



**FACTORS PLAYING ROLE IN THE FROST TOLERANCE OF POPPY
(*PAPAVER SOMNIFERUM* L.)**

DOCTORAL THESIS

CSILLA PAPPNÉ JÁSZBERÉNYI

SUPERVISOR: DR. ÉVA ZÁMBORINÉ NÉMETH (DSC)

BUDAPEST
2014

PhD School

Name: Doctoral School of Horticultural Sciences

Field: Crop Sciences and Horticulture

Head of Ph.D. School: Prof. Dr. Magdolna Tóth
Doctor of the Hungarian Academy of Sciences
Head of Department of Fruit Sciences
Corvinus University of Budapest
Faculty of Horticultural Sciences

Supervisor: Prof. Dr. Éva Zámборiné Németh
Academic Professor, DSc
Head of Department of Medicinal and Aromatic Plants
Corvinus University of Budapest
Faculty of Horticultural Sciences

The applicant met the requirement of the PhD regulations of the Corvinus University of Budapest and the thesis is accepted for the defence process.

.....
Dr. Magdolna Tóth
Head of Ph.D. School

.....
Dr. Éva Zámборiné Németh
Supervisor

1. SCIENTIFIC BACKGROUND AND AIMS OF THE STUDY

Poppy (*Papaver somniferum* L.) is a traditionally consumed food not just in Hungary but in other European countries as well due to the importance in pharmaceutical industry it is one of the species used in the largest scale that accumulate alkaloids. Its importance lies in these two special functions which are confirmed by the increasing volume of researches all over the world.

In Hungary two ecotypes of poppy are grown, one of them is the overwintering winter poppy and the other one is the spring ecotype. Recently the cultivation of winter poppy became the conspicuous, because of the higher yields (compared to the spring ecotype cultivars). Naturally the winter poppy ecotypes possess enhanced frost tolerance in leaf rosette stage and able to overwinter, in contrary to the spring ones (FÖLDESI, 1995). In the Hungarian list of varieties, only six cultivars belong to the winter poppy group while 16 ones are registered as spring poppy cultivars with low or high alkaloid content. Therefore the enhancement of frost tolerance of poppy is the further aim of poppy breeding besides altering the alkaloid content of it (Nemzeti Fajtajegyzék, 2013). The demand of winter poppy cultivars became significant in the last few years for the growers because of the higher yields as well as the earlier sales opportunities that ensure plus income, but in this ecotype the choice is poor.

There are many factors which affect the overwintering of plants and its successfulness. One of the most important environmental factors is the winter temperature and the quantity and the quality of precipitation. In this point of view the genetic endowments and the plants' development are also important. In the suitable developmental stage, the genetically inherited protective mechanisms of the plant are able to manifest in the phenotype. Many biochemical and physiological processes are necessary for the hardening of the plants to endure stress. Freeze causes ice crystals to form in the cells (SZIGETI, 2007), which process the plants try to avoid by accumulating cryoprotectant compounds in the intercellular space. On the other hand, the forming of the different osmolites is a generally induced mechanism caused by stress, where the plant concentrates the extremely cold sensitive cytoplasm and lowers the freeze point and the water content of the cells (KAZUO és KAZUKO, 1996; TAKAGI, 2008; SMIRNOFF, 1998; DELAUNEY és VERMA, 1993). In case of poppy these processes are less researched, the information regarding the poppy's frost tolerance are very sporadic; the basic knowledge is derived from the practice, underpinned only with growing experiments. The long procedure of poppy breeding can be speeded up by identification of marker molecules (soluble sugar, proline) or morphological and histological markers which are connected to stress tolerance in case of several other plant species published in the literature.

Therefore in the present work our aims were to gather more information about winter poppy genotypes, to reveal the biochemical and physiological characteristics related to frost tolerance of poppy and the possible relationship between frost tolerance and other important plant properties.

For this our aims were the followings.

Scientific, theoretical information:

- Gather more information about biochemical and physiological mechanisms playing role in the frost tolerance of poppy, find species-specific characteristics;
- Know more about the possibilities of expansion of winter poppy ecotypes based on the extant core materials, and genetically variability;
- Find relationships between winter hardiness and morphological and production characteristics of poppy and markers.
- Investigate the connection of alkaloid accumulation and winter hardiness.

