Scientific Paper

Effect of controlled conditions on the germination of five *Morus alba* L. varieties♦

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Abstract

The objective of this study was to determine the effect of substrate temperature and light/darkness alternation, on the germination response of fresh seeds (sexually obtained) of five *Morus alba* L. varieties (cubana, tigreada, universidad, universidad mejorada and yu-62) harvested in Matanzas –Cuba–, from two different lots. In the essays a completely randomized design was used; the percentage data of final germination and mean germination time were processed through a simple-classification variance analysis (ANOVA), with factorial arrangement of the treatments. The germination was evaluated at fixed temperature (25 ºC) and in four thermoperiods (25/30 ºC, 25/35 ºC, 25/40 ºC and 25/45 ºC), under two conditions (light and darkness). The seeds did not show dormancy; the highest percentages of final germination for all the varieties were reached at 25/35 ºC with light, although high germination percentages were also obtained in total darkness when the seeds were placed at 25/40 ºC. On the other hand, the fixed temperature of 25 ºC caused in all the varieties a thermoinhibition of germination. The high diversity of germination responses in the studied *M. alba* varieties, in each harvest date, was shown; which indicates that the germination process can occur under different environmental conditions, particularly where there is a wide fluctuation of temperature.

Keywords: light, seeds, temperature

Introduction

Although not enough is known yet about the environmental conditions required for the germination of the cultivated plants, which is partly due to the fact that man's action has favored the selection of fast-germination genotypes for different environments (Smýkal *et al*., 2014; Dürr *et al*., 2015). Thus, plant domestication promotes traits in the seeds which differ from those of their parents or wild relatives. However, having knowledge about the germination requirements of plants helps the development of already established crops and the creation of new varieties/accessions (Shalimu *et al*., 2015).

In Cuba, germination studies of economically important plants for men in agricultural systems are referred to a large extent to such crops as rice (*Oryza sativa* L.) and tomato (*Solanum lycopersicum* L.), in which the effect of substrate salinity and temperature on the germination process and seedling development, has been determined, for some varieties (Sánchez *et al*., 2001; Lamz and González, 2015); this aspect has also been studied, in developing countries as well as in nations of the so-called First World (Górski *et al*., 2013; Tribouillois *et al*., 2016).

Seed germination is a critical stage of the life cycle of plants, which influences crop growth and production, and can be affected by different abiotic and biotic components (Finch-Savage and Bassel, 2015). Among them, harvest conditions and seed obtainment methods; differences among populations, accessions and lots (maternal effects); and light, temperature and substrate moisture, stand out, only to name a few (Baskin and Baskin, 2014).

Variability in germination is so diverse that, sometimes, it is mistakenly classified as the response of one species to an element of the environment; for example, when this process occurs in the presence of light (Górski *et al*., 2013). This indicates, on the one hand, the different regeneration strategies of a plant species to face environmental uncertainty; and, on the other hand, the artificial selection crops have undergone for several centuries (Baskin and Baskin, 2014; Mitchell *et al*., 2016).

♦This result corresponds to the project «Studies with *Morus* sp. for the development of sustainable human and animal feeding and health technologies in Cuba». National Program of Science and Technology «Feed production», of the Ministry of Science, Technology and Environment, Cuba.

Mulberry (*Morus alba* L. Moraceae) is a multipurpose cultivated plant, which originated in India and Central Asia that has been introduced almost throughout the world, mainly because of its importance in the silk industry and its forage value (Martín *et al*., 2014); and the asexual propagation method (by stakes/cuttings) is the most widely used one for this species (Olivera and Noda, 2010).

Nevertheless, propagation by seeds (sexual way) not only constitutes the most economical and simple method, but also ensures genetic diversity and with it the species survival and production under continuous environmental changes (Fenner and Thompson, 2005; Finch-Savage and Bassel, 2015). However, the existing information about the dormancy classes and germination requirements of *M. alba* seeds is contradictory. On the one hand, it is stated that fresh (cultivated/wild) seeds show physiological dormancy (Barbour *et al*., 2008), which is the same one present in its relative *Morus nigra* (Konyuncu, 2005) and in most species of the *Moraceae* family (Willis *et al*., 2014); the presence of physical dormancy in fresh *M. alba* seeds has also been reported (Barbour *et al*., 2008).

