopia). An earlier study [36] was expanded (from 22 to 121 accessions) and deepened to broaden all genetic variability that can be used in breeding programs; representation of different countries of original collection consisting of 231 accessions was maintained in the set utilized.

## 2. Materials and Methods

## 2.1. Plant Materials

This study evaluated 121 lentil accessions from various sources, including: 117 genotypes from the Mediterranean Germplasm Database (MGD), the reference database for agro-food plant germplasm collection stored at the Institute of Biosciences and Bioresources (IBBR) of the Italian National Research Council (CNR—Bari, Italy); two commercial varieties established in Canada (Laird, *macrosperma*) and USA (Eston, *microsperma*); two improved ICARDA genotypes (ILL 7202 and ILL 7535) obtained through classical breeding strategies to realize suitable lines for semi-arid and arid environments. The two ICARDA genotypes were selected for high productivity and stability based on earlier trials on accessions of Lentil International Screening Nursery (LISN-S-94), entry name FLIP92-37L (ILL 7202) and FLIP93-34L (ILL 7535).

The two commercial varieties and the two improved genotypes were used as testers in the agronomic evaluation.

Previous experiments started in 1995 at the Department of AGRARIA (University Mediterranea of Reggio Calabria) on a larger collection of 232 accessions had already demonstrated the improved agronomic performance of those used in this study [19,36–38]. The date passport of each accession preserved at the MDG is available at: https://www.ibbr.cnr.it/mgd/?action=search&p=adv&xkind=genus&xstr=Lens (accessed on 5 September 2023).

The plant material used in this experiment is shown in Table S1 by sequence number, access code, country of origin, and subspecies: *microsperma* (no. 82) and *macrosperma* (no. 39).

The 117 accessions are from 15 different countries: Italy (no. 24), Tunisia (no. 13), Algeria (no. 10), Cyprus (no. 10), Egypt (no. 10), Morocco (no. 10), Pakistan (no. 10), Ethiopia (no. 9), Spain (no. 6), Greece (no. 4), Albania (no. 3), Libya (no. 3), Iran (no. 2), Nepal (no. 2), and South Africa (no. 1) (Table 1).

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Origin <sup>1</sup>	Small	Large	Total
Italy (It)	16	8	24
Tunisia (Tu)	7	6	13
Algeria (Al)	4	6	10
Cyprus (Cy)	4	6	10
Egypt ((Eg)	10	0	10
Morocco (Mo)	4	6	10
Pakistan (Pa)	10	0	10
Ethiopia (Et)	9	0	9
Spain (Sp)	2	4	6
Greece (Gr)	3	1	4
Albania (Ab)	3	0	3
Libya (Li)	2	1	3
Iran (Ir)	2	0	2
Nepal (Ne)	2	0	2
South Africa (SA)	1	0	1
ICARDA genotypes <sup>2</sup>	2	0	2
Varieties <sup>3</sup>	1	1	2
Total	82	39	121

**Table 1.** Number of accessions by country of origin and by subspecies *macrosperma* (large) and *microsperma* (small).

<sup>1</sup> The abbreviation for the country of origin in brackets. <sup>2</sup> ILL 7202 and ILL 7535. <sup>3</sup> Laird (Canada) and Eston (USA), commercial varieties.

## 2.2. Study Location

The experiment was carried out at the experimental farm of the AGRARIA department located in Gallina (38°10′ N, 15°45′ E, 232 m a.s.l.), Reggio Calabria, southern Italy (Figure 1) in two growing seasons (2016/2017 and 2017/2018).



**Figure 1.** Experimental site. The region (Calabria) where the study was conducted is highlighted in red; the red dot indicates the location of the experimental area.

With reference to the 20-year time span from 1997 to 2016 and the October–June crop season, the average temperatures and rainfall ranged between 10 and 23 °C and 400 and 700 mm, with marked interannual rainfall variability. The agricultural soil is classified as "Typic Haploxeralfs" (USDA) with the following physicochemical characteristics (depth 0–30 cm): 36% clay, 24% silt and 40% sand, pH 7.05, organic matter 1.75%, total N [39] 1.02‰, P [40] 12.24 ppm and K 382.18 ppm, bulk density 1.25 g cm<sup>-3</sup>. Water content at field capacity and wilting point was 30.5 and 17.3% moisture (dry weight), respectively.

#### 2.3. Experimental Design and Crop Management

Field experiments were arranged as a  $11 \times 11$  simple lattice design with two replications. The soil that hosted the two-year experiment was previously cultivated with durum wheat. It was then prepared by summer plowing to a depth of about 30 cm and autumn harrowing, during which 100 kg ha<sup>-1</sup> of P<sub>2</sub>O<sub>5</sub>, and 20 kg ha<sup>-1</sup> of N (as starter) were distributed. Sowing was performed manually on the 4th of December (the 1st year) and the 7th of December (the 2nd year) in 6 m<sup>2</sup> plots (1.5 m × 4 m) with 30 cm row spacings. Planned unit investment was 200 viable seeds m<sup>-2</sup>. This seeding density was specifically chosen as it was considered optimal for the growing area in a previous study [41]. Weeds were controlled by mechanical weeding before the canopy was closed. Harvesting was carried out in June at 75% maturity.

Thermo-pluviometric data were acquired by means of an automatic station (Datalogger CR10X, Campbell Sci. U.K., Loughborough, UK) located on the farm that hosted the trial.

#### 2.4. Bio-Agronomic Data Collection

According to IBPGR-ICARDA (1985) descriptors compared with different sources, the following traits were recorded during the crop cycle: flowering (DASF) and maturity (DASM—days after sowing to maturity), number of plants at harvest (NP), biological yield (BY), grain yield (GY), thousand seed weight (TSW), grain protein content (GP), pods per plant (PP), seeds per pod (SP), grain yield per plant (GYP), plant height (PH), and first pod height (FPH). In detail, NP was measured on three central rows of plot; TSW and GP were determined on an adequate sample of grain: in particular, GP was estimated by using the Kjeldahl method described by [42] in dry seeds (%); PP, SP, and GYP were determined on a ten plants plot sample as yield components; PH and FPH were determined as biometric characters.

