

A new species of *Verbascum* L. (Scrophulariaceae) from the Gilan province (Iran), based on morphological and molecular evidences

Arash SOTOODEH

Université de Toulouse, UPS, EDB, UMR 5174,
118 route de Narbonne, F-31062 Toulouse cedex 09 (France)
arash29@gmail.com

Farideh ATTAR

University of Tehran, College of Science,
Central Herbarium of Tehran University, School of Biology, Tehran (Iran)

Laure CIVEYREL

Université de Toulouse, UPS, EDB, UMR 5174,
118 route de Narbonne, F-31062 Toulouse cedex 09 (France)

Published on 24 June 2016

Sotoodeh A., Attar F. & Civeyrel L. 2016. — A new species of *Verbascum* L. (Scrophulariaceae) from the Gilan province (Iran), based on morphological and molecular evidences. *Adansonia*, sér. 3, 38 (1): 127-132. <http://dx.doi.org/10.5252/a2016n1a9>

ABSTRACT

A new endemic species from north of Iran: *Verbascum parsana* Sotoodeh, Attar & Civeyrel, sp. nov. is described. Considering the combination of some characteristics like ebracteolate and single flowers, violet hairs on filaments, pedicel size between 3 to 10 mm and stellate-glandular indumentum, the new species is related to *Verbascum punalense* Boiss. & Buhse, but several differences have been observed between the two species: the shape of the anthers, the petiole of basal leaves, the corolla size and the calyx hairs. We investigate the genetic distance of the new species with close genera using ITS and plastid regions (*trnS-G*, *trnL-F*, *trnH-psbA* and partial *matK*). The new species showed significant molecular and morphological distance from closely related species. Photos and distribution map are presented.

KEY WORDS
Iran,
Gilan,
Scrophulariaceae,
Parsana,
Phylogeny,
new species.

RÉSUMÉ

Une nouvelle espèce de Verbascum L. (Scrophulariaceae) de la province de Gilan (Iran), basée sur des caractères morphologiques et moléculaires.

Une nouvelle espèce endémique de la province de Gilan dans le nord de l'Iran: *Verbascum parsana* Sotoodeh, Attar & Civeyrel, sp. nov. est décrite. Considérant la combinaison de certaines caractéristiques comme des fleurs solitaires et sans bractéoles, des poils violets sur les filaments des anthères, la taille du pédoncule floral entre 3 et 10 mm, les feuilles basales pétioleées et les poils étoilés ou glanduleux, cette nouvelle espèce est proche de *Verbascum punalense* Boiss. & Buhse, mais diffère par plusieurs caractères concernant la forme des anthères, la longueur du pétiole des feuilles basales, la taille de la corolle et les poils du calice. Nous avons réalisé une analyse de distance en comparant les séquences de ces espèces avec ITS et des régions plastidiques (*trnS-G*, *trnL-F*, *trnH-psbA* et une partie de *matK*) et les caractères morphologiques. Les analyses de distance morphologique comme moléculaire avec les espèces proches ont montré une séparation significative avec les autres *Verbascum* et permettent de conclure qu'il s'agit bien d'une espèce distincte. Photos et carte de distribution sont présentées.

MOTS CLÉS
Iran,
Gilan,
Scrophulariaceae,
Parsana,
phylogénie,
espèce nouvelle.

TABLE 1. — Main characters distinguishing *V. parsana* Sotoodeh, Attar & Civeyrel, sp. nov. from *V. punalense* Boiss. & Buhse.

	<i>V. punalense</i>	<i>V. parsana</i>
Anther	decurrent	reniform
Calyx lobes shape	ovate or oblong-elliptic, obtuse	ovate, acute
Calyx indumentum	dense stipitate-glandular hairs	sparse glandular+stellate hairs
Corolla	25-40 mm long, outside sparsely branched hairs	15 mm long, outside densely stellate + glandular hairs
Basal leaves	50-60 × 12 × 15 cm, oblong, irregularly double crenate at margin	10 × 4 cm, elliptic, attenuate at base, indistinctly crenate at margin
Petiole of basal leaves	absent	5 cm long

INTRODUCTION

The genus *Verbascum* L. (Scrophulariaceae) in Iran includes 43 species and three hybrids (Huber-Morath 1981; Sharifnia 2011; Sotoodeh *et al.* 2014, 2015) from which 20 are endemic. The genus *Verbascum* is widespread in Iran, nevertheless most of the species of *Verbascum* have a restricted distribution in Iran, with the exception of *V. cheiranthifolium* Boissier and *V. songaricum* Schrenk ex Fisch & C. A. Meyer, which have the broadest distribution in Irano-Turanian region. Species are more abundant in the Northwest regions (Huber-Morath 1981) especially Irano-Turanian and Hyrcanian regions than in the southern desert regions. Huber-Morath (1981) divided this genus in *Flora Iranica* into two groups based on indumentum type and number of flowers in each axil. The other important characters of *Verbascum* include the number of stamens, the color of hairs's filaments, the size of corolla and the length of pedicels.

