

1 INTRODUCTION

Regeneration and seedling establishment of maritime pine (*Pinus pinaster* Ait.) can potentially be affected by differences in cone, germination and seedling traits among seed sources. Minimum temperatures, which are important germination cues for pine species, are rising in the Iberian Peninsula as a consequence of global warming (Lorenzo et al. 2011), raising the question of how this change will affect natural regeneration of pine forests. In a previous study of seven mature *P. pinaster* populations (Correia et al. 2008), a significant correlation was found between growth and water-use efficiency (WUE), and of both traits with the mean minimal temperature for the coldest month of the populations origin; moreover, significant differences were detected between Bragança and Leiria seed sources, which represent opposite habitats for maritime pine in Portugal: 'inland high-altitude' and 'coastal low-altitude', respectively.

2 OBJECTIVES

- Further investigation of phenotypic variation in Bragança and Leiria provenances, specifically:
- (a) to determine the extent of provenance variation in cone traits, 100-seed weight and germination characteristics
 - (b) to assess the degree of genetic correlation and broad-sense heritability in those traits
 - (c) to examine the provenances' growth potential in the early post-germination period
 - (d) to compare, under nursery conditions, germination success and ontogeny of the two provenances to that of a seed orchard genetically originating from Leiria.

3 MATERIAL AND METHODS



Cone collection in Bragança and Leiria provenances and in a 1st generation seed orchard (open pollinated crosses of phenotypically selected Leiria's plus trees, for growth and form). Approximately 30 trees by population were evaluated for:

- cone and seed traits (cone height, diameter, weight and 100-seed weight;
- germination characteristics under laboratory conditions (8 replicates of 100-seed per mother-tree, parameter description in Table 2);
- early post germination growth under optimum controlled conditions (morphology and biomass allocation);
- germination success and ontogeny under nursery conditions.

Statistical analyses were conducted in a mixed model framework (Correia et al. 2014).

Table 1 - Altitude and climate data for the Bragança and Leiria provenances

| | Altitude (m) | T (°C) | m _{min} (°C) | m _{max} (°C) | P (mm) |
|----------|--------------|--------|-----------------------|-----------------------|--------|
| Bragança | 800 | 12.2 | 1.1 | 24.6 | 742.7 |
| Leiria | 55 | 14.7 | 5.6 | 27.1 | 806.4 |

Note: m_{min} = mean minimal temperature for the coldest 3 months (Dec, Jan, Feb); m_{max} = mean minimal temperature for the hottest 3 months (Jul, Aug, Sep)

Potential of adaptation to a changing climate of *Pinus pinaster* provenances from contrasting altitudes

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Table 2 - F-statistic and least squares mean ± standard error for the analysis of (A) cone and seed traits, and (B) germination parameters, between Bragança and Leiria provenances (Prov). The effect of dormancy-breaking treatment (DB) and of the interaction Prov*DB is shown for germination parameters.

| | | | Mean values±SE | | |
|--|------------------------------|------------|----------------|-------------|-------------|
| | (A) Cone traits | Effect | F-statistic | Bragança | Leiria |
| | Cone height (cm) | Prov | 7.87** | 11.77±0.25 | 12.76±0.25 |
| | Cone width (cm) | Prov | 28.04*** | 5.78±0.06 | 5.31±0.06 |
| | Cone weight (g) | Prov | 0.65 | 151.13±6.57 | 143.59±6.65 |
| | 100-seed weight (g) | Prov | 33.93*** | 6.70±0.18 | 5.20±0.18 |
| | (B) Germination parameters | | | | |
| | FG (%) | Prov | 43.59*** | | |
| | | DB | 1064.80*** | | |
| | | Prov*DB | 1.23 | | |
| | | Prov DB | 36.45*** | 0.89±0.028 | 0.68±0.028 |
| | | Prov no DB | 44.24*** | 0.577±0.028 | 0.291±0.028 |
| | PV (%) | Prov | 48.35*** | | |
| | | DB | 1186.29*** | | |
| | | Prov*DB | 6.27** | | |
| | | Prov DB | 54.53*** | 0.087±0.003 | 0.049±0.003 |
| | | Prov no DB | 34.65*** | 0.032±0.032 | 0.016±0.016 |
| | GV (%) | Prov | 38.41*** | | |
| | | DB | 951.39*** | | |
| | | Prov*DB | 51.28*** | | |
| | | Prov DB | 17.39*** | 0.043±0.001 | 0.032±0.001 |
| | | Prov no DB | 61.10*** | 0.027±0.001 | 0.014±0.001 |
| | T ₅₀ (n. days) | Prov | 21.88*** | | |
| | | DB | 403.15*** | | |
| | | Prov*DB | 0.57 | | |
| | | Prov DB | 26.75*** | 7.88±0.48 | 11.68±0.56 |
| | | Prov no DB | 12.75*** | 15.08±0.51 | 18.36±0.77 |
| | T ₇₅₋₂₅ (n. days) | Prov | 1.55 | | |
| | | DB | 4.72* | | |
| | | Prov*DB | 71.86*** | | |
| | | Prov DB | 16.52*** | 5.98±0.59 | 9.48±0.63 |
| | | Prov no DB | 2.40 | 9.06±0.59 | 7.65±0.69 |
| | TZ (%) | Prov | 20.55*** | 0.908±0.034 | 0.711±0.033 |

