Arch. Biol. Sci., Belgrade, 64 (1), 175-182, 2012 DOI:10.2298/ABS1201175A

INFLUENCE OF PHOTOPERIOD ON VEGETATION PHASES AND TUBER DEVELOPMENT IN TOPINAMBOUR (*HELIANTHUS TUBEROSUS* L.)

S. TERZIĆ[1], JOVANKA ATLAGIĆ[1], IVANA MAKSIMOVIĆ[2], TIJANA ZEREMSKI[1], SOFIJA PETROVIĆ[2] and B. DEDIĆ[1]

[1] *Institute of Field and Vegetable Crops,* 21000 Novi Sad, Serbia
[2]*Faculty of Agriculture,* 21000 Novi Sad, Serbia

Abstract – A topinambour collection was analyzed to determine the genetic variability of 141 accessions in reaction to the length of day (light), its influence on vegetative and flowering phases, tuber number and mass. Day length significantly influenced flowering, which started with the first shorter days (15.6 h) in the third decade of June, while the majority of accessions flowered in the third decade of August (13.3 h). Differences between accessions were statistically significant for the analyzed phenotype traits. A significantly longer vegetative phase was found in Montenegrin accessions in comparison to the other groups of origin. Duration of the vegetative phase was significantly and positively correlated to tuber mass and negatively to their number, while duration of the reproductive phase had an opposite effect. For further work on topinambour breeding, it would be important to describe the mechanism of photoperiodic control of flowering initialization. Only by obtaining accessions neutral to the photoperiod could topinambour become a cultivated crop.

Key words: Genetic variability, *Helianthus tuberosus*, photoperiod, tubers, vegetation phase

INTRODUCTION

Of all the species in the *Helianthus* genus, topinambour (*Helianthus tuberosus* L.) and cultivated sunflower (*H. annuus* var. *macrocarpus* (DC) Ckll.) are among the first cultivated species on the territory of the USA (Heiser, 1995). The life cycle of topinambour can be divided into five basic phases: emergence, canopy development, tuber and rhizome formation, flowering and senescence (McLaurin et al., 1999). The time of flowering and tuber formation is influenced by day length and not by the time of sowing which limits growth to one generation per year (Shoemaker, 1927).

Topinambour, as a short-day plant, first needs long periods of light for vegetative growth, followed by short days that initiate the reproductive phase (Kays and Nottingham, 2008). The biochemical mechanism of plant sensitivity to photoperiodism lies in photomorphogenesis and it depends on a protein pigment phytochrome (Taiz and Zeiger, 2006). The most obvious influence of phytochromes is on cell permeability to K^+ and Cl^-, by which the osmotic pressure and cell size change to cause leaf and stem movement. Phytochromes can also influence gene transcription and cause plant greening (Taiz and Zeiger, 2006). The wide spectra and various mechanisms of phytochrome action imply that to determine the mechanism that influence a trait such as flowering, it is useful to find neutral genotypes or some with significantly different reactions.

There are ten topinambour collections in Europe, of which the largest one is in Serbia, and it is the second largest in the world (Kays and Notting-

ham, 2008). To determine the genetic variability of accessions in reaction to day length we assumed that: (1) because of the distinctively different geographical origin and the nature of accessions, there is significant variability in reaction to the day length; (2) the relation of the analyzed traits can be defined by calculating correlation coefficients and factorial analysis of variance; (3) better understanding of the material could be of significance for future work on the collection and the explanation of photoperiodic influence on flowering initialization in topinambour.

MATERIALS AND METHODS

The trial was performed in Rimski Šančevi, on the quarantine field of the Oil Crops Department of the Institute for Field and Vegetable Crops in Novi Sad. The material consisted of 141 accessions of *H. tuberosus*. The majority of accessions were brought to the collection after several collecting trips to Montenegro (73) and the USA (38). Topinambour cultivars (27) were obtained through exchange with other European collections (TUB BP) and three local populations were collected locally in the vicinity of Novi Sad and Grabovci.