Górski *et al*. (2013) stated that germination is reduced due to the low red:far-red relation and Permán *et al*. (2013) acknowledged that fresh seeds do not require pre-treatment to germinate (Permán *et al*. 2013).

In Cuba, propagation by cuttings is the most widely extended method for *M. alba* (Martín *et al*., 2014), and only recently the procedure for its sexual reproduction was introduced. Thus, the objective of this study was to determine the effect of substrate temperature and light-darkness alternation, on the germination response of fresh seeds of five *M. alba* varieties harvested in the country, from two different lots.

Materials and Methods

Plant material. The fresh (newly harvested) *M. alba* seeds were from the genetic resource bank of the Pastures and Forages Research Station Indio Hatuey. The seeds corresponding to five varieties (cubana, tigreada, universidad, universidad mejorada and yu-62) which had been introduced from Ethiopia, Costa Rica and China (Martín *et al*., 2014), and harvested in March, 2014 (lot 1), and February, 2015 (lot 2), from a plantation aimed at seed production, were used. Seed extraction and cleaning was done manually, through depulping and separation with a sieve and rinsing with tap water; and then they were dried at ambient temperature and under shade during two or three days. Their viability percentage was determined by the tetrazolium test (ISTA, 2007).

Germination requirements. To achieve the germination response under laboratory conditions a completely randomized design was used. Ten treatments were essayed with the combination of five thermoperiods [constant temperature of 25 ºC and in alternate regimes of 25/30 ºC, 25/35 ºC, 25/40 ºC and 25/45 ºC (8 hours for the highest temperature of the thermoperiod and 12 hours at 25 ºC, with 4-hour transition between them)] and the light-darkness alternation [exposure to light during 8 h at 40 µmol m^{-2} s^{-1} approximately and at a length of 400-700 nm, coinciding with the highest temperature period within each treatment; and to total darkness (constant), which was achieved wrapping the plates with two layers of aluminum foil]. The illumination was the one recommended in the internationally established rules (ISTA, 2007). The conditions to which the seeds were subject simulated the possible variations of these climate elements in the soil of a field area, according to the criteria expressed by Sánchez *et al*. (2011). The position of the plates in the incubators was changing regularly.

The seeds, before sowing, were sterilized with mercury bichloride (1 g L^{-1}) to prevent fungal contamination. Five replications of 25 seeds each were used per treatment. The seeding was carried out in hydrostatic agar at 1 % on 9-cm diameter Petri dishes. In the case of the illuminated seeds, the germination count was made daily during 30 days. The germination count in the seeds exposed to continuous conditions of total darkness was determined three days after concluding germination under light. The criterion for germination was radicle emergence. In the treatments under light, the agar was maintained humid throughout the experiment. The final germination percentage under light and darkness was determined, and for the light condition, the mean germination time (days) according to the methodology proposed by Ranal *et al*. (2009).

The final germination percentage under light and darkness was used to calculate the light related germination (LRG) index, which showed the light requirements for germination (Milberg *et al*., 2000). This index was calculated according to the formula LRG = GL/(GD + GL), where GL = germination percentage under light, and GD = germination percentage in darkness. To obtain the LRG index of each variety the optimum temperature range was used (i.e. the one in which the highest germination percentage under light or darkness was recorded). The LRG values varied between 0 (seeds that germinated only in the dark) and 1 (seeds that germinated only under light). If the LRG index was higher than 0,75 it was considered that the species/variety was light dependent (positive photoblastic), if it was lower than 0,25 it was light repellent (negative photoblastic), and if the value was between 0,25 and 0,75 it was established as light indifferent (Funes *et al*., 2009).

