МЕЖДУНАРОДНАЯ КОНФЕРЕНЦИЯ ПАМЯТИ Е.Н. СИНСКОЙ

9 - 11 ДЕКАБРЯ 2009 САНКТ-ПЕТЕРБУРГ

ГЕНЕТИЧЕСКИЕ РЕСУРСЫ КУЛЬТУРНЫХ РАСТЕНИЙ

ПРОБЛЕМЫ ЭВОЛЮЦИИ И СИСТЕМАТИКИ КУЛЬТУРНЫХ РАСТЕНИЙ

Светлой памяти Евгении Николаевны Синской посвящается



1889 - 1965

«...главное не то, что без великих мыслей мы оставались бы дикарями, а главное то, что от великих мыслей когда-нибудь станет человечнее на земле»

Е Н. СИНСКАЯ

(«Воспоминания о Н.И.Вавилове», 1991)

TAXONOMICAL POSITION OF Vavilovia formosa (Stev.) Fed. EVIDENCED FROM MORPHOLOGICAL AND MOLECULAR DATA

A. A. Sinjushin, N. V. Demidenko

Biological Faculty of M.V. Lomonosov Moscow State University, Moscow, Russian Federation e-mail: sinjushin@pisumsativum.org

А.А.СИНЮШИН, Н, В, ДЕМИДЕНКО. ТАКСОНОМИЧЕСКОЕ ПОЛОЖЕНИЕ Vavilovia formosa (Stev.) Fed. ПО ДАННЫМ МОРФОЛОГИЧЕСКИХ И МОЛЕКУЛЯРНЫХ ИССЛЕДОВАНИЙ

The Fabeae Rchb. tribe includes numerous representatives of exclusive agricultural value and seems to be studied precisely. Despite this, some features of its system remain under discussion, namely relations between Orobus and Lathyrus, status of Pisum intrageneric taxa etc. One of the most problematic forms is Vavilovia formosa (Stev.) Fed. which used to be treated as a part of Pisum, Lathyrus and Orobus genera by different authors or recognized as separate genus Vavilovia (Stev.) Fed. a Alophotropis Aschers. et Graebn. (for details see: Makasheva et al., 1973). The most contemporareview on family system also tends to separate it from other representatives of the tribe in generic state (Lock, Maxted, 2005).

Genus Vavilovia inhabits disrupted area in Caucasus, Iran, Anatolia and Lebanon, and characterized as endangered taxon (e.g., Akopian, Gabrielyan, 2008). Regardless of its taxonomic position, it was evaluated as closely related to pea thus being of possible practical interest. If treated as

member within *Pisum*, this represents the only perennial pea species and may be used as a source of perennial character for pea cultivars. However, all attempts to cross *Vavilovia* with *Pisum* failed or produced sterile hybrids (Golubev, 1990). The significant problems arose even with cultivating this rare plant beyond its natural habitat.

The paucity of material seems to be reasonable for deficiency of any detailed information on this plant. Morphological analyses provided somewhat controversial data. No molecular investigations were carried out on this species except work (Jansen et al., 2008) which however provided no additional information on Vavilovia status.

The given work was aimed at obtainment of more detailed results on *V. formosa* taxonomical position with usage of both morphological and molecular data. The herbarium specimens from Moscow State University herbarium (MW) and herbarium of Institute of Botany of the National Academy of Sciences (Republic of Armenia) were used as the material for this study. For comparison, six *Pisum sativum* ssp. *sativum* cultivars from different world regions, four wild *P. sativum* subspecies, and *P. fulvum* accessions from the collection of Genetics Dept. of Moscow State University and John Innes germplasm collection (Norwich, United Kingdom) were used. Except listed forms, 11 morphologically contrast *Lathyrus* species of different sections were analyzed; these were kindly provided by Dr. G.D. Levko (All-Russian Research Institute of Breeding and Seed Growing of Vegetables, Russian Federation, Tesnoi Gorodok, Russian Federation).

The following morphological features were found which separate *Vavilovia* accessions from *Pisum*: perennial life form (*Pisum* annual); narrow sepals with ciliate margins (glabrous and broad with overlapping margins in *Pisum*); leaf rachis ending with cusp (always tendril in *Pisum*); small stipules (of the same size as leaflets or larger in *Pisum*); leathery leaflets (*Pisum* thin); fluted leaf rachis cross-section (rounded in *Pisum*). All listed characters can be found in *Lathyrus* species but their interpretation is ambiguous. The life form (annual or perennial) is not used even to separate sections in *Lathyrus*. Leaf features are known to be very labile in ontogeny of legumes, and first pea leaves are always characterized with inversely-ovate leaflets, fluted rachises ending with short process or even lacking any specialized terminus and even small stipules in *P. fulvum*. Actually, *Vavilovia* leaves combine some features which can be found as juvenile in *Pisum* s. str. The calyx of the latter also possesses ciliate margins before anthesis.