Accessions were grown in plots 1.0 m wide and 7.5 m long with 1 row of 15 plants. Sowing was made by planting the tubers in the part of the quarantine field agrotechnically prepared for cultivated sunflower with a total surface of 1.2 ha. Plants were fertilized with 300 kg/ha of NPK fertilizer type 15:15:15. Plants were drip-irrigated to maintain maximum plant growth.

Phenotypic observations were made in the field during vegetation on ten plants per accession. Dates of emergence, forming of the flower bud (budding), flowering and senescence were noted for every accession. Tuber yield per plant, number, and mass were determined in growth conditions without competition for resources. The mass of individual tubers was determined on three plants with four replications (12 replications per accession).

Because of the various accession origins and levels of human influence on the material used in the trial, the factor of origin was added to the statistical analysis. Obtained raw data were analyzed using descriptive statistics and the variability was analyzed using factorial analysis of variance. Least significant difference (LSD) was calculated by multiplying critical values from the t distribution table with standard error of difference. Interrelationships between analyzed traits were determined by calculating Pearson's linear correlation coefficients.

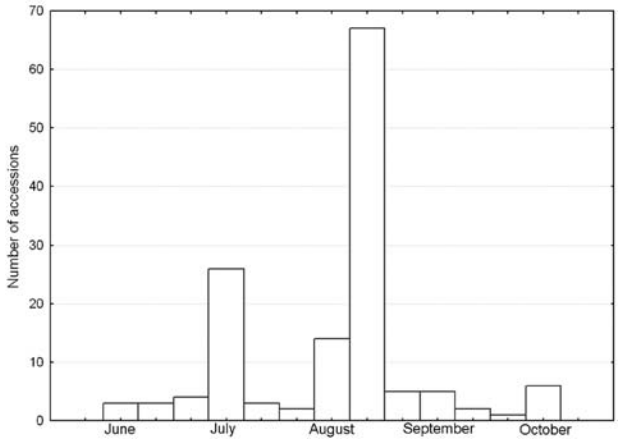
RESULTS

Day shortening starts in the third decade of June when the light period reaches a maximum of 15.5 h. In the third decade of August, the light period is in the range of 13 and 13.5 h, which is considered as a critical value for the start of the reproductive phase for the majority of accessions. Flowering began with the shortening of the day in the majority of the accessions, except for six of them, which did not even form flower buds until senescence. The beginning and duration of the flowering phase were very variable. Two maxima were found when the beginning of flowering was compared; the first was in the middle of July, and the second in the third decade of August (Graphs 1, 2).

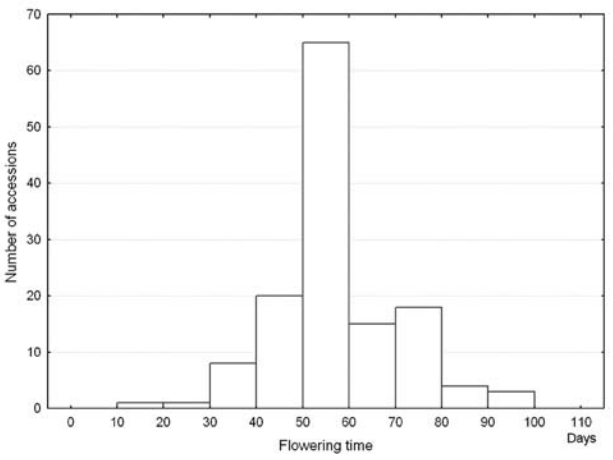
Vegetation phases

Among all the vegetation phases, the duration of budding and flowering was found to be the most variable. The shortest budding phase of 3 days was found for accession TUB 2062, while the longest of 68 days was found for TUB NS 2. Flowering time varied from 5 days in TUB CG 50 to 63 days in TUB 8 (Graph 3).