#### 2.5. Statistical Analysis

RStudio (R version 4.1.0) was used for data analysis and visualization [43]. The collected bio-agronomic data were subjected to analysis of variance (Table 2) according to the selected experimental design ( $11 \times 11$  simple square lattice design) using the *PBIB.test* (Partially Balanced Incomplete Block Design) function available in the agricolae package (v. 1.3.5) [44].

Source of Variation	Degree of Freedo	Sum of Squares	Mean Squares	"F"	
Replication	R-1	1	SSr	MSr	
Block within replication	r(k-1)	20	SSb	MSb	MSb/MSe
Treatment	$k^2 - 1$	120	SSt	MSt	MSt/MSe
Residual	(rk - k - 1)(k - 1)	100	SSe	MSe	
Total	$rk^{2} - 1$	241			

Table 2. Analysis of variance of an experimental design "Simple lattice" with two replications.

Mean values of experimental treatments and interactions were compared using the Least Significant Difference (LSD) test for  $p \le 0.05$ .

The relationships among phenological, biometric, production, and quality traits were studied by Pearson's correlation analysis. The R stats package (v. 4.1.0) [45] was used to compute correlations between all considered variables, and the ggcorrplot package (v. 0.1.3) [46] was used to visualize a correlation matrix using ggplot2 (v. 3.4.2) [47].

Principal component analysis (PCA) was successively carried out using the FactoMineR package (v. 2.4) [48]. The factoextra package (v. 1.0.7) [49] was used to extract and visualize the PCA outputs. Other additional R packages were utilized for data manipulation (dplyr v. 1.1.2 [50]) and to complement ggplot2 for displaying and arranging images (cowplot v. 1.1.1 [51], ggforce v. 0.0.3 [52], and patchwork v. 1.1.1 [53]).

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## 3. Results and Discussion

## 3.1. *Climatic Data*

The thermo-pluviometric trend during the two years is considered typical of the Mediterranean weather and is shown in Table 3 along with the 20-year average (1997–2016).

**Table 3.** Total monthly rainfall and mean air temperatures (maximum and minimum) during the two growing seasons (2016/2017 and 2017/2018) and the long-term average (1997–2016).

	Total Monthly Rainfall (mm)			Temper	ature Maxim	um (°C)	Temperature Minimum (°C)			
Month	2016/2017	2017/2018	1997/2016	2016/2017	2017/2018	1997/2016	2016/2017	2017/2018	1997/2016	
October	35.2	23.6	91.8	24.7	23.4	22.9	18.8	17.5	16.2	
November	80.4	129.2	86.3	20.6	18.7	18.1	15.1	13.3	12.3	
December	27.0	56.4	88.5	16.1	15.1	14.3	11.3	10.0	9.4	
January	105.0	40.6	84.6	12.8	16.3	13.1	7.7	11.4	7.1	
February	44.0	138.0	56.4	16.3	14.2	13.3	10.7	8.9	6.7	
March	28.4	77.8	59.8	17.4	18.3	16.6	11.8	11.8	8.2	
April	10.0	3.4	40.0	19.9	21.5	18.3	13.2	14.5	10.0	
May	92.6	11.0	21.7	23.8	23.9	23.3	16.9	17.2	14.5	
June	6.2	129.2	6.3	28.9	27.1	28.1	21.7	20.6	18.9	

The total amount of rainfall and its distribution were found to be highly varying during the October-June period. Compared to the 20-year average (535 mm) for the experimental site, 429 and 609 mm fell in the first and second growing season, respectively. In detail, 116 and 153 mm fell in pre-sowing period, 204 and 312 mm during the sowing-flowering phase, and 102 and 14 mm in the flowering-maturity phase in the first and the second year, respectively (Figure S1a,b). It should be noted that peaks of 129 mm were registered in June 2018, when harvest had already taken place. For this reason, the lentil did not benefit from this water availability.

The average monthly maximum temperatures in the October–June period were 20.1 and 19.8 °C in 2016/2017 and 2017/2018, respectively. The same average monthly minimum temperature of 14.0 °C was recorded in both years. January 2017 and February 2018 were the coldest months, with minimum temperatures averaging below 10 °C (Table 3). The air temperature gradually increased from January 2017 and March 2018 reaching maximum peaks of 31.4 and 27.3 °C in the third and second ten-day period of June 2017 and 2018, respectively (Figure S1a,b).

#### 3.2. Bio-Agronomic Behavior

The crop productive behavior showed the general high yield potential of lentil over the two-year trial, although it was influenced by substantial climatic variations. The productive threshold of 2 t ha<sup>-1</sup> in semi-arid environments can be considered a more than satisfactory result. The average NP was similar in the two years (188 and 187 plants m<sup>-2</sup>, respectively), slightly lower than the planned investment of 200 plants m<sup>-2</sup>.

The analysis of variance revealed high significance for all studied traits (Table 4), according to the used experimental model.

However, the different rainfall distribution, in particular the low rainfall recorded during the flowering-maturity period in 2018 (Figure S1b), affected the productive determinism and penalized crop vegetative development, as also reported by [54].

The crop biological biomass at harvest (BY, straw + grain) was quite high in the first  $(6.91 \text{ t ha}^{-1})$  and the second year  $(6.57 \text{ t ha}^{-1})$ . The effects of climatic trends on crop bioagronomic behavior were also evidenced through phenological characters, whose range of variation (expressed as annual average data) was quite wide. Particularly, the crop showed the greatest earliness in the second year (116 and 177 days for DASF and DASM, respectively). On the other hand, only an early maturity stage was observed in the first year allowing the crop to achieve the best productive result. In particular, the rainfall that

occurred in the second ten days of May favored seed filling, although resulting in a reduced flowering-maturity period.

**Table 4.** Analysis of variance (variances and test F) for bio-agronomic and qualitative traits of 121 lentil genotypes in the first (2016/2017) and second year (2017/2018).

