

Article

Estimation of Yield and Its Components in White Lupin Lines Under Favorable and Dry (Low-Rainfall) Field Conditions

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Abstract

The present study emphasizes the importance of developing breeding lines that combine productivity with stability under variable environmental conditions. Seed yield and its main quantitative components in 15 white lupin lines were analyzed over two years under favorable and dry environmental conditions. In dry conditions and reduced precipitation for vegetation (by 53 and 63%, respectively, compared to the 30-year norm and favorable conditions in the previous year) in Central Northern Bulgaria, seed yield decreased by an average of 35%. The environment had the strongest influence on the traits of plant height, number of pods and seeds, and seed weight per plant (72.2–81.3% of the total variation). The strength of the environmental factor influence was also decisive for seed yield (51.9%), whereas 1000-seed weight was primarily governed by genetic components (52.4%). The broad-sense heritability coefficient (h^2) for the evaluated quantitative traits ranged from low values for pods per plant (24.27%) and seed weight per plant (24.99%) to high values for plant height (64.58%). With stable expression and values around the group mean, V21 was distinguished for number of pods, number of seeds, and seed weight per plant, and V28/1 for number of seeds and seed weight per plant, while V6, V27/10, and V28/1 had a relatively constant 1000-seed weight, although below the group mean. V4, V1, and V29 can be characterized as high-stemmed and relatively stable across the two seasons. The conducted multifactor stability analysis and the GGE biplot model identify V425 and V28/1 as the most stable and productive among the tested lines, whose yields (3.025 t/ha and 2.495 t/ha, respectively) were above the average for the group and whose assessments of most of the stability parameters indicated a stable response across the two contrasting seasons.



Academic Editor: Yasutomo Hoshika

Received: 30 August 2026

Revised: 2 October 2026

Accepted: 5 October 2026

Published: 7 October 2026

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conditions of the [Creative Commons Attribution \(CC BY\)](https://creativecommons.org/licenses/by/4.0/) license.**Keywords:** white lupin; dry environment; genotype × environment; stability; quantitative traits

1. Introduction

The basis of plant production is the variety. In solving food security problems in modern conditions, the role of developed varieties has increased significantly. According to some researchers [1,2], the contribution of the crop genotype to the productivity of an agrocenosis ranges from 25 to 50–70%.

The challenges that global warming poses to breeding are related to developing varieties characterized by high and stable productivity, regardless of environmental con-

ditions [3–5]. The phenotypic stability of varieties in terms of yield reflects the interaction between individuals' genetic characteristics and the environment. Varieties that are characterized by higher phenotypic stability of yield have a less pronounced genotype–environment interaction, better overall adaptability, and relative constancy in the manifestation of their genetic characteristics [6,7]. White lupin tolerance to dry conditions is very important in Mediterranean-climate areas, where this stress coincides with critical reproductive phases of crop development. It is predicted that drought stress will increase throughout the Mediterranean basin and expand eastward and northward into Europe due to lower precipitation and rising evapotranspiration caused by climate change [8].

One of the crops that demonstrates comparative tolerance to changes in environmental conditions is the white lupin (*Lupinus albus* L.) [9]. In ancient times, it was a staple crop in various regions of the Roman Empire due to its adaptability and ability to improve soil fertility [10–12]. It is traditionally cultivated in the Mediterranean region, where it is used for human consumption, green manure, and as fodder. It is adapted to a wide range of environmental conditions, characterized by low pH, low nutrient availability, high salinity, and excess nitrates [13]. Today, interest in its cultivation as a food or feed crop is also determined by its high protein content (close to 40%) and other favorable quality characteristics of its seeds. It responds to the growing demand for healthy and nutritious plant foods and high-protein feed [10–12].

The use of white lupin seeds as a component of healthy, functional, or vegan food is due to the favorable content of essential amino acids and some of their beneficial functional properties, the positive effects on human health in terms of glycemia, diabetes, hypertension, cardiovascular disease, and obesity, as well as oil content (8–12%) with excellent nutritional characteristics [4,14]. The importance of white lupin as a feed source is confirmed by its high yield of crude protein per unit area compared with other cool-season grain legumes, such as broad bean (*Vicia faba* L.), pea (*Pisum sativum* L.), and narrow-leaved lupin (*Lupinus angustifolius* L.), grown in the Southern European region [15,16].

While the nutritional value and agronomic benefits of white lupin are well documented [10–12], systematic research on the phenotypic stability and genotype-by-environment interactions (GEIs) of its primary yield components remains limited. Most existing studies focus predominantly on final seed yield, often overlooking how individual quantitative traits—such as plant height, pod number, and seed weight per plant—respond to seasonal moisture deficits. Furthermore, the performance and adaptive thresholds of these specific Bulgarian white lupin breeding lines under fluctuating environmental conditions have not been previously evaluated. Assessing these genotypes under contrasting meteorological seasons is essential to identify resilient germplasm capable of sustaining productivity amid increasing climatic unpredictability in South-Eastern Europe.

The present study aimed to evaluate the yield and its components under favorable and dry environmental conditions in white lupin lines with a view to identifying breeding lines with good productivity and increased stability to specific environmental conditions.

2. Results

Environmental conditions (Figure 1) exerted a substantial influence on the phenotypic expression of most studied traits (Table 1, Figure 2). Based on the partitioning of the total sum of squares, environmental variance was the dominant factor for plant height (81.3%), pods per plant (76.9%), seeds per plant (76.0%), and seed weight per plant (72.2%). The variation in seed yield was also highly dependent on the environmental factor, which accounted for 51.9% of the total variation, followed by genotypic effect at 30.3% and genotype-by-environment interaction (GEI) at 15.8%. Conversely, for 1000-seed weight,

the genetic constitution of the breeding lines (52.4%) and the GEI effect (31.9%) played the decisive roles, while the environmental impact was limited to 14.5% (Figure 2).

