NOTE. An Italian language version of this article was previously published in: Revisione botanica di *Pinguicula ramosa* Miyoshi. AIPC Magazine 40 (4): 4-29, Dicembre 2015.

Pinguicula ramosa Miyoshi – a botanical review

Hiro Shimai • Tokyo Metropolitan Board of Education • Shinjuku • Tokyo • Japan • shimaihiro@gmail.com

Keywords: conservation, distribution, ecology, history, morphology, *Pinguicula ramosa*

Abstract: *Pinguicula ramosa* Miyoshi (Lentibulariaceae), endemic to Japan, is one of the most unique *Pinguicula* species often having a branched pedicel. Ecologically it occurs on cliffs formed by tuffs, porous volcanic rocks, which is also unusual as many *Pinguicula* species can be more commonly found on calcareous soils or serpentinite rocks. However, little attention has been previously given to this tiny endangered species with very few references in the literature outside of Japan, or even in there. To reveal the species, observations of the plants in situ, herbarium specimen examinations, and review of the literature have been performed. In addition, comparisons with morphologically and phylogenetically closely related species, *P. variegata* Turcz. and *P. villosa* L., will be summarized. This work is a botanical review overall *P. ramosa* discussing its morphology, distribution, taxonomy, ecology, history, conservation, and other scientific matters.

Introduction

The Genus *Pinguicula* L. (Lentibulariaceae) consists of approximately 90 species (Cieslak *et al*. 2005; Kondo & Shimai 2006), morphologically divided into three subgenera, *Isoloba*, *Pinguicula*, and *Temnoceras*, taking account of corolla shapes, sub-actinomorphic or zygomorphic, and flower colors (Casper 1966). A number of species occur in Mexico and Central America (ca. 46 species), and the rest of them are found in Eurasia, North America, South America, the Caribbean Islands, or the northern part of Africa. Many of them are restricted to a small geographical area and thus they show a higher proportion of endemism (Shimai & Kondo 2007).

At least two *Pinguicula* species are native to Japan. The first one is *P. macroceras* Link, widespread from Hokkaido to Tokushima Prefectures, but it is usually restricted to higher elevations. The scientific name is often treated as *P. vulgaris* L. var. *macroceras* (Link) Herder by Japanese botanists, but it should be segregated from *P. vulgaris* L. as Casper (1962a) discussed. *P. macroceras* is also found widely in far eastern Russia (Kamchatka, Sakhalin Island, and Kuril Islands) and the western part of Canada and the USA (Aleutians, Alaska to California). The second species is *P. ramosa*, endemic to a very small geographical area in Gunma and Tochigi Prefectures. *P. ramosa* can be easily distinguished by its smaller size and morphology from *P. macroceras*. There could be the third possible species, *P. villosa*, in Shikotan Island, one of the South Kuril Islands, lying northeast of the island of Hokkaido, but Japan has a territorial dispute on the South Kuril Islands with Russia. There have been no hitherto collection records of *P. villosa* from the main island of Hokkaido; therefore, two species, *P. macroceras* and *P. ramosa*, are generally recognized in Japan.

Literature regarding *P. ramosa* has been published in only small numbers so far particularly in English even though there are contributions to the species by some Japanese botanists. As a result, *P. ramosa* is still one of the little known species in the world. This work is, thus, attempting to review *P. ramosa*, and some different approaches have been used to achieve it. Firstly, the author

has visited localities of *P. ramosa* many times since the late 1980s to study morphology and ecology of the species in situ. For morphological studies, the plant of *P. ramosa* was photographed in its habitats and those are going to be shown in this work. Based on the material, illustrations were drawn and the author's observations are documented. Secondly, herbarium specimens deposited at A, BM, E, GMNHJ, JE, K, KYO, LE, MAK, PE, SAPS, TI, TNS, Nippon Dental University, and Tochigi Prefectural Museum have been studied and a geographical distribution map will be provided. Also, seeds from herbarium specimens were scanned by Hitachi High-Technologies Miniscope® TM3030Plus, an electron microscope. The diameter of pollen grains was measured by micrometers. Thirdly, literature published by various workers was reviewed to gather as much detail as possible about the botanical characteristics, molecular analyses, ecology, and history of *P. ramosa*.

Pinguicula ramosa Miyoshi, Bot. Mag. Tokyo 4. (1890) 315.
TYPE: JAPAN. Mount Koshin in the Province of Shimotsuke, 9 August 1890, Miyoshi s.n. (holotype?: TI!).
SYNONYM: *P. ramosa* Miyoshi f. *albiflora* Komiya, *P. villosa* L. var. *ramosa* (Miyoshi) Tamura.
DESCRIPTION: Lithophytic. Very small perennial herb (rosette diameter at anthesis 12-25 mm). Cotyledon 1. Root waxy-white, not many, filiform, ca. 15 mm long, very delicate texture. Stem compressed. Leaf dimorphic, summer leaf 3-6, radially lying on ground, yellowish-green, lower surface sometimes faintly suffused with maroon, ovate to elliptic, margin entire, often strongly involute, apex obtuse, base widely cuneate, subpetiolate, 6-15 mm long, 5-8 mm wide, densely covered by stalked and sessile glands only upper surface, winter leaf up to 15, tightly clustered to form a hibernaculum, ovate, apex rather acute, concave, up to 5 mm long. Hibernaculum subglobose, rootless. Pedicel 1, slightly S-shaped, often bifurcate or trifurcate at below middle or near base, 15-90 mm long, densely glandulous. Calyx bilabiate, upper lip 3-lobed, lobes oblong-ovate, ca. 1 mm long, middle lobe slightly longer than laterals, lower lip 2-lobed, ovate, up to 2 mm long, connate at base, glandulous. Flower faintly pale purple to pale purple, purple veins at base of lips to tube, 1-4 yellow spots at base of middle lobe. Corolla bilabiate, 8-10 mm across, zygomorphic, 7-11 mm long including spur, upper lip 2-lobed, lobes ovate-oblong, apex obtuse, 2-3 mm long, 2-3 mm wide, lower lip 3-lobed, lateral lobes ovate to ovate-oblong, apex obtuse, slightly larger than uppers, middle lobe ligulate, convex or with verrucose crests at base, slightly reflexed, apex truncate to emarginate, 4-7 mm long, 3-5 mm wide, numerous white trichomes at base of lower lip to throat, tube purple with darker veins, conical, dorsally compressed, spur yellow, cylindrical, straight or only slightly arcuate, abruptly narrowed at apex, 2-4 mm long. Ovary subglobose. Anther 2, filament ca. 1 mm long. Pollen grain creamy-white, 5- or 6-zonocolporate, 30-33 μm in equatorial diameter. Capsule subglobose, 2.0-2.5 mm, upper margin rather flat. Seed brown, glossy, fusiform-ellipsoid, 630-980 μm long, 195-250 μm wide, surface narrowly reticulate. 2n = 18. Figures 1, 2, and Front Cover.
PHENOLOGY: June to July
ETYMOLOGY: branched (referring to the pedicel)
LOCAL NAME: Koshin-so

1. Distribution

JAPAN. Gunma and Tochigi (1,460-2,300 m alt.).

