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Germination behavior in some Sicilian species of the *Euphorbia* genus

Abstract

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Our work concerns the requirements for germination in five *Euphorbia* species, occurring in uncultivated wet grounds, roadsides, montane pastures or deciduous broadleaf woods. The study taxa are *Euphorbia ceratocarpa*, *E. gasparrinii* subsp. *gasparrinii*, *E. hirsuta*, *E. meuselii*, and *E. platyphyllos*.

Seeds were collected from wild populations both in lowland and mountain areas of Eastern Sicily, at the time of natural dispersal. The germination ability was tested under constant and alternating temperatures, both in light and total dark, at the Catania Germplasm Bank (BGS-CT). The best germination protocols for each species are provided. Specifically, germination response was strongly limited by constant thermal regimes (except for *E. gasparrinii*) and, conversely, improved by fluctuating temperatures, under darkness, in all the study species. *E. gasparrinii* and *E. platyphyllos* reached high germination values also in the light.

Key words: germination protocols, constant temperature, alternating temperature, endemic species, Sicily.

Introduction

The *Euphorbia* genus is the largest one within the *Euphorbiaceae* family and includes approximately 2,000 species (Riina & al. 2013). Seed physiological dormancy has been reported for at least seventeen out of twenty *Euphorbia* species, which were reviewed by Baskin & Baskin (2014).

This work presents new germination data relating to five *Euphorbia* species of the Sicilian flora, which includes approximately thirty of them. Three of the study taxa, i.e., *E. ceratocarpa* Ten., *E. gasparrinii* Boiss. subsp. *gasparrinii*, and *Euphorbia meuselii* Geltman, are Italian endemics, whereas the other two species, *E. hirsuta* L. and *E. platyphyllos* L., have a Mediterranean distribution range. The germination traits of *E. ceratocarpa* and *E. meuselii*, together with other six congeneric species, were investigated in a recent essay (Cristaudo & al. 2019) that focused on Sicilian populations other than the current ones and obtained lower final germinated proportions in both species.

Seeds were collected between June and July 2021 and, within one month from the collection ($22 \pm 2^\circ\text{C}$ and 50% RH storage conditions), germination trials started, except for *E. gasparrinii* (tested within two months and a half). Six different constant (from 5 to 30°C) and five alternating temperature conditions (15/10, 20/10, 20/15, 25/15, and $25/20^\circ\text{C}$) were tested, both in light/dark (12/12 h photoperiod) and darkness (0/24 h). Light and alternating temperatures were assessed because of their well-known role in breaking dormancy and promoting germination (Probert 2000; Catara & al. 2016).

70. *Euphorbia ceratocarpa* Ten. (*Euphorbiaceae*)

Accession data

Si: Belpasso (Catania), Madonna della Roccia (WGS84: 37.601739°N , 15.002731°E), roadside, 650 m a.s.l., 28 Jun 2021, *R. Galesi* (SiMaSeed/CT/21/777, BGS-CT).

Germination data

Pre-treatments: no treatment.

Germination medium: 3 sheets of sterilized filter paper (Whatmann No. 1), imbibed with 6 ml of sterilized distilled water.

Sample size: 100 seeds (25×4 replicates).

Germination	Thermoperiod	Photoperiod [light/dark]	T1 [d]	T50 [d]	Tmax [d]	MTG [d]
87%	alternating 25/15°C	0/24 h	-	-	-	-
85%	alternating 20/10°C	0/24 h	-	-	-	-
83%	alternating 20/15°C	0/24 h	-	-	-	-

Observations

E. ceratocarpa is endemic to Sicily, Apulia, and Calabria, being quite common only in the first of the three Italian regions, where it is frequent, mainly up to 700 m a.s.l., in disturbed plots and along roadsides, keeping a high soil humidity.

Final germination values under constant temperatures did not exceed 10%, regardless of photoperiod conditions, except for 15°C under light/dark (ca. 40%), proving that fresh seeds of *E. ceratocarpa* have a strict requirement for alternating thermal regimes and that, additionally, light can only very slightly substitute them. Moreover, alternating temperatures provided the best performance only under dark conditions, whereas a strong photoinhibition effect appeared in light/dark conditions (final germination less than 30%). The only meaningful T_{50} value (9.5 days at 15°C , light/dark) is in agreement with what has been found in other study species.