At the end of the germination tests, the percentage of non-germinated live seeds was determined by pressing the seeds with a needle to establish whether they contained a white and firm embryo (live seeds) or a soft or gray embryo, the latter indicative of seed unviability –dead seeds (Baskin and Baskin, 2014). For the non-germinated ones in the temperatures 25 ºC and 25/30 ºC, this variable was established through re-seeding in the optimum germination temperature (see Results section) and under white light. Re-seeding was performed to determine whether the seeds acquired thermoinhibition or thermodormancy due to inadequate temperature conditions; it was considered that they acquired thermoinhibition if there were high germination percentages, according to the criterion expressed by Geneve (2005).

Assignation of dormancy classes. It was considered that the species had dormant seeds if their germination started after 28 days under optimum germination conditions (Baskin and Baskin, 2014), or if at least 20 % of the seeds remained alive and without germinating at the end of the germination experiment (Sánchez *et al*., 2015).

Statistical analysis. The statistical processing was made independently for each harvest year (seed lot). The percentage data of final germination and mean germination time were processed through simple classification variance analysis (ANOVA), with factorial arrangement of the treatments; the germination percentage were transformed with the arcsin of the square root of the proportion. No multiple comparison tests of means were applied *a posteriori*, because some independent variables represented combinations of quantitative treatments (light-darkness alternation and temperature). The statistical analyses were made by the program InfoStat v. 2015 (Di Rienzo *et al*., 2015).

Results

Germination requirements. The variance analysis showed that the final germination percentages in both lots of fresh *M. alba* seeds were the result of the highly significant interaction which was established among the varieties, substrate temperatures and light-darkness alternation (table 1). In addition, first-order interactions (variety x temperature, variety x light and temperature x light) and the main effects also played a significant role in germination.

Table 1. ANOVA for the main factors and their interactions in final germination

Variation source	*F*-Test	
	Lot 2014	Lot 2015
Variety	143,3***	271,9***
Temperature	215,9***	308,7***
Light	502,5***	223,7***
Variety x temperature	20,4***	38,3***
Variety x light	15,5**	16,7**
Temperature x light	91,3***	68,9***
Variety x temperature x light	7,0**	8,2**

** $p < 0{,}01$; *** $p < 0{,}001$.

The highest percentages of final germination in all the varieties of each lot were obtained in the alternate temperature range of 25/35 ºC with light (table 2). However, in the case of the tigreada variety of 2014, with light, also the temperatures of 25/40 ºC and 25/45 ºC were adequate to increase germination, although in this last range seedling emergence did not occur. On the other hand, in the university variety of 2014 significant increases of germination were obtained in the seeding at 25/30 ºC under light. Under total darkness conditions high final germination percentages were obtained equally for all varieties when seeding was carried out at 25/40 ºC for the seeds of the 2014 lot, and at 25/35 ºC and 25/40 ºC for the seeds of the 2015 lot.

Instead, the lowest percentages of final germination were reached, for all the varieties and lots, at the constant temperature of 25 ºC and the alternate one of 25/30 ºC, effect that was higher in the cubana and tigreada varieties, under light as well as total darkness conditions (table 2). Under these seeding conditions a large number of non-germinated live seeds also appeared which reached high germination percentages when they were put at alternate temperature of 25/35 ºC.

In general, the fresh seeds of the varieties cubana of 2014 and yu-62 of 2015 showed the lowest percentages of final germination (table 2), performance which was also shown when the effects of the main factors were separately analyzed (table 3);

Table 2. Final germination percentage (mean ± SE) of fresh *M. alba* seeds under different seeding conditions