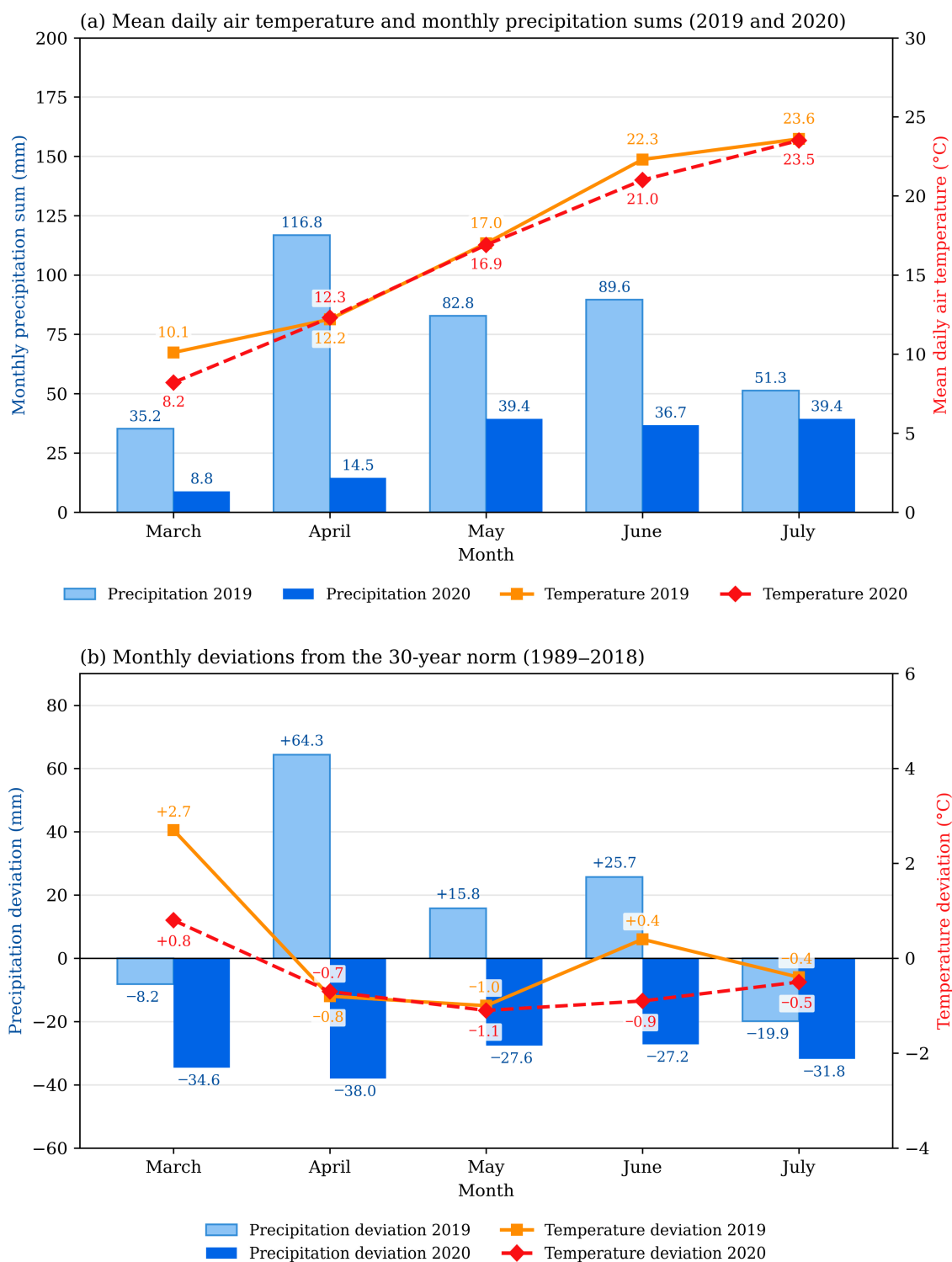
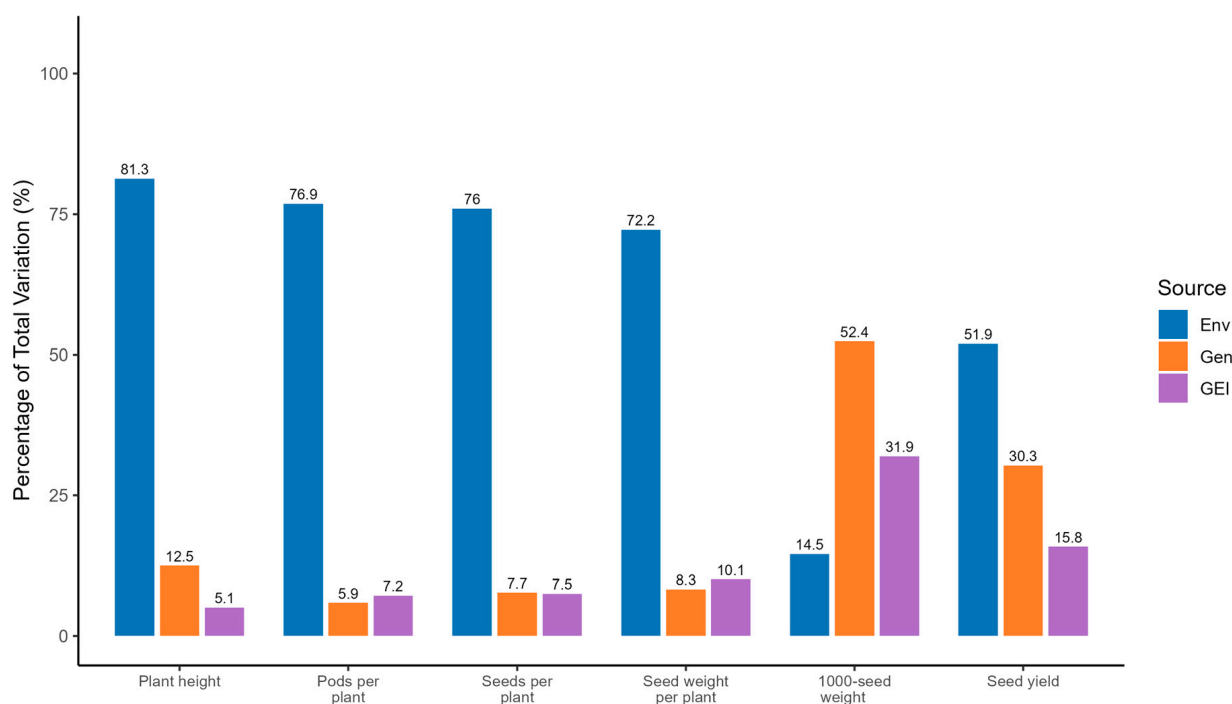


Figure 1. Meteorological conditions during white lupin vegetation: (a) variations in mean daily air temperature and monthly precipitation sums for the two experimental growing seasons (2019 and 2020), and (b) monthly deviations in temperature and precipitation from the 30-year long-term baseline norm (1989–2018) for the period March–July.

Table 1. Analysis of variance (ANOVA) for main quantitative traits and seed yield in 15 white lupin breeding lines.

Source of Variation	Df	Mean Squares					
		Plant Height (cm)	Pods per Plant	Seeds per Plant	Seed Weight per Plant (g)	1000 Seeds Weight (g)	Seed Yield (t/ha)
Environment (E)	1	43,164.401 ***	3285.38 ***	59,077.194 ***	4215.874 ***	26,821.523 ***	252,492.1 ***
Replication	4	29.496 *	59.378 ***	868.183 ***	51.204 ***	188.595 ***	562.642 **
Genotype (G)	14	472.559 ***	17.968 ***	426.428 ***	34.557 ***	6900.442 ***	10,517.832 ***
G × E	14	192.905 ***	21.905 ***	415.786 ***	41.894 ***	4208.604 ***	5500.08 ***
Residuals	56	8.679	3.424	60.083	6.153	23.55	127.981

*/**/** indicate statistical significance at $p \leq 0.05/0.01/0.001$.

**Figure 2.** Percentage contributions of growing season (E), breeding line (G), and their interaction (G × E) to the corrected total sum of squares for plant height (cm), pods per plant, seeds per plant, seed weight per plant (g), 1000-seed weight (g), and seed yield (t/ha). Percentages were calculated from the combined two-year ANOVA. Replication within season and residual error are included in the total but are not displayed; therefore, the plotted bars for each trait do not sum to 100%.

The obtained data on seed yield and its components were subjected to stability analysis for each trait using the most commonly used parametric and non-parametric methods. The importance of plant height as a breeding trait is determined by the fact that the main components of productivity are directly dependent on stem length and the number of branches of first and second order. In the presented group of lupin lines with plant height above the average value, the following breeding lines were distinguished: V27/7, V425, V4, V1, V6, V27/10, V28/1, and V29 (Table 2). In the low-stemmed genotypes, in some lines (V10 and V25), unidirectionality was established in some stability parameters (b_1 and CV), and very different evaluations were observed in others (W^2 and D^2).

