# **English Version**

## Abstract

The first work was executed with the objective to evaluate the better solution in the *Plantago lanceolata* Hook. germination. The seeds were soaked with gibberelic acid (0.01% and 0.02%), thiourea (0.01% and 0.02%) and potassium nitrate (0.2% and 0.4%), plus a control treatment with 4 repetitions, disposed in CRD. The experiments were conduced on gerbox with a constant temperature of 20°C. The evaluations were done every day until 21 days. The 1,000 seeds weight was 2.085 grams. The gibberellic

## Evaluation of english plantain and lemon balm germination and vigor

Renata da Silva Brant<sup>1</sup>; Carlos Juliano Brant Albuquerque<sup>2</sup>; João Almir Oliveira<sup>3</sup>; Carlos Machado dos Santos<sup>4</sup>

acid provided the higher increase of GSI until 15 days. The higher values for germination percentage at seven days were verified in seeds soaked with gibberellic acid in two concentrations and at 21 days there were no differences between the best treatments: control, gibberellic acid (0.01% and 0.02%), thiourea (0.01%) and potassium nitrate (0.2%). In the second experiment, the objective was to evaluate the effects of the temperature and GA<sub>3</sub> on germination and vigor of *Melissa officinalis* L. seeds. It was tested five temperatures (15, 20, 25, 30°C and alternate 20-30°C) in presence and absence of GA<sub>3</sub>. The experiment was carried out in gerbox on blotter paper. Daily counts were performed for 10 days. The experimental design used was completely randomized factorial arrangement (5x2) with four replications, and 50 seeds per treatment. There were no differences in responses of GSI and first germination count of lemon balm's seeds between the temperatures. The presence of GA<sub>3</sub> increased the germination speed index of the seeds, but did not influence in first germination count.

Key words: Plantago lanceolata Hook.; Melissa officinalis L.; seeds; growth regulator

### Introduction

A good crop yield depends on various factors. Initially, the safety of a quality propagation material offers the producer larger chances of success. Accordingly, the germination and the seed vigor occupy prominent place.

Medicinal and aromatic plants present, in practice, great variability in the quality of seeds found in the market, including the unevenness on the germination, problems with diseases and dormancy, empty seeds and others. Thus, it becomes of great importance to know the best conditions of germination and methods to improve it.

Temperature influences the process of germination, especially since it changes the speed of water absorption and modifies the speed of the chemical reactions which will trigger the deployment, the reserve transportation, and the synthesis of substances to the seedlings (BEWLEY e BLACK, 1994).

In accordance with what was cited by Popinigis (1985), the main methods used to overcome the grass seed dormancy are: "disruption of the caryopsis", treatment with potassium nitrate (KNO<sub>3</sub>), exposition to light, use of alternated temperatures, application of precooling, increase on the oxygen tension and treatment with hormones (gibberellins and cytokinins). According to Bewley and Black (1994), gibberellins are involved in the break of the dormancy and promotion of the germination in some species. However, Renner et al. (2007) concluded that the use of GA<sub>3</sub> did not increase the *Pfaffia glomerata* (Spreng) Pedersen seed germination.

English plantain, whose scientific name is *Plantago lanceolata* Hook., originated in Europe has synonyms as ribwort plantain and narrowleaf plantain. The species *Plantago major* and *P tomentosa* are commonly found in Brazil. It is a perennial plant, small, has leaves disposed in rosette, with a long and

2 Researcher, Technological Center of the Triângulo Mineiro and Alto Paranaíba, Agricultural Research Corporation of the State of Minas Gerais (Epamig),Uberlândia, Minas Gerais, Brasil.