Practical purposes:

- Broaden the gene bank collection and the breeding core materials of the Department of Medicinal and Aromatic Plants of the Corvinus University of Budapest with different frost tolerant genotypes;
- Develop a species-specific *in vitro* test method to test the frost tolerance of different genotypes;
- Implement new winter poppy candidate varieties which meet the growing regulations and requirements of the EU.

2. MATERIAL AND METHOD

2.1. GENERAL CONDITIONS OF THE EXPERIMENTS

Open field experiments were carried out in Soroksár, in the Research Field of Corvinus University of Budapest, Faculty of Horticultural Sciences between 2008 and 2012. The seeds were sown in spring in every year by hand, for 50 cm row spacing, using 10 m² parcel size in three replications. In the beginning of the breeding work reciprocal crossing were made. The raw materials of the experiments of further years were ensured by the propagation of seeds derived from these crossings.

In vitro trials were carried out in the Laboratory of the Department of Medicinal and Aromatic Plants, Corvinus University of Budapest. Sanyo MLR-351H climatic chamber was used for the germination and the breeding of plants. The hardening treatment and the frost effects were triggered by RUMED 1000 type programmable light thermostat. The seeds were sown into Blüh-Fix substrate using 7x7 cm size plastic pots and were placed into the climatic chamber with seed trays. The following conditions were used in each cycle: 15/10 °C (day and night temperature), 14000 lux (only day) and 70 % relative humidity.

After germination and a growing period, a hardening treatment was carried out at +2 °C for 4 days. It was followed by the *in vitro* chilling treatment which consists of the following steps: 0 °C (4 days), -2 °C (4 days), -3 °C (5 days). Before evaluation, a regeneration period of 14 days on 15/10 °C took place. The whole program was carried out in presence of moderate illumination. The poppy plants were grown until its 4-6 true leaf stage, which took average 1.5-2 months. This is the earliest phenological stage which is suitable for testing the frost tolerance of plants in controlled environment. 50 plants per genotype were grown.

The three-year experiments were a part of several projects (OTKA K 62732 and industrial errands between 2008 and 2011). The trials were carried out in different periods and on different parcels, in which many poppy cultivars, strains and hybrids were tested. Therefore in this current study the results fit to comprehensive labour processes and were made in diverse populations (*Table 1.*) and more experimental cycle.

Table 1.: *Papaver somniferum* L. cultivars and taxona were tested in the experiments

Registered cultivars, cultivar candidates		Selected strains from the selection work of the Department	Hybrid generations from cross breeding	
spring ecotype	winter poppy		F2	F3
'Al'	'Leila'	„67” (2002)	'Ametiszt' x 'Leila'	'Ametiszt' x 'Leila'
'Ametiszt'	'Kozmosz'		'Kozmosz' x 'Korona'	'Kozmosz' x 'Korona'
Bence	Monte		'Kozmosz' x 'Minoán'	'Kozmosz' x 'Minoán'
'Botond'	'Zeno'		'Kozmosz' x 'Medea'	'Kozmosz' x 'Medea'
'Korona'			'Kozmosz' x 'Przemko'	'Kozmosz' x 'Przemko'
'Medea'			'Korona' x 'Kozmosz'	'Korona' x 'Kozmosz'
'Minoán'			'Korona' x 67	'Korona' x 67
'Óriás kék'			'Leila' x 'Przemko'	'Leila' x 'Przemko'
'Przemko'			'Leila' x 1/172	'Leila' x 1/172
'Tebona'			'Leila' x 'Ametiszt'	'Leila' x 'Ametiszt'
			'Medea' x 'Kozmosz'	'Medea' x 'Kozmosz'
			'Medea' x 67	'Medea' x 67
			'Minoán' x 'Kozmosz'	'Minoán' x 'Kozmosz'
			67 x 'Korona'	'Przemko' x 'Kozmosz'
			67 x 'Minoán'	67 x 'Korona'
			67 x 'Medea'	67 x 'Minoán'
			1/172 x 'Leila'	67 x 'Medea'

2.2. MATERIAL AND METHOD OF THE EXPERIMENTS

2.2.1. Examinations of frost tolerance

For the frost tolerance examinations the seeds were sown in open field in the end of September in 2008, 2009 and 2010. 3 cultivars registered as winter poppy ecotype, 3 spring cultivars and some hybrids from F2 and F3 generation were used for the trials. The assessments of frost tolerance of the plants were made in spring in 2009, 2010 and 2011. In case of bonitation the density of rows was established by visual inspection and categorized in 5 classes (rare: 20 %, rare-medium: 40 %, medium: 60 %, medium-dense: 80 % and dense: 100 %), in 6 replications (2 parcel, 2x3 rows). We determined the overwintering percentage of the plants by this method.