Table 2. Stability parameters for plant height.

Lines	Average (cm)	W ²	b _i (LSD _{0.05})	D ²	σ ²	CV	P _i
V1	78.00	6.94	1.08	232.56	1.53	30.51	22.48
V4	78.92	3.71	0.94	114.26	0.34	26.07	27.84
V6	76.11	61.77	1.25 *	419.17	33.16	36.13	31.97
V10	51.25	103.29	0.67 **	6.01	57.12	28.78	581.59
V21	66.14	97.42	0.68 **	7.53	53.73	22.61	206.21
V23	61.5	0.43	1.02	176.09	0.23	36.42	257.75
V25	58.95	60.2	0.75 *	23.58	32.26	27.91	347.79
V27/7	80.72	186.01	1.44 ***	689.24	104.84	39.12	14.14
V27/10	77.8	78.46	1.29 *	461.07	42.79	36.25	20.8
V28/1	78.03	90.81	1.31 **	490.37	49.92	36.75	19.94
V29	72.53	1.07	1.03	186.24	0.86	31.25	71.9
V125	68.48	17.44	0.87	71.2	7.59	27.72	140.79
V251	70.18	159.13	0.59 ***	0.01	89.33	18.55	159.87
V254	65.78	8.09	0.91	95.45	2.19	30.29	181.19
V425	80.33	26.48	1.17 *	315.42	12.8	31.84	9.01

W²: Ecovalence [17]; b_i: Regression coefficient [18]; D²: Genotypic stability [19]; σ²: Stability variance [20]; CV: Coefficient of variation [21]; P_i: Superiority indexes for all environments [22]; */**/** indicate statistical significance at $p \leq 0.05/0.01/0.001$, based on Student's *t*-test.

The structural responses of the 15 white lupin breeding lines varied significantly between the two meteorologically contrasting seasons (Figure 3). During the low-rainfall 2020 growing season, severe moisture deficits led to a marked reduction across all studied quantitative components compared to the favorable 2019 baseline. On average, plant height, pods per plant, and seeds per plant decreased under stress conditions (Figure 3a–c). This operational reduction directly restricted individual seed weight per plant (Figure 3d), whereas the 1000-seed weight followed an opposite trend, increasing significantly in most lines (13 of 15) under the dry 2020 season (Figure 3e)—a pattern likely reflecting reduced sink competition among fewer, better-provisioned seeds per plant rather than genetic buffering. Consequently, total seed yield decreased by an average of 35% in 2020, with the sharpest declines observed in lines V27/7 and V27/10, which dropped by approximately 53% (Figure 3f).

As we have pointed out in previous studies, differences in the stability assessment of individual parameters are due to the fact that different methods are based on different concepts of stability [23]. Also, the ambiguous assessments of the various parameters show that longer-term studies are needed to determine the ecological stability of this trait.

The values of different stability parameters for the number of pods per plant varied widely (Table 3). Higher values were found for V425, V125, V23, and V27/7. From statistically significant high values of the linear regression coefficient (b_i), as well as the genotypic stability parameter (D²) and stability variance (σ²), it can be assumed that these lines are highly responsive to the seasonal conditions.

V4 took fourth position in terms of number of pods, but like the previous breeding lines, it showed a strong decline in the dry season. Although the parameter *b* was above unity, its estimate was not definitive due to the lack of statistical significance. The additional information from the remaining stability parameters gave reason to believe that V4 is also a highly variable breeding line across environments.

V21 and V28/1 can be considered valuable for their stable response, as indicated by the values of the stability parameters, although the pod number of V28/1 was below the group mean. V29 and V1 were characterized by a slight variation in seed number across both experimental years. The low values of W₂, b_i, and σ₂ indicated that these lines are highly stable.