Pinguicula ramosa is endemic to Gunma and Tochigi Prefectures. The habitat is highly restricted to several mountains and gorges, including Mt. Akanagi-san, Mt. Kesamaru-yama, Mt. Koshin-zan, Mt. Nokogiri-yama, Mt. Nyoho-san, Unryu-keikoku Gorge, and Misawa Rivulet, all of which are

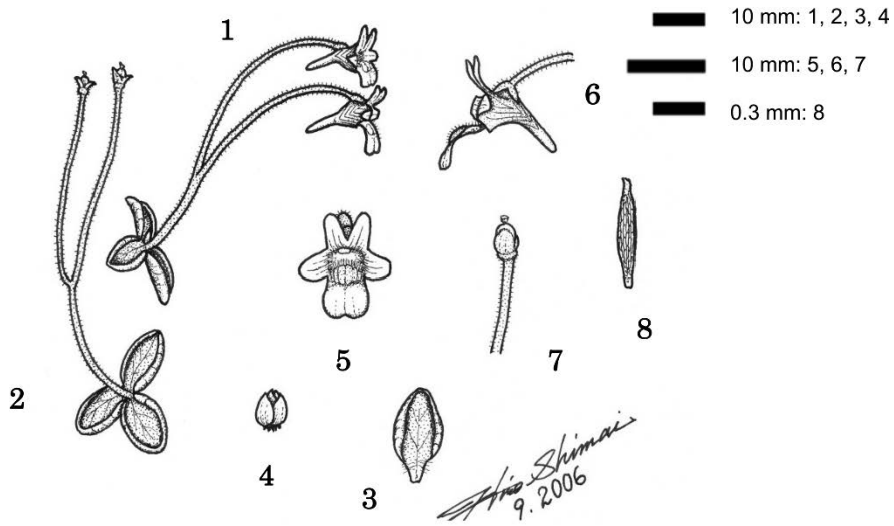


Figure 1: *Pinguicula ramosa* morphological line drawings.

located in Nikko National Park except Mt. Kesamaru-yama. Those habitats are within a boundary of Tochigi Prefecture with the exception of one locality in the Gunma side of Mt. Kesamaru-yama. The type locality mentioned in the original description as the Province of Shimotsuke (or Yashu in other old literature or specimen labels) is almost identical with Tochigi Prefecture today. The distribution pattern is shown in a map (Fig. 3) based on the author's herbarium specimen examinations. It is basically agreed with maps previously presented by Komiya and Shibata (1980, 1999), but additional localities have been indicated.

Uploading the coordinate datasets of habitats mentioned above into GeoCAT (Royal Botanic Gardens, Kew), it showed that Extent of Occurrence (EOO) was 63.098 km^2 and Area of Occupancy (AOO) was 40.000 km^2, implying a very narrow appearance.

Regarding the altitudinal distributions, the lowest height shown on specimen label is 1,250 m, but it is more reliably above 1,460 m. The highest habitat is ca. 2,300 m at Mt. Nantai-san.

2. Taxonomy

P. ramosa is uniform with no morphological variants. Although an unbranched pedicel is commonly produced (53%; investigated at Mt. Nantai-san), the species often has a bifurcate (46%) or trifurcate pedicel, which is absolutely uncommon for the other species in the genus. Branching very likely depends on availability of water and nutrition. Taxonomic studies were previously attempted by a few workers. Tamura (1953) treated as *P. villosa* L. var. *ramosa* (Miyoshi) Tamura without any clear statements. On the other hand, Ernst (1961) thought that *P. ramosa* was synonymous with *P. variegata*, but morphological and ecological differences between the two species were later well-summarized by Komiya and Shibata (1998). Casper (1962b, 1963, 1966) recognized *P. ramosa* as a distinct species. For an infraspecific treatment, Komiya (1972) reported *P. ramosa* Miyoshi f. *al-*

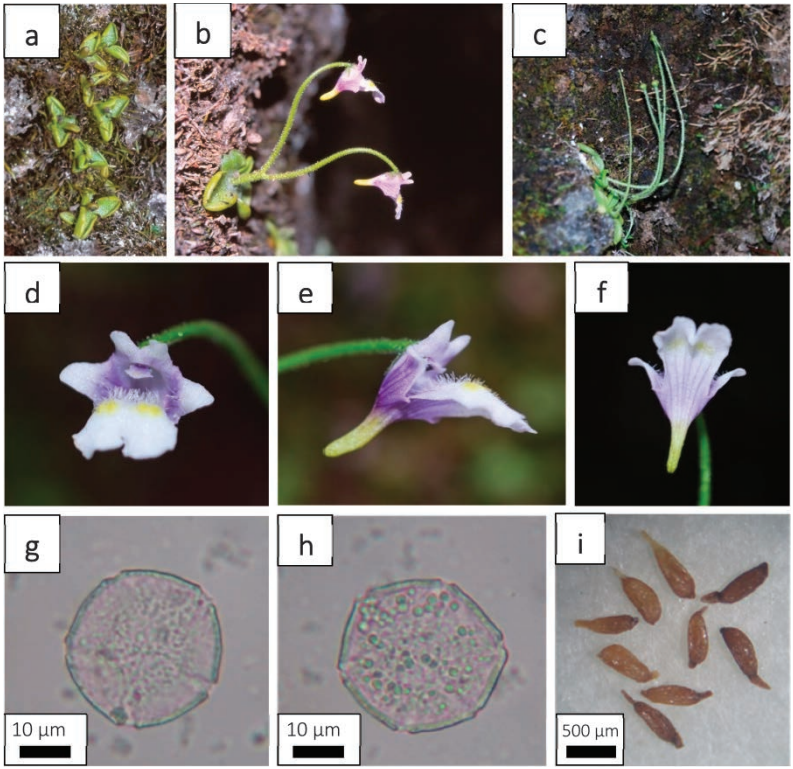


Figure 2: a) rosettes with flower bud at Mt. Nantai-san [9 June 2001], b) *P. ramosa* in flower at Mt. Koshin-zan [10 June 2002]: pedicel developing against rock wall, c) *P. ramosa* in fruit at Mt. Koshin-zan [18 July 2011]: pedicel standing upward parallel to rock wall, d) flower, front view, at Mt. Koshin-zan [17 June 2015], e) flower, lateral view, f) flower, bottom view, g) pollen grain with 5 pores, h) pollen grain with 6 pores, i) seeds.

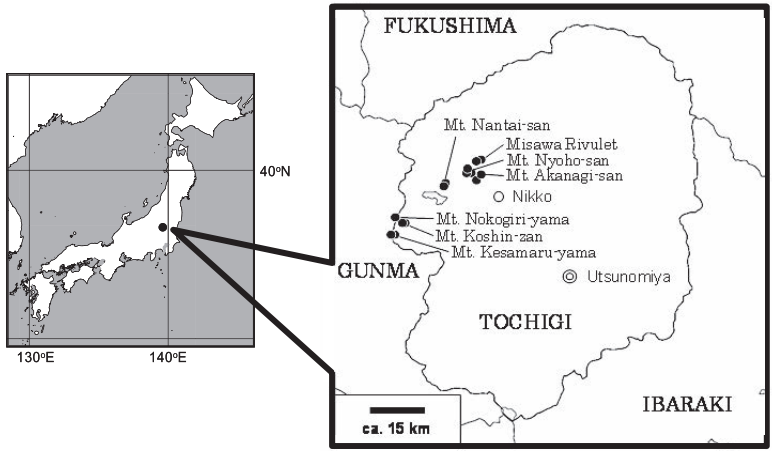


Figure 3: Geographical distribution of *Pinguicula ramosa*.

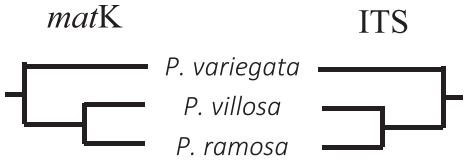


Figure 4: Phylogenetic relationships among *P. ramosa, P. variegata,* and *P. villosa.*

biflora Komiya, a white-flowered form. Casper (1966) mistakenly treated the author's name as *P. ramosa* "Miyoshi ex Yatabe" (citation from the Index Kewensis?) and it has been cited in some literature since then, but it is correctly *P. ramosa* "Miyoshi".

Infrageneric treatments are inconsistent among publications. Ernst (1961), who thought that *P. ramosa* was synonymous with *P. variegata*, did not recognize any subgeneric ranks and placed it into the Section Pionophyllum. Casper (1962b) placed *P. ramosa* into the Section Micranthus in the Subgenus *Micranthus* though Casper (1963, 1966) later moved the Section Micranthus into the Subgenus *Temnoceras*. Since a revision of the Genus *Pinguicula* by Casper (1966) no major taxonomic work over all of the genus with scientific basis has been made and consequently *P. ramosa* is still placed in the Section Micranthus, Subgenus *Temnoceras*.

