**Journal of Applied Botany and Food Quality 89, 156 - 162 (2016), DOI:10.5073/JABFQ.2016.089.019**

[1]Department of Plant Anatomy and Cytology, University of Maria Curie-Sklodowska, Lublin, Poland
[2]Institute of Natural Fibres and Medicinal Plants, Poznań, Poland
[3]Poznań University of Life Sciences, Poznań, Poland

# Vitality and germination of lemon balm (*Melissa officinalis* L.) seeds

**Krystyna Winiarczyk[1], Katarzyna Seidler-Łożykowska[2], Joanna Gębura[1], Jan Bocianowski[3]**

(Received September 22, 2015)

## Summary

Between 2012-2014, the germination abilities of lemon balm seeds were tested. The investigated seeds were obtained from lemon balm breeding project conducted in Institute of Natural Fibres & Medicinal Plants (INF&M), of Poznań, Poland. The germination rates were calculated by using International Rules for Seed Testing (ISTA) seed germination rules and a high fluctuation of germination rates was observed. A low rate of germination usually devalues the seeds. Therefore, the anatomy of the investigated lemon balm seeds was analyzed by scanning electron microscopy (SEM) and a tetrazolium test was done. SEM analysis showed correct morphological structures for all seed parts. In particular, the embryos, endosperm and testa were well and correctly developed and abnormal seeds parts were not found. The tetrazolium test revealed a high rate of vitality for the investigated strain seeds. Thus, before seed are disqualified because of low germination rate, we recommended that other useful tests should be done.

## Introduction

Lemon balm (*Melissa officinalis* L.) is a valuable medicinal plant native to southern Europe (near the Mediterranean) and western Asia. The species is commonly cultivated in Europe; in Poland it occupies an area of approximately 5000 ha. Lemon balm herb (*Melissa herba*) and lemon balm leaves (*Melissa folium*), the components of herbal mixtures, are used as a digestive, antispasmodic, sedative and antiviral remedies. Lemon balm is recommended in case of sleep disturbances, headache, colds and functional gastrointestinal disorders (Aziz and El-Ashry, 2009). Most of its pharmacological properties have been attributed to its essential oil components (Uyanik and Gurbuz, 2014). Lemon balm essential oil (*Melissa oleum*), with its fresh lemon odor and light yellow colour, is obtained from the leaves or the herb itself by hydro-distillation, and consists mainly of citronellal (2-40%) and citral (10-30%). Citral, itself a mixture of the two monoterpenes geranial and neral (Aziz and El-Ashry, 2009), is widely used in the food and cosmetic industries thanks to its lemon aroma.

In addition, the essential oil can be used as antioxidant, and has antimicrobial and free radical scavenging capacities. It may be also used as an antiviral agent against herpes simplex virus type 2 (Saeb and Gholamrezaee, 2012). Lemon balm oil is widely used by the pharmaceutical, cosmetic and food industries (Menezes et al. 2015). Finally, lemon balm may also be useful as an insect repellent (Abbaszadeh et al., 2009).

Lemon balm is a perennial plant that reaches maximum of 1 m with hairy, deeply veined, heart-shaped leaves which are 2 - 8 cm long. The flowers, 4 to 12 in small clusters, are white or pale pink. Lemon balm is a cross-pollinating species and its ovate seeds are black or dark brown. The weight per thousand seeds varies from 0.5 to 0.7 g. Lemon balm has a hairy root system with many lateral branches which adapt well to different environmental conditions (Moradkhani et al., 2010), but during cultivation, lemon balm plants prefer warm, sunny sites. Plant can grow rapidly at the temperature from 15 °C to 35 °C and require 500 to 600 mm of precipitation well distributed over the growing season (Saeb and Gholamrezaee, 2012). The seeds germinate at 10-12 °C, but the proper temperature for germination is 18-20 °C (Tobeh et al., 2013). Many factors (environmental and agricultural) affect herb yield, essential oil content and composition, and seed yield and quality (Patora et al., 2002; Farahani et al., 2009; Vaverkova et al., 2012; Sharopov et al., 2013; Nurzyńska-Wierdak et al., 2014; Singh et al., 2014; Uyanik and Gurbuz, 2014). Seeds are the main propagation method. It is recommended to use mature, well developed seeds with a high germination rate. While there have been many studies determining essential oil composition under different conditions, information about lemon balm seed quality are very limited. Seidler-Łożykowska et al. (2013), who evaluate the variability of lemon balm genotypes, reported that seed yield yield varied from 22 g to 182 g/m$^2$ in the analyzed collection, and the weight per thousand seed ranged from 0.356 g to 0.632 g (seed yield being positively correlated with the weight per thousand seeds). Wahl and Plescher (2014) analyzed lemon balm seed quality on the German seed market, and found that in 2009-2011, 33-82% of seed batches fulfilled the suggestions according to germination standards.