3 Professor Adjunto\*, Sector of Seeds, Department of Agriculture, Federal University of Lavras, Lavras, Minas Gerais, Brazil. 4 Professor Adjunto\*, Institute of Agricultural Sciencies, Federal University of Uberlândia, Uberlândia, Minas Gerais, Brazil. \*Brazilian academic degree

<sup>1</sup> Invited Professor, Institute of Agricultural Sciences, Federal University of Uberlândia. Uberlândia, Minas Gerais, Brazil. Rua Varginha 1214. Bairro: Daniel Fonseca. Uberlândia, Minas Gerais. CEP: 38400-322. e-mail: renataplantasmedicinais@yahoo.com.br. Author for correspondence.

winged petiole and the flowers are small and in spike. Among the main chemical constituents are tannins, mucilage, organic acids, salts of potassium, vitamin C and others. Its indications are: expectorants, diarrheal (leaf), healing, astringent, emollient and purifying. It is also used as fodder, and its new leaves are edible, as vegetables (MARTINS et al., 2000). It is propagated by seeds (FURLAN, 1998).

Lemon balm (*Melissa officinalis* L.) belongs to the family Lemiacae. It is a perennial plant, Herbaceous (0.5 to 1.0 m) with fibrous roots and branched rhizomes, tender, square, upright, or less prost, hairy. It is an aromatic herb, with a characteristic smell. Its leaves are white and are hidden under the foliage. It has tannin and an essential oil which contains citral (soothing). All the plant is anti-spasmodic, sedative, digestive, stomachic, carminative and stimulant (LORENZI and MATOS, 2002).

Considering the process of propagation of this species, Martins et al. (2000) defined that it is multiplied by cutting and seeds. The seeds found in the Brazilian market are imported and many times, in practice, there have been many problems with germination.

The work was performed with the objective to evaluate the English plantain seed germination in different growth regulator substances and the effect of the temperature and the gibberellin in germination and vigor of lemon balm seeds.

## Material and methods

#### Experiment with Plantago lanceolata Hook.

The experiment was conducted in an incubator in the Laboratory of Seed Analysis of the ICIAG (Institute of Agricultural Sciences), of the Federal University of Uberlândia (UFU). English plantain seeds (*Plantago lanceolata* Hook.) were collected from plants located on the garden of medicinal plants of the Federal University of Lavras. The seeds were cleaned using seed blower, Dakota model, and performed the weighing of 1,000 seeds. Later they were placed in alcohol 70% and then treated with hypochlorite for one minute. After this period they were washed with deionized water to remove the excess of the substances cited and put to dry in room temperature on absorbent paper.

In order to determine the capacity of seed germination the seeds were put to germinate, soaked in gibberellic acid solution (GA<sub>3</sub>) at 0.01 and 0.02%, thiuram at 0.01 and 0.02% and potassium nitrate at 0.2 and 0.4%, which totalized, with the control (non-treated seed), seven treatments. Each treatment consisted of a hundred seed (divided in four replications of 25) which were distributed in gerbox lined with two sheets of filter paper, previously moistened in the proportion 2:1 (volume of water in mL and paper weight in grams). Then, they were put to germinate in a germination chamber, at a constant temperature of 20 °C, with direct light. During the performance of the test, the moisture of the substrate was completed with deionized water applied once.

The evaluations of the seedlings were daily at the same time, from the beginning of the germination to the 21<sup>st</sup> day. The seeds considered germinated were those which presented radicle and shoot emergence, according to the criteria adopted on the Rules of Seed Analysis (BRASIL, 1992). The parameters evaluated were the following ones: weight of 1,000 seeds; percentage of germination on the seventh and 21<sup>st</sup> days; Germination Speed Index (GSI) to the 15<sup>th</sup> day, according to the formula proposed by Maguire (1962).

## Experiment with Melissa officinalis L.

The experiment was conducted in the Laboratory of Seed Analysis of the Agricultural Department of the Federal University of Lavras. The experimental design used was completely randomized to the first count of germination with five temperatures and presence and absence of GA<sub>3</sub>, arranged in factorial scheme, with four replications.

