



Dianthus pseudocrinitus (Caryophyllaceae), a new species from Northeast of Iran identified by morphological and molecular data

JAMIL VAEZI¹*, MARYAM BEHROOZIAN², FARSHID MEMARIANI² & MOHAMMAD REZA JOHARCHI²

¹Department of Biology, Faculty of Sciences, Shahid Chamran University of Ahvaz, Ahvaz, Iran

²Department of Botany, Research Center for Plant Sciences, Ferdowsi University of Mashhad, Mashhad, Iran

*Corresponding author: J.vaezi@scu.ac.ir

Abstract

A new species from southwest of Bojnord, NE of Iran is described and illustrated here as *Dianthus pseudocrinitus* (Caryophyllaceae). This species is morphologically similar to *D. crinitus* subsp. *turcomanicus*, but can be distinguished from the latter by bract number, width of lower and upper leaves, margin of thickness of outermost bracts, length of calyx, length and width of anther, branches of stem, type of sheath of lower leaf, and tip shape of petal fimbria. The new species is also similar to *D. orientalis* subsp. *stenocalyx* in terms of floral characters, but they differ by several non-overlapping morphological characters. Results obtained from the morphological data are consistent with those obtained from the molecular phylogenetic trees based on sequences of the two copies of DFR1 gene, confirming phylogenetic affinity of the new species to *D. crinitus* subsp. *turcomanicus*.

Key words: Caryophyllaceae, *Dianthus*, morphology, NE of Iran, phylogenetic analysis, species delimitation

Introduction

The genus *Dianthus* Linnaeus (1753: 409) (Caryophyllaceae) with more than 300 species consists of a group of annual or perennial taxa. The genus is widespread in Eurasia and Africa, but the Mediterranean area acts as a diversity center for the whole genus (Constantinidis 1999, Valente *et al.* 2010).

The taxonomy of *Dianthus* is notoriously problematic due to, in particular, highly morphological diversity making species delimitation to be difficult (Tutin & Walters 1993, Balao *et al.* 2010). In addition, low molecular variation together with morphological diversity is characteristic of rapid species diversification (Hughes & Eastwood 2006, Meudt & Simpson 2006, Balao *et al.* 2010, Valente *et al.* 2010). The last process is a consequence of the evolutionary radiation which is commonly occurred in *Dianthus* (Balao *et al.* 2010, Valente *et al.* 2010). On the other hand, many researchers showed that polyploidy is a common phenomenon in *Dianthus* (Balao *et al.* 2009 and the references therein). This evolutionary force along with speciation often takes place via hybridization and genome duplication (Weiss *et al.* 2002).

The plant identification in recently evolved taxa such as *Dianthus* has become an important controversial issue in plant taxonomy (Rieseberg *et al.* 2006, Fazekas *et al.* 2009, Yan *et al.* 2011, Valente *et al.* 2010). The recognition of a new species in *Dianthus* still relies on overall similarities of certain morphological traits (Yilmaz *et al.* 2011, Shaulo & Erst 2012, İlçim *et al.* 2013). Nowadays, the use of a high number of morphological characters in a multivariate morphometric approach in combination with other techniques such as cytological, anatomical, and molecular criteria could provide valuable insights into the exact taxonomic status of an unknown taxon (Johnson *et al.* 2012, Qiu *et al.* 2012).

The Irano-Turanian floristic region comprises a large area of SW and Central Asia and it is connected to the Mediterranean, Euro-Siberian (boreal) and Saharo-Sindian regions. This area is an important center of diversity for plants, as highlighted by the high number of endemisms (see e.g. Takhtajan 1986, Zohary 1973). Several biogeographical units can be distinguished. One of them is the Khorassan-Kopetdagh floristic province that is

100 km² (calculated 53 km² using GeoCAT tool; Bachman *et al.* 2011) with severely fragmented populations. The habitats on the mountain steppes of the area are threatened mainly by overgrazing. Thus, an urgent planning is required for conservation of the fragile ecosystem and the threatened species.

Etymology:—Refers to the vegetative similarity to *D. crinitus* subsp. *turcomanicus* (Latin, *pseudocrinitus* = apparently similar to *crinitus*).