Seed dormancy is a major factor influencing the timing of seedling. Initiation of this process depends on many environmental and physiological factors (Nonogaki, 2014). The proportional amounts of two antagonistic plant hormones, ABA and gibberellins regulate seed germination in most plant species. Maternal ABA especially is involved in the induction of seed dormancy, and it inhibits seed germination (Baskin et al., 2000; Kanno et al., 2010).

Seed permeability by water or gases also plays an important role in maintaining seed viability and germination. The initiation of germination depends on many environmental factors, including thermal cardinal points: the minimal, maximal and optimal temperatures for initiating this process. Other environmental factors, such as the substrate and the availability of water, also affect the process of seed germination.

The tetrazolium test is widely recognized as an accurate method for estimation of seed viability and providing labeling information for immediate shipment of seed lots (without waiting for completion of germination test) is necessary. It is also a valuable technique for determining reasons for deficiencies of germination.

At the Institute of Natural Fibres and Medicinal Plants, an on-going breeding project, with multidirectional evaluation of morphological, chemical and yielding traits, is being conducted to obtain new, valuable cultivar of lemon balm. During this project the high fluctuation of seed germination rates was observed, which inspired the authors to closely analyze the anatomy of the lemon balm seeds in question.

## Materials and methods

In 2012, the plant breeding nursery consisting of four breeding strains (7, 28, 33, 33/II) was established in the field and in the spatial isolation. The strains were selected from the lemon balm breeding

project of INF&MP. The seeds to be analyzed were collected in the second year (2013) and in the third year (2014) of the nursery growing program. They were collected by hand at ripeness, and then dried, threshed and cleaned. Finally, seed yield of and weight per thousand seeds were estimated, before the seeds themselves were stored in refrigeration at the controlled temperature of 5 °C in the paper bags. The germination analysis of seeds harvested in 2013 was done in February 2014 and 2015 and seeds harvested in 2014 were analyzed in February 2015. In 2014, the evaluation of seed germination of lemon balm cv. 'Quedlinburger Niederliegende' was also done. These seeds had been collected and stored in 2013 and originated from Pharmasaat (Germany).

The evaluation of germination was done according to ISTA rules (ISTA, 2010) which for lemon balm seed recommended germination on filter paper in a temperature range of 20-30 °C. After 4-7 days the first counting of germinated seeds is recommended and the last counting after 21 days. In case of low germination rate, seed cooling is advised.

**Germination on filter paper**

In accordance with ISTA rules, 100 seeds of each evaluated strain were placed on 90 mm Petri dishes on two disk of filter paper in four repetitions. Then the distilled water was added.

The germination experiment was done at 24 °C, and after four days the germinated seeds were counted and removed from the dishes. The last counting of the germinated seeds was done in the 21st day of examination.

**Germination in peat media**

In April 2015, further seed samples of strain seeds were germinated in a peat medium. One hundred seeds of each lemon balm strain under investigation were sown in one row on the seedling tray filled with 15 cm layer of peat, in three repetitions. The trays were placed in greenhouse and keep watered. After 21 days the seedlings were removed from the peat and counted.

**Scanning electron microscopy (SEM)**

The biological material used for scanning microscopy observation consisted of 10 seeds of the each lemon balm strain and was fixed in a 5:5:90 (v:v:v) mixture of glacial acetic acid:formalin (40%):ethanol (70%) and dehydrated in a graded acetone series (40%, 70%, 80%, 100%). Samples were analyzed in the scanning electron microscope (LEO1430VP) with an accelerating potential of 15 kV.

**Tetrazolium test for seed viability and vigor**

The analyses were also based on ISTA'S rules, recommendations and methodologies for seed testing. Sixty seeds were soaked in sterile distilled water at 20 °C for 18 hours. Each seed was then cut in half and immersed in 1% tetrazolium solution (pH 6.5). The samples were incubated for 18 hours at 30 °C. The dried seeds were observed and examined under a microscope. Each analysis was done in four repetitions. Two-way analysis of variance (ANOVA) was used to analyze germination viability and germination rate with years of collection and age of seeds as the two fixed factors. The least significant differences (LSD) test was used to distinguish significant differences.