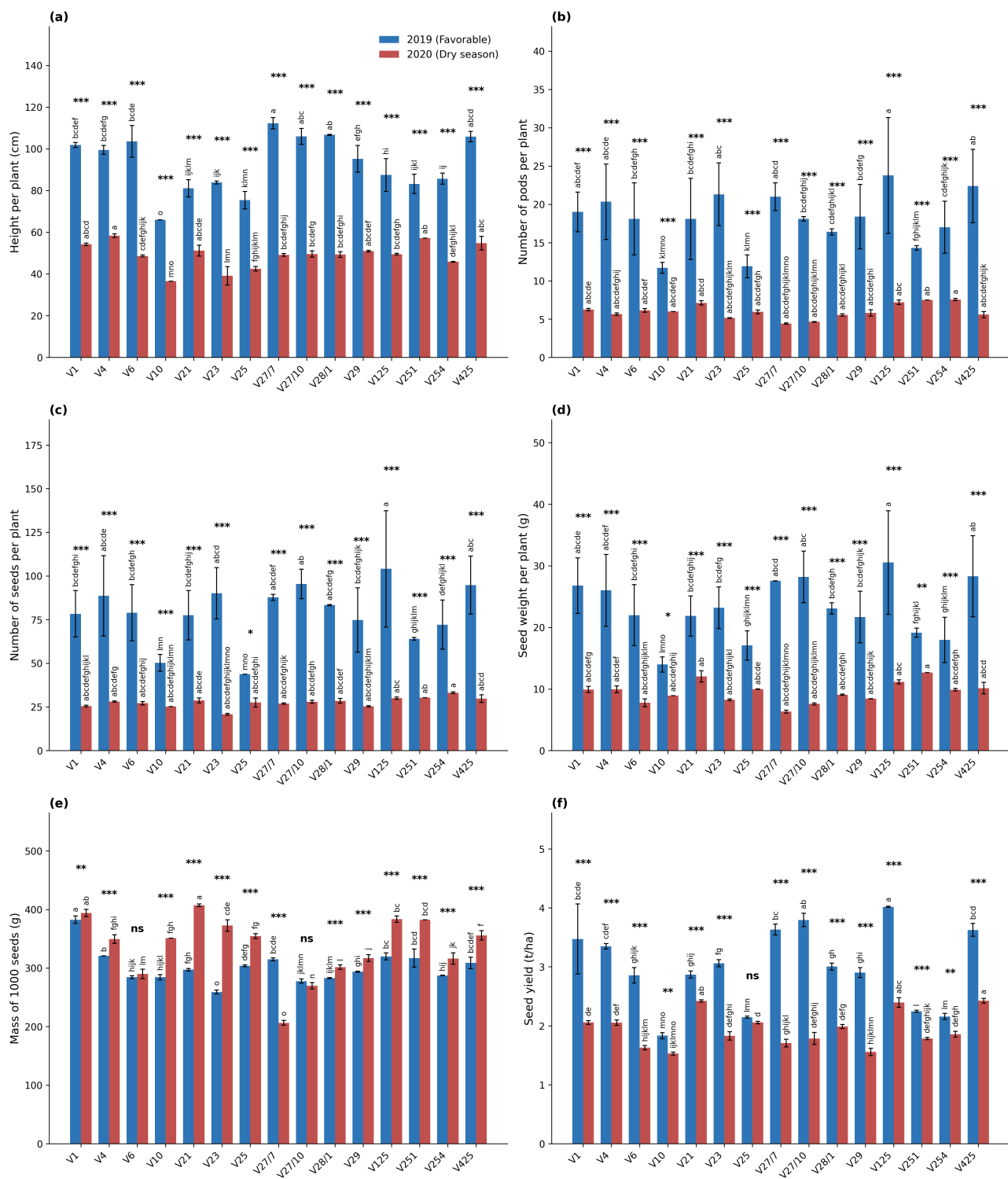


Figure 3. Mean values of main quantitative traits and seed yield in 15 white lupin breeding lines evaluated during the favorable 2019 season (blue bars) and the dry 2020 season (red bars): (a) plant height (cm), (b) pods per plant, (c) seeds per plant, (d) seed weight per plant (g), (e) 1000-seed weight (g), and (f) seed yield (t/ha). Vertical error bars represent the standard error of the mean (SEM). Lowercase letters compare genotypes within the same year only (2019 vs. 2019, or 2020 vs. 2020; Tukey's HSD, $p \leq 0.05$); bars not sharing a letter differ significantly. Asterisks above each genotype pair indicate the significance of the favorable (2019) vs. dry (2020) difference for that genotype (*— $p \leq 0.05$, **— $p \leq 0.01$, ***— $p \leq 0.001$; ns = not significant).

Table 3. Stability parameters for number of pods per plant.

Lines	Average	W ²	b _i (LSD _{0.05})	D ²	σ ²	CV	P _i
V1	12.62	0.21	1.05	24.85	0.16	50.50	6.10
V4	12.99	3.34	1.21	40.35	1.64	56.51	3.85
V6	12.12	0.01	0.99	19.61	0.28	49.36	8.53
V10	8.85	20.48	0.47 ***	0.10	11.53	32.20	37.00
V21	12.61	0.64	0.91	13.89	0.09	23.49	8.07
V23	13.22	8.2	1.33 *	54.6	4.45	61.06	2.96
V25	8.93	18.91	0.49 ***	0.03	10.63	33.33	35.84
V27/7	12.71	10.05	1.37 **	59.22	5.52	65.25	4.37
V27/10	11.38	0.91	1.11	30.03	0.25	59.12	10.13
V28/1	10.97	0.78	0.90	13.26	0.17	29.43	14.57
V29	11.99	0.26	1.06	25.36	0.13	53.48	7.96
V125	15.48	9.98	1.37 **	59.04	5.48	53.50	0.03
V251	10.90	14.04	0.56 **	0.60	7.82	31.19	22.4
V254	12.28	3.51	0.78	7.03	1.74	38.49	11.45
V425	14.00	11.05	1.39 **	61.6	6.09	60.00	1.42

W²: Ecovalence; b_i: Regression coefficient; D²: Genotypic stability; σ²: Stability variance; CV: Coefficient of variation; P_i: Superiority indexes for all environments. */**/** indicate statistical significance at $p \leq 0.05/0.01/0.001$, based on Student's *t*-test.

Their main disadvantage was the lower number of seeds per plant. In V21 and V28/1, the number of seeds was around or above the average for the studied group, despite differences in trait manifestation in 2019 and 2020. In them, the stability parameters' values (with small variations, Table 4) were similar to the previous two lines, but the advantages of V21 and V28/1 were in the higher trait values.

Table 4. Stability parameters for number of seeds per plant.

Lines	Average	W ²	b _i (LSD _{0.05})	D ²	σ ²	CV	P _i
V1	51.90	1.08	1.03	667.95	4.7	50.87	180.61
V4	58.35	42.04	1.18	979.03	18.93	51.84	66.18
V6	53.01	0.10	1.01	631.46	5.27	48.85	167.45
V10	37.73	347.95	0.49 ***	37.85	195.42	33.07	741.01
V21	53.12	3.31	0.95	528.36	3.42	45.89	181.30
V23	55.45	161.46	1.35 *	1407.15	87.82	62.49	86.90
V25	35.67	615.3	0.32 ***	0.04	349.66	22.78	915.86
V27/7	57.29	45.10	1.19	993.59	20.7	53.09	76.82
V27/10	61.7	129.12	1.31 *	1308.16	69.17	54.62	25.41
V28/1	55.9	6.02	1.07	743.05	1.85	49.02	113.22
V29	49.43	0.17	0.99	594.93	5.23	51.33	229.19
V125	67.01	259.62	1.44 **	1674.28	144.45	55.30	2.56
V251	47.17	156.00	0.66 *	151.67	84.67	35.69	403.32
V254	52.62	76.63	0.76	257.64	38.88	37.01	255.47
V425	62.25	94.81	1.27	1193.16	49.37	52.29	24.44

W²: Ecovalence; b_i: Regression coefficient; D²: Genotypic stability; σ²: Stability variance; CV: Coefficient of variation; P_i: Superiority indexes for all environments. */**/** indicate statistical significance at $p \leq 0.05/0.01/0.001$, based on Student's *t*-test.

In terms of seed productivity per plant (Table 5), the stability variance (σ²), which accounts for linear interactions, gave a unidirectional high estimate for V6, V21, V28/1 and V29. A similar trend was found for W² and b_i. On the other hand, the stability indices such as D² and CV did not support this conclusion. This apparent disagreement is not contradictory but reflects that these parameters quantify stability relative to different

reference points. In two environments, W^2 and σ^2 are mathematically near-equivalent measures of deviation from $b_i = 1$ (the group's average responsiveness), whereas D^2 measures deviation from the least-responsive genotype in the trial (here, V10, $b_i = 0.37$); consequently, genotypes such as V6, V28/1, and V29, whose b_i values are close to 1, are classified as highly stable by W^2 , σ^2 , and b_i but as comparatively unstable by D^2 . Similarly, CV is a mean-normalized measure, so V6 and V29—which have among the lowest average seed weight per plant in the group—show inflated CV values despite minimal absolute variability, whereas W^2 and σ^2 are unaffected by trait mean. Of this group, only V21 and V28/1 were distinguished by better productive potential (over 16 g), while the rest were below average.

Table 5. Stability parameters for seed weight per plant.

Lines	Average (g)	W^2	b_i (LSD _{0.05})	D^2	σ^2	CV	P_i
V1	18.35	4.94	1.23	69.31	2.31	45.93	5.40
V4	17.98	2.75	1.17	60.24	1.05	44.67	6.98
V6	14.88	0.13	1.04	41.82	1.46	47.82	24.24
V10	11.44	37.25	0.37 **	0.03	20.95	22.22	72.07
V21	16.95	7.58	0.72	11.23	3.83	28.98	18.95
V23	15.72	0.76	1.09	48.62	1.10	47.54	18.35
V25	13.53	21.98	0.52 **	2.03	12.14	26.19	47.15
V27/7	16.95	28.28	1.55 **	130.43	15.78	62.66	12.21
V27/10	17.88	23.73	1.50 **	120.44	13.15	57.62	7.83
V28/1	16.08	0.04	1.02	39.74	0.51	43.53	17.11
V29	14.85	0.02	1.004	37.14	1.54	46.15	24.03
V125	20.85	16.07	1.41 *	102.24	8.73	46.48	0.56
V251	15.89	26.17	0.47 **	0.97	14.56	20.39	32.55
V254	13.93	15.89	0.59 *	4.48	8.63	29.00	41.44
V425	19.23	9.87	1.32 *	85.46	5.16	47.23	2.83

W^2 : Ecovalence; b_i : Regression coefficient; D^2 : Genotypic stability; σ^2 : Stability variance; CV: Coefficient of variation; P_i : Superiority indexes for all environments. */** indicate statistical significance at $p \leq 0.05/0.01$, based on Student's *t*-test.