The requirement for alternating thermal regimes had not been highlighted in the fresh seeds of this species. In addition, the present work provides the first data on the germination behavior of a further Sicilian population of *E. ceratocarpa*, in addition to that from hilly ranges of Strasatto (Palermo), for which lower germination values were obtained (Cristaudo & al. 2019).

71. *Euphorbia hirsuta* L. (*Euphorbiaceae*)**Accession data**

Si: Augusta (Siracusa), Tenuta Grande-Gelsari (WGS84: 37.334078°N, 15.081472°E), uncultivated wet grounds, 1 m a.s.l., 06 Jul 2021, *R. Galesi* (SiMaSeed/CT/21/779, BGS-CT).

Germination data

Pre-treatments: no treatment.

Germination medium 3 sheets of sterilized filter paper (Whatmann No. 1), imbibed with 6 ml of sterilized distilled water.

Sample size: 100 seeds (25 × 4 replicates).

Germination	Thermoperiod	Photoperiod [light/dark]	T1 [d]	T50 [d]	Tmax [d]	MTG [d]
100%	alternating 25/15°C	0/24 h	-	-	-	-
97%	alternating 20/10°C	0/24 h	-	-	-	-
93%	alternating 20/15°C	0/24 h	-	-	-	-
87%	alternating 25/20°C	0/24 h	-	-	-	-
83%	alternating 25/15°C	12/12 h	9	10.6	23	11.8

Observations

E. hirsuta is a Mediterranean species, ranging from the Macaronesian region to the Caucasus (Georgia) (POWO 2021). It grows in damp and uncultivated places, as well as along riverbanks.

The germination process in the fresh seeds of this species strictly required alternating thermal regimes, providing a germination percentage over 80% also at 25/20°C in the dark, unlike all the other study species. Photoinhibition characterized germination behavior in the light/dark conditions under alternating regimes, except at 25/15°C, where seeds showed a T_{50} value of approximately 11 days. The presence of light was not able to substitute alternating regimes under constant temperatures. Finally, *E. hirsuta* appeared as one of the two study species germinating across the widest thermal range, together with *E. platyphyllos* (see report n. 73, below), since a quite high germination value (67%) has been reached also at 15/10°C in the dark.

72. *Euphorbia meuselii* Geltman (*Euphorbiaceae*)**Accession data**

Si: Messina (Messina), Pizzo Chiarino (WGS84: 38.192111°N, 15.484194°E), mixed reforestation with coniferous and broadleaf trees, 788 m a.s.l., 10 Jun 2021, *A.*

Cristaudo, F. Carruggio, M. Castrogiovanni & R. Galesi (SiMaSeed/CT/21/757, BGS-CT).

Germination data

Pre-treatments: no treatment.

Germination medium: 3 sheets of sterilized filter paper (Whatmann No. 1), imbibed with 6 ml of sterilized distilled water.

Sample size: 100 seeds (25 × 4 replicates).

Germination	Thermoperiod	Photoperiod [light/dark]	T1 [d]	T50 [d]	Tmax [d]	MTG [d]
93%	alternating 20/15°C	0/24 h	-	-	-	-
85%	alternating 20/10°C	0/24 h	-	-	-	-

Observations

E. meuselii is endemic to the main Italian islands (Sicilia and Sardinia) and two southern regions, Basilicata and Calabria. It grows in deciduous oak and beech woods, at an elevation from 600 to 1500 m a.s.l.

Our tests highlighted an overall germination behavior similar to those of the other study species, i.e., an obligate requirement for alternating thermal regimes. However, a general preference for lower temperatures could be noted (79 vs. 54% final germination percentage, at 15/10 and 25/15°C, respectively, in darkness). In addition, rather high germination values have been recorded under alternating temperatures also in the light/dark (71% final germination percentage, at 20/10 and 20/15°C), although in connection with T_{50} values of 13-15 days. Our data do not confirm the requirement of mechanical manipulations and GA_3 in promoting seed germination, differently from some protocols reported by the Seed Information Database (Royal Botanic Gardens Kew 2021). Conversely, they support the results obtained by Cristaudo & al. (2019) for a different population from Madonie Mountains (Palermo), which reached the highest germination value (ca. 60%) at 8/25°C (16.5°C average) in long-stored seeds.