2016/2017													
Source	DF	N	Р	G	Y	В	Y	DA	SF	DA	SM	PI	Н
		MS	F	MS	F	MS	F	MS	F	MS	F	MS	F
Replication	1	158.74	ns	0.035	ns	1.901	*	1.491	ns	155.5	*	12.5	*
Block	20	73.27	*	0.016	ns	0.205	ns	3.042	ns	20.02	*	4.94	*
Accession	120	88.00	**	0.634	***	1.106	***	80.05	***	110.01	***	52.90	***
Residual	100	42.32		0.019		0.142		2.18		11.30		2.44	
Mean—CV		187.9–	-3.46	2.31—	-5.94	6.91–	-5.44	118.9—	-1.241	174.4-	-1.93	43.9-	-3.56
Source	GL	FP	Н	Pl	Р	S	Р	GY	ſΡ	TS	W	G	Р
		MS	F	MS	F	MS	F	MS	F	MS	F	MS	F
Replication	1	5.355	*	16.06	**	0.002	ns	0.021	**	0.102	ns	1.083	*
Block	20	2.156	*	1.991	ns	0.001	ns	0.003	ns	0.059	*	0.152	ns
Accession	120	22.894	***	236.8	***	0.132	***	0.188	***	4.001	***	3.697	***
Residual	100	1.063		1.431		0.001		0.002		0.0295		0.154	
Mean—CV		28.9—	-3.57	31.9—	-3.75	1.23-	-1.66	1.06-	-3.72	37.5–	-4.58	27.7—	-1.38
						2017/2018	8						
Source	DF	N	Р	G	Y	В	Y	DA	SF	DAS	SM	PI	Н
		MS	F	MS	F	MS	F	MS	F	MS	F	MS	F
Replication	1	54.65	ns	0.235	ns	0.353	ns	23.87	*	3.719	ns	1.981	ns
Block	20	35.92	*	0.085	ns	0.184	ns	2.54	ns	12.21	*	2.497	ns
Accession	120	128.42	***	0.522	***	1.575	***	90.98	***	36.22	***	70.84	***
Residual	100	15.25		0.053		0.124		2.44		4.960		1.586	
Mean—CV		186.7–	-2.09	2.22—	-7.64	6.57—	-4.98	116.2-	-1.35	176.8-	-1.26	42.6-	-2.96
Source	GL	FP	Н	P	Р	S	Р	G١	ſΡ	TS	W	G	Р
		MS	F	MS	F	MS	F	MS	F	MS	F	MS	F
Replication	1	1.309	ns	18.583	*	0.001	ns	0.018	*	0.179	ns	1.118	*
Block	20	1.157	*	1.838	***	0.001	ns	0.002	ns	0.043	ns	0.153	ns
Accession	120	72.84	***	206.20	ns	0.134	***	0.159	***	4.336	***	3.730	***
Residual	100	0.809		1.717		0.001		0.001		0.021		0.156	
Mean—CV		28.8—	-3.15	29.7—	-4.41	1.21-	-1.69	0.98-	-3.72	36.3—	-4.01	27.9—	-1.41

Abbreviations: ns = not significant; \* = significant at 0.05 significance level; \*\* = significant at 0.01 significance level; CV = coefficient of variance; DF = degrees of freedom; NP = plant density (no m<sup>-2</sup>); GY = grain yield (t ha<sup>-1</sup>); BY = biological yield (t ha<sup>-1</sup>); DASF = flowering, days after sowing (dd); DASM = maturity, days after sowing (dd); PH = plant height (cm); FPH = First pod height (cm); PP = pods per plant (no); SP = seeds per pod (no); GYP = grain yield per plant (g d.m.); TSW = thousand seed weight (g); GP = grain protein content (% d.m.).

The importance of the earliness character in lentil, as "escape" mechanism from stress in the final phase of its biological cycle, was also confirmed by the close negative and significant relationship (r = -0.61 \*\*\*) between GY and DASF (Figure 2). The early flowering time and a shorter vegetative phase can be very important strategies for the production of autumn–spring cycle crops in the Mediterranean environment increasingly in conditions of terminal drought. This has been demonstrated for many herbaceous crops, in particular for wheat and some grain legumes [36,55,56].

Particularly in lentil, yield reduction can range from 24%, if stress occurs during the reproductive phase, up to 70% during the pod filling phase [57,58]. Probably the drought, together with the high temperatures occurring in the final stage of the biological cycle in 2018, negatively affected the crop's production behavior.



**Figure 2.** Correlation matrix and level of significance for 11 bio-agronomic traits. Abbreviations:  $GY = Grain yield (t ha^{-1})$ ;  $BY = biological yield (t ha^{-1})$ ; DASF = flowering, days after sowing (dd); DASM = maturity, days after sowing (dd); PH = plant height (cm); FPH = First pod height (cm); PP = pods per plant (no); SP = seeds per pod (g d.m.); GYP = grain yield per plant (g d.m.); TSW = thousand seed weight (g); GP = grain protein content (% d.m.). Insignificant correlation coefficients are crossed out (non-significant at <math>p < 0.05).

The identification of early genotypes is crucial in the Mediterranean environment, which is often characterized by drought and heat stresses during the grain filling stage of legumes; moreover, these adverse environmental conditions have become more frequent and intense as a consequence of climate change. Studies conducted in Morocco have shown the negative effect of both heat and drought stresses on the bio-agronomic behavior and nutritional quality of lentil as a result of the consequent shortening of phenological stages [59,60]. These effects have also been previously confirmed by studies conducted on a variety of legumes such as lentil [61], fava bean [62], chickpea [63], and common bean [64,65].

Moreover, the flowering character was found to be highly correlated with first pod height (r = 0.46 \*\*\*); this result would suggest that early flowering and lower height of the first fertile node are correlated with genetic structure and that this occurs with high frequency in the Mediterranean Basin, as shown by [66].

The height of both plant and first fertile pod is negatively correlated to grain yield (r = -0.31 \*\*, r = -0.40), in agreement with a previous study [36].