According to the interpretation of the regression coefficient, genotypes with higher values are assessed as more variable because they interact more strongly with environmental conditions.

The most productive lines, V1, V4, V125, and V425, showed a stronger decline in the dry season. In them, the coefficient b was considerably above unity, and the parameters D^2 , σ^2 , and CV confirmed this assessment.

Regarding the trait of mass per 1000 seeds (Table 6), the coefficient of linear regression can not provide a final assessment of the stability of the studied lupin lines due to its insignificance. The data show that the large-seeded lines, such as V21 and V125, were too labile under abiotic stress conditions. Similar assumptions can be made for V125 and V251 based on the estimates of the ecovalence W^2 , genotypic stability D^2 and stability variance σ^2 . The trait instability was mainly due to considerable differences in the 1000-seed weight during the experimental years, as meteorological conditions had an essential influence on it.

Stable breeding lines can be identified from the group of medium-seeded genotypes (with a 1000-seed weight ranging between 270 and 295 g), such as V27/10 and V28/1, for which most of the stability parameters were consistent in their assessments. These lines showed a relatively constant 1000-seed weight across the two seasons.

Table 6. Stability parameters for 1000-seed weight.

Lines	Average (g)	W ²	b _i (LSD _{0.05})	D ²	σ ²	CV	P _i
V1	388.26	266.84	0.33	7188.01	99.99	1.47	44.22
V4	335.29	15.97	0.84	9432.51	4.74	4.31	1779.72
V6	287.19	419.09	0.16	6503.7	187.83	0.97	5848.18
V10	317.82	522.41	1.94	15,369.4	247.43	10.52	3193.06
V21	352.24	2853.8	3.19 **	23,882.1	1592.47	15.63	1821.16
V23	315.92	3122.37	3.29 **	24,647.6	1747.41	17.97	4105.75
V25	329.26	131.20	1.47	12,672.3	21.73	7.70	2239.46
V27/7	260.81	1224.69	0.14 **	0.001	5844.9	20.8	11,209.18
V27/10	273.84	896.82	0.23	5065.21	463.44	1.43	7462.02
V28/1	292.29	125.63	0.54	8083.56	18.52	3.19	5270.52
V29	305.48	63.58	0.67	8675.74	7.28	3.81	3999.81
V125	351.73	424.09	1.84	14,813.5	190.71	9.05	1121.97
V251	349.78	478.76	1.90	15,128.4	222.25	9.36	1225.77
V254	301.88	15.55	0.84	9442.82	4.99	4.80	4330.22
V425	332.31	79.36	1.36	12,105.7	8.17	7.09	2022.1

W²: Ecovalence; b_i: Regression coefficient; D²: Genotypic stability; σ²: Stability variance; CV: Coefficient of variation; P_i: Superiority indexes for all environments. ** indicate statistical significance at $p \leq 0.01$, based on Student's *t*-test.

The average seed yield of white lupin lines in 2020 was 35% lower compared to 2019 (Figure 3). Most of the stability parameters indicate that genotypes with high seed yield, such as V125, V27/10, V1, and V4, showed larger yield losses in the dry season but, at the same time, good responsiveness under favorable environmental conditions (Table 7). Accordingly, their yield reduction in 2020 was greater (Figure 3f). On the other hand, less productive genotypes (V251, V254, V10 and V25) showed smaller relative yield reductions in the dry season, despite the lack of absolute unidirectionality in the assessment of stability parameters. All stability parameters (except σ² and P_i) identified the lowest-yielding lines (V10/1.680 t/ha/, V251/2.014 t/ha/, and V254/2.008 t/ha/) as ecologically stable.

Table 7. Stability parameters for seed yield.

Lines	Average (t/ha)	W ²	b _i (LSD _{0.05})	D ²	σ ²	CV	P _i
V1	2.764	638.38	1.34	8803.32	297.78	25.63	1084.6
V4	2.701	278.05	1.22	7264.95	89.90	23.98	1463.92
V6	2.242	138.31	1.16	6451.34	9.28	27.33	4958.66
V10	1.680	2859.44	0.29 **	227.59	1579.17	9.02	13,948.66
V21	2.645	1880.28	0.42 *	634.93	1014.26	8.43	3304.83
V23	2.445	148.03	1.16	6516.82	14.89	25.19	3176.72
V25	2.101	4700.46	0.08 ***	0.01	2641.29	2.14	9100.54
V27/7	2.669	3738.67	1.82 **	16,823.3	2086.41	36.04	1655.55
V27/10	2.790	4510.76	1.90 ***	18,420.5	2531.85	36.00	1143.15
V28/1	2.495	8.58	0.96	4307.31	5.56	20.40	3049.89
V29	2.229	403.19	1.27	7856.94	162.09	30.13	4996.59
V125	3.206	1594.24	1.53 *	11,769.6	849.24	25.33	2.33
V251	2.014	1781.65	0.44 *	694.34	957.36	11.48	8877.45
V254	2.008	2885.59	0.28 **	220.29	1594.25	7.46	9452.13
V425	3.025	101.41	1.13	6182.72	2.01	19.86	383.67

W²: Ecovalence; b_i: Regression coefficient; D²: Genotypic stability; σ²: Stability variance; CV: Coefficient of variation; P_i: Superiority indexes for all environments. */**/** indicate statistical significance at $p \leq 0.05/0.01/0.001$, based on Student's *t*-test.

Of particular interest were V425 and V28/1, whose seed yields (3.025 t/ha and 2.495 t/ha, respectively) were above the group average (Table 7) and, according to most stability parameters, they showed a stable response across the two contrasting seasons. In this regard, particularly indicative parameters were the ecovalence (W^2) of Wricke [17], the linear regression coefficient (b_i) of Finlay and Wilkinson [18], and the stability variance (σ^2) of Shukla [20]. These lines may exhibit good adaptive abilities in dry environmental conditions and should be confirmed in multi-year and multi-location trials.

Through the GGE biplot model, each genotype receives an estimate for the effects of genotype (G) and genotype–environment interaction (GEI) [24]. For an evaluation of each genotype, the starting point is the small circle in the center of the biplot, which represents the average environmental coordinate (AEC). AEC is the arithmetic mean of the PC1 and PC2 estimations of the environment (E) (Figure 4). The identification of breeding lines with positions close to the ideal genotype confirms their superiority regarding both yield and stability. The closer the point of a given genotype is to the beginning of the concentric circle, the weaker its variation under experimental conditions. None of the white lupin lines fell in the center of the concentric circles. V425, V28/1 and V21, which are located between the second and fourth circles, had little variation. V1, V4, and V25 were more distant from the starting point, and it can be assumed that they have an average degree of variation, while V10 and V29 were most susceptible to changes in environmental conditions. The ranking of lines presented through this visualization does not strictly correspond to the obtained estimates of stability parameters. It is important in this case that much of the information provided by the different statistical models for the tested genotypes is relatively similar. The similar arrangement of the lines in terms of yield and stability supports this statement.