Key to three similar *Dianthus* species occurring in NE of Iran

1. Stems slender; petal limb fimbriate to one-third, rarely dentate, pink; calyx 20–25 mm long, shape of dent tip mucronate *D. orientalis* subsp. *stenocalyx*
- Stems strong; petal limb fimbriate from the middle to base, white to pale pink; calyx 20–32 mm long, shape of dent tip acute or acuminate..... 2
2. Petal limb fimbriate for 1/2 its length; bracteoles 4(–6), broad-membranous at margin; calyx 20–22 mm long *D. pseudocrinitus*
- Petal limb fimbriate for more than 1/2 its length; bracteoles (4)–6–8(–10), narrowed-membranous at margin; calyx 25–32 mm long..... *D. crinitus* subsp. *turcomanicus*

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References

- Akaike, H. (1973) Information theory as an extension of the maximum likelihood principle. In: Petrov, B.N. & Csaki, F. (eds.) *Second International Symposium on Information Theory*. Akademiai Kiado, Budapest, pp. 267–281.
- Altschul, S.F., Gish, W., Miller, E.W. & Lipman, D.J. (1990) Basic local alignment search tool. *Journal of Molecular Biology* 215: 403–410. Available from <http://blast.ncbi.nlm.nih.gov> (accessed: 01 April 2006).
- Assadi, M. (1985) The genus *Dianthus* L. (Caryophyllaceae) in Iran. *Iranian Journal of Botany* 3: 9–54.
- Bachman S., Moat J., Hill A.W., de la Torre, J. & Scott, B. (2011) Supporting Red List treat assessment with GeoCAT: geospatial conservation assessment tool. *ZooKeys* 150: 117–126.
<http://dx.doi.org/10.3897/zookeys.150.2109>
- Balao, F., Casimiro-Soriguer, R., Talavera, M., Herrera, J. & Talavera, S. (2009) Distribution and diversity of cytotypes in *Dianthus broteri* as evidenced by genome size variations. *Annals of Botany* 104: 965–973.
- Balao, F., Valente, L.M., Vargas, P., Herrera, J. & Talavera, S. (2010) Radiative evolution of polyploid races of the Iberian carnation *Dianthus broteri* (Caryophyllaceae). *New Phytologist* 187: 542–551.
<http://dx.doi.org/10.1111/j.1469-8137.2010.03280.x>
- Behroozian, M., Vaezi, J. & Joharchi, M.R. (2013) A karyological study of some *Dianthus* L. species (Caryophyllaceae) in Northeast of Iran. *Feddes Repertorium* (in press).
- Boissier, P.E. (1867) *Flora Orientalis*, Vol. 1, Geneva, 1017 pp.
- Constantinidis, T. (1999) *Dianthus haematocalyx* subsp. *Phitosianus* (Caryophyllaceae), a New Serpentine Endemic from Greece. *Phyton* 39: 277–291.
- Cronn, R.C., Small, R.L., Haselkorn, T. & Wendel, J.F. (2003) Cryptic repeated genomic recombination during speciation in *Gossypium gossypioides*. *Evolution* 57: 2475–2489.
<http://dx.doi.org/10.1554/02-705>
- Dellaporta, S.L., Wood, J. & Hicks, J.B. (1983) A plant DNA minipreparation: Version 2. *Plant Molecular and Biological Reporter* 1: 19–21.
<http://dx.doi.org/10.1007/bf02712670>
- Djamali, M., Akhani, H., Khoshravesh, R., Andrieu-Ponel, V., Ponel, P. & Brewer, S. (2011) Application of the Global Bioclimatic Classification to Iran: implications for understanding the modern vegetation and biogeography. *Ecologia Mediterranea* 37(1): 91–114.
- Doyle, J.J. & Doyle, J.L. (1987) A rapid DNA isolation procedure for small quantities of fresh leaf tissue. *Phytochemistry Bulletin* 19: 11–15.
- Farsi, M., Behroozian, M., Vaezi, J., Joharchi, M.R. & Memariani, F. (2013) The evolution of *Dianthus polylepis* complex