According to molecular phylogenetic analyses, *P. ramosa* seems to be a close relative to *P. variegata*, distributed in Siberia to Kamchatka, and *P. villosa*, very widely distributed in the subarctic regions of Eurasia and North America, inferred from *mat*K in chloroplast DNA (Cieslak *et al*. 2005; Beck *et al*. 2008) and ITS in nuclear ribosomal DNA sequences (Kondo & Shimai 2006). Those are partially extracted and presented together in Figure 4.

3. Seed

Seed shapes may vary but mostly fusiform-ellipsoid (rarely subclavate) with narrowly reticulated surface (Fig. 5). The mean seed size measured was 753 µm in length and 219 µm in width which was much narrower than that of *P. variegata* 1,042 × 300 µm or *P. villosa* 1,079 × 656 µm (Shimai, unpublished). Those seed characteristics are summarized in Table 1 as well as other morphological features.

The mean number of seeds per capsule was 56.4 ± 20.3 which contained mature seeds 77.7%, immature seeds 3.8%, and sterile seeds 18.6% (Oba *et al*. 2013). This amount was more than that in *P. villosa* (Swedish materials), which was 29 ± 11 (Karlsson 1986).

More than 80% of seeds stored at either –20ºC or 20ºC both maintained in dried air humidity had germination ability after 10 months (Oba *et al*. 2013). It is questionable, but Oba *et al*. (2013) also reported that the seed totally lost its germination ability after one month stored under 20ºC without air humidity controls (i.e. higher air humidity).

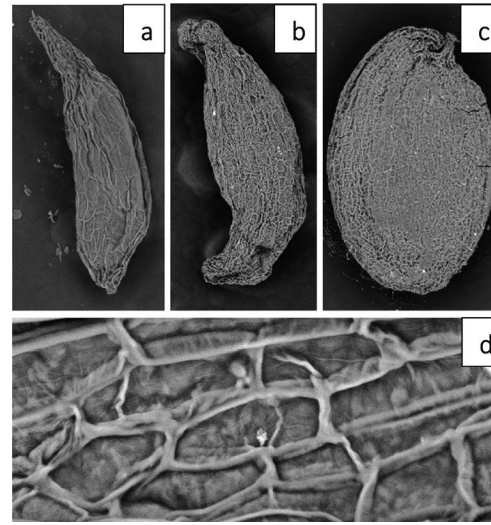


Figure 5: Seed morphology of three *Pinguicula* species. a) *P. ramosa* (ca. 750 µm long × 200 µm wide) from Mt. Koshin-zan, b) *P. variegata* (ca. 950 µm long × 330 µm wide) from Sakhalin Island, Russia, c) *P. villosa* (ca. 950 µm long × 580 µm wide) from Sweden, d) seed coat of *P. ramosa.*

Table 1. Comparisons among *Pinguicula ramosa*, *P. variegata*, and *P. villosa*.				
		P. ramosa	*P. variegata*	*P. villosa*
Subgenus		*Temnoceras*	*Temnoceras*	*Pinguicula*
Section		Micranthus	Micranthus	Nana
Stem L		very short	very short	up to 4 cm
Summer leaf	number	3-6	3-7	1-5
	lamina shape	ovate to elliptic	ovate to orbicular	elliptic
	size L × W (mm)	6-15 × 5-8	6-20 × 5-7	7-13 × 3-7
	color	yellowish-green	yellowish-green or maroon	yellowish-green
	base shape	widely cuneate	widely cuneate	narrowly cuneate
Flower	pedicel L (mm)	15-90	30-250	15-95
	pedicel shape	branched	unbranched	unbranched
	color	pale purple	pale purple	reddish-purple
	corolla L with spur (mm)	7-11	4-17	5-11
Seed	capsule shape	subglobose	obovoid	subglobose to obovoid
	shape	fusiform-ellipsoid	fusiform-ellipsoid	ellipsoid
	size L × W (μm)	630-980 × 195-250	970-1,180 × 230-330	990-1,140 × 610-720
Chromosome number		2n = 18	2n = 64	2n = 16
Distribution area		Japan	Eastern Russia	Eurasia & North America
Altitude (m)		1,460-2,300	0-1,800	0-1,400
Climate zone		Temperate	subarctic	subarctic
Environment		forest	bog, streamside	bog, streamside
Soil types		tuff	gravelly soil or sphagnum mat	sphagnum mat

4. Cytology

The chromosome number for *P. ramosa* has been counted as 2n = 18 by Yoshimura (1973), using materials from Mt. Nyoho-san (Unryu-keikoku Gorge?). Casper and Stimper (2009) also counted 2n = 18 using materials from Mt. Nyoho-san. However, it will need a critical review since the basic chromosome number of x = 9 is unusual in the genus (mostly x = 8 or 11). Yoshimura (1973) suggested that 2n = 18 might have originated from 2n = 22 as a result of fusion, but no species with 2n = 22 have been known so far from Eurasia. Chromosome number reported in *P. variegata* is 2n = 64 (Zhukova & Tikhonova 1971) and that in *P. villosa* is 2n = 16 (Löve & Löve 1982).

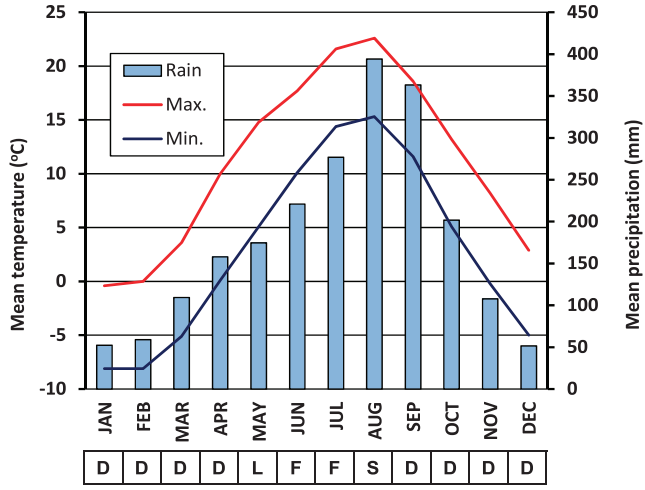


Figure 6: Monthly mean precipitation and maximum and minimum temperatures at Oku-Nikko. D: dormancy, F: flowering, L: leaf development, S: seed dispersal.

5. Natural hybrid

No natural hybrid of *P. ramosa* has been recorded though *P. ramosa* occurs sympatrically with *P. macroceras* at Mt. Nantai-san. The best explanation for this could be that they are genetically not close to each other to produce a hybrid.

6. Climates

The distribution is highly restricted to subalpine zones of several volcanic mountains and their environments lying within ca. 30 km, including Mt. Nantai-san, Mt. Nyoho-san, Mt. Kesamaru-yama, and a few other mountains as well as the type locality of Mt. Koshin-zan, around the City of Nikko (ca. 120 km north of Tokyo). Like the majority of Japan, the Nikko region has four very clear seasons. The Nikko region is much cooler in summer and colder in winter than coastal regions because of higher elevation. Monthly mean precipitation, maximum and minimum temperatures, recorded at Oku-Nikko Meteorological Station (36°44.3'N 139°30.0'E, 1,292 m alt.), on the eastern shore of Lake Chuzenji-ko, are shown in Figure 6. Annual mean temperature between 1981 and 2010 recorded at the station was 6.9°C and annual mean precipitation was 2,176.3 mm in the same period. More rainfall is generally apparent in summer than other seasons and there is some snowfall in winter. It should be noted that all localities of *P. ramosa* are 150-1,000 m higher than the meteorological station, so that the temperature at the habitat is lower than the data presented in the chart. In winter, the minimum temperature sometimes drops below –20°C and the rock walls are not covered by snow.