Lot/variety	Germination percentage				
	25 ºC	25/30 ºC	25/35 ºC	25/40 ºC	25/45 ºC
2014	Light				
Cubana	28,0 ± 4,6	33,3 ± 9,3	34,6 ± 5,8	24,0 ± 4,6	24,0 ± 2,3
Tigreada	5,3 ± 1,3	81,3 ± 2,6	96,0 ± 2,3	96,0 ± 0,8	96,0 ± 0,9
Universidad	68,0 ± 1,1	92,0 ± 2,3	93,3 ± 2,6	81,3 ± 4,8	86,6 ± 1,3
Universidad mejorada	46,6 ± 8,1	70,6 ± 10,6	82,6 ± 4,8	82,7 ± 7,4	44,0 ± 4,6
Yu-62	50,6 ± 5,3	81,3 ± 1,3	90,6 ± 3,5	82,6 ± 1,3	88,0 ± 6,1
2014	Darkness				
Cubana	5,3 ± 1,3	10,6 ± 2,6	25,3 ± 2,6	32,0 ± 10,0	22,3 ± 5,0
Tigreada	0	0	21,3 ± 1,3	98,6 ± 1,3	82,6 ± 2,6
Universidad	8,6 ± 4,3	18,0 ± 1,0	54,6 ± 3,5	94,6 ± 2,6	64,0 ± 4,0
Universidad mejorada	1,3 ± 0,8	2,6 ± 1,3	20,0 ± 8,3	89,3 ± 1,3	28,0 ± 9,6
Yu-62	9,3 ± 1,3	6,6 ± 2,6	69,3 ± 7,0	96,0 ± 2,3	72,0 ± 4,0
2015	Light				
Cubana	2,6 ± 1,3	86,6 ± 1,3	100	96,0 ± 2,3	90,6 ± 3,5
Tigreada	9,3 ± 2,6	53,3 ± 1,3	73,3 ± 5,3	52,0 ± 4,0	5,3 ± 1,3
Universidad	61,3 ± 2,6	80,0 ± 8,3	89,3 ± 4,8	70,6 ± 1,3	8,0 ± 2,3
Universidad mejorada	41,3 ± 6,6	73,3 ± 4,8	78,6 ± 1,3	73,3 ± 3,5	18,6 ± 4,8
Yu-62	10,6 ± 1,3	9,3 ± 3,5	18,0 ± 1,3	8,0 ± 4,0	1,3 ± 0,2
2015	Darkness				
Cubana	0	8,0 ± 2,3	97,0 ± 1,5	97,3 ± 2,6	84,0 ± 4,6
Tigreada	0	2,6 ± 1,0	48,0 ± 4,6	29,3 ± 3,5	1,3 ± 1,0
Universidad	4,0 ± 2,3	13,3 ± 1,3	70,6 ± 1,3	84,0 ± 2,3	10,6 ± 3,5
Universidad mejorada	1,3 ± 1,3	4,0 ± 2,3	62,6 ± 1,3	70,6 ± 8,7	40,0 ± 4,6
Yu-62	1,3 ± 1,0	10,6 ± 2,6	10,3 ± 1,3	12,6 ± 3,5	0

The percentage data shown are in correspondence with the original ones.

the tigreada and universidad varieties of the 2014 lot showed a higher performance than those collected in 2015. Universidad mejorada showed a similar performance between the years. Finally, the effect of the light factor, as germination stimulator, was higher for both years, although its result was more marked in the 2014 seeds (table 3).

The mean germination time (or necessary time to reach 50 % of the germinated seeds) for both seed lots was significantly affected by the interaction of the factors variety and temperature, and also by the main effects (table 4). The lowest time to germinate, independently from the variety and collection time, was always reached at the alternate range of 25/35 ºC (table 5), with a range from 2,4 ± 0,1 days (in yu-62 of 2014) to 7,1 days in tigreada and yu-62 of 2015. It was also observed that from 25/35 ºC germination was slower, particularly in the range of 25/45 ºC, in which it tended to concentrate in a few days. The above-explained facts showed that the 25/35 ºC thermoperiod was the optimum one for the germination of all varieties, because with it the maximum percentages of final germination were achieved in the lowest possible time. It can be stated that, for all the varieties of 2014, the onset of germination started after the second day of seeding at 25/30 ºC and 25/35 ºC; and for the 2015 varieties, at the same temperatures, germination started on the third day for cubana and universidad, on the fourth day for tigreada and universidad mejorada, and on the fifth day for yu-62.