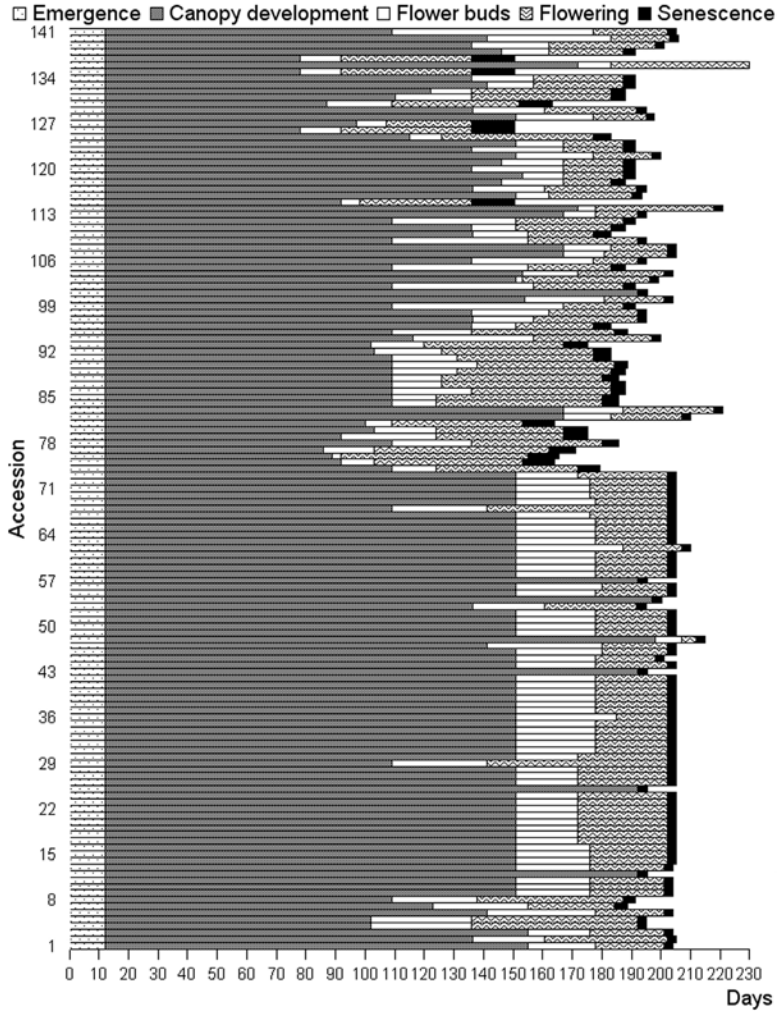
Five accessions from Montenegro (TUB CG 14, 27, 45, 56, 59) and one from the USA (TUB 2052) did not flower or form flower buds until senescence, although they all had a vegetative phase equal to or longer than 180 days (Graph 3).