Besides plant height (+1.3 cm), the better environmental conditions in the first year were evidenced by the numerical values of production components: PP (+2.2), SP (+0.02), and GYP (+0.1 g d.m.). In particular, PP appears to be a key character for lentil production determinism: in fact, it appears to be positively correlated with SP (r = 0.55 \*\*\*) and GYP (r = 0.65) and negatively correlated with TSW (r = -0.58 \*\*\*). Similar results were found by [67] on a collection of Spanish lentil (27 accessions) and also for chickpea and common bean [33,68].

The lowest TSW value was recorded in the year with the highest yield (2.31 t  $ha^{-1}$ , 2016–2017). This result was likely due to the presence of a greater number of seeds that

allowed an optimal distribution of assimilates in the sinks in the presence of adequate water availability into the soil. However, TSW was found to be negatively correlated with GY (-0.26 \*\*), in contrast to the report of other authors [69,70].

Grain protein content of seed was found to be quite high and did not differ significantly between the two years (27.9 and 27.7% d.m., respectively).

Table 5 shows the average agronomic performances of the 22 better genotypes (16 *microsperma* and 6 *macrosperma*) in comparison with the two commercial varieties and the ICARDA genotypes over the two years. The above-mentioned accessions placed at the top of the production ranking during the two-year trial, demonstrating high production stability.

**Table 5.** Average performances over the two years of the best *microsperma* and *macrosperma* genotypes for grain yield and other agronomic traits comparing commercial varieties and ICARDA genotypes (indicated in bold).

Origin	AC	ssp	NP (no m <sup>-2</sup> )	GY <sup>1</sup> (t ha <sup>-1</sup> )	BY (t ha <sup>-1</sup> )	DASF (dd)	DASM (dd)	PH (cm)	FPH (cm)	PP (no)	SP (no)	GYP (g d.m.)	TSW (g)	GP (g d.m.)
Algeria	106365	S	197	3.50	8.07	112	169	39.3	24.3	36.0	1.79	1.54	26.1	27.9
Tunisia	106692	S	196	3.48	7.26	111	170	36.0	22.4	61.1	1.75	1.72	21.2	27.4
Italy	116218	S	196	3.23	8.26	113	174	47.4	28.1	25.8	1.66	1.22	28.7	28.3
Tunisia	106748	S	197	3.19	6.93	112	170	37.1	23.0	47.1	1.78	1.39	20.8	28.3
Tunisia	106682	S	192	3.18	7.00	112	171	35.5	22.3	48.2	1.78	1.42	20.4	28.5
Pakistan	107386	S	198	3.16	7.23	112	171	38.5	24.5	44.8	1.83	1.30	20.6	29.0
Algeria	107516	S	194	3.15	6.70	111	170	39.1	26.2	49.6	1.66	1.44	22.5	30.1
Cyprus	112375	S	192	3.11	8.00	113	173	44.5	28.7	37.1	1.10	1.60	42.4	28.4
Tunisia	106746	S	197	3.07	6.94	112	170	36.9	22.0	32.6	1.79	1.13	20.8	28.3
Egypt	111820	S	197	2.99	7.53	112	171	47.1	28.5	48.3	1.56	1.44	25.4	29.0
Italy	116221	S	193	2.97	7.65	112	170	47.5	28.8	33.2	1.30	0.97	25.7	28.6
Egypt	111819	S	198	2.97	7.46	112	171	44.8	28.1	51.7	1.41	1.45	24.6	29.7
Greece	107585	S	189	2.92	7.45	113	176	44.5	29.3	26.4	1.06	0.96	39.2	26.5
Morocco	112163	S	198	2.87	7.79	112	176	45.7	31.7	27.2	0.98	1.06	39.9	26.1
Egypt	110830	S	194	2.81	7.15	112	170	40.7	26.8	39.4	1.27	1.02	25.6	26.9
Italy	116222	S	195	2.73	7.19	112	173	37.7	22.8	24.4	1.40	1.01	24.3	30.3
ICARDA	ILL 7202	S	198	3.09	7.12	112	174	40.2	25.4	36.3	1.20	1.48	39.8	30.0
ICARDA	ILL 7535	S	192	2.86	6.45	112	177	41.0	25.6	29.0	1.09	1.26	38.4	30.2
USA	Eston	S	188	1.79	6.24	125	185	44.1	24.3	34.5	1.10	1.06	30.3	29.2
Cyprus	112366	L	192	2.96	7.63	113	175	44.6	28.9	27.2	1.11	1.43	50.9	27.9
Morocco	112114	L	192	2.96	7.52	112	175	44.9	29.0	28.1	1.30	1.23	40.3	28.1
Morocco	112112	L	190	2.82	7.13	113	174	44.4	28.0	28.9	1.09	1.17	40.5	28.1
Cyprus	112374	L	190	2.77	7.29	113	173	46.6	30.5	24.5	1.13	1.02	43.7	27.8
Cyprus	112377	L	188	2.74	6.79	112	171	44.9	27.9	40.5	1.20	1.50	43.6	28.7
Algeria	106402	L	189	2.72	8.06	112	173	48.2	36.5	23.6	1.11	1.54	70.6	28.0
Canada	Laird	L	188	1.94	8.32	128	184	57.9	33.0	29.8	0.96	1.07	66.3	28.6

 $^{1}$  Within types, the ranking was made by decreasing average yield.

Limited to the two-year trial, yield and stability of productions are important factors to be considered in order to help plant breeders make accurate choices in the selection of genotypes suited to specific environment [71].

In particular, the *micro* accessions 106365 (Algeria), 106692 (Tunisia), and 116218 (Italy) were the most productive (>3.20 t ha<sup>-1</sup>), while the commercial variety Eston was clearly below all the other accessions considered, with a grain yield of 1.79 t ha<sup>-1</sup>. The two ICARDA genotypes showed a good yield (2.98 t ha<sup>-1</sup>, on average), while the commercial variety Laird was the least productive (1.94 t ha<sup>-1</sup>) among the *macro* subspecies. Overall, the most productive *macro* accessions were 112366 (Cyprus), 112114, and 112112 (Morocco), showing average yields greater than 2.80 t ha<sup>-1</sup>.

## 3.3. The Bio-Agronomic Behaviour of Accessions from Different Countries

The analysis of the average productive response between the two years showed a rather wide range of values (Table S2a,b).