7. Ecology

Pinguicula ramosa is basically found on weathered tuffs, such as welded tuffs, formed by volcanic activities, often vertical or overhanging cliffs with or without mosses on the surface. Soil pH measured at Tsuru-iwa rock, Mt. Koshin-zan, was 6.0 and that at Mt. Nantai-san was 6.5. The soil is not too wet or rather dry, but frequent fogs and higher air humidity provide moisture to the plant

and the rock surface. The irradiance level at the habitats is relatively low since those are mostly located below forest lines. It is commonly seen together with *Saxifraga forunei* var. *alpina*, *Primula farinosa* subsp. *modesta*, *Parnassia palustris*, or *Schizocodon soldanelloides* at microhabitats. The soil composed by tuff contains poor organic substances restricting to grow only a few plant species including *P. ramosa* and those mentioned above. Usually *P. ramosa* is not exposed to direct sunlight, hence it is tolerant to low light intensity and higher air humidity. In contrast, *P. variegata* and *P. villosa* are often found on sphagnum mats in bogs or along streams in open areas.

8. Pollination

The flower of *P. ramosa* is very likely entomophily but what kind of insects are actually pollinating has remained unknown (flies are seen at the habitat). Despite that, most of flowers are successfully pollinated. Yellow spots at the base of middle lobe of lower lip and nectar held in a spur may have important roles to attract pollinators. Ultraviolet reflection patterns on flowers might also have a role to attract pollinators as suggested by Gloßner (1992). In any case, it is probable that there is almost no chance of gene flow among isolated habitats.

9. Growth cycles

In general, a mature plant of *P. ramosa* breaks its dormancy in May and starts to expand summer leaves. A flower bud usually becomes visible when the third summer leaf is expanding. A pedicel elongates gradually against the rock wall where the plant grows, and it reaches anthesis between the beginning of June and the beginning of July. Two flowers open almost at the same time on the bifurcate pedicel. Peak flowering season varies among localities, but it normally ends within 10 days at each microhabitat. When seed setting is successfully completed, the base of pedicel curves upward and the pedicel stands nearly parallel to the rock wall. Some pedicel elongation occurs simultaneously as seeds are developing. The seed is mature approximately 1.5-2 months after anthesis and the seed capsule is naturally open vertically when it is dried. Seed dispersal is gravitational whereas only a small number of seeds are successfully deposited on the rock wall. Those stages may slightly vary year to year due to weather conditions (see also Fig. 6).

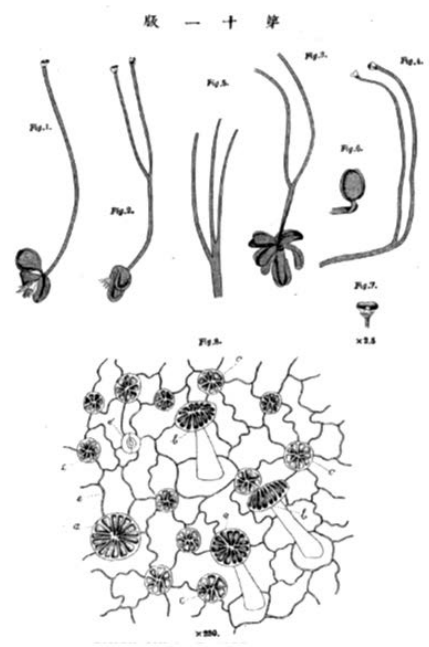


Figure 7: Drawings of *P. ramosa* in the original description by Miyoshi (1890).

After seed germination, it normally takes three years to reach a reproductive phase in situ. Seed germination takes place in the first year. This may occur immediately after seed dispersal, or the next growth season in the following year. Seedlings become larger but remain vegetative in the second year. They are tolerant enough to low temperature at this stage when they have formed a hibernaculum since the rock walls are often not

covered by snow and are exposed to harsh weather conditions during winter. Flower primordia may occur when a hibernaculum is formed but further flower bud development may not take place unless it experiences low temperature in a certain period. Finally the plant may be able to flower in the third year. The mean rosette diameter at anthesis was 17.7 mm at Tsuru-iwa rock, Mt. Koshin-zan, and 18.5 mm at Mt. Nantai-san. The seed does not require low temperature (ca. 0ºC) before germination, but the seedling germinated immediately after seed dispersal tends to have a lower survival rate for the next growth season. Although *P. ramosa* is perennial, how many years mature plants normally can live remain uncertain, but it seems that they could survive three years or so.

10. Hibernaculum induction

According to Oba *et al*. (2013), *P. ramosa* formed a hibernaculum under either short-day length (SD; 8 hours) or low temperature (LT; 5ºC). The hibernaculum is usually formed by the end of August in situ under long-day length (LD) and high temperature (HT). It is unclear whether SD or LT is one of the factors affecting hibernaculum formation, but it is probable that after developing a certain number of summer leaves (e.g. 5-6), the plant would naturally form a hibernaculum regardless of day length or temperature that has been confirmed *in vitro* conditions under LD (18 h) and HT at 20ºC throughout. Once the hibernaculum has been formed, it will stay in dormancy tolerant to LT (around 0ºC or lower) and/or dryer soil moisture. It will, however, break dormancy within 40-60 days if those conditions are not fulfilled.

Asexual reproduction by gemmae or daughter hibernacula formed at the base of the main hibernaculum as seen in some other *Pinguicula* species is not active.

11. Carnivory

Like those of other *Pinguicula* species, the upper surface of leaves and a pedicel are densely covered by two types of tiny glands, i.e. stalked and sessile glands. Stalked glands secrete mucilage to capture prey, and sessile glands secrete digestive enzyme and absorb digested substance (Heslop-Harrison 1970). During the flowering season, the margins of leaves are often strongly incurved and only a small number of prey can be captured by the leaf itself. However, the pedicel may have a more important role to capture prey. After ending the flowering season, the leaf tends to be open and it captures more prey than during the flowering season. The size of prey captured is usually very minute, such as mites or springtails. No mycorrhizal organisms associated with the root have been detected so far for *Pinguicula* (Juniper *et al*. 1989; Heslop-Harrison 2004), implying that carnivory may play an important role for nutrient uptake though its efficiency is unknown.

12. History

It is believed that the first discovery of *P. ramosa* was made by Manabu Miyoshi (1862-1939), at Tokyo Imperial University (The University of Tokyo), during his botanical expedition in 1890. While on his one-month-long expedition, Miyoshi saw *P. macroceras* in Nagano. At Mt. Koshin-zan, his final destination, Miyoshi found a number of tiny *Pinguicula* growing on a rock cliff named Fuji-sengen on the mountain trail between Koshin-sanso Hut and the summit. He was not sure if it was certainly a new species since the flowering season had been over at the time of discovery in August. However, he was reasonably confident that it was likely a new species having a branched pedicel. As soon as returning to the university, Miyoshi (1890) described this taxon as *Pinguicula*

ramosa sp. nov. in Botanical Magazine Tokyo, Vol. 4, No. 43, pp. 314-319 with drawings (Fig. 7), issued in September 1890, and at the same time gave a Japanese name, koshin-so (literally "koshin plant") derived from the name of mountain. In the article, Miyoshi (1890) stated the discovery of *P. ramosa* at Mt. Koshin-zan in details both in English and Japanese. The type specimens (Fig. 8) have been preserved at Botanical Gardens, Koishikawa, the University of Tokyo (TI). Miyoshi (1890) expected to observe the flower in the next flowering season and to compare anatomically with the flower of *Pinguicula macroceras* (as *P. vulgaris* in his paper), more widely distributed in Japan, but it does not seem such a study has been published. An additional sampling was made by Miyoshi in July 1891 at Mt. Koshin-zan (the specimen without flowers has been deposited at TNS), but it is probable that he did not have a chance to observe the flower at the habitat.

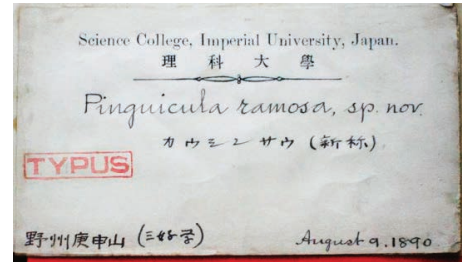
Prior to Miyoshi's type specimen (9 August 1890), Kinashi collected *P. ramosa* in July 1890, which was initially identified as *P. vulgaris* and the specimen has been deposited at KYO. If the date was correct, Kinashi's material had been collected a few weeks earlier than Miyoshi's type specimen. Nothing was, however, documented in the literature regarding Kinashi's specimen and it has remained unclear. According to Komiya (pers. com.), it was very likely that Kinashi misdated the label. Therefore, it is normally considered that Miyoshi was the first botanist, who collected the species. Shortly after the discovery of *P. ramosa* by Miyoshi (1890), a number of botanists visited the habitat at Mt. Koshin-zan to collect the plant. By early in the 20th century, a few other habitats, such as Mt. Nyoho-san and Mt. Nantai-san, were discovered by other botanists. More recently, new localities were found along the Misawa Rivulet, on the northern slope of Mt. Akanagi-san.