Graph. 1. Beginning of flowering phase for 141 topinambour accessions



Graph. 2. Flowering phase duration of topinambour accessions

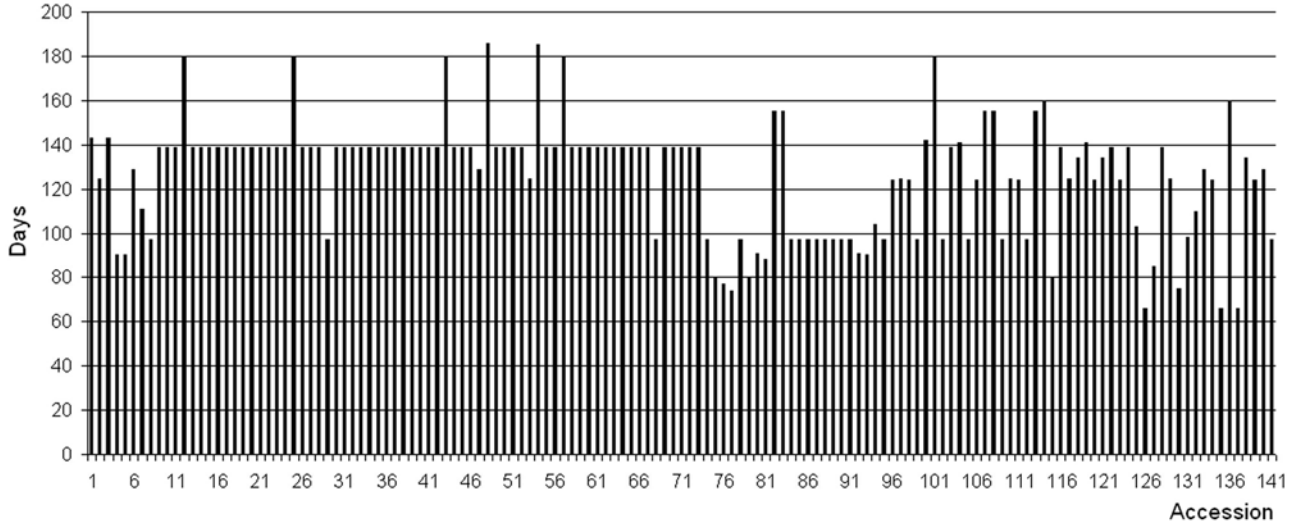


Graph. 3. Length of vegetation phases for topinambour accessions sorted according to their origin and given in days from planting to senescence (1-73 Montenegro, 74-111 USA, 112-138 cultivars, 139-141 local populations)

Table 1. Basic statistical parameters for vegetative phase duration of topinambour

	*N**	Mean	Min.	Max.	*Std. Dev.*	*CV* (%)	LSD (0,05)
Vegetative phase duration (days)	141	126	66	186	21	26	28

* *N* – total number of samples, *Std. Dev* – standard deviation, *CV* – coefficient of variation

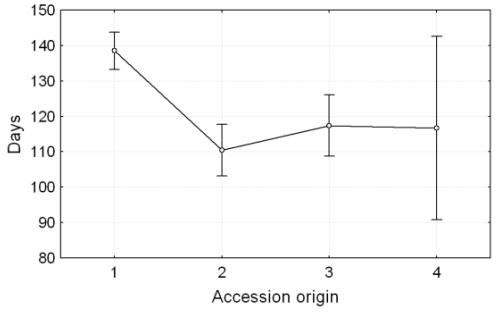


Graph. 4. Vegetative phase duration of topinambour (1-73 Montenegro, 74-111 USA, 112-138 cultivars, 139-141 local populations)

Vegetative phase

The longest phase in the vegetation of all accessions was vegetative (canopy development, from emergence to budding). It was shortest in the cultivars TUBBP 15, 24 and 26 with 66 days, and longest in accession TUBCG50 with 186 days (Tab. 1; Graph 4).

A significantly longer vegetative phase was found in the accessions from Montenegro in comparison to the cultivars and accessions collected in the USA. Variability within the groups varied, but it was two-fold times larges in the cultivars and accessions collected in the USA (CV=24 and 25%) than it was in the Montenegrin accessions (CV=13%) (Graph 4, 5).



Origin	1. (139)	2. (111)	3. (117)	4. (117)
1. Montenegro		0,001**	0,001**	0,104
2. USA	0,001**		0,225	0,648
3. Cultivars	0,001**	0,225		0,957
4. Local populations	0,104	0,648	0,957	

Graph. 5. Vegetative phase variability of topinambour in relation to the origin

Table 2. Vegetation phase length and tuber yield component correlations in topinambour

Vegetation phase	Accessions	Correlation coefficient	$P^{\S}$
Reproductive phase / Tuber mass	141	-0,442**	0,0001
Reproductive phase / Yield per plant	141	-0,382**	0,0001
Flowering / Tuber mass	141	-0,355**	0,0001
Flowering / Yield per plant	141	-0,354**	0,0001
Budding / Tuber mass	141	-0,261**	0,0018
Vegetative phase / Tuber number	141	-0,247**	0,0031
Budding / Yield per plant	141	-0,220**	0,0087
Budding / Tuber number	141	-0,085	0,3189
Reproductive phase / Tuber number	141	0,200*	0,0173
Vegetative phase / Yield per plant	141	0,311**	0,0002
Flowering / Tuber number	141	0,331**	0,0001
Vegetative phase / Tuber mass	141	0,347**	0,0001

§ p – risk of rejecting the null hypothesis, ** significant at the 0,01 level, * at 0,05

Vegetation phase length and tuber yield correlations

The reproductive phase (budding and flowering) was in negative correlation to the tuber mass and tuber yield per plant. The only positive correlation was found in relation to tuber number per plant. The length of the vegetative phase positively influenced tuber yield through increased tuber mass, even though it was negatively correlated to tuber number (Tab. 2).