The highest grain yields were recorded for accessions from Egypt (no. 10), Cyprus (no. 10), and Nepal (no. 2), always exceeding the threshold of 2.50 t  $ha^{-1}$ .

The productive behavior of landraces from Iran (no. 2), Algeria (no. 10), Tunisia (no. 13), and Morocco (no. 10) was also remarkable: in this case, yields resulted above the average value for the two-year period ( $2.26 \text{ t ha}^{-1}$ ). The 24 Italian accessions showed an average productivity of 1.99 t ha<sup>-1</sup> in the two-year. A lower and identical yield ( $1.89 \text{ t ha}^{-1}$ ) was observed for Spain (six accessions) and Albania (three accessions). These were followed by the only accession from South Africa ( $1.60 \text{ t ha}^{-1}$ ), which was found to be the least productive.

Net relative inferiority in yield was observed for all accessions when compared to the two ICARDA lines (2.97 t  $ha^{-1}$  the two-year average). In contrast, about 90% of the assayed material yielded more than the two commercial varieties on average (1.86 t  $ha^{-1}$ ).

Analysis of the annual results revealed a considerable interaction between year and geographic origin (Figure 3a), however confirming the consistent yield superiority of accessions from Egypt, Cyprus, Algeria, Nepal, and Tunisia, whose seed yields always exceeded the field average with values even higher than 20% (Egypt).



**Figure 3.** Barplots showing the mean values of four traits for each country of origin and year (blue for 2016/2017 and orange for 2017/2018): (a) GY—Grain yield; (b) PP—Pods per plant; (c) SP—Seeds per pods; (d) GYP—Grain yield per plant. The dashed line within each subplot indicates the two-year average. The error bars indicate the standard deviation of the mean.

Much more variability was clearly evident in the case of accessions from Pakistan and Iran. Yield values over the general average were observed only in the first year. Accessions from Italy yielded 10–15% lower than the field average. Despite that, they reached a good production level.

Overall, accessions from Algeria, Libya, Tunisia, Italy, and Morocco showed a high variability, which was amplified by the presence of *micro* and *macro* genotypes within each group.

Studies conducted in the Mediterranean basin by [72] on the genetic diversity of accessions from different countries (some of which are present in our work) showed that Moroccan landraces, followed by those of Italian and Greek origin, presented higher diversity and differentiation among individuals within the landraces. This evidence could be exploited to select genotypes with better adaptation to biotic and abiotic stresses, which are increasingly frequent in the Mediterranean environment.

High genetic and phenotypic variability relative to Italian accessions, and Sicilian in particular, has also been demonstrated in a study conducted by [73].

The production behavior of the testers differed markedly, even though with little interannual variability: ICARDA lines achieved a production level higher than the field average by about 30%; on the contrary, the commercial varieties yielded below the same average in both years.

On average, all lentil accessions showed a fairly high range of variability regarding total biomass production, varying from 7.23 t ha<sup>-1</sup> (Algeria, 10 accessions) to 4.96 t ha<sup>-1</sup> (Nepal, 3 accessions). The 24 Italian accessions were among the most productive (7.06 t ha<sup>-1</sup>), outperforming commercial varieties (7.03 t ha<sup>-1</sup>) and ICARDA lines (6.54 t ha<sup>-1</sup>). The year factor again exerted a high impact also on this character, especially for accessions from Spain, Morocco, and for ICARDA.

Thousand-seed weight presented quite a wide range of values due to different numerosities of *micro* and *macro* types and their ratio within individual country. It varied from 19.8 g for the only *micro* accession of South Africa to 49.1 g for Spanish accessions (four out of six are *macro* types). Accessions from Algeria and Cyprus ranked among the best having a greater number of large seeds (Table 1). Italy ranked among the medium-to-high weight countries, despite the double number of small seed accessions (Table 1). The gap between the commercial varieties and ICARDA lines, greater than 10 g among the two-year averages, is to be considered an effect of the different composition and lesser number of the two groups: in fact, ICARDA lines are *micro* genotypes while the two commercial varieties include both types. The higher value is attributed to a higher percentage incidence of *micro* versus *macro* accessions.

Flowering time (days after sowing) showed wide variability among the groups with a difference of 22 days between the earliest and the late accession, on average. Four countries (Ethiopia, Nepal, Egypt, and Cyprus) showed flowering time shorter than 115 days after sowing; good earliness was also found in ICARDA lines (112 days). Compared to the material assayed, the commercial varieties resulted considerably late (127 days). Their flowering time was slightly earlier than Albania (129 days). Italian accessions showed a late average flowering time (122 days).

The ranking of countries for maturity time was quite similar to flowering time, but with a smaller range of variability (16 days). Commercial varieties confirmed their lateness (184 days, the highest value among the assayed groups). However, they presented a rather long flowering-maturity period.

The average plant height in the two-year was 43.2 cm, with a range of variability ranging from 30.7 (Nepal) to 47.2 cm (Morocco). The commercial varieties presented the highest values in both growing seasons: 49.0 cm in 2017 and 53.0 cm in 2018.

The average height of the first pod insertion was 28.7 cm, with a range of variability between 20.5 (Nepal) and 32.3 cm (Morocco).

Yield components are influenced by agronomic management, genotype, and environment and contribute to explain yield production. Egli (2017) suggested using two components to simplify the analysis [74]. We decided to use the three standard components: pods per plant, seeds per pod, and grain yield per plant.

Yield components showed rather a wide range of variability among the different countries, also due to the different *micro/macro* ratio within them (Figure 3). The average number of pods per plant varied from 23.3 (Libya) to 43.9 (Egypt), where all accessions are *micro* types. Number of pods per plant above 40 was also shown by Albania (three *micro* accessions). Italy was well below the average among the groups, with 24.5 pods per plant. The commercial varieties and the ICARDA lines showed, on average, more pods per plant than the two-year average (32.2 and 32.6, respectively).