The light-related germination index (LRG) in the temperature of 25/35 ºC was higher in all the 2014 varieties, except in yu-62, with regards to the

Table 3. Effect of the main factors (variety, temperature and light) on the final germination percentage (mean ± SE)

Variety	Final germination (%)	
	2014 lot	2015 lot
Cubana	23,3 ± 2,2	66,2 ± 7,7
Tigreada	57,7 ± 7,8	27,4 ± 4,8
Universidad	66,1 ± 5,5	49,2 ± 6,3
Universidad mejorada	46,8 ± 6,1	46,4 ± 5,3
Yu-62	64,6 ± 5,8	8,4 ± 1,3
Temperature	2014 lot	2015 lot
25 ºC	22,3 ± 4,4	13,2 ± 3,7
25/30 ºC	39,6 ± 6,7	34,1 ± 6,2
25/35 ºC	70,8 ± 5,6	68,5 ± 5,6
25/40 ºC	68,7 ± 4,9	64,5 ± 5,6
25/45 ºC	60,7 ± 5,2	27,8 ± 6,1
Illumination	2014 lot	2015 lot
Light	66,4 ± 3,3	48,0 ± 4,1
Darkness	37,2 ± 4,0	31,0 ± 3,9

The percentage data shown are in correspondence with the original ones.

Table 4. ANOVA for the main effects and their interactions in the mean germination time.

Variation source	*F*-test	
	2014 lot	2015 lot
Variety	9,4**	21,5***
Temperature	121,1***	71,5***
Variety x temperature	2,2**	3,7***

** $p < 0{,}01$; *** $p < 0{,}001$.

Table 5. Mean germination time (mean ± SE) under light and different substrate temperature.

Lot/variety	Seeding temperature				
2014	25 ºC	25/30 ºC	25/35 ºC	25/40 ºC	25/45 ºC
Cubana	4,6 ± 0,7	3,5 ± 0,7	3,3 ± 0,3	4,7 ± 0,4	10,5 ± 0,3
Tigreada	4,8 ± 0,9	5,1 ± 0,8	3,9 ± 0,1	4,7 ± 0,1	10,1 ± 0,2
Universidad	4,9 ± 0,0	3,7 ± 0,1	2,9 ± 0,1	4,3 ± 0,1	9,5 ± 0,4
Universidad mejorada	5,2 ± 0,6	3,2 ± 0,1	3,1 ± 0,1	4,8 ± 0,2	8,9 ± 0,5
Yu-62	4,6 ± 1,1	3,0 ± 0,1	2,4 ± 0,1	3,8 ± 0,2	6,2 ± 0,2
2015	25 ºC	25/30 ºC	25/35 ºC	25/40 ºC	25/45 ºC
Cubana	5,3 ± 3,1	5,8 ± 0,2	4,3 ± 0,1	5,4 ± 0,1	9,2 ± 0,3
Tigreada	7,5 ± 0,4	7,3 ± 0,4	7,1 ± 0,2	8,7 ± 0,1	18,3 ± 0,6
Universidad	6,5 ± 0,4	6,1 ± 0,2	5,3 ± 0,31	6,7 ± 0,1	12,0 ± 1,0
Universidad mejorada	8,5 ± 0,7	5,8 ± 0,1	5,5 ± 0,2	6,9 ± 0,9	11,1 ± 0,4
Yu-62	9,6 ± 1,2	7,4 ± 0,2	7,1 ± 0,8	8,17 ± 1,8	19,6 ± 0,3

The percentage data shown are in correspondence with the original ones.

values obtained for the 2015 seed lots (fig. 1). The varieties tigreada and universidad mejorada showed LRG values higher than 0,75; thus, they were photoblastic. In the other varieties, independently from the collection time, there were LRG values from 0,50 to 0,63, for which they were considered indifferent photoblastic.