The new species was encountered during a systematic revision of Iranian *Verbascum* as part of the first author's Ph.D thesis. Examining *Verbascum* specimens in the Central Herbarium of Tehran University (TUH after Holmgren *et al.* 1990), an unnamed specimen was noticed (collected by: Moazzeni & Keshvari, TUH 34941, 12.VIII.2004). It did not match any known species and was different of the other Iranian species. We used all the published taxonomic and molecular works available for *Verbascum* for comparison (Murbeck 1933; Parsa 1952; Feinbrun-Dothan 1978; Huber-Morath 1978; Huber-Morath 1981; Fedtschenko 1997; Karaveliogulları & Aytaç 2008; Sharifnia 2011; Sotoodeh *et al.* 2014, 2015; Ghahremaninejad *et al.* 2015) and it remained distinct to all species checked. Also we investigated the distance of the new species with close genera using ITS and plastid regions (*trnS-G*, *trnL-F*, *trnH-psbA* and partial *matK*).

MATERIAL AND METHODS

The specimens compared with different Iranian *Verbascum* collections in various European (BM, K, MPU, P) and Iranian (TUH, TEH, HKS, IRAN, TARI) herbaria and also from

specimens scanned from other Herbaria on Jstor Global Plants (<https://plants.jstor.org>). When we compared the unidentified specimen, the only species that could be mistaken with this new species was *V. punalense* Boiss. & Buhse, because of having single flowers, stellate and glandular hairs and the same number of stamens. Nevertheless it differs in corolla size and its hairs, anther shape, basal leaves and petioles. Also we incorporated all available species of *Verbascum* in the phylogenetic analysis (Sotoodeh pers. comm.).

To discriminate *Verbascum parsana* sp. nov., *V. punalense* and *V. shahsavarensis*, we performed hierarchical clustering (R function hclust) on the characters and visualized the results as a dendrogram. The dendrogram was constructed based on the five genes sequences and also their morphological characteristics. Morphological characters considered are given in Table 1 and the matrix formed by morphological and molecular data in Appendix 1. The distance tree created using the “dist” (euclidean distance) and hclust (average or UPGMA method) of R function (Dunn & Everitt 2004; Parikh *et al.* 2010; Paradis 2011; Baselga & Orme 2012; Murtagh & Legendre 2014).

RESULTS

Verbascum parsana Sotoodeh, Attar & Civeyrel, sp. nov.
(Fig. 1)

Planta verosimiliter biennis, indumento cinereo e pilis stellato-ramosis, glandulosa. Folia basalia oblonga-elliptica, margine crenata, petiolata; caulina basilibus conformia sed minora, non decurrentia. Inflorescentia paniculato-ramosa; bractee lineares-acuminatae; pedicellus usque 8 mm longus; calyx 4-6 mm longus; corolla lutea, 15 mm diam., extus stellato-glandulosa; stamina 5; filamenta dense villosa-velutina violacea; filamenta antica apice nuda; stigma. Capsula elliptica, obtus, 5-7 mm longa, stellato-glandulosa.

TYPUS. — Iran. Gilan, Asalem to Khalkhal (Asbdavani), 1876 m, 37°37'4.68"N, 48°46'2"E, 12.VIII.2004, Moazzeni & Keshvari 34941 (holo-, TUH!; iso-, P[P04022112]).

HABITAT AND ECOLOGY. — *Verbascum parsana* sp. nov. is known only from the type specimen and one locality (type location) in north of Iran (Fig. 2). Based on present study it is a narrow endemic taxon. The new species grows on porous and soft sandy-clay soils at elevation of 1876 m in transition zone out of forest.

ETYMOLOGY. — The epithet is to honor Dr Ahmad Parsa (1907-1997), the first Iranian botanist who produced important concepts for flora of Iran.