2.2.2. Soluble sugar content experiments

The analyse of soluble sugar content as one of the possible marker of frost tolerance of poppy was investigated on open field in different cultivars in case of spring and autumn sown populations using several replications. The seeds were sown in spring of 2009. Spring ecotypes and winter poppy cultivars, as well as hybrids belonging to the F3 generation were used. For determining the quantity of soluble sugar content present in different genotypes without triggering

any stress, the samples were taken in May of 2009. In this period the plants were in leaf rosette stage.

To ascertain the dynamic of accumulation of sugar content during winter caused by frost stress, the seeds were sown in September of 2009. We used spring and winter poppy cultivars and hybrids belonging to the F2 and F3 generations. The samples were taken in the leaf rosette stage of poppy plants. Samples were taken three times: before frost (when the average temperature did not fall down under 0°C), after the frost set in (when the average temperature was under zero for one week at least) and in the next year, in spring (when the frost has passed and when the plants could regenerate).

After the cleaning and grinding of the experimental material which contained the whole plant (root, stem and leaf as well), we measured 3 g per genotype in three replications. The determination of fructose, glucose and saccharose content was carried out with HPLC (high-performance liquid chromatography) method.

2.2.3. Examinations regarding proline content

The proline content of 5-6 true leaf poppy plant was determined *in vitro* in the summer of 2009. The plants were grown in a climatic chamber and a light thermostat in the Department of Medicinal and Aromatic Plants. For this we chose two characteristically frost tolerant winter poppy cultivars and two sensitive spring ecotype poppy cultivars. For the examination we collected the samples six times in six different temperatures of the chilling treatment: 15/10 °C (06.08.), +2 °C (10.08.), 0 °C (14.08.), -2 °C (17.08.), -3 °C (21.08.) and 10/15 °C (24.08.).

The proline content of poppy plants was analysed in open field also, in case of the different cultivars, both in spring sown and overwintering autumn sown populations. The seeds were sown in the spring of 2009. Several winter poppy cultivars and spring ecotype cultivars as well as hybrids belonging to the F3 generation were used for the experiment. From the spring sown populations the samples were taken in May of 2009, when the poppy plants were in a leaf rosette stage.

For determining the proline accumulation in the autumn sown, overwintering populations, spring and winter poppy ecotype cultivars were sown in September of 2010. To analyse the proline content of poppy plants, the samples were taken in the leaf rosette stage, three different times. Before frost, after the frost set in, and in the spring of the next year.

The proline content of the samples (root, stem and leaf) was determined on the bases of study of BATES et al. (1973) using ninhydrin reagent.

2.2.4. Histological experiments

For the histological experiments spring and winter poppy ecotype cultivars were used. The seeds were sown into 10 m² size parcels, in three replications, in the end of September 2010. The samples were taken in the next year (23.05.2011). The histological experiments were carried out in the Department of Botany and in the Soroksár Botanical Garden. Leaves were cross sectioned at the basal portion in the lamina (through the midrib) using a freezing microtome (Leitz Wetzlar). For the measurements all the chosen leaves were of the same stage of maturity. The anatomical features of the 25-30 µm thick sections were studied under a light microscope (Zeiss, Axio Imager A2) with dark-field illumination in 200 times magnification. Thickness of adaxial and abaxial epidermis cells and cuticle thickness of both epidermis layers were measured. For each variety 30 different measured data of the leaves were documented.

2.2.5. Relative water content experiments

In our experiment the relative water content of six poppy cultivars with 4-6 true leaflets were determined *in vitro*. The samples were taken during the 15/10 °C treatment. The relative water content was studied in case of ten randomly chosen individual plants on the bases of SCHONFELD et al. (1988) revised study.

2.2.6. Alkaloid content tests

We measured the alkaloid content of capsules of hybrids belonging to the F2 and F3 generations with different frost tolerance. Samples were taken both in the spring and autumn sown populations, under the same production site conditions. The seeds were sown in September 2009 and March 2010 into open field and the capsules were collected in July 2010. The samples were taken in 12 replications.

The accumulated alkaloid content of leaves of poppy plants were measured in case of several spring and winter poppy ecotype cultivars as well as F3 hybrids in spring sown populations. The samples were taken from leaf rosette stage poppy plants in the middle of May in three replications per genotype.