Lemon balm seeds were imported from France and submitted to five temperatures (15°C, 20°C, 25°C, 30°C and alternate 20-30°C), in the presence and absence of GA<sub>3</sub>.The fixed temperature (15°C, 20°C, 25°C, 30°C) were effected in Thermogradient Table (Seed Processing type 5001.10.00, series 15893) monitored by the computer program Wizcons for Windows. To evaluate the alternate temperature was performed in BOD (20-30° C).

The germination test was conducted in gerbox

Pesquisa Aplicada & Agrotecnologia v2 n2 Mai. – Ago. 2009 Print-ISSN 1983-6325 (On line) e-ISSN 1984-7548 with four replications of 50 seeds per treatment. The seeds were equally divided in a blotting paper, moistened with water in quantity equivalent to 2.5 times their weights. It was applied gibberellin in the concentration of 250 mg of  $GA_3$  by liter of water in the treatments with its presence. The evaluations were made during 10 days.

The germination speed index was calculated together with the germination test. Daily count of the germinated seeds (radicle protrusion) was performed during 10 days. To the calculation of the GSI it was used the formula proposed by Maguire (1962).

#### Statistic analysis

The analysis of the data was performed through the Sistema de Análise Estatística para Microcomputadores (Statistical Analysis System for Microcomputers) – SANEST (ZONTA e MACHADO, 1984).

## **Results and discussion**

#### Experiment with Plantago lanceolata Hook.

The weight of 1,000 seeds was 2.085 grams. The germination was influenced by the treatments (Table 1). The Germination Speed Index was higher when the seeds were treated with gibberellic acid, independent on the concentration. Thiuram and potassium nitrate influenced negatively on the germination speed, so that they obtained values inferior to the control.

Concerning the percentage of germination on the seventh day, the best response was found with

the application of gibberellin in both concentrations. The potassium nitrate, though, provided the lowest responses to the English plantain.

To the percentage of germination on the 21st day, it was observed that the control obtained higher value together with the application of gibberellic acid at 0.01% and 0.02%, thiuram at 0.01% and 0.02% and potassium nitrate at 0.2%. Thus, it was observed that the control, the thiuram and the potassium nitrate influenced in a lower speed of germination to the species, nonetheless, they provided a similar germination, in quantity, after 21 days of the sowing. This results are in accordance with Ayoama et al. (1999), studying the effect of the gibberellic acid in lavender (Lavandula angustifolia Miller), and Ehlert (2000), in Ocimum gratissimum L., who verified that gibberellin provided an acceleration on the process and on the other hand, in these species, it provides an increase in the germination percentage, which was not verified in the English plantain.

The use of substrate soaked with solution of potassium nitrate ( $\text{KNO}_3$ ) did not present significant responses to the germination count 21 days after the sowing of the seeds of *Brachiaria decumbens* cultivar Marandu when submitted to the accelerated aging (MESCHEDE et al., 2004).

#### Experiment with Melissa officinalis L.

The characteristics evaluated pointed out that the seeds of *Melissa officinalis* L. are not sensitive to the different temperatures evaluated (Figures 1 and 2).

There was no difference between the characteristics of the first germination count and

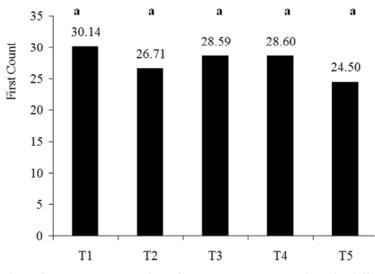
Service and Servic			
Treatments	GSI 15 days	% Germ. 7 days	% Germ. 21 days
GA <sub>3</sub> 0.01%	16.75 ab	59.50 ab	68.75 ab
GA <sub>3</sub> 0.02%	21.25 a	64.75 a	69.50 a
Thiuram 0.01%	12.75 bcd	48.00 cd	63.25 abc
Thiuram 0.02%	10.50 cd	40.25 de	58.25 с
KNO <sub>3</sub> 0.2%	9.75 cd	32.00 ef	67.50 ab
KNO <sub>3</sub> 0.4%	7.75 d	25.00 f	60.50 bc
Control	15.00 bc	51.75 bc	66.75 abc

**Table 1.** Germination Speed Index (GSI) until 15 days, germination percentage in seventh and 21° days of *Plantago lanceolata* Hook. in germinator.