FG = Final germination % on day 21; PV = Peak value (maximum % cumulative germination / day); GV = Germination value (final % germination / 21 days); T50 = Number of days until 50% germination ; T75-25 = Number of days with 25% to 75% germination. TZ = Viable seed % estimated by the tetrazolium test. Significant differences are represented by P < 0.05; *P < 0.01; **P < 0.001; ***P < 0.0001.

Figure 1 - Variance components as the % of total variance for (A) cone traits and 100-seed weight and (B) germination parameters. Family heritabilities (±SE) were estimated by provenance and, for germination parameters FG, PV, GV, T50 and T75-25, are presented only for pre-treated seeds. σ²_{prov} = provenance variance, σ²_{fam} = among-family variance, σ²_{res} = residual variance, σ²_{DB} = dormancy-breaking variance, h²_{fam} = among-family heritability, estimated by provenance. Germination acronyms as in Table 2.

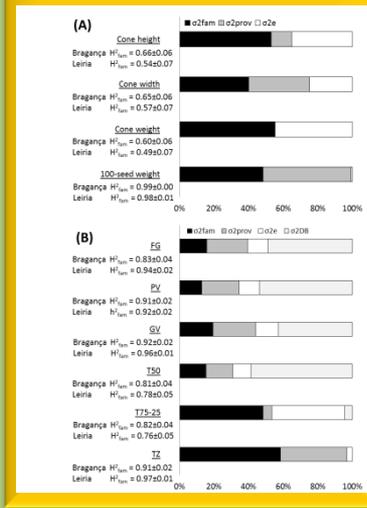


Figure 2 - Allometric relationships for log-transformed root:shoot ratio for Bragança and Leiria (3 months after seeding). The allometric slopes (±SE) are included. No significant differences at P<0.05 were detected between the fitted intercepts or slopes.

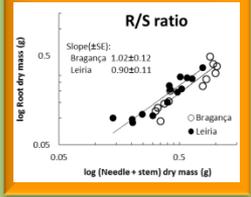
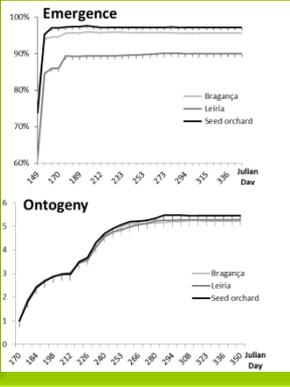


Figure 3 - Weekly population mean values in emergence % and ontogenic stage (notation scale) in the nursery, for Bragança and Leiria provenances and a seed orchard (phenotypically selected Leiria trees); at July day 350, 16 December, the plants were 7-months old.



4 RESULTS

Significant differences were found between the two provenances in most traits. High-altitude Bragança, where minimum temperatures possibly exert a selection pressure, had larger cones and higher seed weight, higher germination capacity and speed than coastal Leiria. Cone and germination traits were genetically correlated (data not shown) and were under a strong genetic control, indicating an evolutionary potential to cope with a changing climate. Early growth under optimal conditions pointed to higher biomass values for Bragança, but did not reveal allometric differences between the two provenances. Under nursery conditions, the seed orchard reached the highest germination success (97.1%) and differed marginally from Leiria, its genetic origin; population differences in the ontogenic development were not detected.

5 CONCLUSIONS

The high- and low-altitude provenances, which showed, in a previous study, significant differentiation in adaptive traits at a mature stage (growth and WUE) also differ in traits directly affecting regeneration (cone, seed and germination traits), probably as a response to different minimum temperatures at the provenances origin, mediated by maternal effects. The distinct strategies displayed by these provenances can be explored when allocating genetic resources at a regional level, for management or conservation purposes. To potentiate resilience in the face of climate change, *in situ* conservation areas and seed orchards should be established in high- and low-altitude sites, and, when applicable, seed zones should be defined by elevation bands.

REFERENCES

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