OTHER SPECIMENS EXAMINED. — *Verbascum hervieri* Degen., Spain: Le Pozo, 1500 m, 1905, *Elisée Reverchon 1375* (P[P03803835, P03803836, P03803837, P03803838], images!).

Verbascum microcarpum Benth., Mesopotamia: 1837, *Aucher-Eloy 2458* (P[P03287023], image!).

Verbascum punalense Boiss. & Buhse, Iran: Tonekabon, 1085 m, 2012, *Arash Sotoodeh 43467* (TUH!); Tonekabon, 250 m, 2012, *Arash Sotoodeh 43464* (TUH!); Gilan, 1893, *Punal Buhse – no number* (K!); Mazandaran, Kujur, 1800 m, 1948, *Rechinger 6543* (BM!).

Verbascum shahsavarensis Sotoodeh, Attar & Civeyrel, Iran: Tonekabon (Shahsavar), road of Sehezar, 36°27'59"N, 50°51'13"E, 1570 m, 30.VI.2014, *Arash Sotoodeh et al. AS29* (holo-, TUH!; iso-, P[P04021689]!, K).

Verbascum siculum Tod. Ex Lojac., Italy: Bosco Bauli Siracusa, 1976, *Brullo 057058* (CAT, image!).



FIG. 1. — *Verbascum parsana* Sotoodeh, Attar & Civeyrel, sp. nov.: **A**, herbarium specimen; **B**, **C**, a part of inflorescence and fruits, bract and no bracteole; **D**, corolla and androecium; **E**, arrows show two anterior anthers. Photographs from the type specimen by A. Sotoodeh. Scale bars: 1 cm.

Verbascum spectabile M. Bieb., Turkey: Giresum-Susehri, 1700 m, 1977, *Max Nydegger 12948* (P[P03836032], image!); Tauria, 1875, *no number* (P[P03416378, P03416379], image!).

Verbascum subnivale Boiss. & Hausskn. ex Boiss., Turkey: Montis Berytdagh, 9000 ped, 1865, *Hausknecht – no number* (iso-, JE[JE00007237], P[P03287723], image!).

Verbascum virgatum Stokes, India: Nilgiri, 1885, *Beddome 5631* (BM!). — France. Rennes, *Legris* (P[P00700061], image!); Cognac-le-Froid, 7.VIII.1954, *H. Bouby 3190* (P[P00687503], image!). — Spain. Huelva Galarosa, 28.V.1931, *E. Gros* (P[P03986576], image!).

Verbascum wiedemannianum Fisch. & C.A.May:—Turkey. Karasar-

Gecidi, 1650 m, 6.VI.1987, *Nydegger 42651* (P[P03807346], image!); Merzifoun, 1891/92, *Manissadjian 182* (P[P03287104, P03287105], images!); Paphlagonia, Wilajet Kastambuli Tossia Seke, *Sintenis 4010* (P[P03287101, P03287103, P03287106], images!); Arac-Kastamonu, 1985, *Norman 7* (K!); SE of Bayburt, 1150m, 1965, *Mathew & Tomlinson 4340* (K!); Keyseri-Bonyan, 1954, *Davis 21793* (K!).

DESCRIPTION

Plant, probably biennial, covered by sparse gray stellate and glandular hairs on basal parts and mostly glandular on