Seeds number per pod also had a significant influence on crop productive determinism by showing significant differences among accessions of different geographical origins. The highest values were recorded for Nepal and Pakistan (1.69 and 1.48, respectively); values above 1.30 seeds per pod, on average, were recorded for accessions from Egypt, Tunisia, Iran, and Algeria. In contrast, Ethiopian accessions averaged the lowest value (0.99), slightly lower than commercial varieties (1.03). Grain yield per plant ranged from 0.54 g d.m. for South Africa to 1.42 g d.m. for Iran; values above 1.10 g were recorded for Algeria (1.31 g d.m.), Nepal (1.31 g d.m.), Cyprus (1.20 g d.m.), Tunisia (1.14 g d.m.), and Egypt (1.12 g d.m.). High grain yield per plant was shown by ICARDA improved genotypes (1.37 g d.m.), while commercial varieties presented a value of just over 1 g d.m.

Grain protein content was overall high (27.8% d.m.) with a fairly narrow range of variability (4%). Also noteworthy for this character was the behavior of improved ICARDA genotypes, which showed the highest value (30.1% d.m.).

#### 3.4. Bio-Agronomic Behaviour of Micro and Macro Types

The evaluation of lentil accessions was also carried out considering small-seeded separately from large-seeded types on country origin basis (Table S3a–c). Analysis of the assayed material made it possible to assess the differences in bio-agronomic behavior of the two types and their contribution by country of origin.

The unit investment at harvest was closer to that planned in *micro* types  $(-11 \text{ plants m}^{-2})$  than in *macro* types  $(-18 \text{ plants m}^{-2})$ , on average.

The average yield of *micro* types was higher than that of *macro* types in both years  $(+0.31 \text{ and } +0.41 \text{ t ha}^{-1}, \text{ respectively})$ , exceeding the 2 t ha<sup>-1</sup> threshold, on average.

The number of pods per plant proved to be the most important agronomic trait in determining the grain yield. It was found to be significantly higher in *microsperma*, on average (+10.7 *micro* vs. *macro*). These results confirm what has already been shown in previous studies conducted on a smaller number of genotypes in Calabria region [36,75].

In addition, *micro* types accessions grouped by country of origin were on average more productive than the commercial variety Eston, with the exception of the South African accession in the second year.

Among small-seeded accessions from different countries, productive response (as an average value of trial period) recorded a range between 1.60 t ha<sup>-1</sup> for South Africa (one accession) and 2.80 t ha<sup>-1</sup> for Tunisia (seven accessions). The latter presented yields always higher than 2 t ha<sup>-1</sup> in the two crop seasons, exceeding the average field yield for *micro* by 19%. High productivity was also noted in Cyprus (4 accessions), Algeria (4 accessions), Egypt (10 accessions), and Nepal (2 accessions), with yields exceeding 2.5 t ha<sup>-1</sup>.

*Micro* types accessions of all countries revealed a lower productivity than improved material in both years. The *micro* types of Italian origin presented an average productivity (2.08 t ha<sup>-1</sup>) slightly lower than the average of all micro types. Italian accessions were always inferior to the general accessions average (-10%). Compared to ICARDA genotypes, the difference reached 30%.

Among large-seeded accessions and considering the average for the two-year period, yields of different countries (eight) presented a range of values from 1.59 for Libya (one

accessions) to 2.35 t ha<sup>-1</sup> for Cyprus (six accessions). The productivity of Algerian and Moroccan *macros* was relatively high (>2.00 t ha<sup>-1</sup>).

With reference to large-seeded only, the Italian group was below field average in both years. Laird yield was just below (-0.18 t ha<sup>-1</sup>, 2017) and above (+0.10 t ha<sup>-1</sup>, 2018) the average of the large-seeded types.

Total biomass production, in two-year average, was lesser in *micro* than *macro* types at 6.84 and 7.31 t ha<sup>-1</sup>, respectively. Among *micro*, on the other hand, the differences between the groups of different geographical origins were more pronounced with a range of variability found between 7.64 in Greece and 5.21 t ha<sup>-1</sup> in Nepal. Among *macro*, the highest value in both years was recorded by Laird (8.32 t ha<sup>-1</sup>).

Thousand-seed weight was more differentiated in small-seeded types. In this case, the range of variability was between 19.8 (South Africa) and 39.9 g (Cyprus), on average; ICARDA genotypes (39.1 g) and Morocco (38.5 g) were above 35.0 g; Italy (32.2 g) was just above Eston variety (30.3 g). Among the lowest values were Nepal, Pakistan, and Tunisia with values less than 22 g.

In large-seeded types, Morocco and Greece showed the lowest weights (44.6 and 47.0 g, respectively), while Spain and Algeria exceeded 60 g; the remaining countries, including Italy, showed values ranging from 51.6 (Cyprus) to 57.2 g (Tunisia). The commercial variety Laird presented the highest value of 66.3 g.

Flowering time revealed a significantly wider range of variability among countries in *micro* (22 days versus 14 days in *macro*). Within the *micro*, Ethiopia showed the earliest flowering time as the average of the two-year period (107 days after sowing); in contrast, Albania presented the highest value among the assayed groups (129 days). Moreover, among the earliest groups, in addition to the ICARDA genotypes (112 days), the two accessions of Nepal (112 days) were identified, followed by the accessions of Egypt and Cyprus (113 days); good earliness also showed the accessions of Tunisia and Libya (114 days).

In *macro*, the differences observed among countries, in flowering time, were generally smaller than in *micro*; the group with the highest earliness was Cyprus (114 gg), the remaining seven countries showed flowering time varying from 117 gg (Greece and Morocco) to 122 gg (Spain and Italy). The commercial variety, Laird, (128 gg) was the most delayed overall.

The differences recorded at flowering time between and within the countries were found to be smaller at maturation (14 gg of *micro* vs. 10 gg of *macro* types).

Plants height, as well as the insertion of the first fertile pod, on the whole, was greater in *macro* types (+5.4 cm and +4.4 cm, on average, respectively); however, it should be emphasized especially for this last character how many countries presented, on average, higher values than both commercial varieties and improved genotypes. These are nonnegligible details that should be considered in genetic improvement programs aimed at the realization of genotypes suitable for harvest mechanization.