DISCUSSION

Some phenotypic traits significantly depend on environmental conditions, where competition for resources has considerable influence. This is why phenotype evaluation was performed in controlled conditions where plants were grown with equal spacing and on a uniformly prepared field. Differences between the topinambour accessions were statistically significant for all evaluated phenotypic traits (duration of vegetative and reproductive phase, budding and flowering, tuber mass, tuber number and yield per plant), which confirms the existence of genetic variability among accessions.

Most of the analyzed accessions originated from Montenegro. By analyzing the influence of day length on flowering initiation and the length of the vegetative phase, it was found that their variability was lower than in accessions from the USA and cultivars. Accessions originating from the USA can be considered as typical representatives of the species from its natural habitat and the center of origin. That is why it could be expected for them to show greater variability than a group of accessions that after introduction, remained on the relatively small area of Montenegro (Dozet et al., 1993; Vasić et al., 2002). The cultivated accessions that were analyzed in this trial were obtained after substantial breeding efforts and it was expected they would differ significantly from the other accessions.

Flowering initiation and duration

The key phase of topinambour vegetation from which tuber yield is directly dependent is the vegetative. The initiation of the reproductive phase is induced by the photoperiod and because of this the photoperiodic response is one of the most analyzed traits in topinambour breeding. Ideally, a cultivar

should be neutral to the photoperiod because this would allow for easier planning of breeding efforts. The flowering of such cultivars would only depend on the rate of plant development, or, in other words, the planting date. Two groups of accessions were found for flowering initiation in regard to the day length. The first corresponds to the defined critical day length for topinambour of 13 to 13.5 h (Zhou et al., 1984) and these were mostly accessions from Montenegro and cultivars. The other group flowered when day shortening started at 15.5 h, and it contained mostly wild accessions from the US. There are two possible causes for the existence of the second flowering group. The first is that those are accessions neutral to photoperiod and that they flowered approximately in the same time because of the same planting date and similar growth conditions. The other possibility is that for those accessions, as short-day plants, either the critical day length is greater and close to 15 h, or that they react on the first shortening day. By early flowering these accessions ensure enough time for flowering, pollination and seed production, which is a characteristic for wild accessions and opposite to cultivars. Six accessions remained in the vegetative phase during the whole vegetation. It is possible that for these accessions the critical photoperiod was shorter than 10 h, which was the length at the beginning of November and the start of senescence. The reproductive phase of the other accessions started in mid June and lasted until the first half of October, which can point to photoperiod neutrality, especially for early flowering ones (Kays and Kultur, 2005), and variability in the duration of the vegetation phases.

Vegetation phase length and tuber yield correlations

Breeding had a substantial influence on the analyzed tuber traits. Selection pressure was made for developing larger tubers closer to the stem to obtain higher yield and easier harvest (Le Cochec, 1990). Wild accessions from the USA had a significantly higher tuber number and the lowest tuber yield per plant in comparison to the other groups. This implies a multiplication strategy where yield is not as important as the ability to survive and occupy new territory. In this respect a larger tuber number is beneficial (Swanton, 1986).

The calculated correlation coefficients for the duration of the vegetative and reproductive phases in relation to tuber mass and number per plant, confirmed different multiplication strategies between wild and cultivated accessions. Tuber mass and yield per plant was affected negatively by the duration of the reproductive phase and positively by the duration of the vegetative phase.

High yield per plant and tuber mass are traits by which cultivated accessions could be differentiated from other groups and confirm the effect of breeding. A few accessions with similar yield and tuber mass were only found in the Montenegrin group. On average, the plants formed 44 tubers, which classifies them as spreading, while most of the accessions in the fast-spreading group, with more than 69 tubers per plant (Pasko, 1973), were wild USA accessions.