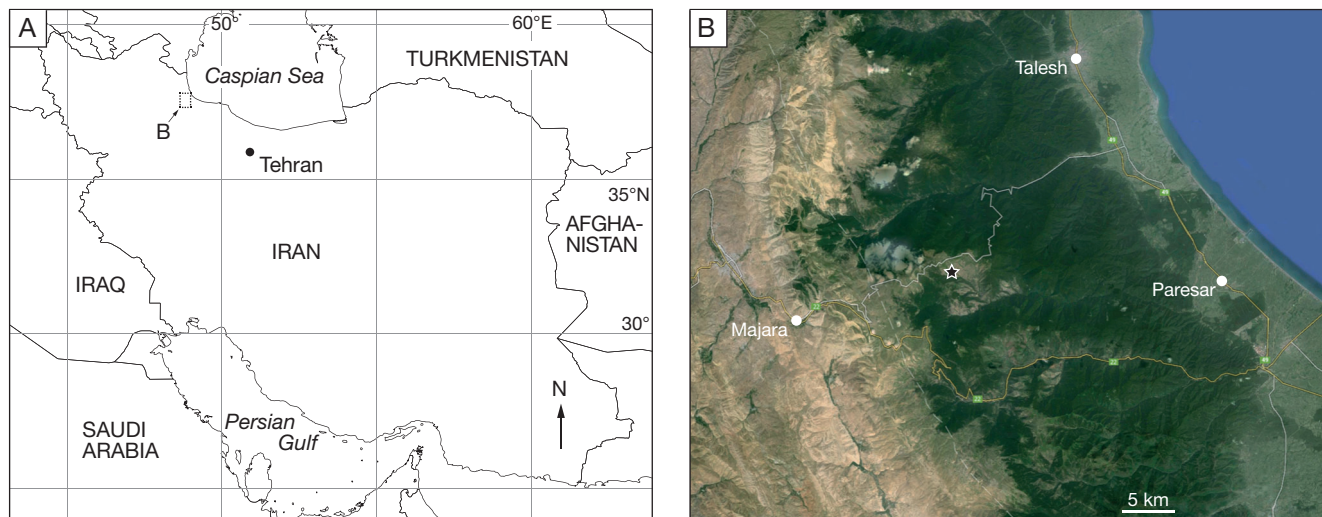


Fig. 2. — Distribution of *Verbascum parsana* Sotoodeh, Attar & Civeyrel, sp. nov. (★) The magnified view on the right is from Google map (maps.google.com).

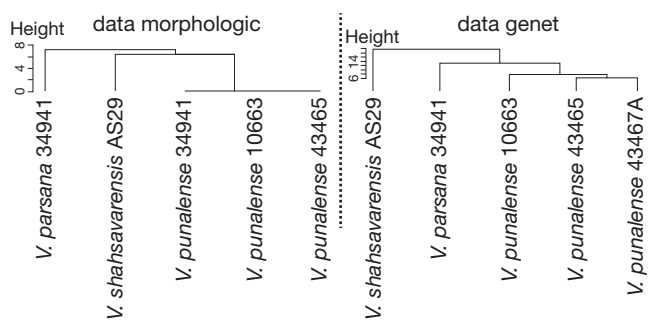


Fig. 3. — Distance based trees on morphological (left) and genetic (right) characters used to differentiate species of *Verbascum parsana* Sotoodeh, Attar & Civeyrel, sp. nov., *V. punalense* Boiss. & Buhse and *V. shahsavarensis* Sotoodeh, Attar & Civeyrel.

inflorescence region. Stem up to 60 cm long, terete. Basal leaves oblong-elliptic, semi-crenate at margin, with up to 5 cm long petiole, tapering toward apex, 5-8 × 2-3 cm; cauline leaves are similar to basal ones but smaller, not decurrent. Inflorescence paniculiform, lax; flowers single at the axil of each bract; bracts linear-acuminate; bracteoles absent; pedicel up to 8 mm long (Fig. 1B, C), covered with stellate and glandular hairs; calyx 4-6 mm long, divided, with elliptic segments (Fig. 1B, C); corolla yellow, 15 mm in diam (Fig. 1D), densely stellate and glandular hairy outside; stamens 5; hairs of filaments violet; 2 anterior stamens naked from the top to $\frac{3}{4}$ (blue arrows Fig. 1E), only the last quarter of the filaments close to the corolla hairy (Fig. 1E); anthers reniform, 4-5 mm long; stigma 4-5 mm (Fig. 1D). Capsule elliptic-obtuse, 5-7 mm long, covered with dense stellate and glandular hairs (Fig. 1C).

DISCUSSION AND CONCLUSION

The new species described as *V. parsana* sp. nov. has a combination of characters that is uncommon within the genus

Verbascum. Only a few species have both stellate and glandular hairs together such as *V. punalense* and *V. sinuatum* L. var. *adenosepalum* Murb.. If we consider the combination of characteristics: ebracteolate and single flowers, violet hairs on filaments, pedicel size between 3 to 10 mm and stellate-glandular indumentum, the new species is closely related to *V. punalense* but cannot be mistaken with it because it has no decurrent anthers, the basal leaves are narrow, the corolla is smaller, its calyx lobes are acute and have sparse glandular-stellate hairs (Table 1).

Based on Murbeck (1933), the new species belongs to the subsection II, Singuliflora, and to the group ebracteolata. Seven species have been described in this section. But they all differ from *V. parsana* sp. nov.

There is no species looking like this new species in any of the recent floras published since then (Ferguson 1972; Feinbrun-Dothan 1978; Huber-Morath 1978; Huber-Morath 1981; Fedtschenko 1997; Ibn Tattou 2007; Benedi 2009; Sharifnia 2011).