13. Ethnobotany and iconic uses

No ethnobotanical use of *P. ramosa* has been recorded since the species occurs in mountains isolated from areas of human activity. In the recent years, apart from using the plant itself, illustrations of *P. ramosa* are sometimes used for symbolic icons of the Ashio region. For example, *P. ramosa* is painted on local pottery called Ashio-yaki, or local shops sell posters of *P. ramosa* photographed at Mt. Koshin-zan for visitors. In 1978, a commemorative postage stamp with illustrations of *P. ramosa* (Fig. 9), one of the Nature Conservation Series, was issued by the Ministry of Post and Telecommunications. In 1982, Ashio Town designated that *P. ramosa* was a symbolic flower for the town and it continued until the town was merged into Nikko City in 2006.

14. Conservation

No *Pinguicula* species are listed in CITES Appendixes for international commercial trade (von Arx *et al*. 2001, and also see CITES website for an updated checklist). *P. ramosa* is, however, one of the threatened species. GeoCAT indicated that Critical (CR) for EOO and Endangered (EN) for AOO while Red List assessment by GeoCAT does not consider the number of individuals at each habitat. *P. ramosa* is actually listed in the Red List as Vulnerable (VU) status by the Ministry of the Environment. Similarly it is also listed as the same status in local versions of Red Lists issued by Gunma and Tochigi Prefectures, to protect the species under bylaws. Most of *P. ramosa* habitats are located in "special areas" of Nikko National Park (114,753 ha), environmentally protected by the Natural Park Law. Furthermore, each forest area is under control by either Forestry Agency or local administrations depending on the area. The habitat of *P. ramosa* on Mt. Koshin-zan was designated a Natural Monument in 1921 to protect not only the species itself but also the environment where the species occurs, and it has been elevated to a "special" rank since 1952. Therefore, it is heavily



Science College, Imperial University, Japan

Pinguicula ramosa, sp. nov.
Koshin-so (new name)
[TYPUS]
Yashu, Koshin-zan (Miyoshi Manabu)
August 9, 1890

Figure 8: *Pinguicula ramosa* type specimen, collected by Miyoshi on 9 August 1890 at Mt. Koshin-zan, deposited at Botanical Gardens, Koishikawa, University of Tokyo (TI), which provided this image. Although the status of type is not specified, it is normally recognized as the holotype.

Figure 9: *Pinguicula ramosa* postage stamp and its First Day Cover.

protected by different regulations and laws directly or indirectly. Apart from legal protections, little has been attempted with effective and practical approaches for conservations of the species.

The number of *P. ramosa* individuals may occasionally decrease suddenly at the microhabitats. One possible reason could be that it is stolen by plant vendors or amateur plant collectors. Wild animals, e.g. deer or serows, may devour it together with other plant species when their food is scarce. However, most plants grow on unreachable overhanging rock cliffs and therefore such activities may not necessary be direct causes. It is likely that environmental changes at microhabitats such as dryness or corruption of rocks may be possible direct causes. Global warming and acid rain could also be factors that cannot be ignored. Recently, the population at the Gunma side of Mt. Kesamaru-yama is nearly critical (Ohmori, pers. com.).

15. Cultivation

P. ramosa forms relatively large colonies (up to a thousand individuals on a rock) on slightly wet weathered tuff cliffs, but has a higher risk of rapid decline of the population number at the habi-

Table 2. Composition of medium for *Pinguicula* culture.	
Compound	g/l
HYPONeX® 6.5-6-19	3.0
sucrose	32.0
agar	8.4
pH 5.8	

tat due to environmental stresses and/or climate changes as already stated. Long-term cultivation of *P. ramosa* ex situ is difficult since it is not possible to provide an appropriate environment artificially. Moreover, it is strongly prohibited to collect the plants, seeds, or any segments of the plant at the habitats, and it is basically very difficult to obtain permissions. However, European vendors occasionally sell *P. ramosa* plants or seeds for a commercial purpose. It would be worth establishing cultivation methods for ex situ conservation of the species.

Concerning a type of compost, using soils from the habitat may be an option, but it seems, for some reason, that *Pinguicula* does not always grow well for long periods on the soil under cultivation. In Japan, sphagnum moss is very widely used for cultivation of *Pinguicula*. Also some other types of compost are commercially available, but it is not easy to say which one is the most suitable. It is not simply a question of soil types, but a whole set of environments including optimum temperatures, moistures, irradiance levels, ventilations, and pest and disease controls. Under cultivation, grey mold is the most common disease and the slug is the most problematic pest.

Another possible and more successful approach is *in vitro* culture. Many *Pinguicula* species grow successfully and multiply profusely *in vitro* condition. It is much easier to obtain plants from seeds than buds. The seeds in a tube can be disinfected by shaking for a few minutes in 10% kitchen bleach solution, and should be carefully rinsed with sterile distilled water. Kano's medium (Kano 1965), or more often called Hyponex medium, was modified for *Pinguicula* culture (Shimai, unpublished). The modified Kano's medium component is very simple, which includes granule fertilizer HYPONeX® N-P-K = 6.5-6-19 (HYPONeX Japan Corp. Ltd., Osaka, Japan) 3.0 g, sucrose 32.0 g, and agar 8.4 g per liter (Table 2). No hormones are basically required. It appears that pH level is not an absolute factor, but it should be adjusted to pH 5.8. It produces clones and multiplies very quickly on this medium, so using wider jars is much better than test tubes. Those should be maintained under 18 hours of fluorescent light. Illuminating white LEDs may perform similarly. Optimum temperature appears to be 18-20°C, but no problems have been observed under 25°C. Acclimatization is rather difficult, but to take out the *in vitro* cultured plant that has formed a hibernaculum in late autumn and to overwinter as a dormant condition in a pot may result in higher survival rates than in other seasons.

16. Representative localities

As shown in the distribution map (Fig. 3), *P. ramosa* occurs in several mountains and those environments which are mostly in Nikko National Park. Two representative localities, Mt. Koshin-zan and Mt. Nantai-san, will be described.

1) Mt. Koshin-zan (1,892 m) is located near Ashio, an old copper mining town, almost at the western edge of Tochigi Prefecture. Mt. Koshin-zan forms the Ashio Mountain Range together with Mt. Kesamaru-yama (1,961 m), Mt. Sukai-san (2,144 m), and some other mountains. Mt. Koshin-zan itself has been worshipped as a deity for more than 1,200 years. It was also known as a place where shugendo followers practiced mental and physical training because of a rocky mountain. As mentioned earlier, the first discovery of *P. ramosa* was made at Fuji-sengen rock along the mountain trail between Koshin-sanso Hut and the summit. It

is, however, not very common to see *P. ramosa* along the trail anymore. Today a number of populations are more frequently observed along Oyama-meguri trail (Miyoshi also visited a part of this trail in 1890). This loop trail goes up from Old Sarutahiko-jinja Shrine site, ca. 200 m below Koshin-sanso Hut and meets the trail to the summit above the hut through very rocky slopes and cliffs. There are many habitats along Oyama-meguri trail, but one of the best places to observe a number of *P. ramosa* plants could be on vertical or overhanging rock walls of Kame-iwa (Turtle rock) and Tsuru-iwa (Crane rock), which are almost next to each other, at an altitude of ca. 1,750 m. Those habitats are more or less facing south. Peak flowering season at the habitats is usually in the first half of June (It used to be around the 20th of June in the 1980s, but it is around the 10th of June in recent years though it may vary year to year).