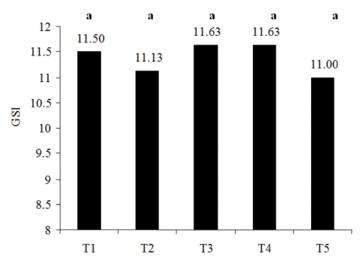
Original means followed by different letters differ by the Tukey Test at a level of 5% probability.

Pesquisa Aplicada & Agrotecnologia v2 n2 Mai.- Ago. 2009 Print-ISSN 1983-6325 (On line) e-ISSN 1984-7548 the GSI to the temperatures (alternated 20-30 °C, 15 °C, 20 °C, 25 °C and 30 °C). Possibly, the company which produces the seed, despite being located in temperate climate has directed lots with highest plasticity of germination in different temperature conditions to the Brazilian marked, since Brazil is a country with major climate differences from North to South. Different results were found by Silva et al.

(2004), who used the temperatures 20° C, 25° C, 30° C and 35° C and verified that there was an increase in the GSI with the increase in the temperature, which was less expressive only from 25° C to 30° C. This can be due to differences in the productive process of the seed, so that nowadays there is a higher demand of this species in Brazil, and thus, a greater preoccupation from the exporting companies to



**Figure 1.** Average values of germination count first of *Melissa officinalis* L. seeds under different temperatures (T1 – Alternated temperature 20-30 °C, T2 – 15 °C, T3 – 20 °C, T4 – 25 °C, T5 – 30 °C).



**Figure 2.** Average values of Germination Velocity Index (GSI) of *Melissa officinalis* L. seeds under different temperatures (T1 – Alternated temperature 20-30 °C, T2 – 15 °C, T3 – 20 °C, T4 – 25 °C, T5 – 30 °C).

Pesquisa Aplicada & Agrotecnologia v2 n2 Mai.- Ago. 2009 Print-ISSN 1983-6325 (On line) e-ISSN 1984-7548 provide input of greatest quality.

Another factor might be the origin of the seeds used. Wanderer et al. (2007) evaluated the germination percentage of lots of *Melissa officinalis* L. seeds coming from France and Netherlands and concluded that seeds coming from France obtained best results (91%) than those from Netherlands (78%). There was no interaction between temperature and GA<sub>3</sub>, according to the GSI and first germination count.

To Germination Speed Index it was verified a significant effect of the factor gibberellin (Table 2). With its application, there was a higher GSI. Thus, this growth regulator has influence on the metabolism of the seed of some species, providing the acceleration of their germination (TAIZ e ZEIGER, 2004). Aragão et al. (2001) applied bioregulators in seeds of sweet corn and concluded that the presence of GA<sub>3</sub> provided better germination and higher number of seedlings emerged in the first germination count.

There were no differences on the responses of

GSI and first germination count of *Melissa officinalis* L. seeds between the temperatures. The presence of  $GA_3$  increased the Germination Seed Index, however it did not influence in the first germination count.

## Conclusions

1. The presence of gibberellin caused higher speed on the English plantain seed germination, however, on 21 days the percentage of germination of all the treatments with growth regulator was similar to the control.

2. The different temperatures did not influence in the lemon balm germination, while the presence of gibberellin increased the speed of germination. However, it did not influence the average values of first germination count.

## Acknowledges

The authors thank CAPES, CNPq and FAPEMIG by the financial support.

**Table 2.** Average values of first germination count and Germination Seed Index (GSI) of *Melissa officinalis*L. in presence and absence of gibberellins.