Some characters which distinguish *Vavilovia* from all studied *Lathyrus* species (and hence drawing with peas together) can be evaluated as more significant. These are: elliptic seed hilum (linear in *Lathyrus*); absence of pubescence on all organs; dilated filaments of outer whorl stamens (as they are in *Pisum*; somewhat dilated filaments are also found in *L. maritimus*); plicate ptyxis (supervolute in *Lathyrus*, this trait is reported as one of high significance by F. Kupicha (1981)). It should be noted that these characters had never been found altered even in mutational variation of *Pisum* and hence can be referred to as strongly constant for genus identity. Standard (vexillum) shape of *Vavilovia* also resembles one of *Pisum* rather than of *Lathyrus*. As a whole, *Vavilovia* seems to be morphologically closer to proboid representatives of *Lathyrus*.

Two traits were cited to separate *Vavilovia* from *Pisum*, viz. pistil groove gaping only at base (at base and apex in *Pisum*) and non-cristate keel (cristate in pea) (see Gunn, Kluve, 1976; these authors reject these differences). Pistils seem to be grooved in the same way in both genera, as do other pistil features such as hair pattern (adaxial). As for keel crest, this feature is difficult to analyze on herbarium material and needs more detailed investigations.

At least two features distinguish *Vavilovia* from all other studied species. These are leathery leaflets and specific leaflet shape. In all studied plants, proximal leaflet half (i.e. one directed to leaf assement) is wider than distal (directed to leaf rachis tip) or has almost the same width. In all *Vavilovia* plants distal half is notably wider than proximal.

The Vavilovia specimens are not completely uniform in studied sample. All Armenian plants of the from the rest ones having leaflets with keel-like basement (rounded base in other specimens). This feature was used by A.A. Grossheim (1949) to separate two different species within Alophotropis (Vavilovia), namely A. formosa and A. ausheri, but in our opinion the whole genus should be treated as monotypic (at least basing on available material), probably with contrast forms interpreted as varieties.

Polymerase chain reaction (PCR)-based DNA markers were also used to reveal polymorphism between studied samples. Few types of markers (RAPD, CAPS, SSR, and IRAP) were compared, and ones with multiple amplicon seem to be more informative. SSR (microsatellite) markers were rejected as products with the expected molecular weight were obtained for *Pisum* samples only. Some polymorphism was also found in length of *trnH-psbA* spacer of chloroplast genome. This work is still in progress but some preliminary results point at certain similarity of *Vavilovia* and *Pisum* s. str.

For more detailed molecular analysis, the internal transcribing spacer region (ITS1-5.8 rRNA-ITS2) of nuclear DNA was sequenced from *Vavilovia* herbarium specimen and then compared with numerous representatives of Fabeae tribe (including *Pisum*, *Vicia*, *Lathyrus*, and *Lens*; *Cicer* (*Cicereae* Alef.) chosen as an outgroup) obtained from GenBank database. It had been shown that *V. formosa* can be generally accepted as a member of *Pisum* clade being more distant from *P. sativum* than *P. fulvum*. This fact indicates that taxonomical analysis of these two genera needs to be performed, including both pea species, not only *P. sativum*.

Conclusively, the original results somewhat contradict ones discussed in literature (see Lock, Maxted, 2005) and evidence for unity of *Pisum* and *Vavilovia* genera. Hence, the analyzed species should be treated as *Pisum formosum* (Stev.) Alef., which is monotypic but perhaps needs to be separated in monotypic section, as it was proposed by some authors (see: Makasheva et al., 1973).

Authors express their gratitude to Prof. S.A. Gostimskii (Moscow State University, Russian Federation) for helpful comments, Dr. J.A. Akopian (Institute of Botany of the National Academy of Sciences, Republic of Armenia) for providing herbarium specimen from territory of Armenia; Dr. M. Ambrose (John Innes Centre, Great Britain) and Dr. G.D. Levko (All-Russian Research Institute of Breeding and Seed Growing of Vegetables, Russian Federation) for kindly provided seeds of *Pisum* and *Lathyrus* species.

REFERENCES

Akopian J.A., Gabrielyan I.G. On high-mountain pea, Vavilovia formosa (Stev.) Fed. (Fabaceae) in Armenia. Crop Wild Relative. 2008. V. 6. P. 26-27.

Golubev A.A. Habitat, collections, cultivation and hybridization of *Vavilovia formosa* Fed. Bull. Appl. Bot., Genet. Pl. Breed. 1990. V. 135. P. 67-75.

Grossheim A.A. Identification of plants of the Caucasus. Moscow, 1949. P. 162.

Gunn C.R., Kluve J. Androecium and pistil characters for tribe Vicieae (Fabaceae). Taxon. 1976. V. 25. P. 563-575.

Jansen R.K., Wojciechowski M.F., Sanniyasi E., Lee S.-B., Daniell H. Complete plastid genome sequence of the chickpea (*Cicer arietinum*) and the phylogenetic distribution of *rps12* and *clpP* intron losses among legumes (Leguminosae). Mol. Phylogenet. Evol. 2008. V. 48. № 3. P. 1204–1217.

Kupicha F. Vicieae. Advances in legume systematics. Kew, 1981. P. 377-381. Lock J.M., Maxted N. Fabeae. Legumes of the world. Kew, 2005. P. 504-509.

Makasheva R.K., Drozd A.M., Adamova O.P., Golubev A.A. Perennial pea. Bull. Appl. Bot., Genet. Pl. Breed. 1973. V. 51. № 1. P. 44-56.