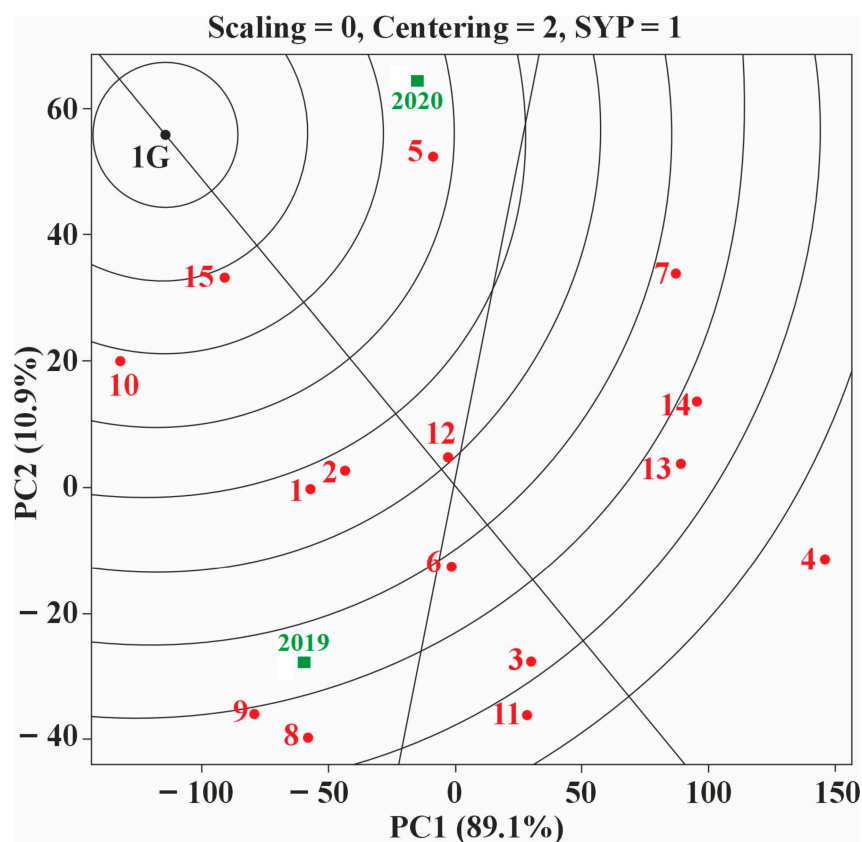


Figure 4. GGE biplot model showing the seed yield performance and macro-environmental stability of 15 white lupin breeding lines across the contrasting 2019 (favorable) and 2020 (dry) growing seasons. Breeding lines are coded as follows: 1—V1, 2—V4, 3—V6, 4—V10, 5—V21, 6—V23, 7—V25, 8—V27/7, 9—V27/10, 10—V28/1, 11—V29, 12—V125, 13—V251, 14—V254, and 15—V425.

The genotypic (V_g) and phenotypic variance (V_p), and genotype $\times$ environment interaction variance (V_{ge}), are presented in Table 8. Phenotypic variance accounted for the largest share, exceeding the influence of other factors. The share of total variation due to V_g and V_{ge} was considerably smaller, as the differences between them were non-significant. For traits such as plant height, 1000-seed weight, and yield, the influence of V_g dominated over V_{ge} . The highest genotypic and phenotypic variance was found for seed yield, followed by 1000-seed weight and number of seeds per plant. The greatest variance, determined by environmental influence, was found in seed yield and 1000-seed weight. The variance of the same factor was characteristic of the number of pods and seed weight per plant.

Table 8. Variance components and heritability of quantitative traits in white lupin lines.

Traits	V_g	V_{ge}	V_p	h^2 (%)
Plant height	67.89	29.67	105.12	64.58
Pods per plant	1.78	2.71	7.32	24.27
Seeds per plant	48.32	52.58	150.89	32.03
Seed weight per plant	3.47	5.25	13.90	24.99
1000-seed weight	916.26	694.76	1631.05	56.18
Seed yield	1447.41	881.70	2434.06	59.46

V_g —genotypic variance, V_{ge} —genotype $\times$ environment interaction variance, V_p —phenotypic variance, h^2 —broad-sense heritability.

Heredity is an important selection parameter and an effective method for improving varieties. Selecting one genotype can be very effective for a particular trait that is highly heritable, compared to another that has a lower heritability. The broad-sense heritability coefficient for the six quantitative traits ranged from 24.27–24.99% (for number of pods and seed weight per plant) to 64.58% (plant height). The traits seeds per plant, 1000-seed weight, and seed yield were characterized by a medium heritability coefficient.

3. Discussion

Water deficit has an essential negative impact on crop habitus, metabolism, and development. Dehydration is associated with physiological changes that slow growth and reduce yield stability. The impact of drought is particularly pronounced in certain stages of crop development [25,26]. According to data from Perdigão et al. [27], the amount and distribution of precipitation during the growing season play an important role in seed yield in *Lupinus*. In a study aimed at reducing the knowledge gap regarding drought tolerance variations in white lupin, yield was evaluated in 21 landraces, 2 breeding lines, and one cultivar. Drought stress decreased seed yield by 79%, and some local accessions provided a better yield than the improved germplasm under stressful or favorable conditions [28]. In our study, the yield reduction under conditions of insufficient rainfall (in 2020) was 35% (on average), ranging from 4.2% (V25) to approximately 53% (V27/7 and V27/10). During this year, only 6 lines (V1, V4, V21, V25, V125, and V425) achieved yields above 2.000 t/ha.

Annicchiarico et al. [29] reported significant variation in grain yield among *L. albus* accessions of different geographical origins from a global collection grown in the Mediterranean region. A strong genotype $\times$ environment interaction was found for grain yield in dry and moisture-favorable years. The authors recommended a set of local varieties with good performance and adaptation to these conditions. According to Valente et al. [30], in *L. albus*, seed yield is strongly influenced by genotype, environment, and genotype $\times$ environment interaction, with environment being the main factor accountable for variability (73% of the total variation). Under the conditions of the present experiment, the environment accounted for 51.9% of the total variation for seed yield, 14.5% for the 1000-seed weight,

and ranged from 72.2% to 81.3% for the remaining quantitative traits (plant height, pods per plant, seeds per plant, and seed weight per plant). Other researchers [31,32] also noted that white lupin productivity is influenced by genotype $\times$ environment interactions, with a significant environmental effect confirmed by ANOVA for the GEI model. Atnaf et al. [33] point out that the interaction between genotype and environment is an important issue both in genotype evaluation and for cultivar recommendation in a given area.

When testing white lupin cultivars in climatically contrasting environments in Italy, Annicchiarico and Carroni [34] found specific and widely adapted cultivars. The distinction between environments defined as limiting and favorable further emphasizes the dominant role of the environment. Previous investigations in both legume and cereal species have shown that moisture stress or uneven rainfall distribution increases varietal variability. The higher variability established under adverse conditions increases the expression of GEI, making genotype assessment more complex [35–37]. Stable genotypes are usually distinguished by lower interaction effects and constant yields across different environments, while specifically adapted genotypes have higher responsiveness [38,39]. Julier et al. [31] and Valente et al. [30] determined cultivars belonging to *L. albus* as less stable (compared to *L. angustifolius* and *L. luteus*) in different environments, although higher average values for seed productivity were obtained from them.