The alkaloid content analysis of capsules and leaves was carried out with TLC (thin layer chromatography).

2.2.7. Fatty oil content experiment

For the experiment spring and winter poppy cultivars were used. The seeds were sown in the end of March 2011 and the samples were taken in July 2011. Determination of the fatty oil content

of seeds was carried out with using Soxhlet extractor and Rotadest equipments (BAJPAI et al., 1999) in three replications.

2.2.8. Germination experiments

Spring and winter poppy cultivars were used for the germination experiments in spring of 2012. 3x50 pieces of seeds were studied per cultivar in two replications. The germination process was carried out following MSZ 6354-3:1991 numbered Hungarian Standard. 10 °C and 25 °C were chosen to the basis of comparison of spring ecotype and winter poppy cultivars.

2.3. STATISTICAL ANALYSIS

The data were processed using Microsoft Excel and Microsoft Word program. The statistical analysis were carried out with Pasw 18 statistical program. One-way and multifactor Variance Analysis, Two-Sample t-Test, Welch's t-test and Spearman's nonparametric regression analysis were used. The equality of variances was determined by Levene's test. The results were analysed on 95 % confidence level in all cases.

3. RESULTS AND CONCLUSIONS

Regarding the frost tolerance of poppy cultivars in open field, it can be established that the overwintering genotypes showed better frost tolerance in the mean of three years than the spring ecotype varieties (average of 276 %). The two groups are separated statistically, but there are differences within the groups also. According to overwintering data, the frost tolerance of hybrids made of the crossings of winter and spring ecotypes show intermediate character. It can be presumed also that the frost tolerance of the winter ecotype parent can be manifested in other ways in the progenies which depends on the spring ecotype parent (*'Kozmosz' x 'Korona'* shows better frost tolerance than *'Kozmosz' x 'Minoán'*). There aren't any significant changes between the different generations (F2-F3) based on data.

The leaves of poppy adapt to environmental conditions, thus anatomical changes appear in the tissue structure of the mesophyll as well as of the epidermis. We could observe significant difference between the cuticle thickness of spring ecotype and winter poppy varieties, which means that the genotypes that are able to overwinter have thicker cuticle than the spring varieties. This difference occurred both on the abaxial and the adaxial sides of the leaves, in an average of 1,858 and 2,349 μm . We can conclude that cuticle thickness can be a predictive characteristic of frost tolerant genotypes, although thinner cuticle is not necessary related to intolerance. Cuticle thickness as one of the markers of frost tolerance may offer the possibility of a rapid screening method for new cold tolerant winter ecotypes of poppy.

We proved that the relative water content of poppy cultivars, measured in 5-6 leaf stage and under 15/10 °C (which represents the average conditions in case of an autumn or spring sown population), do not differ significantly (winter poppy cultivars: 59,139 %, spring ecotype cultivars: 59,280 %). No relationship between the relative water content of genotypes and their prospective frost tolerances could be found.

Results showed that the winter poppy cultivars suitable for overwintering could accumulate more fatty oil than the spring ecotypes cultivars. Based on the tested inland cultivars it can not be excluded that the higher seed oil content is a typical feature of a better overwintering cultivars.

Generally the winter ecotype cultivars could germinate on every tested temperature (10 and 25 °C) and the lower temperatures only slowed down the germination of seeds but did not block it. In case of spring ecotype cultivars 25 °C seemed to block the germination, although in the reaction given on this temperature were strongly species-specific. Compared to the data of 28 years ago this means a remarkable difference suggesting a certain adaptation. Probably the genotypes of winter poppy cultivars have changed because of presumable selection pressure and their ecological amplitudes have increased.

Based on our study, in 4-6 leaf stage poppy plants a correlation can be assumed between the total soluble sugar content and the frost tolerance which can be proved by the r^2 values in case of both the cultivars ($r^2=0,774$) and the hybrids ($r^2=0,781$). At the same time it seems that the sugar accumulation (as a significant osmolytic) is specific not only for ecotype but for cultivar as well. These results may offer a possibility to develop a rapid screening method, which might facilitate the selection for overwintering in the course of breeding winter ecotype cultivars.