2) Mt. Nantai-san (2,484 m), formerly called Mt. Futara-san, is one of the mountains belonging to the Nikko Volcanic Mountain Range, together with Mt. Akanagi-san (2,010 m), Mt. Nikko-Shirane-san (2,578 m), Mt. Nyoho-san (2,483 m), and some other mountains. Mt. Nantai-san itself is considered as a deity of Futarasan-jinja Shrines, situated at Chugushi at the base of the mountain and in Nikko city center. Mt. Nantai-san is located at approximately 12 km west-northwest of Nikko city center and just north of Lake Chuzenji-ko, which is a dammed lake formed by volcanic activities of the mountain ca. 20,000 years ago. Mt. Nantai-san, a stratovolcano, is conical shaped with a broken crater at the top and many radial hollows between ridges. The main habitat of *P. ramosa* is found in Hanage-no-nagi hollow, just east side of a mountain trail at the northern slope of the mountain, below the summit around 2,200-2,300 m alt. The habitat is not visible from the mountain trail because it is hidden by birch trees (*Betula ermanii*). Peak flowering season at this habitat is the beginning of July. Plant size of the population at Mt. Nantai-san is slightly larger and the pedicel is more frequently branched compared with those at Mt. Koshin-zan. This is probably due to environmental factors such as availability of water and/or soil nutrition. Apart from altitude, the environment is very similar to that of Mt. Koshin-zan (i.e. soil type, light intensity, etc.). It is the only known microhabitat where *P. ramosa* and *P. macroceras* occur sympatrically.

Key to species

1. Flower color reddish-purple, tube narrowly cylindrical, base of leaf narrowly cuneate (Eurasia & N. America) ---------- *P. villosa*
1. Flower color faintly pale purple to pale purple, tube conical, dorsally compressed, base of leaf broadly cuneate ---------- 2
   2. Middle lobe of lower lip broadly rotundate, pedicel not branched, often suffused with maroon, up to 25 cm long, leaf yellowish-green or maroon entirely, ovate to orbicular (E. Russia) ---------- *P. variegata*
   2. Middle lobe of lower lip ligulate, tip truncate to emarginate, pedicel often bifurcate or trifurcate, yellowish-green, up to 9 cm long, leaf yellowish green, lower surface sometimes faintly suffused with maroon, ovate to elliptic (Japan) -- *P. ramosa*

Specimens examined

Pinguicula ramosa Miyoshi

JAPAN. Gunma: Midori, Mt. Kesamaru-yama, 14 June 1974, Sudo s.n. (GMNHJ); 29 June 2003, Shibata s.n. (Herb. Dept. Biol., Nippon Dental Univ.). Midori, Mt. Kesamaru-yama, Ato-Kesamaru-yama, source of Konaka River, 1,780 m, 28 June 2009, Yoshii *et al*. s.n. (GMNHJ). Tochigi: Ashio (Nikko), Mt. Kesamaru-yama, 1,830 m, 24 July 1985, Ogura *et al*. s.n. (Tochigi Pref. Museum).

Ashio (Nikko), Mt. Koshin-zan, July 1890, Kinashi s.n. (KYO; as *P. vulgaris*); 30 July 1891, Hori s.n. (MAK); July 1891, Miyoshi s.n. (TNS); July 1891, Sakurai s.n. (TNS); 20 July 1909, Sakurai s.n. (LE); 18 June 1910, Kurihara s.n. (GMNHJ); 5 July 1912, Shimazu s.n. (TI); 2 August 1912, Koidzumi 69204 (TNS); 15 August 1913, Imai s.n. (TNS); July 1914, Takamatsu s.n. (SAPS); 20 July 1918, Shimazu s.n. (TI); 31 July 1921, Ogura s.n. (TI); 2 August 1921, Ogura s.n. (TI); 15 July 1952, Haginiwa s.n. (TNS); 22 July 1952, leg. ign. s.n. (K); 22 June 1954, Higuchi s.n. (GMNHJ); 6 August 1954, Moriya s.n. (Tochigi Pref. Museum); 31 June 1957, Komiya s.n. (TNS); 1,750 m, 26 June 1966, Michikawa 8154-8155 (TNS); 1,400-1,800 m, 4 July 1970, Furuse 49227 (PE); 2 June 1982, Nemoto s.n. (Tochigi Pref. Museum); 1,700 m, 3 June 1984, Ogura s.n. (Tochigi Pref. Museum); 1,720 m, 11 July 1984, Ogura & Noguchi s.n. (Tochigi Pref. Museum); 15 June 1997, Shibata s.n. (Herb. Dept. Biol., Nippon Dental Univ.); Ashio (Nikko), Mt. Koshin-zan, SE slope, 1,700 m, 22 June 1986, Ogura s.n. (Tochigi Pref. Museum). Ashio (Nikko), Mt. Koshin-zan, near summit, 1,850 m, 1 September 1989, Ogura s.n. (Tochigi Pref. Museum). Ashio (Nikko), Mt. Nokogiri-yama, 14 August 1984, Hasegawa s.n. (Tochigi Pref. Museum); 1,950 m, 7 August 1986, Noguchi *et al*. s.n. (Tochigi Pref. Museum). Nikko, Mt. Akanagi-san, near Akanagi-okusha-ato, 2,100 m, 30 June 2001, Nazuka s.n. (Tochigi Pref. Museum). Nikko, Mt. Akanagi-san, Hinata, 7 September 1904, Takeda s.n. (TNS). Nikko, Mt. Akanagi-san, Oku-Akanagi-san, 2,000 m, 11 August 1984, Ogura s.n. (Tochigi Pref. Museum). Nikko, Kami-Kuriyama, upper stream of Misawa Rivulet, 1,460 m, 23 August 1989, Noguchi *et al*. s.n. (Tochigi Pref. Museum); 1,460 m, 23 August 1989, Ogura *et al*. s.n. (Tochigi Pref. Museum); 1,950 m, 20 July 1992, Yamashita s.n. (Tochigi Pref. Museum); 1,950 m, 29 July 1992, Yamashita s.n. (Tochigi Pref. Museum). Nikko, Mt. Nantai-san, 2 August 1921, Ogura s.n. (TI); 7 July 1924, leg. ign. s.n. (TNS); July 1929 & 3 July 1931, leg. ign. s.n. (TNS); 3 July 1931, Hara s.n. (TI); 4 August 1931, Shimada s.n. (TNS); 2 July 1934, Ito s.n. (TI); 10 September 1934, Koidzumi 94467 (TNS); 5 July 1964, Shibusa s.n. (Tochigi Pref. Museum); 2,300 m, 15 June 1982, Ogura s.n. (Tochigi Pref. Museum); 2,200 m, 22 June 1982, Ogura s.n. (Tochigi Pref. Museum); 2,240 m, 24 July 1983, Ogura & Noguchi s.n. (Tochigi Pref. Museum); 2,300 m, 9 August 1997, Hasegawa s.n. (Tochigi Pref. Museum); 2,240 m, 15 October 2002, Suzuki s.n. (Tochigi Pref. Museum). Nikko, Mt. Nantai-san, Hanatate-sawa, 27 June 1997, Shibata s.n. (Herb. Dept. Biol., Nippon Dental Univ.). Nikko, Mt. Nantai-san, above Shizu-goya Shelter, 4 June 2001, Shibata s.n. (Herb. Dept. Biol., Nippon Dental Univ.). Nikko, Mt. Nyoho-san, June 1901, Matsumura s.n. (TNS); 4 July 1903, Takeda s.n. (MAK, TI); July 1905, Sakurai s.n. (TNS); 2 September 1905, Hayakawa s.n. (TI); 10 August 1908, Sakurai s.n. (E); July 1913, Koidzumi s.n. (TI); 6 August 1914, leg. ign. 140 (KYO); 20 June 1916, Kurihara s.n. (GMNHJ); 28 May 1924, Kishida s.n. (TNS); 26 July 1924, Narita 1376 (A, BM, PE, TI); July 1924, Hayakawa 8 310 (TI); July 1924, Hayakawa T609 (TI); 2 July 1933, Ito s.n. (TI); 2 July 1933, Tsuyama s.n. (TI); 1,900 m, 24 July 1951, Furuse 23871 (PE); 6 July 1952, Kubota s.n. (Tochigi Pref. Museum); 2,000 m, 6 July 1952, Okuyama 16772 (TNS); 1,800 m, 7 July 1952, Yamazaki 3122 (TI); 2 June 1961, Shibusa s.n. (Tochigi Pref. Museum); 5 July 1965, Okuyama & Okuyama 24712 (TNS); 2,090 m, 26 September 1984, Ogura s.n. (Tochigi Pref. Museum). Nikko, S of Mt. Nyoho-san, 1,850 m, cultivated material vouchered on 15 July 2001 (originally collected at the habitat on 12 October 1999 by Steiger), Steiger s.n. (JE). Nikko, Mt. Nyoho-san, Karasawa, s.d., Zyoo s.n. (TI). Nikko, Mt. Nyoho-san, above Nanataki, 20 June 1891, Zyoo s.n. (TI). Nikko, Unryu-keikoku Gorge, 1,500 m, 30 July 1972, Nakayama s.n. (Tochigi Pref. Museum); 23 June 1981, Sato s.n. (Tochigi Pref. Museum); 1,450 m, 3 June 1983, Ogura & Noguchi s.n. (Tochigi Pref. Museum); 1,500 m, 14 August 1996, Hasegawa s.n. (Tochigi Pref. Museum); 29 June 1999, Hasegawa s.n. (Tochigi Pref. Museum); 1,250 m, 13 July 2005, Ogura s.n. (Tochigi Pref. Museum). Nikko, locality not specified, June 1901, leg. ign. s.n. (MAK); August 1908, Yokohama