#### 3.5. Multivariate Analysis for Whole Collection and for the Two Subspecies Micro and Macro

Principal component analysis (PCA) was used to extract relevant information from our dataset and highlight similarities and differences between countries and *micro* and *macro* subspecies. A first PCA was conducted on the whole dataset after grouping the variables of interest by genotype, origin, and subspecies. The mean values were then computed for all genotypes over the two years and the four repetition. In addition, two variables (plant density and biological yield) were not further considered to simplify the analysis. The data were standardized prior to PCA, as they were measured on different scales. The first three principal components accounted collectively for 77.7% of the total variability in the data (Table S4).

The first principal component (PC1) explained 46.2% of the variability and was mainly associated with both yield and its components (DASM, SP, PP, TSW), and biometric traits (FPH and PH) (Table 6).

PC-1	PC-2	PC-3	PC-4	PC-5
-0.778	-0.007	0.488	-0.053	0.084
0.637	0.559	-0.335	0.305	-0.062
0.773	0.378	-0.163	0.223	-0.199
0.757	0.319	0.429	-0.104	0.283
0.822	0.239	0.337	-0.015	0.329
-0.697	0.528	0.059	0.165	-0.139
-0.731	0.380	-0.159	0.183	0.358
-0.533	0.394	0.638	0.278	-0.192
0.676	-0.280	0.560	0.044	-0.260
-0.070	0.669	-0.044	-0.705	-0.176
	PC-1 -0.778 0.637 0.773 0.757 0.822 -0.697 -0.731 -0.533 0.676 -0.070	PC-1         PC-2           -0.778         -0.007           0.637         0.559           0.773         0.378           0.757         0.319           0.822         0.239           -0.697         0.528           -0.731         0.380           -0.533         0.394           0.676         -0.280           -0.070         0.669	$\begin{array}{ c c c c c c c c c c c c c c c c c c c$	$\begin{array}{ c c c c c c c c } \hline PC-1 & PC-2 & PC-3 & PC-4 \\ \hline -0.778 & -0.007 & 0.488 & -0.053 \\ 0.637 & 0.559 & -0.335 & 0.305 \\ 0.773 & 0.378 & -0.163 & 0.223 \\ 0.757 & 0.319 & 0.429 & -0.104 \\ 0.822 & 0.239 & 0.337 & -0.015 \\ -0.697 & 0.528 & 0.059 & 0.165 \\ -0.731 & 0.380 & -0.159 & 0.183 \\ -0.533 & 0.394 & 0.638 & 0.278 \\ 0.676 & -0.280 & 0.560 & 0.044 \\ -0.070 & 0.669 & -0.044 & -0.705 \\ \hline \end{array}$

Table 6. Correlation matrix with PCA components of whole collection.

The second principal component (PC2) was responsible for 17.2% of the variability. It was associated with grain protein content and flowering time (GP and DASF). The third principal component (PC3) contributed to 14.3% of the total variability and was mainly associated with GYP and TSW. Overall, Figure 4 highlights that DASF, DASM, PH, and FPH are located close to each other.



**Figure 4.** Biplot of principal component analysis of bio-agronomic traits in 121 lentil genotypes (dots and numbers in the biplot). The different shapes of the dots differentiate the *microsperma* type from the *macrosperma* type. The yellow circle highlights the cluster of the Ethiopian genotypes.

The same pattern can be observed for GY, SP, PP, and GYP. These variables are indeed correlated to each other within the distinct groups they shape, while TSW and GP seem to be less correlated between them and the other variables.

The graphical representation of the relationship between genotypes and variables clearly shows the different distributions of *micro* and *macro* types, with most of the latter clustered on the positive side of the PC1. It can be clearly seen that these genotypes tend to gradually localize toward the variable TSW. Indeed, the genotypes belonging to the *macro* category were characterized by a high value of TSW. On the contrary, the *micro* genotypes were mainly characterized by higher values for GY, SP, PP, and GP. Their distribution appears to be quite scattered, which can be attributed to the larger number of genotypes and country of origin considered if compared to the *macro* group. In particular, all Ethiopian genotypes form a clearly distinct cluster on the negative side of PC2, at the lowest values. Studies conducted by [76] on genetic variability of a lentil collection (some accessions are also evaluated in this work) did not show a clear grouping pattern in relation to geographic origin or for belonging to *macro* or *micro* types, with the exception of accessions (only one in this work) from Ethiopia.

To deepen the extent of the contribution of *micro* and *macro* types on the same variables, PCA was also conducted separately for the two subspecies (Figure 5).



**Figure 5.** Biplot of principal component analysis of bio-agronomic traits: (**a**) 82 lentil genotypes (*microsperma*); (**b**) 39 lentil genotypes (*macrosperma*). The different colors and shapes identify the various countries of origin, as depicted in the legend.

In this case, data have been filtered for subspecies, then grouped by genotype and origin. The first three principal components for *micro* and *macro* subspecies contributed 76.9% and 72.1% of the total variability, respectively (Tables S5 and S6). Specifically for small-seeded accessions, PC1 explained 41.7% of the variability and was associated (Table 7) with grain yield and its components (DASM, SP, DASF, GYP, and PP), and biometric characters (FPH and PH). PC2 explained 20.7% of the variability. It was associated with PP, DASF, and GP. The PC3 contributed 14.5% and was exclusively associated with TSW and GY.