According to our data, one week of frost on open field does not induce an excessive proline accumulation in poppy plants. An average of 185 % change was detected. The frost effect triggered under artificial circumstances, in controlled temperature stages resulted proline changes in poppy genotypes. Accordance with our data of *in vitro* experiments, the frost tolerant winter ecotype cultivars accumulated an average of 221 % more osmoprotectant proline than the spring cultivars, which correlates with the frost tolerance of the cultivars. Based on the results, it can be concluded that proline may play a role in the frost tolerance of poppy but it is not the only factor determining this feature. The role of the cryoprotectant proline also depends on the strength and length of the frost effect and on the acclimation conditions before frost.

In an eight stages experiment an *in vitro* test method adapted for poppy was developed for the reliable and rapid separation of the 'winter' and 'spring' features of poppy genotypes. It ensures standard conditions for the separation of the ecotypes and produce statistically reliable result. We determined the phenological stage (4-6 leaf stage) of the plants which is optimal for testing frost tolerance; the parameters of the growing period (day/night temperature: 15/10 °C, 14000 lux light intensity, 70 % relative humidity); the conditions of cold treatment (+2 °C: 4 days, 0 °C: 4 days, -2 °C: 3 days, -3 °C: 5 days) as well as the regeneration condition of poppy plants (15/10 °C: 14 days).

It could be concluded that there is no definite correlation between the frost tolerance of poppy genotypes and the alkaloid content of capsules, what is very important result for the practice. The low alkaloid content poppy strains did not prove to be more frost tolerant than the ones with high alkaloid content. Populations showed lower total alkaloid content on the experimental plots selected by frost than the ones which derived from the spring sowing and have the same genetic composition. At the same time we found some combination in the crossed generations with very high individual alkaloid content (3.10 %) in the overwintered plots also, therefore we can assume that with targeted breeding, both high and low alkaloid content winter poppy genotypes can be produced.

We detected significant correlation between the leaf and the capsule alkaloid content with moderate strength ($r^2= +0,488$). The alkaloid content of the leaf can be the distinctive character for the different poppy genotypes (culinary or industrial purposes). Beside the total alkaloid content we can implicate the prospective main component in the capsule also, that occurs mainly in case of

those cultivars which have characteristically different alkaloid content (with narcotine or thebaine content). In the future this method may play a practical role in making the selection targeted and in speeding up the generations in the breeding process. Moreover, we can lay down the basics of a cultivar identification system assisting the official controls as well.

According to the results of the breeding process, the spectrum of cultivars suitable for both culinary and industrial purposes was broadened with valuable genotypes which can be used safely in overwintering cultivation. The frost tolerance observed in the F3 generation stabilized throughout the three generations of the selected strains. Due to industry interests, one of strains with thebaine content (thebaine content: 1.88 %, morphine content: 0,86 %) was announced to cultivar notification in the autumn of 2013 as '*Hunor*'.

4. NEW SCIENTIFIC RESULTS

Our research work is ended with the following scientific and practical results:

1. For the first time, data were reported regarding the frost tolerance of poppy in open field. It can be concluded that the genotypes bred for overwintering showed better frost tolerance in the average of three years than the spring ecotype cultivars.
2. The histological characteristics of poppy cultivars regarding frost tolerance were measured for the first time. It can be established that the winter ecotype cultivars of poppy have thicker abaxial and adaxial cuticle.
3. It was proved that the water content of the leaves in 5-6 leaf stage and on 15/10 °C did not differ significantly in case of the tested cultivars.
4. It can be stated that the winter poppy cultivars suitable for overwintering could accumulate more fatty oil than the spring ecotypes cultivars.
5. It was determined that the lower temperatures did not block the germination of seeds of winter ecotype cultivars.
6. A correlation can be assumed between sugar content measured under frost and frost tolerance.
7. It can be concluded that proline may play a role in the frost tolerance of poppy but it is not the only factor determining this feature.
8. An *in vitro* test method adapted for poppy was developed for the reliable and rapid separation of the 'winter' and 'spring' features of poppy genotypes.
9. No definite correlation could be found between the frost tolerance of poppy genotypes and the alkaloid content of capsules.
10. Significant correlation between the leaf and the capsule alkaloid content was detected; furthermore we can implicate the prospective main component in the capsule also.
11. According to the results of the breeding process, the spectrum of cultivars suitable for both culinary and industrial purposes was broadened with valuable genotypes. One of strains with thebaine content was announced to cultivar notification in the autumn of 2013 as '*Hunor*'.