Although there are cultivars neutral to photoperiod initiation of flowering (Kays and Kultur, 2005), there are still no cultivars in which tuber formation is not influenced by the shortening of the day (Kays and Nottingham, 2008). For the purpose of breeding, flowering is even artificially controlled by shading (Sawicka and Wadysaw, 2005).

To establish the possibility of controlling the influence of photoperiod on topinambour development, it is necessary first to determine the signal transduction pathway, starting from the phytochrome as the receptor of light. Photoperiod as a factor has multiple effects on the topinambour, such as the initiation of flowering, certain aspects of tuberization and vegetative growth (Hackbarth, 1937). This is why it is necessary to determine the point at which the photoperiodic influence on flowering can be interrupted without changing other processes.

A transduction pathway described on *Arabidopsis* indicates that several genes interact prior to flowering initialization (Velverde et al., 2004; An et al., 2004; Abe et al., 2005; Lifschitz et al., 2006). In cul-

tivated sunflower, temperature and photoperiod are found to be the key environmental factors affecting the time from emergence to floral initiation. Genetic investigations on the number of days from emergence to flowering suggested polygenic inheritance patterns. Some evidence was also found for genetic factors with major, qualitative effects on flowering and the transduction pathway (Leon et al., 2001; Dezar et al., 2011).

It is possible to apply various manipulations on the molecular level, such as antisense mRNA for the genes involved in the signal transduction pathway, to interrupt flowering but also to help explain their effect. Locating the genes that are responsible for the photoperiodic response in the topinambour could be initiated by comparing accessions that are neutral to the photoperiod with the wild type.

CONCLUSION

Differences between accessions and, in most cases, between the groups of origin were statistically significant for all the studied phenotypic traits (duration of the vegetative and reproductive phases, budding and flowering, tuber mass, tuber number and yield per plant), which confirmed the existence of genetic variability in the collection.

The vegetative phase duration can be regarded as an important trait for cultivated topinambour because it is positively correlated to tuber mass and yield and negatively to tuber number, which simplifies harvest. On the other hand, the reproductive phase in general, and the flowering phase, had the highest negative influence on tuber mass and yield per plant.

Wild and cultivated accessions differ in multiplication strategies. Cultivated accessions form smaller numbers of larger tubers during a long vegetative phase, while the wild-type forms large numbers of small tubers and have a long reproductive phase.

The transition from the vegetative to reproductive phase is influenced by the photoperiod in the topinambour. Therefore, it would be of great advantage for a cultivated form to be neutral to the photoperiodic initiation of flowering, because this would allow easier breeding and the topinambour could be further developed as a cultivated crop.

Acknowledgments - This research was supported by the Ministry of Science and Technological Development of the Republic of Serbia (Projects TR20080 and TR31025).

REFERENCES

Abe, M., Kobayashi, Y., Yamamoto, S., Daimon, Y., Yamaguchi, A., Ikeda, Y., Ichinoik, H., Notaguchi, M., Goto, K., and *T. Araki* (2005). FD, a bZIP protein mediating signals from the floral pathway integrator FT at the shoot apex. *Science* **309**, 1052–1056.

An, H., Roussot, C., Suarez-Lopez, P., Corbesier, L., Vincent, C., Pineiro, M., Hepworth, S., Mouradov, A., Justin, S., Turnbull, C., and *G. Coupland* (2004). CONSTANS acts in the phloem to regulate a systemic signal that induces photoperiodic flowering of *Arabidopsis*. *Development*. **131**, 3615–3626.

Dezar, C.A., Giacomelli, J.I., Manavella, P.A., Re´. D.A., Alves-Ferreira, M., Baldwin, I.T., Bonaventure, G., and *R.L. Chan* (2011). HAHB10, a sunflower HD-Zip II transcription factor, participates in the induction of flowering and in the control of phytohormone-mediated responses to biotic stress. *Journal of Experimental Botany*. **62**, 1061–1076.