- (Caryophyllaceae) inferred from morphological and nuclear DNA sequence data: one or two species? *Plant Systematics and Evolution* 299: 1419–1431.
<http://dx.doi.org/10.1007/s00606-013-0804-z>
- Fazekas, A.J., Kesanakurti, P.R., Burgess, K.S., Percy, D.M., Graham, S.W., Barrett, S.C.H., Newmaster, S.G., Hajibabaei, M. & Husband, B.C. (2009) Are plant species inherently harder to discriminate than animal species using DNA barcoding markers? *Molecular Ecology Resources* 9: 130–139.
<http://dx.doi.org/10.1111/j.1755-0998.2009.02652.x>
- Felsenstein, J. (1988) Phylogenies from molecular sequences: Inference and reliability. *Annual Review of Genetics* 22: 521–565.
<http://dx.doi.org/10.1146/annurev.genet.22.1.521>
- Fet, V. (1994) Biogeographic position of Khorassan-Kopetdagh. In: Fet, V. & Atamuradov, K. I. (eds.) *Biogeography and Ecology of Turkmenistan*. Kluwer Academic Publisher, Dordrecht, pp. 197–204.
http://dx.doi.org/10.1007/978-94-011-1116-4_12
- Goldman, D.H., Klooster, M.R., Griffith, M.P., Fay, M.F. & Chase, M.W. (2011) A preliminary evaluation of the ancestry of a putative *Sabal* hybrid (Arecaceae: Coryphoideae), and the description of a new nothospecies, *Sabal × brazoriensis*. *Phytotaxa* 27: 8–25.
- Hall, T.A. (1999) BioEdit: a user-friendly biological sequence alignment editor and analysis program for Windows 95/98/NT. *Nucleic Acids Symposium Series* 41: 95–98.
- Henderson, A. (2006) Traditional morphometrics in plant systematics and its role in palm systematics. *Botanical Journal of Linnean Society* 151: 103–111.
<http://dx.doi.org/10.1111/j.1095-8339.2006.00526.x>
- Hollander, M. & Wolfe, D.A. (1999) Nonparametric Statistical Methods. 2nd ed. New York: John Wiley & Sons, 816 pp.
- Huelsenbeck, J.P. & Ronquist, F. (2001) MrBayes: Bayesian inference of phylogeny. *Bioinformatics* 17: 754–755.
<http://dx.doi.org/10.1093/bioinformatics/17.8.754>
- Hughes, C. & Eastwood, R. (2006) Island radiation on a continental scale: exceptional rates of plant diversification after uplift of the Andes. *Proceeding of the National Academy of Sciences, USA* 103: 10334–10339.
<http://dx.doi.org/10.1073/pnas.0601928103>
- Ilçim, A., Behçet, L. & Müksamre, M. (2013) *Dianthus vanensis* (Caryophyllaceae), a new species from Turkey. *Turkish Journal of Botany* 37: 219–224.
- IUCN (2013) *Guidelines for Using the IUCN Red List Categories and Criteria, version 10 Prepared by the Standards and Petitions Subcommittee of the IUCN Species Survival Commission*. Available from <http://intranet.iucn.org/webfiles/doc/SSC/RedList/RedListGuidelines.pdf> (accessed: February 2013).