The phylogenetic dendrograms constructed using distance method on molecular data and on morphological data, confirm the difference of our new species with *V. punalense* and *V. shahsavarensis* on both analyses (Fig. 3).

Acknowledgements

This research was supported by a grant from Campus France in the program Gundishapur (27907RL). The authors thank Christophe Andalo for his help with the R software, the herbarium keepers from Europe (MPU, K, BM), and Iran (TUH, TEH, HKS, IRAN, TARI) and Mr. Lhote for his support. They also thank the referees of this articles, Thierry Deroin and Farrokh Ghahremaninejad for their usefull comments on the manuscript. At last, SimpleMapp (Shorthouse, David P. 2010), has been used to produce the Figure 2A (<http://www.simplemapp.net>).

LINKS

Verbascum: <http://www.verbascum.org>
 International Plant Names Index (INPI): <http://www.ipni.org>
 The plant list: <http://www.theplantlist.org>
 Flora of Iran: <http://www.flora-iran.com>
 (last consultation 7th April, 2016).

REFERENCES

- BASELGA A. & ORME C. D. L. 2012. — Betapart: an R package for the study of beta diversity. *Methods in Ecology and Evolution* 3: 808-812.
- BENEDI C. 2009. — *Verbascum*. *Flora Iberica* 13: 49-71.
- DUNN G. & EVERITT B. S. 2004. — *An Introduction to Mathematical Taxonomy (Dover Books on Mathematics)*. Dover Publications, New York, 160 p.
- FEDTSCHENKO B. A. 1997. — *Verbascum* L., in SCHISCHKIN B. K. & BOBROW E. G. (eds), *Flora of USSR* 22: 132-197.
- FEINBRUN-DOTHAN N. 1978. — *Verbascum* L., in ZOHARY M. & FEINBRUN-DOTHAN N. (eds), *Flora Palaestina* 3: 282-302.
- FERGUSON I. K. 1972. — *Verbascum* L., in TUTIN T. G. et al. (eds), *Flora Europaea* 3: 205-216.
- GHAHREMANINEJAD F., RIAHI M., BABAEI M., ATTAR F., BEHÇET L. & SONBOLI A. 2015. — Monophyly of *Verbascum* (Scrophulariaceae): evidence from nuclear and plastid phylogenetic analyses. *Australian Journal of Botany* 62: 638-646. <http://dx.doi.org/10.1071/BT14159>
- HOLMGREN P. K., HOLMGREN N. H. & BARNETT L. C. 1990. — *Index Herbariorum. Part I: The Herbaria of the World. 8th Edition*. New York Botanical Garden, New York, 693 p.
- HUBER-MORATH A. 1978. — *Verbascum* L., in DAVIS P. H. (ed.), *Flora of Turkey and the East Aegean Islands* 6: 461-603.
- HUBER-MORATH A. 1981. — *Verbascum* L., in RECHINGER K. H. (ed.), *Flora Iranica* 147: 1-51.
- IBN TATTOU 2007. — *Scrophulariaceae*, in FENNANE M., TATTOU M. I., OUYAHYA A. & EL OUALIDI J. (eds), *Flore Pratique du Maroc*. Travaux de l'Institut scientifique, Série Botanique, Rabat, vol. 2: 507-514.
- KARAVELIOGULLARI F. A. & AYTAÇ Z. 2008. — Revision of the genus *Verbascum* L. (Group A) in Turkey. *Botanical Research* 1: 9-32.
- MURBECK S. 1933. — *Monographie Der Gattung Verbascum*. Lunds Universitets Arsskrift, Lund, 630 p.
- MURTAGH F. & LEGENDRE P. 2014. — Ward's hierarchical agglomerative clustering method: which algorithms implement Ward's criterion? *Journal of Classification* 31 (3): 274-295. <http://dx.doi.org/10.1007/s00357-014-9161-z>
- PARADIS E. 2011. — *Analysis of Phylogenetics and Evolution with R*. Springer Science & Business Media, 386 p.
- PARIKH A., MIRANDA E. R., KATOH-KURASAWA M., FULLER D., ROT G., ZAGAR L., CURK T., SUCGANG R., CHEN R., ZUPAN B., LOOMIS W. F., KUSPA A. & SHAULSKY G. 2010. — Conserved developmental transcriptomes in evolutionarily divergent species. *Genome Biol* 11: R35. <http://dx.doi.org/10.1186/gb-2010-11-3-r35>
- PARSA A. 1952. — *Verbascum* L. and *Celsia* L., in *Flore de l'Iran*. Publications de l'Université de Téhéran, Téhéran, vol. 4: 232-361.
- SWOFFORD D. L. 2002. — PAUP* Phylogenetic analysis using parsimony (*and other methods), v. 4.0 beta 10. Sunderland: Sinauer Associates.
- SHARIFNIA F. 2011. — *Scrophulariaceae*, in *Flora of Iran*. Edition of Organization of Forest and Pasture Research (in Persian), vol. 68: 7-74.
- SOTOODEH A. 2015. — *Histoire biogéographique et évolutive des genres Verbascum et Artemisia en Iran à l'aide de la phylogénie moléculaire*. Phd Thesis, Université Paul Sabatier de Toulouse, Toulouse, 193 p.
- SOTOODEH A., ATTAR F. & CIVEYREL L. 2015. — *Verbascum shahsavarensis* (Scrophulariaceae), a new species for Flora of Iran. *Phytotaxa* 203 (1): 76-80. <http://dx.doi.org/10.11646/phytotaxa.203.1.8>
- SOTOODEH A., CIVEYREL L., ATTAR F. & ZAMANI A. 2014. — *Verbascum oreophilum* var. *oreophilum* and *Verbascum cheiranthifolium* var. *asperulum* (Scrophulariaceae), two new records for the flora of Iran. *Phytotaxa* 178: 205-210. <http://dx.doi.org/10.11646/phytotaxa.178.3.6>