## Results and discussion

The results of analysis of variance for comparison age of seeds and year of collection indicated that the main effects of strains were significant for both evaluated traits: germination viability and germination rate (Tab. 1). According to ISTA rules, the germination viability and germination rate of the investigated strain seeds collected in 2013 and 2014 were very low (Tab. 2). The seeds collected in 2013 after six months of storage germinated at rates varying from 22% (strain 7) to 87% (strain 33/II), while those which were harvested in 2014 after six-month storage germinated at rates between 5% (strain 7) and 65% (strain 33/II) only. In both group of lemon balm seeds, seeds of strain 7 and 33 had the lowest rate of germination. After 18 months of cool storage the rate of 2013 seed germination decreased from 14% (strain 33) to 80.7% (strain 33/II). Germination rate of the seeds of German cultivar 'Ouedlinburger Niederliegende', done in 2014, was high (86%), but the age of these seeds was unknown (Tab. 2).

In our long experience with medicinal plant seed germination the lemon balm seeds of breeding strains collected in 2013 and 2014 had the lowest rate of germination compared to the average of those which were germinated in years 2004 - 2014 (Tab. 3).

Using SEM, lemon balm was observed to have produced. ovate black or dark brown seeds with an average length 1.82 mm (Fig. 1a). The lemon balm seeds were covered by a thin testa with a raphe visible on its surface (Fig. 1a). The seed coat was made of uniform and tight packed cells. The testa surface was characteristically sculptured with numerous cavities and convex (Fig. 1b). At the narrow end, the abscission tissue consisted of sclerenchymatous cells was observed (Fig. 1c, d). The seed interior was filled with the endosperm, a nutrinal tissue containing store substances for the embryo. After discontinuation of testa differentiated endosperm has been released (Fig. 2a, d). The seed coat was made of three layers. The surface and subepidermal layer was consisted of flattened, sclerenchymatous cells. The middle layer was made of subsurface, elongated sclerenchymatous cells, which was arranged anticlinal to other layers (Fig. 2b). Immediately under the testa was an aleuron grain endosperm (Fig. 2 c, d, e) and deeper amorphous proteins were observed (Fig. 2 d, e, f). The embryo was centrally located and surrounded by the nutritive tissue developed inside the seed (Fig. 2c).

Tetrazolium test revealed the viability of the embryos developing inside the seeds (Fig. 3). Red colouration was assigned to viable seeds

**Tab. 1:** Mean squares from two-way analysis of variance for observed traits to compare age of seeds and year of collecting

| Source of variation | d.f. | Comparison of age of seeds | | Comparison of years of seed collection | |
|---|---|---|---|---|---|
| | | Germination viability | Germination rate | Germination viability | Germination rate |
| Experiment | 1 | 130.67** | 400.17** | 1568.167*** | 3432.04*** |
| Strain | 3 | 6399.28*** | 5490.33*** | 4072.444*** | 5131.38*** |
| Experiment × Strain | 3 | 3.11 | 19.61 | 223.278*** | 145.26* |
| Residual | 16 | 13.54 | 37.5 | 8.417 | 30.54 |