Assignation of seed dormancy classes

The seeds did not show dormancy; in the optimum temperature the germination process started before seven days, and reached for most varieties more than 70 % of final germination, according to the initial viability of the lots (97 %). In addition, in the temperature considered as optimum the seeds which did not germinate were dead, according to the results of the cutting test.

Discussion

Under the experimental conditions, it was shown that the fresh *M. alba* seeds did not show physical or physiological dormancy, because they were hydrated and their germination started in less than 28 days; result that coincides with the report by Permán *et al*. (2013) for this species. It was also proven that the germination of the varieties can occur in a wide range of temperatures under light and darkness conditions. This aspect is possibly due to the domestication process of the species, which during centuries has favored that the seeds germinate faster and that there is a loss of dormancy. Similar domestication center has been reported for a large number of crops (Fuller and Allbay, 2009: Dürr *et al.,* 2015) and represents an advantage for the management and fast establishment of the harvested plants (Smýkal *et al.,* 2014). However, the diversity in the germination response could also be an advantage to face the uncertainty or environmental stress the seeds can find once they are planted under nursery conditions (Cochrane *et al.,* 2015; Mitchell *et al*., 2016). In fact, the stress conditions, in productive systems as well as natural systems, are the ones which the seeds commonly find when they reach the soil (Finch-Savage and Bassel, 2015; Gardarin *et al*., 2016).

The results of this study showed that the germination of *M. alba* can be reduced by the inadequate conditions of substrate temperature and illumination. The final germination significantly decreased at fixed temperature of 25 ºC and alternate temperature of 25/30 ºC, but when reseeding was done at 25/35 ºC high germination percentages were achieved, which proved the thermoinhibition process, as has been reported for many species (Geneve, 2005; Baskin and Baskin, 2014). Likewise, it can be stated that germination was always stimulated under white light conditions, which is due to the acknowledged photosensitivity for the seeds of different *M. alba* genotypes (Smýkal *et al*., 2014).

The substrate temperature seems to be the main factor that regulates the germination of the studied varieties, because thermoinhibition occurred in the seeds sown under white light as well as in those under total darkness conditions. Nevertheless, it was

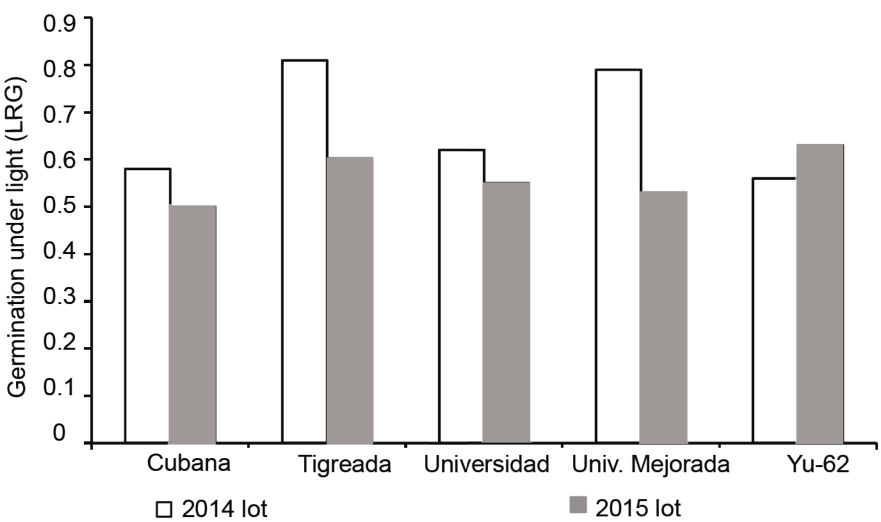

Figure 1. Light relative germination index for fresh *Morus alba* seeds.

also proven that at the optimum seeding temperature (25/35 ºC) the fresh seeds from tigreada and universidad mejorada behaved as positive photoblastic, according to the light relative germination index (Milberg *et al*., 2000). The above-explained facts prove that such varieties were the most sensitive to the light stress and to the low substrate temperatures, result which could have high practical implication in the reproduction of these varieties under nursery and field conditions.