The identification of relatively high-yielding and stable genotypes, such as V425 and V28/1 under the experimental conditions, is consistent with earlier studies that also reported genotypes combining high productivity and stability. In testing 12 local white lupin cultivars in northwestern Ethiopia, Atnaf et al. [33] found that the highest-yielding genotype (G2) was also the most stable. G2 was recommended for production. Similarly, region-specific white lupin cultivars with high yield and adaptability have been reported by researchers for different parts of the world, such as Western Europe [40], Southern Europe [29,34,41], and North Africa [33,42].

Valente et al. [30] reported different results in a comparative evaluation of 7 lupin varieties. The researchers found that the highest-yielding variety, cv. Estoril (2.42 t seeds/ha) was the least stable regarding three stability parameters (CV, coefficient of variation; W, rank of the ecovalence stability index [17]; rank of Shukla's variance [20]). Danakumara et al. [43] and Khan et al. [44] point out that genotypes with high yield in different environments are particularly valuable in selection programs because they show the presence of favorable allelic combinations related to both productivity and resistance. In contrast, genotypes characterized by high productivity only under favorable conditions but with low yields in stressful environments are defined as specifically adapted. They can be recommended for targeted cultivation rather than for widespread production [38]. Today, under changing climatic conditions, identifying genotypes with high yield and stability is particularly important and relevant, as environmental unpredictability is increasing [35,45].

Several researchers have found stability not only for seed yield but also for other agronomic characteristics (flowering time, longevity of reproductive phase, aboveground biomass, etc.) under different environmental conditions in white lupin [29,34,41,46]. However, information remains limited in the scientific literature assessing the stability of the main quantitative traits related to seed yield. In our previous study [47], in another annual legume crop (*Vicia faba*), an assessment of biological manifestations, stability of some quantitative characteristics (plant/height, number of pods and seeds/plant, mass of 100 seeds), and productivity was made in 17 Bulgarian and introduced accessions. Regarding the trait of plant height, FbH 16 and FbH 13 were determined as tall and ecologically stable ($b_1 = 0.76$; $b_1 = 0.79$). BGE 029055 was low-variable, with high values for the number of pods and seeds per plant. From a breeding point of view, of particular interest was BGE 041470,

which was characterized by high values of mass per 100 seeds and productivity/plant, and relative stability ($b_1 = 1.10$).

GGE biplots are suitable for analyzing and visualizing the $G \times E$ model [33]. According to Yan and Kang [24], the ideal genotype should be stable and have the highest possible productivity. In a study of 12 genotypes conducted by the authors, the desirability of each accession was determined by its proximity to the “ideal” genotype. G2 was the most desirable genotype, and G4 was the least desirable. The remaining accessions, based on the distance to the ideal genotype, were ordered as follows: $G10 > G5 > G8 > G3 > G12 > G7 > G6 > G1 > G11 > G9$. According to some researchers [38,44,45], GGE biplot analysis is an effective means for identifying genotypes with broad and specific adaptation, as well as for assessing the representativeness of test environments. The GGE biplot statistical model under the experimental conditions positioned lines V425, V28/1, and V21 outside the zone of high variability, defining them as relatively stable and productive in the two tested seasons. In the context of the increased aridity in the continental climate of South-Eastern Europe, these lines represent promising breeding material, whose broader adaptability should be verified in multi-year and multi-location trials.

Beyond the aggregate seed yield response, individual yield components followed largely parallel seasonal patterns across the tested lines, offering limited scope for single-trait explanations of differential performance. Plant height and pod number declined by a broadly similar proportion in nearly all lines (mean reduction 46.6% and 65.1%, respectively, and SD 6.9 and 10.3 percentage points), including in the two lines identified as high-yielding and stable, V425 and V28/1. In contrast, seed weight per plant showed more line-specific responses: V27/10, despite an above-average seed number in 2019, exhibited one of the steepest reductions in seed weight per plant (−73.1%, compared with a 15-line mean of −57.0%), consistent with its overall low stability ranking, whereas V425 and V28/1 tracked close to the group average (−64.2% and −60.7%, respectively). Notably, 1000-seed weight increased under the dry season in 13 of the 15 lines, including V425 (+15.3%) and V125 (+19.9%)—the latter despite its classification as an unstable, favorable-season-dependent line—indicating that this trait alone does not discriminate stable from unstable genotypes and that its apparent conservatism (Figure 3e) reflects a general seasonal response rather than a genotype-specific buffering mechanism.

The evaluation of variance components revealed that broad-sense heritability (h^2) was low for pods per plant (24.27%) and seed weight per plant (24.99%), indicating that these traits are highly susceptible to environmental fluctuations and less responsive to direct phenotypic selection in early generations. Conversely, plant height (64.58%) and 1000-seed weight (56.18%) exhibited moderate-to-high heritability, suggesting stronger genetic control. From a breeding perspective, these findings imply that selection strategies for improving white lupin productivity in stress-prone environments should prioritize highly heritable traits, such as 1000-seed weight, as indirect selection criteria, or employ index-based selection methods to effectively manage the substantial environmental noise affecting unstable yield components.

In conclusion, the results in the present study emphasize the importance of developing breeding lines that combine high productivity with stability in different environments. Identifying such breeding lines is essential for creating varieties with consistent characteristics under variable environmental conditions. Given the growing challenges of climate change, including greater frequency of stress and environmental variability, the selection of stable genotypes is increasingly important for sustainable white lupin production.

Finally, several limitations of the present study must be acknowledged. This evaluation was conducted over two consecutive growing seasons at a single macro-environmental location and lacked a widely cultivated commercial check cultivar for baseline comparison.

Furthermore, the moisture deficit in 2020 arose from naturally occurring low rainfall rather than an experimentally imposed drought treatment, and no direct measurements of soil moisture, evapotranspiration, water balance, or a standardized drought index were taken. While these conditions provided highly contrasting meteorological settings to assess early-stage breeding materials, multi-year and multi-location trials across diverse agro-ecological zones are required to conclusively confirm the stability and adaptability thresholds of the identified high-yielding lines prior to commercial recommendation.

4. Materials and Methods

4.1. Experimental Site and Environmental Conditions

The field experiment was conducted during two distinct growing seasons (2019 and 2020) at the experimental field of the Institute of Forage Crops located in Pleven, Central Northern Bulgaria (43°25' N, 24°37' E, altitude 145 m a.s.l.). The soil type at the experimental site is characterized as a leached chernozem with moderate organic matter content and a neutral to slightly acidic pH.

Meteorological data, including daily air temperature and precipitation sum, were obtained from the automated weather station located at the Institute of Forage Crops—Pleven. Long-term (1989–2018) monthly precipitation and temperature norms were calculated as simple arithmetic means from the same weather-station records. The two experimental years represented highly contrasting meteorological environments. The 2019 growing season (March–July) was characterized by optimal rainfall distribution and cumulative precipitation of 376 mm, serving as a favorable baseline. Conversely, the 2020 growing season experienced severe moisture deficits, with a cumulative precipitation of 139 mm during vegetation, representing a 63% and 53% reduction compared to 2019 and the 30-year long-term norm (1989–2018), respectively. Because these conditions were environmental rather than managed, the 2020 season is described throughout this study as a dry or low-rainfall growing season (Figure 1).