d.f. - number of degrees of freedom
** P<0.01; *** P<0.001

**Tab. 2:** Germination viability and germination rate of lemon balm strain seeds collected in 2013 and 2014

| Strain number | Year of collecting | Year of germination trial | Age of seeds [months] | Germination viability [%] | s.d. | Germination rate [%] | s.d. |
|---|---|---|---|---|---|---|---|
| 7 | 2013 | 2014 | 6 | 10.3 | 5.512 | 22.0 | 4.583 |
| 28 | 2013 | 2014 | 6 | 8.3 | 2.082 | 46.7 | 12.014 |
| 33 | 2013 | 2014 | 6 | 16.3 | 6.11 | 24.7 | 7.506 |
| 33/II | 2013 | 2014 | 6 | 75.3 | 1.528 | 87.0 | 1 |
| $LSD_{0.05}$ | | | | 4.50 | | 7.49 | |
| cv. 'Quedlinburger Niederliegende' | unknown | 2014 | unknown | 65.0 | - | 86.0 | - |
| 7 | 2013 | 2015 | 18 | 5.7 | 2.082 | 18.0 | 2.646 |
| 28 | 2013 | 2015 | 18 | 3.0 | 1 | 35.0 | 3.606 |
| 33 | 2013 | 2015 | 18 | 10.3 | 1.528 | 14.0 | 1.732 |
| 33/II | 2013 | 2015 | 18 | 72.7 | 6.658 | 80.7 | 7.371 |
| $LSD_{0.05}$ | | | | 3.19 | | 5.3 | |
| 7 | 2014 | 2015 | 6 | 0.7 | 0.577 | 5.0 | 1 |
| 28 | 2014 | 2015 | 6 | 2.0 | 1 | 8.3 | 3.055 |
| 33 | 2014 | 2015 | 6 | 1.3 | 0.577 | 6.3 | 2.082 |
| 33/II | 2014 | 2015 | 6 | 41.7 | 3.055 | 65.0 | 2.646 |
| $LSD_{0.05}$ | | | | 3.55 | | 6.76 | |

**Tab. 3:** Germination viability and germination rate of lemon balm seeds collected in years 2001 - 2010

| Year of collecting | Year of germination trial | Age of seeds [years] | Germination viability [%] | Germination rate [%] |
|---|---|---|---|---|
| 2001 | 2004 | 3 | 50 | 72 |
| 2002 | 2004 | 2 | 68 | 78 |
| 2003 | 2004 | 1 | 55 | 74 |
| 2003 | 2005 | 2 | 69 | 80 |
| 2005 | 2006 | 1 | 63 | 71 |
| 2006 | 2009 | 3 | 73 | 82 |
| 2007 | 2009 | 2 | 56 | 63 |
| 2009 | 2010 | 1 | 52 | 62 |
| 2006 | 2011 | 5 | 79 | 87 |
| 2007 | 2011 | 4 | 59 | 67 |
| 2009 | 2011 | 2 | 86 | 92 |
| 2010 | 2011 | 1 | 66 | 74 |
| 2005 | 2012 | 7 | 31 | 40 |
| 2006 | 2012 | 6 | 30 | 36 |
| 2009 | 2012 | 3 | 82 | 88 |
| 2010 | 2012 | 2 | 60 | 66 |
| 2009 | 2014 | 5 | 73 | 84 |
| 2010 | 2014 | 4 | 51 | 67 |

(Fig. 3 a, b, c) and white to non-viable ones (Fig. 3d). In viable seeds both embryos and endosperm were uniformly stained by phormazane (Fig. 3a, b). The viability of the tested seeds was high in each investigated lemon balm strain (96.67 - 98.89 %) and it was not depend on age of seeds (Tab. 4).

The results of two-way analysis of variance indicated that main effect of strain and effect of strain × year of collection interaction were significant for the germination rate of lemon balm seeds done in peat medium (Tab. 5). The main effect of year of collection was not statistically significant.

The germination rate of strains seeds done in peat medium was high and varied from 83 % (strain 33/II, 2013) to 95 % (strain 7, 2013) (Tab. 6). These results confirmed data obtained with tetrazolinum test.

The recent study done by WAHL and PLESCHER (2015), who analyzed the impact of temperature to the seed germination of chamo-