Publications in relation to the PhD Thesis

Book chapter:

JÁSZBERÉNYI Cs.; KUROLI G.; NÉMETH L.; REISINGER P.; CSATHÓ P.; ÁRENDÁS T.; NÉMETH T.; FODOR N. (2012): Mák. In: Radics László (szerk): Fenntartható szemléletű szántóföldi növénytermesztéstan 2. Agroinform Kiadó, Budapest p. 411–421.

Publications with IF:

JÁSZBERÉNYI Cs.; LUKÁCS L.; INOTAI K.; NÉMETH É. (2012): Soluble sugar content in poppy (*Papaver somniferum* L.) and its relationship to winter hardiness. Zeitschrift für Arznei- und Gewürzpflanzen, 17(4): 169-174. **IF: 0,296**

JÁSZBERÉNYI Cs.; NÉMETH É. (2012): Connection of frost tolerance and alkaloid accumulation potential in poppy (*Papaver somniferum* L.). Journal of Applied Botany and Food Quality, 85(1): 116-119. **IF: 0,34**

NÉMETH-ZÁMBORI É.; **JÁSZBERÉNYI Cs.**; RAJHÁRT P.; BERNÁTH J. (2011): Evaluation of alkaloid profiles in hybrid generations of different poppy (*Papaver somniferum* L.) genotypes. Industrial Crops and Products, 33: 690-696. **IF: 2,469**

Publication without IF:

JÁSZBERÉNYI Cs.; NÉMETH É. (2010): Megfigyelések a mák (*Papaver somniferum* L.) jellemző morfológiai bélyegeinek öröklődéséről. Kertgazdaság. 43(2): 53-62.

International conference papers (full paper):

JÁSZBERÉNYI Cs.; VARGA D.; NÉMETH É.; ERŐS-HONTI Zs. (2012): Histological differences between the leaves of spring and winter ecotypes of poppy (*Papaver somniferum*) varieties. 47th Croatian & 7th International Symposium on Agriculture. Opatija (2012.02.13-17.) Proceedings p. 359-362.

International conference papers (abstract):

JÁSZBERÉNYI Cs.; NÉMETH É. (2011): *In vitro* test method of frost tolerance of poppy (*Papaver somniferum* L.). 1. Transilvanian Horticulture and Landscape Studies Conference, 8-9. April 2011., Marosvásárhely, Book of Abstracts p. 38.

JÁSZBERÉNYI Cs.; NÉMETH É. (2011): Frost tolerance of spring and winter ecotypes of poppy (*Papaver somniferum* L.). International Symposium on Papaver, 7-11, February, Lucknow, India, Book of Abstracts p. 9.

NÉMETH É.; **JÁSZBERÉNYI Cs.**; VARGA D.; BERNÁTH J. (2012): Background and results of development of winter poppy (*Papaver somniferum* L.) varieties in Hungary. 5th International Symposium Breeding Research on Medicinal and Aromatic Plants, Vienna, Book of Abstracts, 22.

Conference papers (Hungarian, abstract):

JÁSZBERÉNYI Cs.; VÉGVÁRI Gy.; NÉMETH É. (2009): Az oldható cukortartalom alakulása őszi és tavaszi mák (*Papaver somniferum* L.) ökotípusokban. Lippay-Ormos-Vas Tudományos Ülésszak. Budapest (2009. október 28-30.) Összefoglalók p. 108-109.

JÁSZBERÉNYI Cs.; CSONTOS L.; ÁRVAY K.; LUKÁCS N.; ZÁMBORINÉ N. É. (2010): Prolin felhalmozódása őszi és tavaszi ökotípusú mákban (*Papaver somniferum* L.). XVI. Növénynevelési Tudományos Napok. Budapest (2010.03.11.) Összefoglalók p. 81.

JÁSZBERÉNYI Cs.; ZÁMBORINÉ N. É. (2011): *In vitro* fagyteszt kidolgozása mák növényre (*Papaver somniferum* L.). XVII. Növénynevelési Tudományos Napok. Budapest (2011.04.27.) Összefoglalók p. 126.

VARGA D.; **JÁSZBERÉNYI Cs.**; NÉMETH É.; ERŐS-HONTI Zs. (2012): Tavaszi és őszi ökotípusú mákfajták (*Papaver somniferum* L.) leveleinek szövettani összehasonlítása. Kihívások és megoldások a XXI. század élelmiszertudományában. Záró konferencia 2012. január 18-19.