Submitted on 23 March 2015;
 accepted on 14 October 2015;
 published on 24 June 2016.

APPENDIX 1. — Matrix formed by morphological and molecular data.

	1 – 1, Anther decurrent; 0, reniform	2 – 1, Calyx shape ovate; 0, Calyx shape Triangular	3 – 1, Calyx end obtuse; 0, Calyx end acute	4 – Calyx'hairs glandular	4 – Calyx'hairs stelate	4 – Calyx'hairs glandular-stelate	5 – 1, Corolla 15 mm; 0, Corolla 25-40 mm	6 – Corolla hairs outside branched	6 – Corolla hairs outside stellate	6 – Corolla hairs outside stellate+glandular	7 – 1, Bas. leaves elliptic; 0, Bas. leaves oblong	8 – Bas. leaves margin irregularly double crenate	8 – Bas. leaves margin entire	8 – Bas. leaves margin indistinctly crenate	9 – Petiole bas. Leaves Absent	9 – Petiole bas. Leaves up to 3	9 – Petiole bas. Leaves up to 5
<i>V. parsana</i> , sp. nov. 34941	0	1	0	0	0	1	1	0	0	1	1	0	0	1	0	0	1
<i>V. punalense</i> 10663	1	1	1	1	0	0	0	1	0	0	0	1	0	0	1	0	0
<i>V. punalense</i> 43465	1	1	1	1	0	0	0	1	0	0	0	1	0	0	1	0	0
<i>V. punalense</i> 43467A	1	1	1	1	0	0	0	1	0	0	0	1	0	0	1	0	0
<i>V. shahsavarensis</i> AS29	1	0	0	0	1	0	0	0	1	0	0	0	1	0	0	1	0

	153	647	667	760	874	900	1009	1358	1370	1474	1609	1773	2166	2260	2318	2717	3144	3411	3514	3563	3734	3749	3848	3974	4082	4218	4219	4419	
<i>V. parsana</i> , sp. nov. 34941	C	C	T	G	C	A	T	T	C	G	T	C	T	C	A	A	T	T	A	A	T	G	C	C	A	C	T	C	G
<i>V. punalense</i> 10663	T	C	C	G	C	G	T	T	C	G	G	C	T	C	A	A	A	T	G	G	C	G	C	C	G	C	T	A	G
<i>V. punalense</i> 43465	T	C	C	G	C	G	T	T	C	G	G	T	T	C	A	A	A	T	A	A	C	G	C	C	G	C	T	A	G
<i>V. punalense</i> 43467A	T	C	C	G	C	G	T	T	T	G	G	C	T	C	A	A	A	T	A	A	C	G	C	C	G	C	T	A	G
<i>V. shahsavarensis</i> AS29	T	A	C	A	T	G	C	G	C	A	G	C	C	G	T	G	A	C	A	A	C	C	T	A	G	C	C	C	C