Nursery Co., LTD. s.n. (E). Misc. (Prefecture not specified): Mt. Kesamaru-yama, 2 July 1978, Sudo s.n. (GMNHJ). (Locality not specified): July 1910, Mochizuki s.n. (E).

[Note] Old local names have been revised. All specimens at Department of Biology, Nippon Dental University were transferred to TNS in 2013.

Acknowledgements: The author is indebted to Drs. Anthony Brach (A; Harvard University), Jonathan Gregson (BM; Natural History Museum), Lesley Scott (E; Royal Botanic Garden Edinburgh), Takehiro Ohmori (GMNHJ; Gunma Museum of Natural History), Joern Hentschel (JE; Friedrich Schiller University Jena), David J. Goyder (K; Royal Botanic Gardens, Kew), Hiroaki Setoguchi (KYO; Kyoto University), Roman Ufimov (LE; Russian Academy of Sciences), Noriaki Murakami (MAK; Tokyo Metropolitan University), Zhi-Rong Yang (PE; Chinese Academy of Sciences), Hideki Takahashi (SAPS; Hokkaido University Museum), Tetsuo Toma (TI; University of Tokyo), Atsushi Ebihara (TNS; National Museum of Nature and Science), Sadashi Komiya and Chiaki Shibata (Nippon Dental University), and Naoto Hoshi (Tochigi Prefectural Museum) for their curatorial assistances, Eri Tobe (Waseda Junior & Senior High School) for mineral identifications, and Asako Tsuda (Ministry of the Environment) and Satoru Honma (Agency for Cultural Affairs) for informative comments concerning the current legal statuses. Similarly, the author would like to express his sincere thanks to Mr. Takayuki Uzawa and Ms. Kaori Ichikawa (Hitachi High-Technologies Corporation) for their technical support using Miniscope® TM3030Plus, and to Dr. Amy Hinsley (University of Kent) for linguistic corrections and critical reading of the manuscript.

References

Beck, S., Fleischmann, A., Huaylla, H., Müller, K.F., and Borsch, T. 2008. *Pinguicula chuquisacensis* (Lentibulariaceae), a new species from the Bolivian Andes, and first insights on phylogenetic relationships among South American *Pinguicula*. Willdenowia 38: 201-212.

Casper, S.J. 1962a. On *Pinguicula macroceras* Link in North America. Rhodora 64: 212-221.

Casper, S.J. 1962b. Revision der Gattung *Pinguicula* in Eurasien. Feddes Repertorium 66:1-148.

Casper, S.J. 1963. Gedanken zur Gliederung der Gattung *Pinguicula* L. Botanisch Jahrbücher 82(3): 321-335.

Casper, S.J. 1966. Monographie der Gattung *Pinguicula* L. Bibliotheca Botanica 127/128.

Casper, S.J., and Stimper, R. 2009. Chromosome numbers in *Pinguicula* (Lentibulariaceae): survey, atlas, and taxonomic conclusions. Plant Systematics and Evolution 277: 21-60.

Cieslak, T., Polepalli, J.S., White, A., Müller, K., Borsch, T., Barthlott, W., Steiger, J., Marchant, A., and Legendre, L. 2005. Phylogenetic analysis of *Pinguicula* (Lentibulariaceae): chloroplast DNA sequences and morphology support several geographically distinct radiations. American Journal of Botany 92(10): 1723-1736.

Ernst, A. 1961. Revision der Gattung *Pinguicula*. Botanisch Jahrbücher 80(2): 145-194.

Gloßner, F. 1992. Ultraviolet pattern in the traps and flowers of some carnivorous plants. Botanisch Jahrbücher 113(4): 577-587.

Heslop-Harrison, J. 1970. Scanning electron microscopy of fresh leaves of *Pinguicula*. Science 167: 172-174.

Heslop-Harrison, J. 2004. *Pinguicula* L. Journal of Ecology 92: 1071-1118.

Juniper, B.E., Robins, R.J., and Joel, D.M. 1989. The Carnivorous Plants. Academic Press, London.

Kano, K. 1965. Studies on the media for orchid seed germination. Memoirs of Faculty of Agriculture, Kagawa University 20: 1-68.

Karlsson, P.S. 1986. Seasonal pattern of biomass allocation in flowering and nonflowering specimens of three *Pinguicula* species. Canadian Journal of Botany 64: 2872-2877.
Komiya, S. 1972. Systematic studies on the Lentibulariaceae. Dissertation, Nippon Dental College, Tokyo.
Komiya, S., and Shibata, C. 1980. Distribution of the Lentibulariaceae in Japan. Bulletin of Nippon Dental University, General Education 9: 163-212.
Komiya, S., and Shibata, C. 1998. A comparative-study of *Pinguicula ramosa* Miyoshi and *P. variegata* Turcz.; Ecology and morphology. Second conference of the International Carnivorous Plant Society, Botanischer Garten Bonn, Germany, Proceedings, Schlauer, J., and Meyers-Rice, B. (ed.), p. 23.
Komiya, S., and Shibata, C. 1999. Japanese *Pinguicula* (Lentibulariaceae). [in Japanese]. Bulletin of the Nippon Dental University, General Education 28: 117-146.
Kondo, K., and Shimai, H. 2006. Phylogenetic analysis of the northern *Pinguicula* (Lentibulariaceae) based on transcribed spacer (ITS) sequence. Acta Phytotaxonomica et Geobotanica 57(2): 155-164.
Löve, A., and Löve, D. 1982. IOPB chromosome number reports LXXV. Taxon 31(2): 342-368.
Miyoshi, M. 1890. Notes on *Pinguicula ramosa* sp. nov. Botanical Magazine Tokyo 4: 314-319.
Oba, H., Shimizu, J., Ayabe, M., and Tateno, M. 2013. Winter bud induction and optimal seed storage conditions of *Pinguicula ramosa* Miyoshi (Lentibulariaceae). [in Japanese with English abstract]. Bulletin of Japan Association of Botanical Gardens 47: 84-90.
Shimai, H., and Kondo, K. 2007. Phylogenetic analysis of Mexican and Central American *Pinguicula* (Lentibulariaceae) based on internal transcribed spacer (ITS) sequence. Chromosome Botany 2: 67-77.
Tamura, M. 1953. Key to the insectivorous plants of Japan. Acta Phytotaxonomica et Geobotanica 15(1): 31-32.
von Arx, B., Schlauer, J., and Groves, M. 2001. CITES Carnivorous Plants Checklist. Royal Botanic Gardens, Kew.
Yoshimura, K. 1973. Studies on the chromosome number and karyotype of *Pinguicula ramosa* Miyoshi (Lentibulariaceae). Journal of Japanese Botany 48(10): 289-294.
Zhukova, P.G., and Tikhonova, A.D. 1971. Chromosome numbers of certain plant species indigenous to the Chukotskiy Province. Bot. Zhurn. 56: 868-875.