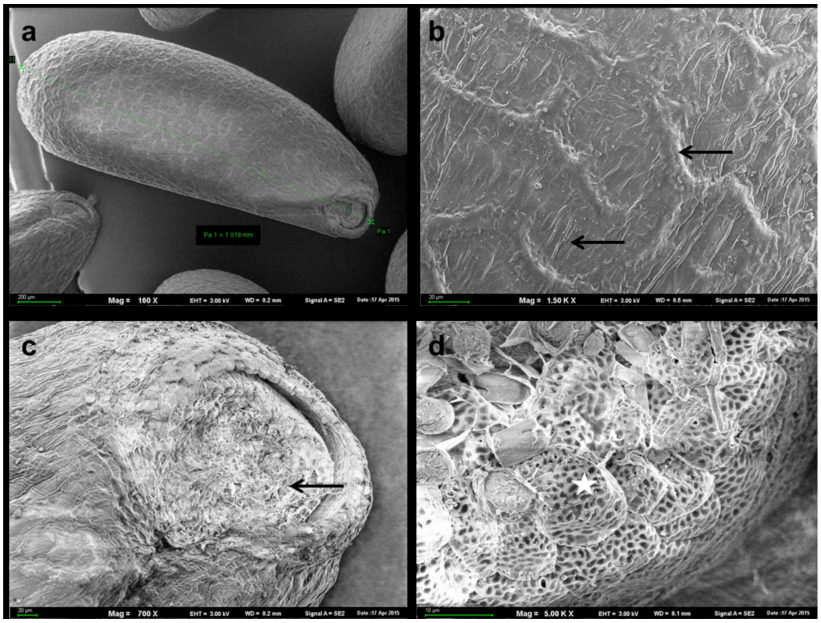


**Fig. 1:** Picture from scanning electron microscope (SEM). Morphology of lemon balm seed of the strain 7/2014. (A) The shape and size of seeds, 200 μm. (B) The characteristic sculpture of the seed coat texture (testa) with numerous cavities and convex (arrows), 20 μm. (C) The abscission tissue (cut-off) (star), 20 μm consisted of (D) sclerenchymatous cells (star), 10 μm.

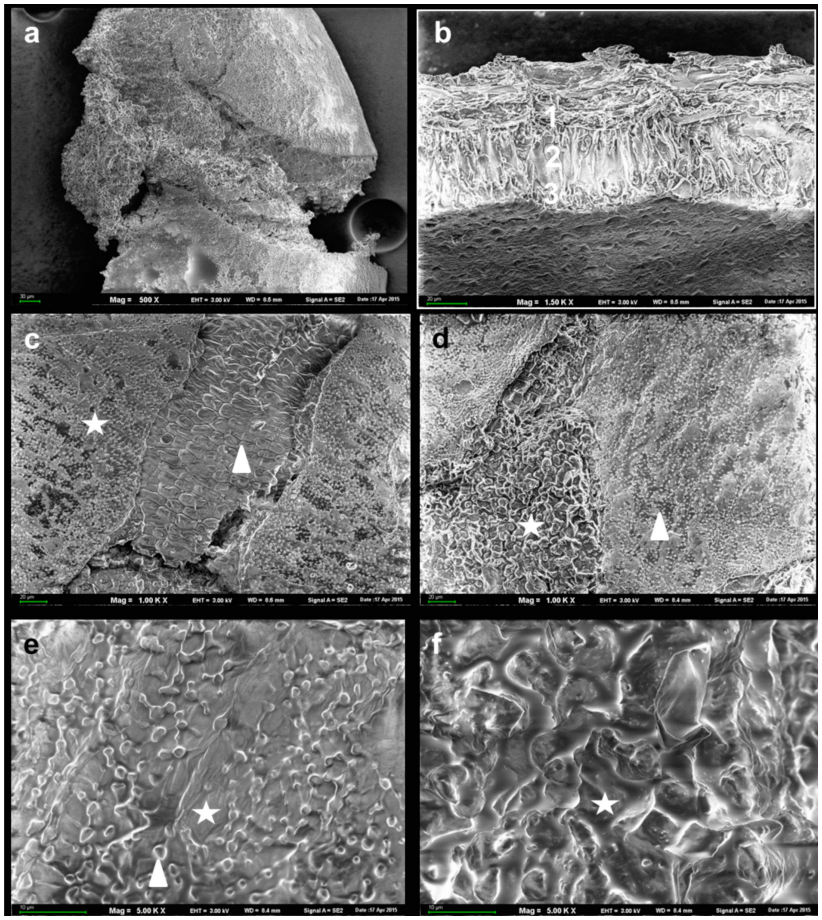


**Fig. 2:** Picture from scanning electron microscope (SEM). Structure of the lemon balm seed, strain (28/2013). (A) The seed with the damaged testa 30 μm. (B) Longitudinal section of the seed coat, 1 - crushed sclerenchymatous cells surface, 2 - subsurface cells sclerenchymatous cells arranged anticlinal to other layers 3 - sclerenchymatous cells arranged subepidermal side 20 μm. (C) Part of embryo tissue (arrow) and endosperm layers below testa (asterisk), 20 μm. (D) A different structure of the endosperm of the seeds lemon balm, aleurone layer of grain (arrow) and amorphous protein (asterisk), 20 μm. (E) Endosperm layers in the mature seed: aleurone grains (arrow) amorphous protein (asterisk) 10 μm. (F) The endosperm in the form of an amorphous protein under the surface of the seed, of 10 μm (F).