PC-1	PC-2	PC-3	PC-4	PC-5
-0.737	0.034	0.571	-0.018	0.158
0.643	0.574	-0.352	0.260	-0.060
0.759	0.394	-0.087	0.305	-0.159
0.750	0.320	0.410	-0.120	0.314
0.835	0.258	0.274	-0.037	0.323
-0.571	0.626	0.110	0.155	-0.237
-0.642	0.508	-0.227	0.013	0.422
-0.589	0.548	0.457	0.282	-0.108
0.513	-0.367	0.675	0.173	-0.209
0.164	0.582	0.101	-0.714	-0.301
	PC-1 -0.737 0.643 0.759 0.750 0.835 -0.571 -0.642 -0.589 0.513 0.164	PC-1         PC-2           -0.737         0.034           0.643         0.574           0.759         0.394           0.750         0.320           0.835         0.258           -0.571         0.626           -0.642         0.508           -0.589         0.548           0.513         -0.367           0.164         0.582	PC-1PC-2PC-3-0.7370.0340.5710.6430.574-0.3520.7590.394-0.0870.7500.3200.4100.8350.2580.274-0.5710.6260.110-0.6420.508-0.227-0.5890.5480.4570.513-0.3670.6750.1640.5820.101	PC-1PC-2PC-3PC-4-0.7370.0340.571-0.0180.6430.574-0.3520.2600.7590.394-0.0870.3050.7500.3200.410-0.1200.8350.2580.274-0.037-0.5710.6260.1100.155-0.6420.508-0.2270.013-0.5890.5480.4570.2820.513-0.3670.6750.1730.1640.5820.101-0.714

Table 7. Correlation matrix with PCA components for ssp microsperma.

On the other hand, regarding *macro* accessions, PC1, which explained 40.0% of the variability, was associated (Table 8) mainly with grain yield, phenological traits, and yield components (DASF, GY, DASM, GYP, PP, and SP). PC2, responsible for 19.1% of the variability, was associated with plant height and first pod insertion height (FPH and PH), while PC3 contributed 13.0% and was associated with TSW.

**Table 8.** Correlation matrix with PCA components for *ssp macrosperma*.

Variable	PC-1	PC-2	PC-3	PC-4	PC-5
GY	0.847	0.138	-0.231	0.031	0.082
DASF	-0.886	0.078	0.140	0.089	0.031
DASM	-0.791	-0.197	0.355	0.093	-0.036
PH	-0.278	0.802	0.425	0.079	0.072
FPH	-0.365	0.812	0.073	-0.264	0.181
PP	0.639	0.271	0.472	0.204	-0.436
SP	0.606	-0.046	0.257	-0.511	0.407
GYP	0.790	0.499	-0.103	0.125	-0.106
TSW	-0.314	0.422	-0.673	0.383	0.160
GP	0.399	-0.227	0.398	0.635	0.456

## 4. Conclusions

Based on the results of this study, the ten accessions from Egypt (all belonging to the *micro* type) were the most productive. They expressed a high yield level of 2.69 t ha<sup>-1</sup>, as average for the two-year period (+16% compared to the average of all accessions assayed), accompanied by small fluctuations in annual values and low variability among accessions. The Egyptian group also stood out for flowering and maturity earliness.

Accessions from Cyprus ranked in second position regarding yield, with an average value of 2.53 t ha<sup>-1</sup>, although influenced by a clear variation through the years. The four *micro* accessions presented a higher average productivity than *macro* (+0.4 t ha<sup>-1</sup>), and both groups still showed a yield level above the field average (+20%). All Cypriot accessions were also distinguished by earliness (113 days).

Earliness can undoubtedly be considered one of the causes of the higher yield of the *micro* than the *macro* in some countries.

The *micro* accessions from Nepal and Iran presented a productivity (2.56 and 2.49 t  $ha^{-1}$ , respectively) similar to the North African group, with yields well above the field average in both years.

The yield of the 24 Italian accessions was below the field average (-10%), with a mean value of 1.99 t ha<sup>-1</sup> for the two-year period. A late flowering time (122 days) was also observed for both *macrosperma* and *microsperma* types.

The analysis of yield components revealed a different productive determinism within the *micro* and *macro* types. In fact, the first one presented a significantly higher number of pods per plant and seeds per pod (+10.8 and +0.26, respectively) on average, although with

considerable variability between countries of origin. The grain protein content was also higher in micro types (+0.5%) than in macro.

Lentil is of potential interest to Southern Italy, and its cultivation must nonetheless face the increasingly pressing issue of environmental change. Research must address this ongoing challenge by suggesting sustainable agronomic practices and genotypes that can thrive in specific environments. Our results conclude that there is considerable genetic variation for phenological and morphological traits in the core collection, which can be used to breed higher biomass and seed yield cultivars. Further studies will be required on the best performing accessions identified in this first screening in the test area, considering the interaction between genotype and environment. Other authors have already provided findings under high-temperature conditions using stress tolerance indices [31] or focusing on production stability and quality traits specifically in Southern Italy [77]. In this research context, it will therefore be appropriate to further evaluate abiotic stress phenomena and their influence in order to identify the most suitable genotypes in this particular environment.

Supplementary Materials: The following supporting information can be downloaded at: https: //www.mdpi.com/article/10.3390/agronomy14020303/s1, Figure S1: Thermo-pluviometric trend in the two growing seasons: (a) 2016/2017; (b) 2017/2018. The three ten-day periods are shown on the *x*-axis (I, II, III) for each month. Maximum temperatures are shown in red; minimum temperatures are shown in blue; rainfall is represented as green bars. Data are divided by cultivation phases (presowing and harvesting) and plant development stages (sowing-flowering and flowering-maturity). The arrows specify: the sowing time (4th of December (a); 7th of December (b)); the mean flowering time (2nd of April (a); 5th of April (b)); the mean maturation time (29th of May (a); 3rd of June (b)); Table S1: List of test accessions by country of origin, accession code (AC), and subspecies; Table S2a: Bio agronomic and quality characteristics of genotypes sampled by country of origin and by year (average values); Table S2b: Bio agronomic and quality characteristics of genotypes sampled by country of origin and by year (average values); Table S3a: Bio agronomic and quality characteristics of genotypes sampled by country of origin, by year, and subspecies microsperma (L) and macrosperma (S) (average values); Table S3b: Bio agronomic and quality characteristics of genotypes sampled by country of origin, by year, and subspecies microsperma (L) and macrosperma (S) (average values); Table S3c: Bio agronomic and quality characteristics of genotypes sampled by country of origin, by year, and subspecies microsperma (L) and macrosperma (S) (average values); Table S4: Eigenvalue and cumulative variance for the entire collection; Table S5: Eigenvalue and cumulative variance for ssp micro; Table S6: Eigenvalue and cumulative variance for ssp macro.

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