Gibberellin	First count	GSI
Without	26.06 a	11,05 b
With	29.34 a	11,70 a

## References

ARAGÃO, C.A.; LIMA, M.W.P.; MORAIS, O.M.; ONO, E.O.; BOARO, C.S.F.; RODRIGUES, J.D.; NAKAGAWA, J.; CAVARIANI, C. Fitorreguladores na germinação de sementes e no vigor de plântulas de milho super doce. **Revista Brasileira de Sementes**, Brasília, v.23, n. 1, p.62-67, 2001.

AYOAMA, E.M.; ONO, E.O.; FURLAN, M.R. Estudo da germinação de sementes de lavanda (*Lavandula angustifolia* Miller). **Scentia Agrícola**, Piracicaba, v. 53, n. 2, n.3, 1996.

BEWLEY, J. D.; BLACK, M. Seeds, physiology and germination. 2. ed. New York : Plenum Press, 1994. 445 p.

BRASIL, Ministério da Agricultura. Secretaria Nacional de Defesa Agropecuária. **Regras para Análise de Sementes.** Brasília, 1992. 365p.

EHLERT, P.A.D. A**spectos agronômicos da alfavaca-cravo**. Dissertação (Mestrado em Fitotecnia-Plantas Medicinais) – Universidade Federal do Ceará, Fortaleza, 2000. 44p.

FURLAN, M. R. Cultivo de Plantas Medicinais. Coleção agroindústria, v.13. 1998. p.130.

LORENZI, H.; MATOS, F.S.A. **Plantas medicinais do Brasil: nativas e exóticas.** Nova Odessa: Plantarum, 2002. p.245.

Pesquisa Aplicada & Agrotecnologia v2 n2 Mai.- Ago. 2009 Print-ISSN 1983-6325 (On line) e-ISSN 1984-7548 MAGUIRE, J.D. Speed of germination-aid in selection and evaluation for seedling emergence and vigor. **Crop Science**, Madison, v.2, p.176-177, 1962.

MARTINS, E. R.; CASTRO, D.M.; CASTELLANI, D.C.; DIAS, J.E. **Plantas Medicinais.** Viçosa: UFV, 2000. 220p.

MESCHEDE, D.K.; SALES, J.G.C.; BRACCINI, A.L.E; SCAPIM, C.A.; SCHUAB, S.R.P. Tratamentos para superação da dormência das sementes de capim-braquiária cultivas Marandu. **Revista Brasileira de Sementes**, Pelotas, v.26, n.2, p.76-81. 2004.

POPINIGIS, F. Fisiologia da semente. Brasília: AGIPLAN, 1985, 289p.

RENNER, G.D.R.; CAMACHO, F.; PEIXE, S. Ação da temperatura, ácido giberélico e luz na germinação de sementes de fáfia – *Pfaffia glomerata* (Spreng.) Pedersen. **Semina: Ciências Agrárias**, Londrina, v.28, n.3, p.349-354. 2007.

SILVA, P. A.; SANTOS NETO, A. L.; MEDEIROS FILHO, S.; BLANK, A.F. Efeito da temperatura e da luz na germinação e no vigor de sementes de *Melissa officinalis* L. **Horticultura Brasileira**, Campo Grande, v.22, n.2, jul., 2004. Suplemento CD ROM.

TAIZ, L.; ZEIGER, E. Fisiologia vegetal. 3.ed. Porto Alegre: Artmed. 2004. 719p.

WANDERER, M; FRANKE, L.B.; BARROS, IBI. Germinação de sementes de melissa com diferentes origens. In: II Congresso Brasileiro de Agroecologia, 2007. **Resumos...** Revista Brasileira de Agroecologia, Porto Alegre, v.2, n.1, p.1114-1117, fev., 2007.

ZONTA, E.P.; MACHADO, A.A. Sistema de análise estatística para microcomputadores – Sanest. Pelotas: UFPel. 1984. 109p.