- Jafari, A. & Behroozian, M. (2010) A Cytotaxonomic Study on *Dianthus* L. Species in North Eastern Iran. *Asian Journal of Plant Sciences* 9: 58–62.
- Jiang, J., Zhu, M. & Ai, X. (2013) Molecular evidence for a natural diploid hybrid between *Misanthus sinensis* (Poaceae) and *M. sacchariflorus*. *Plant Systematics and Evolution* 299: 1367–1377.
- Johnson, L.A., Chan, L.M., Burr, K. & Hendrickson, D. (2012) *Navarretia furnissii* (Polemoniaceae), a new diploid species from the intermountain western United States distinguished from tetraploid *Navarretia saximontana*. *Phytotaxa* 42: 51–61.
- Joly, S., Starr, J.R., Lewis, W.H. & Bruneau, A. (2006) Polyploid and hybrid evolution in roses east of the Rocky Mountains. *American Journal of Botany* 93: 412–425.
<http://dx.doi.org/10.3732/ajb.93.3.412>
- Kass, R.E. & Raftery, A.E. (1995) Bayes factors. *Journal of the American Statistical Association* 90: 773–795.
<http://dx.doi.org/10.1080/01621459.1995.10476572>
- Linnaeus, C. (1753) *Species Plantarum* 1. Laurentii Salvii, Stockholm, 560 pp.
- Memariani F., Zarrinpour V. & Akhani H. (2014) A review of plant diversity, vegetation and phytogeography of Khorassan-Kopetdagh floristic province in Irano-Turanian region (Northeastern Iran, Southern Turkmenistan). In: Box E. O. & Fujiwara K. (eds.) *Warm-temperate Deciduous Forests around the Northern Hemisphere*. Geobotany Studies, Springer-Verlag, Berlin (in press).
- Meudt, H.M. & Simpson, B.B. (2006) The biogeography of the austral, subalpine genus *Ourisia* (Plantaginaceae) based on molecular phylogenetic evidence: South American origin and dispersal to New Zealand and Tasmania. *Biological Journal of Linnean Society* 87: 479–513.
<http://dx.doi.org/10.1111/j.1095-8312.2006.00584.x>
- Mráz, P., Chrtk, J., Fehrer, J. & Pláčková, I. (2005) Rare recent natural hybridization in *Hieracium* s. str. – evidence from morphology, allozymes and chloroplast DNA. *Plant Systematics and Evolution* 255: 177–192.
<http://dx.doi.org/10.1007/s00606-005-0329-1>
- Müller, K. (2005) SeqState-primer design and sequence statistics for phylogenetic DNA data sets. *Applied Bioinformatics* 4: 65–69.
<http://dx.doi.org/10.2165/00822942-200504010-00008>
- Nylander, J.A.A. (2004) MrModeltest v2. Program distributed by the author, Evolutionary Biology Centre, Uppsala University. Available from <http://www.abc.se/~nylander/> (accessed: 01 February 2005).
- Page, D.M. (2001) TreeView (Win32) Version 1.6.6. Available from <http://taxonomy.zoology.gla.ac.uk/rod> (accessed: 03