HIRO SHIMAI

Born in Higashi-Hiroshima, Hiroshima, Japan. Botanical curator. Postgraduate research works at the University of Nottingham (in horticulture) and the University of Kent (in conservation). The author has extensive field observations of *Pinguicula* at habitats in Europe, Turkey, Cyprus, North and South Americas, Cuba, and Japan as well as specimen examinations at herbaria. Currently, the author is reviewing geographical distribution patterns of *Pinguicula* taxa based on specimens. Other interests are tropical plants, wetland plants, and plant conservations at botanical gardens.

Hiro Shimai searching for *Pinguicula crystallina* in Mugla Province, Turkey.

Getting to Mt. Koshin-zan

The closest train station to Mt. Koshin-zan is Haramuko Station on the Watarase Keikoku Railway line, connecting Kiryu and Mato stations along the upper half of the Watarase-gawa River (Fig. 1). To access from Tokyo, Tobu Railway's the "Ryomo" limited express train (seat reservation required) departs hourly from Asakusa Station in central Tokyo to Aioi Station, a transfer station

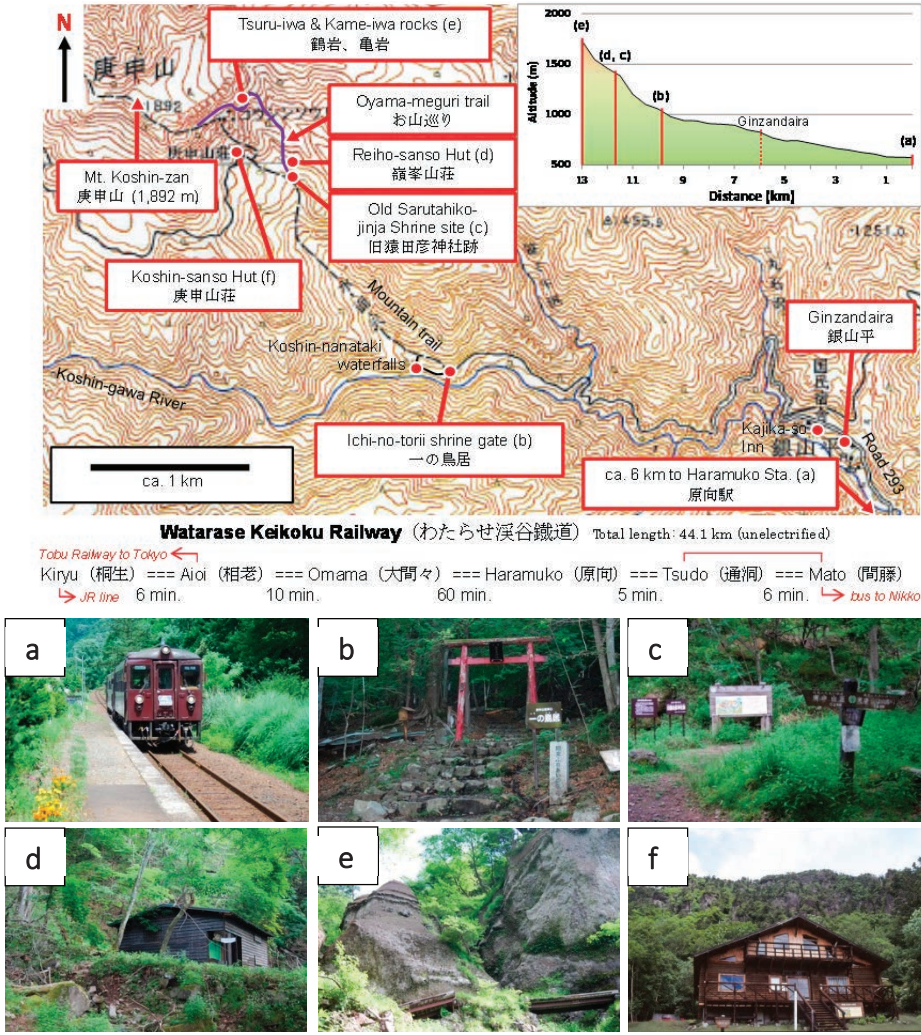


Figure 1: Map around Mt. Koshin-zan and views. The map was reproduced from 1:50,000 topographic maps "Ashio (NJ-54-30-9)" and "Nantai-san (NJ-54-29-12)" issued by Geospatial Information Authority of Japan. a) Haramuko Station, b) Ichi-no-torii shrine gate, c) Old Sarutahiko-jinja Shrine site, d) Reiho-sanso Hut, e) *P. ramosa* habitat of Kame-iwa rock (right) and Tsurigane-iwa rock (left). Tsuru-iwa rock, not shown in the picture, is located just on the left hand side of Tsurigane-iwa rock, f) Koshin-sanso Hut.

to the Watarase Keikoku Railway line, taking ca. 1 h 45 min. Then it takes further ca. 70 min. from Aioi Station to Haramuko Station.

The distance from Haramuko Station to Ginzandaira, a trailhead area, is ca. 6 km following on Road 293. There are no public transportations to reach Ginzandaira, but a taxi is available at Tsudo Station, or a mini-bus service may be arranged by Kajika-so Inn in Ginzandaira if staying there at least one night with an advanced booking. Then the road, mostly unpaved, continues approximately 4 km from Ginzandaira (ca. 840 m alt.) to Ichi-no-torii shrine gate (ca. 1,050 m alt.) along a deep gorge of the Koshin-gawa River, but no vehicles are permitted to enter beyond Kajika-so Inn (gate closed).

From Ichi-no-torii shrine gate, where an actual mountain trail starts, it will take approximately 1 h 20 min. for ca. 2 km to reach Old Sarutahiko-jinja Shrine site (the shrine buildings were destroyed by fire in 1946). A branch trail to Koshin-nanataki waterfalls diverging on the left from the main trail at ca. 150 m above Ichi-no-torii shrine gate is not for the mountain. The mountain trail runs mostly in a forest but signs and artificial objects indicate the route. At Old Sarutahiko-jinja Shrine site (ca. 1,450 m alt.), the trail diverges into two directions, the left trail goes for Koshin-sanso Hut or the summit of Mt. Koshin-zan, and the right trail goes a counterclockwise direction of Oyama-meguri trail. It will take further a few hours for a complete trekking on Oyama-meguri trail (ca. 2.5 km long). Tsuru-iwa and Kame-iwa rocks, almost the middle way on the loop trail, are good places to observe *P. ramosa*. To reach those rocks, the zigzagged steep trail, a counterclockwise direction on the loop, goes up from Old Sarutahiko-jinja Shrine site through behind Reiho-sanso Hut (not open to public). There are ladders and chains to climb up rocks or steep slopes on some parts of the trail. Immediately after passing beneath the overhanging rock of Kame-iwa, the *P. ramosa* habitats are just there.

Koshin-sanso Hut, ca. 200 m above Old Sarutahiko-jinja Shrine site, has basically no manager except weekends during peak seasons and only very basic facilities just for sleeping are provided. A number of people visit the mountain to see *P. ramosa* during the flowering season in June and it tends to be congested in the hut on weekends, but no reservations are accepted.

Alternatively, there is a local bus service between Nikko Stations and Tsudo Station (no rail link between those) taking about 50 min. though only 6 times per day. Nikko is a very popular holiday destination so that two different railway operators, JR and Tobu, connect Tokyo and Nikko. Car rental is also available in Nikko.

Because of higher elevations, the habitat is not easy to reach. The flowering season in the first half of June is usually about the beginning of rainy season. Appropriate shoes for trekking, a waterproofed jacket, large-scale maps (1:50,000 or 1:25,000), food and water, and very careful planning are required. There could be a higher risk of heavy rain particularly in the afternoon on the mountain. Under poor weather conditions, it is very dangerous to get there.