Conclusions

There was high diversity of germination response in the studied *M. alba* varieties, in each harvest date; which indicates that the germination process can occur under different environmental conditions, particularly where there is wide temperature fluctuation. In addition, it could be observed that very high temperature ranges, such as 25/45 ºC, could be effective for germinating, but not for achieving seedling emergence. These elements should be taken into consideration if the species is to be sexually reproduced, because such diversity could attempt against obtaining uniform plantations, and it could also favor the adaptation of the species to different climate conditions.

It is recommended to determine the effects of the pre-germination hydration-dehydration treatments of these seeds, to synchronize and increase the germination and establishment of the species under stressful conditions (heat, drought, salinity, among others).

Bibliographic references

Barbour, J. R.; Read, R. A. & Barnes, R. L. *Morus* L. In: F. T. Bonner and R. P. Karrfalt, eds. *The woody plant seed manual*. Agriculture Handbook 727. Washington, DC: Department of Agriculture, Forest Service. p. 728-732, 2008.

Baskin, Carol C. & Baskin, J. M. *Seeds: ecology, biogeography and evolution of dormancy and germination*. 2 ed. San Diego, USA: Academic Press, 2014.

Cochrane, A.; Yates, C. J.; Hoyle, G. L. & Nicotra, A. B. Will among-population variation in seed traits improve the chance of species persistence under climate change? *Glob. Ecol. Biogeogr*. 24:12-24, 2015.

Di Rienzo, J. A.; Casanoves, F.; Balzarini, Mónica G.; Gonzalez, Laura; Tablada, Margot & Robledo, C. W. InfoStat versión 2015 Córdoba, Argentina: Grupo InfoStat, FCA, Universidad Nacional de Córdoba. http://www.infostat.com.ar. [13/12/2015], 2015.

Dürr, Carolyne; Dickie, J. B.; Yang, X. Y. & Pritchard, H. W. Ranges of critical temperature and water potential values for the germination of species worldwide: Contribution to a seed trait database. *Agric. For. Meteorol*. 200:222-232, 2015.

Fenner, M. & Thompson, K. *The ecology of seeds*. Cambridge, United Kingdom: Cambridge University Press, 2005.

Finch-Savage, W. E. & Bassel, G. W. Seed vigour and crop establishment: extending performance beyond adaptation. *J. Exp. Bot*. 67 (3):567-591, 2015.

Fuller, D. Q. & Allbay, R. Seed dispersal and crop domestication: shattering, germination and seasonality in evolution under cultivation. *Ann. Plant Rev*. 38:238-295, 2009.

Funes, G.; Díaz, Sandra & Venier, Paula. La temperatura como principal determinante de la germinación en especies del Chaco seco de Argentina. *Ecol. Austral*. 19 (2):129-138. http://www.scielo.org.ar/scielo.php?script=sci_arttext&pid=S1667-782X2009000200005&lng=es. [13/12/2015], 2009.

Gardarin, A.; Coste, F.; Wagner, Marine-Héléne & Dürr, Carolyne. How do seed and seedling traits influence germination and emergence parameters in crop species? A comparative analysis. *Seed Sci. Res*. 26 (4):317-331, 2016.

Geneve, R. L. *Some common misconceptions about seed dormancy*. Combined Proceedings International Plant Propagators' Society. USA: International Plant Propagators' Society, 2005.

Górski, T.; Górska, Krystyna & Stasiak, H. Inhibition of seed germination by far red radiation transmitted through leaf canopies. *Pol. J. Agr.* 13:10-38, 2013.

ISTA. *International rules for seed testing*. Bassersdorf, Switzerland: International Seed Testing Association, 2007.

Koyuncu, Fatma. Breaking seed dormancy in black mulberry (*Morus nigra* L.) by cold stratification and exogenous application of gibberellic acid. *Acta Biol. Cracov. Series Botánica*. 47 (2):23-26, 2005.