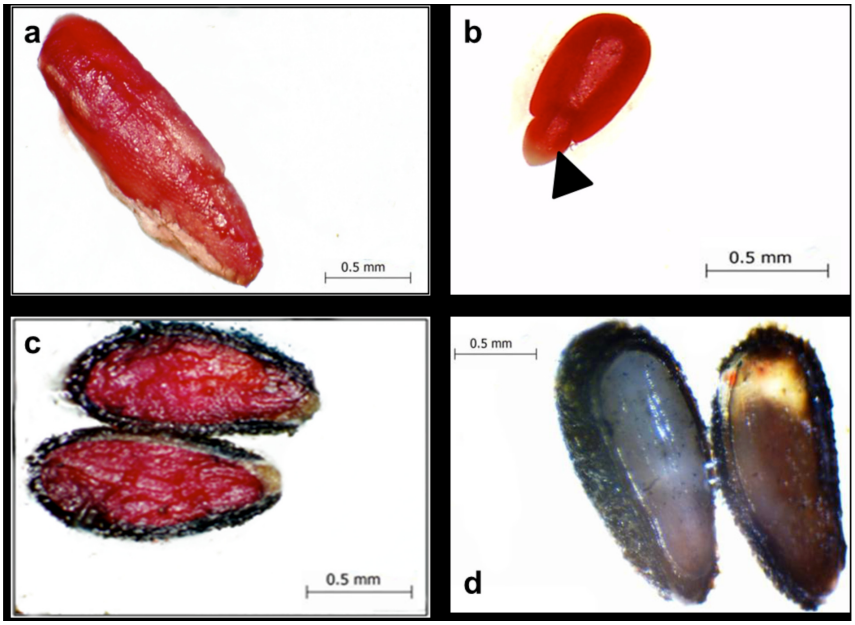


**Fig. 3:** Pictures of stereoscopic microscope. Tetrazolium test viability of the embryos developing inside the lemon balm seeds (scale bar = 0.5 mm). (A) Viable endosperm and embryo stained in red without the seed coat, strain 7/2013, (B) strain 7/2014. (C) Longitudinal section through viable seeds lemon balm with embryo stained in red, strain 28/2013. (D) Longitudinal section through the dead seed lemon balm with non coloured embryo, strain 33 / II / 2014.

**Tab. 4:** Viability and vigour of lemon balm seeds collected in 2013 and 2014 by tetrazolinum test

| Strain number | Year of collecting | Year of test trial | Age of seeds [months] | Viable seeds [%] |
|---|---|---|---|---|
| 7 | 2013 | 2015 | 19 | 95.56 |
| 28 | 2013 | 2015 | 19 | 98.33 |
| 33 | 2013 | 2015 | 19 | 98.88 |
| 33/II | 2013 | 2015 | 20 | 97.78 |
| 7 | 2014 | 2015 | 8 | 98.89 |
| 28 | 2014 | 2015 | 7 | 96.67 |
| 33 | 2014 | 2015 | 8 | 96.67 |
| 33/II | 2014 | 2015 | 8 | 96.67 |

**Tab. 5:** Mean squares of two-way analysis of variance for germination rate done in peat medium

| Source of variation | Germination rate [%] | | | |
|---|---|---|---|---|
| | d.f. | Sum of squares | Mean squares | F-statistic |
| Strain (S) | 3 | 141.458 | 47.153** | 5.99 |
| Year of collection (Y) | 1 | 30.375 | 30.375 | 3.86 |
| S × Y | 3 | 150.125 | 50.042** | 6.35 |
| Residual | 16 | 126 | 7.875 | |

d.f. - number of degrees of freedom
** p<0.01

mile, lemon balm and valerian showed that lemon balm germinates at warm temperature which affects on duration of germination. In lower temperature 15/8 °C (day/night) germination started between 9 and 13 days after sowing. For quick (after 4-5 days) lemon balm seed germination the optimal temperature was between 27/18 °C and 33/20 °C (day/night). The authors tested six cultivars of lemon balm, which showed a germination capacity range from 69.5% (cv. 'Offstein') to 100% (cv. 'Erfurter Aufrechte').