73. *Euphorbia platyphyllos* L. (*Euphorbiaceae*)

Accession data

Si: Augusta (Siracusa), Tenuta Grande-Gelsari (WGS84: 37.334078°N, 15.081472°E), uncultivated wetlands, 1 m a.s.l., 08 Jun 2021, *R. Galesi* (SiMaSeed/CT/21/751, BGS-CT).

Germination data

Pre-treatments: no treatment.

Germination medium: 3 sheets of sterilized filter paper (Whatmann No. 1), imbibed with 6 ml of sterilized distilled water.

Sample size: 100 seeds (25 × 4 replicates).

Germination	Thermoperiod	Photoperiod [light/dark]	T1 [d]	T50 [d]	Tmax [d]	MTG [d]
100%	alternating 20/10°C	0/24 h	-	-	-	-
98%	alternating 20/10°C	12/12 h	6	6.2	8	6.8
97%	alternating 20/15°C	12/12 h	5	4.9	8	5.6
95%	alternating 25/15°C	12/12 h	5	4.8	9	5.6
92%	alternating 20/15°C	0/24 h	-	-	-	-

Observations

E. platyphyllos is an annual Euri-Mediterranean weed and has spread in both North and South America (POWO 2021).

Alternating temperatures proved to be efficient in promoting seed germination also in this species, providing good performance within a wide thermal range, as in *E. hirsuta*. Indeed, in addition to the data shown in the table above, final germination values exceeded 70% also at 15/10 and 25/15°C in the dark. Alternating temperatures and light in combination provided very high germination values, in addition to T_{50} values of 5-6 days. However, the light confirms itself as an inefficient trigger for germination under constant temperatures also in *E. platyphyllos*. The only information available on germination requirements in this species concerns the combined use of GA₃ and alternating temperatures (33/19°C, light/dark 12/12, 78% final germination; Royal Botanic Gardens Kew 2021).

74. *Euphorbia gasparrinii* Boiss. subsp. *gasparrinii* (Euphorbiaceae)

Accession data

Si: Alcara Li Fusi (Messina), Lago Maulazzo (WGS84: 37.942736°N, 14.672809°E), montane pastures, 1453 m a.s.l., 10 Jul 2015, *A. Cristaudo* (SO.PRO.ME/CT/15/985, BGS-CT).

Germination data

Pre-treatments: no treatment.

Germination medium: 3 sheets of sterilized filter paper (Whatmann No. 1), imbibed with 6 ml of sterilized distilled water.

Sample size: 100 seeds (25 × 4 replicates).

Germination	Thermoperiod	Photoperiod [light/dark]	T1 [d]	T50 [d]	Tmax [d]	MTG [d]
90%	alternating 20/10°C	12/12 h	7	9.7	20	10.9
85%	alternating 20/15°C	12/12 h	7	8.7	18	9.5
85%	alternating 15/10°C	12/12 h	9	10.9	15	11.5

Observations

E. gasparrinii is an Italian endemic species showing a disjunct distribution across two main mountain ranges, i.e., the central Apennines and the Calabrian-Sicilian area, growing between 800 and 1850 m a.s.l. Central Italian populations have been ascribed to the subsp. *samnitica* (Fiori) Pignatti and are rather numerous (Cresti & al. 2019). Conversely, the southern populations have been attributed to the subsp. *gasparrinii* and have become rarer over the last decades, to the point that the study population of Lago Maulazzo (Nebrodi Mountains) may be probably considered the only existing one (Cresti & al. 2019).

The germination behavior of the present population confirmed, as in the other species, the good performance of alternating temperature regimes. Our results suggested that light has an amplifying effect on germination under fluctuating temperatures, although final germination values higher than 70% were recorded also in the dark (73 and 77% at 15/10 and 20/10°C, respectively). Fresh seeds required ca. 10 days to reach 50% germination. *E. gasparrinii* strongly differed from all the other study species because of high germination values under some constant temperature regimes, regardless of the photoperiod applied. Indeed, at 15 and 20°C, values of 73 and 70% in the light/dark and 58 and 64% in the dark were recorded, respectively. This behavior showed that light partially compensated for the lack of alternating regimes.

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