Lamz, A. & González, María C. Indicadores de crecimiento inicial y del estado nutricional para la selección temprana de genotipos de arroz (*Oryza sativa* L.) tolerantes a la salinidad. *Cultivos Tropicales*. 36 (2):41-48, 2015.

Martín, G. J.; Pentón, Gertrudis; Noda, Yolai; Contino, Y.; Díaz, Maykelis; Ojeda, F. *et al.* Comportamiento de la morera (*Morus alba* L.) y su impacto en la producción animal y la crianza de gusanos de seda en Cuba. *Rev. cubana Cienc. agríc*. 48 (1):73-78, 2014.

Milberg, P.; Andersson, L. & Thompson, K. Large-seeded species are less dependent on light for germination than small-seeded ones. *Seed Sci. Res*. 10 (1):99-104, 2000.

Mitchell, J.; Johnston, I. G. & Bassel, G. W. Variability in seeds: biological, ecological, and agricultural implications. *J. Exp. Bot*. 68 (4):809-817, 2017.

Olivera, Yuseika & Noda, Yolai. Origen, distribución, características botánicas, especies y variedades. En: Milagros Milera, comp. *Morera. Un nuevo forraje para la alimentación del ganado*. Matanzas, Cuba: EEPF Indio Hatuey. p. 9-19, 2010.

Permán, J.; Navarro, R.; Nicolás, J. L.; Prada, María & Serrada, R. *Producción y manejo de semillas y plantas forestales*. Madrid: Organismo Autónomo Parques Nacionales, Ministerio de Agricultura, Alimentación y Medio Ambiente, 2013.

Ranal, Marli A.; Santana, Denise G. de; Ferreira, Wanessa R. & Mendes-Rodrigues, C. Calculating germination measurements and organizing spreadsheets. *Revista Brasil. Bot*. 32 (4):849-855, 2009.

Sánchez, J. A.; Montejo, Laura; Gamboa, A.; Albert-Puentes, Delhy & Hernández, F. Germinación y dormancia de arbustos y trepadoras del bosque siempreverde de la Sierra del Rosario, Cuba. *Pastos y Forrajes*. 38 (1):11-28, 2015.

Sánchez, J. A.; Muñoz, Bárbara & Fresneda, J. Combined effects of hardening hydration-dehydration and heat shock treatments on the germination of tomato, pepper and cucumber. *Seed Sci. Technol*. 29 (3):691-697, 2001.

Sánchez, J. A.; Suárez, A. G.; Montejo, Laura & Muñoz, Bárbara C. El cambio climático y las semillas de las plantas nativas cubanas. *Acta Bot. Cub*. 214:38-50, 2011.

Shalimu, D.; Li, K.; Baskin, Carol C.; Baskin, J. M. & Liu, Y. Seed germination biology of four Pomegranate (*Punica granatum*) cultivars from Xinjiang, China. *HortScience*. 50 (6):826-829, 2015.

Smýkal, P.; Vernoud, Vanessa; Blair, M. W.; Soukup, A. & Thompson, R. D. The role of the testa during development and in establishment of dormancy of the legume seed. *Front. Plant Sci*. 35:1-19. https://www.ncbi.nlm.nih.gov/pmc/articles/PMC4102250/pdf/fpls-05-00351.pdf, 2014.

Tribouillois, Hélène; Dürr, Carolyne; Demilly, D.; Wagner, Marine-Hélène & Justes, E. Determination of germination response to temperature and water potential for a wide range of cover crop species and related functional groups. *PLoS One*. 11 (8):e0161185, 2016.

Willis, C. G.; Baskin, Carol C.; Baskin, J. M.; Auld, J. R.; Venable, D. L.; Cavender-Bares, Jeannine *et al*. The evolution of seed dormancy: environmental cues, evolutionary hubs, and diversification of the seed plants. *New Phytol*. 203 (1):300-309, 2014.

Received: November 24, 2016

Accepted: September 20, 2017