Hassanzadeh et al. (2014), who analyzed the effect of salicylic acid on lemon balm seed germination under salinity stress, determined the seed germination rate varying from 73% to 91%. The investigated seeds were nine months old and have been stored in the paper bags in temperature of 4 °C and 20% relative humidity. These seeds were thus stored in the conditions similar to our investigated of lemon balm strains seed. The authors reported that germination rate was significantly increased by salicylic acid under salinity conditions compared to the non-treated ones.

Karimian (2011), in determining the germination rate of lemon balm seeds after different time of hydropriming, reported the low germination rates (from 21% to 63%) and the highest rate was achieved after 21 hours of hydropriming. The author did not give any information about seed origin and storage conditions.

The investigation done by Hoseini et al. (2013) showed that the magnetic priming of lemon balm seeds increased the germination rate to 86% (for seeds treated by magnetic field) compare to the untreated control, with had a rate of 58%.

**Tab. 6:** Germination rate of lemon balm strain seeds done in peat medium in greenhouse 2015

| Strain number | Year of collection | Germination rate [%] | Standard deviation | Year of collection | Germination rate [%] | Standard deviation |
|---|---|---|---|---|---|---|
| 7 | 2013 | 95 | 2 | 2014 | 91 | 3 |
| 28 | 2013 | 93 | 2.517 | 2014 | 87 | 5 |
| 33 | 2013 | 93 | 1 | 2014 | 87 | 1.528 |
| 33/II | 2013 | 83 | 3 | 2014 | 89 | 2.517 |

$LSD_{0.05}$ S: 3.435; Y:2.429; S × Y: 4.857

## Conclusion

SEM analysis of lemon balm seeds showed correct morphological structure of all seed parts. The embryos, endosperm and testa were well and correctly developed. Abnormal seed parts were not found. The tetrazolium test revealed a high rate of vitality of the investigated seeds. Despite these positive results of morphology and viability, their germinate rate was very low following the ISTA rules of lemon balm germination. Plant breeder and seed producer should therefore declare the germination rate of the seeds they offer. Low rate of germination usually devalues the seeds, even though other tests (SEM observation, tetrazolium test or *in vivo*) might show completely different results. Thus, before seed disqualification in case of low germination rate, other useful tests should be recommended.

## References


Abbaszadeh, B., Farahani, H.A., Valadabadi, S.A., Darvishi, H.H., 2009: Nitrogenous fertilizer influence on quantity and quality values of balm (*Melissa officinalis* L.). J. Agri. Ext. Rural Dev. 1, 31-33.

Aziz, E., El-Ashry, S.M., 2009: Efficiency of slow release urea fertilizer on herb yield and essential oil production of lemon balm (*Melissa officinalis* L.) plant. Am. Eur. J. Agric. Environ. Sci. 5, 141-147.

Baskin, J.M., Baskin, C.C., Li, X., 2000: Taxonomy, anatomy and evolution of physical dormancy in seeds. Plant Species Biol. 15, 139-152.

Farahani, H.A., Valadabadi, S.A., Daneshian, J., Khalvati, M.A., 2009: Evaluation changing if essential oil of balm (*Melissa officinalis* L.) under water deficit stress conditions. J. Med. Pl. Res. 3, 329-333.

Hassanzadeh, K., Ahmadi, M., Shaban, M., 2014: Effect of pre-treatment of lemon balm (*Melissa officinalis* L.) seeds on seed germination and seedlings growth under salt stress. International Journal of Plant, Animal and Environmental Sciences 4, 260-265.

Hoseini, M., Rahimzadeh-Khoei, F., Mirshekari, B., 2013: Seed priming techniques improve germination and yield in two landraces of lemon balm in laboratory experiment and field study. International Journal of Indigenous Medicinal Plants 29, 1144-1150.

International Rules for Seed Testing, 2010: International Seed Testing Association (ISTA) Bassersdorf, Switzerland ISBN 83-891172-39-9. Part 5: Determination of seed germination.

Kanno, Y., Jikumaru, Y., Hanada, A., Nambara, E., Abrams, S.R., Kamiya, Y., Seo, M., 2010: Comprehensive hormone profiling in developing Arabidopsis seeds: examination of the site of ABA biosynthesis, ABA transport and hormone interactions. Plant Cell Physiol. 51, 1988-2001. doi: 10.1093/pcp/pcq158. Epub 2010 Oct 19.

Karimian, D., 2011: Determination of germination percentage in balm (*Melissa officinalis* L.) under hydropriming. Research Journal of Fisheries and Hydrobiology 6, 495-498.