Dozet, B., Marinković, R., Vasić, D., and *A. Marjanović* (1993). Genetic similarity of the Jerusalem artichoke populations (*Helianthus tuberosus* L.) collected in Montenegro. *Helia*. **18**, 41-48.

Hackbarth, J. (1937). Versuche über Photoperiodismus. IV. Über das Verhalten einiger Klone von Topinambur. *Der Züchter*. **9**, 113–118.

Heiser, C.B. (1995). Sunflowers: *Helianthus* (*Compositae*). In: *Evolution of Crop Plants*, 2nd ed., (Eds. J. Smartt, and N.W. Simonds), 1-531. Longman Scientific, Harlow, U.K.

Kays, S.J., and *F. Kultur* (2005). Genetic variation in Jerusalem artichoke (*Helianthus tuberosus* L.) flowering date and duration. *Hort. Science*. **40**, 1675–1678.

Kays, S.J., and *S.F. Nottingham* (2008). *Biology and chemistry of Jerusalem artichoke* (*Helianthus tuberosus* L.). CRC press, Boca Raton, FL, 1-478.

Le Cochec, F. and *D.G. de Barreda* (1990). Hybridation et production de semences a partir de clones de topinambour (*Helianthus tuberosus* L.). Rapport EN3B-0044 F, Com-

mission des Communautés Européennes Energie, CECA-Bruxelles.

Leon, A.J., Lee, M., and *F.H. Andrade* (2001). Quantitative trait loci for growing degree days to flowering and photoperiod response in Sunflower (*Helianthus annuus* L.). *Theor. Appl. Genet.* **102**, 497–503.

Lifschitz, E., Eviatar, T., Rozman, A., Shlit, A., Goldshmidt, A., Amsellem, Z., Alvarez, J.P., and *Y. Eshed* (2006). The tomato FT ortholog triggers systemic signals that regulate growth and flowering and substitute for diverse environmental stimuli. *Proc. Nat. Acad. Sci., USA*. **103**, 6398–6403.

McLaurin, W.J., Somda, Z.C., and *S.J. Kays* (1999). Jerusalem artichoke growth, development, and field storage. I. Numerical assessment of plant development and dry matter acquisition and allocation. *J. Plant Nutr*. **22**, 1303–1313.

Pas'ko, N.M. (1973). [Basic morphological features for distinguishing varieties of Jerusalem artichoke]. *Trudy po Prikladnoy Botanike, Genetike i Selektsii.* **50**, 91–101.

Sawicka, B., and *M. Wadysaw* (2005). Evaluation and productivity of *Helianthus tuberosus* L. in the conditions of central-east Poland. Electron. *J. Pol. Agric. Univ. Hort.* 8, http://www.ejpau.media.pl/volume8/issue3/art-42.html.

Shoemaker, D.N. (1927). The Jerusalem artichoke as a crop plant. *Technical bulletin.* **33**, 1-32.

Swanton, C.J. (1986). Ecological Aspects of Growth and Development of Jerusalem Artichoke (*Helianthus tuberosus* L.). *Ph.D. thesis,* University of Western Ontario, London.

Taiz, L., and *E. Zeiger* (2006). *Plant Physiology.* 4th ed., Sinauer Associates, Inc. 1-765.

Vasić, D., Miladinović, J., Marjanović-Jeromela, A., and *D. Škorić* (2002). Variability between *Helianthus tuberosus* L. accessions collected in the USA and Montenegro. *Helia*. **37**, 79-84.

Velverde, F., Mouradov, A., Soppe, W., Ravenscroft, D., Samach, A., and *G. Coupland* (2004). Photoreceptor regulation of CONSTANS protein in photoperiodic flowering. *Science*. **303**, 1003–1006.

Zhou, K.F., Zhang, A.H., Zou, S.B., and *M.L. Li* (1984). Studies on the photoperiodic responses in short-day plant *Helianthus tuberosus*. *Acta Bot. Sin.* **26**, 392–396.