4.2. Plant Material and Crop Management

The experimental germplasm consisted of 15 advanced white lupin (*Lupinus albus* L.) breeding lines developed at the Institute of Forage Crops—Pleven: V1, V4, V6, V10, V21, V23, V25, V27/7, V27/10, V28/1, V29, V125, V251, V254, and V425. Due to the historical baseline nature of this early-stage evaluation, a commercial standard or check cultivar was not included, which represents a recognized limitation of the study design.

The trials were established using a randomized complete block design (RCBD) with three independent replications. Sowing was performed manually on 5 March in both seasons (2019 and 2020) at a sowing depth of 4–5 cm, utilizing a plot size of 4.0 m² consisting of 4 rows, each 2 m long. The row spacing was set at 50 cm with an intra-row plant spacing of 10 cm, achieving a targeted density of 20 plants per m² (80 plants per plot). No artificial irrigation or synthetic fertilizers were applied during either growing season to evaluate performance under natural rainfed conditions. Standard agronomic procedures for weed control and crop protection were applied uniformly across all plots in accordance with established crop production technologies. Harvesting was conducted manually upon full maturity on 8 July 2019 and 15 July 2020.

4.3. Data Collection and Trait Measurements

At the fully mature developmental stage (BBCH 89), ten representative plants were randomly sampled from the inner rows of each plot to minimize edge effects. These sampled plants were treated as subsamples to calculate the plot mean, avoiding pseudo-replication. Quantitative traits were evaluated based on standardized terminology: plant

height (cm), pods per plant, seeds per plant, seed weight per plant (g), and 1000-seed weight (g). Measurements for pods and seeds included all primary and higher-order structural branches per plant.

To determine the 1000-seed weight, two random samples of 100 seeds were counted per plot, weighed, and extrapolated. Total seed yield per plot was determined by harvesting the full plot area (4.0 m²). Seed weights per plant and net plot yields were corrected to a standard moisture content of 13% using a digital moisture analyzer. Plot-level clean seed weights were converted to total seed yield expressed in tons per hectare (t/ha). Plot yield (kg/plot) was converted to seed yield per hectare using the formula: Yield (t/ha) = [Plot yield (kg) × 10]/Plot area (m²), based on the harvested area of 4.0 m².

4.4. Statistical Analysis

The primary data were subjected to a two-factor analysis of variance (ANOVA) to assess the main effects of genotype (G), environment (E), and their corresponding genotype-by-environment interaction (GEI). Prior to performing the ANOVA, the underlying statistical assumptions, including the normality of residuals and homogeneity of variances, were verified using the Shapiro–Wilk and Levene’s tests, respectively. The relative contribution of each source of variation to the total phenotypic variation was estimated as the percentage of its sum of squares (SS) relative to the total corrected sum of squares (Environment + Replication + Genotype + G × E + Residual error), calculated from the ANOVA. Effect sizes for each factor were quantified to complement the significance levels. Mean separations were determined using Tukey’s Honestly Significant Difference (HSD) test at ($p \leq 0.05$).

Phenotypic stability across the contrasting environments was evaluated using a combination of parametric and non-parametric indices: Wricke’s ecovalence (W^2) [17], Finlay and Wilkinson’s linear regression coefficient (b_i) [18], Hanson’s genotypic stability (D^2) [19], Shukla’s stability variance (σ^2) [20], Francis and Kannenberg’s coefficient of variation (CV, %) [21], and Lin and Binns’ superiority index (P_i) [22]. Genotypic variance (V_g), phenotypic variance (V_p), and GEI variance (V_{ge}) components, along with broad-sense heritability (h^2), were estimated following the variance partitioning methods of Johnson et al. [48] and Fehr [49]. The graphical visualization of yield performance and macro-environmental stability was constructed using the GGE biplot model based on singular value decomposition (SVD) of the first two principal components [50]. All statistical procedures were executed using GENES software (version 2009.7.0 for Windows) [51] and R (version 4.4.3) [52], using the packages *dplyr* and *tidyr* for data handling and *ggplot2* and *patchwork* for graphical visualization.

5. Conclusions

In dry environmental conditions and reduced precipitation for vegetation (by 53 and 63%, respectively, compared to the 30-year norm and favorable conditions in the previous year) in Central Northern Bulgaria, seed yield decreased by an average of 35%. The results of the present study showed different performances of 15 white lupin lines under favorable environmental and dry conditions, which proves the importance of the G × E interaction in the evaluation of breeding lines. Based on the partitioning of the total sum of squares, the environment exerted the strongest influence on plant height, pods per plant, seeds per plant, and seed weight per plant, accounting for 72.2% to 81.3% of the total variation. The environmental factor was also a major driver for seed yield (51.9%), whereas 1000-seed weight was primarily governed by genetic components, with environmental impact limited to 14.5%. A stable expression with values around the group mean was found in V21 for number of pods, number of seeds, and seed weight per plant, and in V28/1 for number of seeds and seed weight per plant. V6, V27/10, and V28/1 had a relatively

constant 1000-seed weight across the two seasons, although below the group mean. V4, V1, and V29 can be characterized as high-stemmed and relatively stable under changing environmental conditions.

The conducted multifactor stability analysis and the GGE biplot model identified lines V425 and V28/1, whose yields (3.025 t/ha and 2.495 t/ha, respectively) were above the average for the group and whose assessments of most of the stability parameters (especially in terms of ecovalence W^2 , linear regression coefficient (b_1), and stability variance σ^2) indicated a stable response across the two contrasting seasons. Given the growing challenges of climate change, including greater frequency of stress and environmental variability, the selection of stable genotypes is increasingly important for sustainable white lupin production.

Given that this evaluation was limited to two growing seasons at a single location without a standard check cultivar or direct soil-moisture measurements, future work should prioritize multi-year, multi-location trials incorporating a reference cultivar and soil-moisture monitoring to confirm the drought-adaptive value of V425 and V28/1 and to clarify the physiological basis of the seasonal increase observed in 1000-seed weight.

Author Contributions: Conceptualization, N.G., V.I.K., H.P.S. and A.I.A.; methodology, N.G., V.I.K. and H.P.S.; software, N.G. and V.I.K.; validation, N.G., V.I.K. and H.P.S.; formal analysis, N.G. and V.I.K.; investigation, N.G. and V.I.K.; resources, N.G. and V.I.K.; data curation, N.G. and V.I.K.; writing—original draft preparation, N.G., V.I.K. and H.P.S.; writing—review and editing, N.G., V.I.K., H.P.S. and A.I.A.; visualization, N.G. and V.I.K.; supervision, N.G., V.I.K. and H.P.S.; project administration, N.G., V.I.K. and H.P.S.; funding acquisition, N.G., H.P.S., V.I.K. and A.I.A. All authors have read and agreed to the published version of the manuscript.

Funding: This research received no external funding.

Institutional Review Board Statement: Not applicable.

Informed Consent Statement: Not applicable.

Data Availability Statement: The original contributions presented in this study are included in the article. Further inquiries can be directed to the corresponding author.

Acknowledgments: The authors are very grateful to the anonymous reviewers whose valuable comments and suggestions improved the quality of the paper.

Conflicts of Interest: The authors declare no conflicts of interest.

Abbreviations

The following abbreviations are used in this manuscript:

GGE Genotype plus genotype $\times$ environment interaction
GEI Genotype $\times$ Environment Interaction

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