Menezes, C., Guerra, F.Q., Pinheiro, L.S., Trajano, V.N., Pereira, F., de Souza, V.G., de Souza, F.S., Lima, E., 2015: Investigation of *Melissa officinalis* L. essential oil for antifungal activity against *Cladosporium carrionii*. Int. J. Trop. Dis. & Health 8, 49-56.

Moradkhani, H., Sargsyan, E., Bibak, H., Naseri, B., Sadat-Hosseini, M., Fayazi-Barjin, A., Meftahizade, H., 2010: *Melissa officinalis* L., a valuable medicine plant: A review. J. Med. Pl. Res. 25, 2753-2759.

Nonogaki, H., 2014: Seed dormancy and germination-emerging mechanisms and new hypotheses. Front Plant Sci. 28, 5:233. doi: 10.3389/fpls.2014.00233.

Nurzyńska-Wierdak, R., Bogucka-Kocka, A., Szymczak, G., 2014: Volatile constituents of *Melissa officinalis* L. leaves determined by plant age. Nat. Prod. Comm. 9, 703-706.

Patora, J., Majda, T., Góra, J., Klimek, B., 2003: Variability in the content and composition of essential oil from lemon balm (*Melissa officinalis* L.) cultivated in Poland. Acta Pol. Pharm. 60, 395-400.

Saeb, K., Gholamrezaee, S., 2012: Variation of essential oil composition of *Melissa officinalis* L. leaves during different stages of plant growth. Asian Pac. J. Trop. Biomed. 32, 547-549.

Seidler-Łożykowska, K., Bocianowski, J., Król, D., 2013: The evaluation of the variabilty of morphological and chemical traits of the selected lemon balm (*Melissa officinalis* L.) genotypes. Ind. Crops Prod. 49, 515-520.

Sharopov, F.S., Wink, M., Khalifaev, D.R., Zhang, H., Dosoky, N.S., Setzer, W.N., 2013: Composition and bioactivity of the essential oil of *Melissa officinalis* L. growing wild in Tajikistan. Int. J. Trad. Nat. Med. 2, 86-96.

Singh, S., Haider, S.Z., Chauhan, N.K., Lahani, H., Sah, S., Yadav, R.K., 2014: Effect of time of harvesting on yield and quality of *Melissa officinalis* L. in doon Valley, India. Ind. J. Pharm. Sci. 76, 449-452.

Tobeh, A., Moghanlu, F.S., Imani, A.A., 2013: Effect of different planting and harvest dates on quantitative yield of lemon balm as a medicinal plant. Int. J. Agron. Pl. Prod. 4, 839-847.

Uyanik, M., Gurbuz, B., 2014: Chemical diversity in essential oil composition of leaf, herb and flower in lemon balm (*Melissa officinalis* L.). Turk. J. Agricul. Nat. Sc. 1, 210-214.

Vaverkova, S., Mistrikova, I., Farkas, P., 2012: Qualitative properties of *Melissa officinalis* L. after the application of Rastim 30 DKV. Botanica Serb. 36, 81-84.

Wahl, S., Plescher, A., 2014: Saatgutqualitat von Kamille (*Matricaria recutita* L.), Melisse (*Melissa officinalis* L.) und Baldrian (*Valeriana officinalis* L.) am deutschen Saatgutmarkt 2009 bis 2011. Z. Arznei-Gewürzpfla. 19, 70-78.

Wahl, S., Plescher, A., 2015: Einfluss der Temperatur auf die Keimung der Samen von Kamille, Melisse und Baldrian. Z. Arznei-Gewürzpfla. 20, 54-60.



Address of the author:

Krystyna Winiarczyk, Joanna Gębura, Department of Plant Anatomy and Cytology, University of Maria Curie-Sklodowska, 20-033 Lublin, Akademicka Str. 19, Poland

E-mail: krystyna.winiarczyk@poczta.umcs.lublin.pl

E-mail: joanna.gebura@tlen.pl

Katarzyna Seidler-Łożykowska, Institute of Natural Fibres and Medicinal Plants, 60-630 Poznań, Wojska Polskiego Str. 71B, Poland

E-mail: katarzyna.lozykowska@iwnirz.pl

Jan Bocianowski, Poznań University of Life Sciences, Wojska Polskiego 28, 60-637 Poznań, Poland

E-mail: jboc@up.poznan.pl