September 2001).

- Popov, S.V., Shcherba, I.G., Ilyina, L.B., Nevesskaya, L.A., Paramonova, N.P., Khondkarian, S.O. & Magyar, I. (2006) Late Miocene to Pliocene palaeogeography of the Paratethys and its relation to the Mediterranean. *Palaeogeography Palaeoclimatology Palaeoecology*, 238 (1–4): 91–106.
<http://dx.doi.org/10.1016/j.palaeo.2006.03.020>
- Posada, D. (2004) Collapse: describing haplotypes from sequence alignments, Ver.1.2. Available from <http://darwin.uvigo.es/software/collapse> (accessed: 09 May 2004).
- Qiu, Z.J., Wang, X.L., Liu, Z.Y., Yang, J.F. & Zhang, S.Z. (2012) Cytological and phylogenetic study of *Petrocosmea hexiensis* (Gesneriaceae), a new species from Chongqing, China. *Phytotaxa* 74: 30–38.
- Rambaut, A. & Drummond, A.J. (2007) Tracer, version 1.4. Available from: <http://beast.bio.ed.ac.uk/tracer> (accessed: 11 October 2007).
- Rechinger, K.H. (1983) Acht neue Arten der Gattung *Dianthus* (Caryophyllaceae) aus dem Gebiet der Flora Iranica. *Plant Systematics and Evolution* 142: 239–246.
- Rechinger, K.H. (1986) *Dianthus crinitus* und *D. orientalis*, zwei polymorphe Arten und ihre geographischen Rassen im Gebiet der Flora Iranica. *Plant Systematics and Evolution* 151: 281–293.
<http://dx.doi.org/10.1007/bf00985901>
- Rechinger, K.H. (1988) *Flora Iranica* 163. Caryophyllaceae II. Akademische Druck– u. Verlagsanstalt, Graz.
- Rieseberg, L.H., Wood, T.E. & Baack, E.J. (2006) The nature of plant species. *Nature* 440: 524–527.
- Schischkin, B.K. (1936) *Dianthus turcomanicus*. In: Komarov, V.L. (eds.) *Fl. USSR*. vol. 6. Izdatel'stvo Akademii Nauk SSSR, Moscow, Leningrad, p. 853.
- Shaculo, D.N. & Erst, A.S. (2012) A new species of *Dianthus* L. (Caryophyllaceae) from the West Sayan, Altai-Sayan Mountains, Russia. *Feddes Repertorium* 122: 344–350.
<http://dx.doi.org/10.1002/fedr.201100007>
- Simmons, M.P. & Ochoterena, H. (2000) Gaps as characters in sequence-based phylogenetic analyses. *Systematic Biology* 49: 369–381.
- Smith, J.E. (1794) Remarks on the genus *Dianthus*. *Transactions of the Linnean Society* 2: 292–304.
<http://dx.doi.org/10.1111/j.1096-3642.1794.tb00262.x>
- Swofford, D.L. (2002) PAUP*: phylogenetic analysis using parsimony (*and other methods), version 4.0b10. Sinauer Press, Sunderland, Mass.
- Takhtajan, A. (1986) Floristic Regions of the World. University of California Press, California, 544 pp. (English translation from Russian).
- Thompson, J.D., Higgins, D.G. & Gibson, T.J. (1994) ClustalW: improving the sensitivity of progressive multiple sequence alignment through sequence weighting, position-specific gap penalties and weight matrix choice. *Nucleic Acids Research* 22: 4673–4680.
<http://dx.doi.org/10.1093/nar/22.22.4673>
- Tutin, T.G. & Walters, S.M. (1993) *Dianthus* L. In: Tutin, T.G., Burges, N.A., Chater, A.D., Edmondson, J.R., Heywood, V.H., Moore, D.M., Valentine, D.H., Walters, S.M. & Webb, D.A. (eds.) *Flora Europaea*, vol. 1. Cambridge, UK, Cambridge University Press, pp. 227–246.
- Vaezi, J. & Brouillet, L. (2009) Phylogenetic relationships among diploid species of *Symphyotrichum* (Asteraceae: Astereae) based on two nuclear markers, ITS and GAPDH. *Molecular Phylogenetics and Evolution* 51: 540–553.
<http://dx.doi.org/10.1016/j.ymp.2009.03.003>
- Valente, L.M., Savolainen, V. & Vargas, P. (2010) Unparalleled rates of species diversification in Europe. *Proceedings of the Royal Society B: Biological Sciences* 277: 1489–1497.
<http://dx.doi.org/10.1098/rspb.2009.2163>
- Weber, F. & Mohr, M.H. (1805) *Beiträge zur Naturkunde* 1. Kiel, in der Neuen akademischen Buchhandlung, 400 pp.
- Weiss, H., Dobes, C., Schneeweiss, G.M. & Greimler, J. (2002) Occurrence of tetraploid and hexaploid cytotypes between and within populations in *Dianthus* sect. *Plumaria* (Caryophyllaceae). *New Phytologist* 156: 85–94.
<http://dx.doi.org/10.1046/j.1469-8137.2002.00500.x>
- Yan, H.F., Hao, G., Hu, C.M. & Ge, X.J. (2011) DNA barcoding in closely related species: A case study of *Primula* L. sect. *Proliferae* Pax (Primulaceae) in China. *Journal of Systematics and Evolution* 49: 225–236.
<http://dx.doi.org/10.1111/j.1759-6831.2011.00115.x>
- Yilmaz, O., Kaynak, G., Daşkin, R. & Mericlioğlu, A. (2011) *Dianthus goekayi* (Caryophyllaceae), a new species from Turkey. *Annales Botanici Fennici* 48: 74–78.
<http://dx.doi.org/10.5735/085.048.0111>
- Zohary, M. (1973) *Geobotanical Foundations of the Middle East*. 2 Vols. Gustav Fischer Verlag, Stuttgart, 765 pp.