

Wild grape germplasms in Japan

H. Yamashita*, R. Mochioka**

* Faculty of Life and Environmental Sciences, University of Yamanashi, 4-4-37, Takeda, Kofu, Yamanashi, Japan.

** Faculty of Agriculture, Kagawa University, 2393 Ikenobe, Miki-cho, Kita, Kagawa, Japan.

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Abstract: In Japan, seven species and eight varieties of wild grapes were identified, among which the main species are *Vitis coignetiae* Pulliat, *V. flexuosa* Thunb., and *V. ficifolia* Bunge var. *lobata* (Regel) Nakai (syn. *V. thunbergii* Sieb. et Zucc.). This paper summarizes the identification and classification of wild grapes native to Japan based on the past reports. Their distributions in Japan and physiological and ecological traits are also reviewed for effective practical use for grape breeding in the future.

1. Introduction

It is thought that ancestors of grape (genus *Vitis*) appeared during the first half of the Cretaceous period. They then spread around the world according to environmental and anthropogenic influences, and now comprise three major groups of species: European, North American, and East Asian species, which differ in their physiological and ecological characteristics (Horiuchi and Matsui, 1996). Wild grapes native to Japan belong to the group comprising the East Asian species. Only a few reports on wild grape species, including classification, physiological, and ecological characterizations, have been published so far (Horiuchi and Matsui, 1996). Nevertheless, the importance of wild grapes as genetic resource for grape breeding has gradually been recognized because some wild grapes show superior traits towards global warming in terms of sustainable berry production under hot and humid conditions.

This paper describes the identification and classification of wild grapes native to Japan. Their physiological and ecological traits, as well as their utilization are also reviewed by focusing on the latest research findings regarding wild grapes native to Japan.

2. Geographical distribution

This, and previous studies, found that seven *Vitis* species and eight varieties are distributed throughout Japan, from Hokkaido (northern region) to Okinawa (southern region) (Table 1) (Nakagawa *et al.*, 1991). Of these, Yamabudo, Ebizuru, and Sankakuzuru are the three main spe-

cies found in Japan. Many other species exist locally in limited areas. In addition, researchers from Osaka Prefecture University discovered Shiohitashibudou (tentative name) (Nakagawa *et al.*, 1991). The geographical distribution of the wild grapes native to Japan are shown in figures 1-4. These figures were created from a site survey from Hokkaido to Okinawa starting in 1973, and were made based on past records and reports using conserved (pressed) leaf specimens from Hokkaido University, Tokyo Metropolitan University, Kyoto University, Niigata University, Kumamoto University (Japan), and Taiwan University (Taiwan).

Yamabudou, *Vitis coignetiae* Pulliat (Fig. 1)

This species is widely distributed from level ground to the lowest mountain areas in Hokkaido; from the lowest areas in the mountains to the mountain zone in the Tohoku district (northeastern region of Japan); from the mountain zone to the alpine region in the Chubu district (central region of Japan); and in the alpine regions of the Kinki, Chugoku, and Shikoku districts. It is thought that this species is also present in a limited area of the alpine regions in the Kyushu district, but it has not yet been discovered around Mt. Aso, which is consistent with the fact that we could not find any pressed leaf specimen in the universities located in the Kyushu district. It is noteworthy that this species is not distributed in South Korea, China, and neighboring countries; including Far Eastern Russia. However, it has been confirmed that Yamabudou grows naturally in the South Chishima and Sakhalin districts (Horikawa, 1972).

Sankakuzuru (*Gyojanomizu*), *V. flexuosa* Thunb. (Fig. 2)

This species is distributed in the mid regions of the Yamabudou (Fig. 1) and Ebizuru (Fig. 3) ranges, overlap-

Table 1 - Systematic and geographical distribution of wild grapes native to Japan (Nakagawa *et al.*, 1991)

Species or varieties	Japanese name	Locality where grown
<i>Vitis coignetiae</i> Pulliat	Yamabudou	Hokkaido, Honshu, Shikoku
<i>Vitis coignetiae</i> Pulliat var. <i>glabrescens</i> Hara	Takeshimayamabudou	Hokkaido, Honshu
<i>Vitis flexuosa</i> Thunb.	Sankakuzuru (Gyojanomizu)	Honshu, Shikoku, Kyusyu
<i>Vitis flexuosa</i> Thunb. var. <i>rufo-tomentosa</i> Makino	Kesankakuzuru	Southern Honshu, Shikoku
<i>Vitis flexuosa</i> Thunb. var. <i>tsukubana</i> Makino	Usugesankakuzuru	Northern Honshu
<i>Vitis flexuosa</i> Thunb. var. <i>crassifolia</i> Hara	Atsubasankakuzuru	Shikoku
<i>Vitis saccharifera</i> Makino	Amazuru (Otokobudou)	Southern Honshu, Shikoku
<i>Vitis yokogurana</i> Makino	Yokogurabudou	Shikoku (Kochi Pref.)
<i>Vitis ficifolia</i> Bunge var. <i>lobata</i> (Regel) Nakai (<i>Vitis thunbergii</i> Sieb. et Zucc.)	Ebizuru ⁽²⁾	All over Japan
<i>Vitis ficifolia</i> Bunge var. <i>izu-insularis</i> Hara	Shititoubizuru	Izu Islands
<i>Vitis ficifolia</i> Bunge var. <i>sinuata</i> Hara	Kikubaebizuru	Southern Honshu, Shikoku, Kyusyu
<i>Vitis ficifolia</i> Bunge var. <i>ganebu</i> Hatusima	Ryuukyuganebu	Amami Islands, Okinawa Islands, Yaeyama Islands
<i>Vitis austrokoreana</i> Hatusima	Kenashiebizuru	Tsushima Islands
<i>Vitis kiusiana</i> Momiyama	Kumagawabudou	Kyushyu (Kumamoto pref., Kagoshima pref.)
<i>Vitis shiragai</i> Makino	Shiragabudou	Honshu (Okayama pref.)
<i>Vitis</i> sp.	Shiohitashibudou (tentative)	Kyusyu (Kagoshima pref.)

⁽²⁾ used in some classifications as a species (*thunbergii*).



Fig. 1 - Geographic distribution of *Vitis coignetiae* Pulliat (Nakagawa *et al.*, 1986).



Fig. 2 - Geographic distribution of *Vitis flexuosa* Thunb. (Nakagawa *et al.*, 1986).

ping with the two species, and is found in slightly lower altitude areas than Yamabudou (Fig. 1). We can usually find this species from the lowlands to the mountainous area of the Tohoku district or the Chubu district; it does not grow naturally in Hokkaido.

Ebizuru, V. ficifolia Bunge var. lobata (Regel) Nakai (Fig. 3)

This species is one of the most widespread *Vitis* species in Japan. Its distribution extends from the southern Hokkaido region to the flatlands and mountainous terrain in the Okinawa district; it can be found in a wide variety of habitats, including both the seashore and urban districts. This species is considered to be highly adaptable to the environment, thus it has a wide distribution compared with other *Vitis* species. As a result, many variants of morphological and physiological traits are found in this species as a result of adaptation to local climates. Ryuukyuuganebu, Shichituebizuru, and Kikubaebizuru are varieties belonging to *V. ficifolia*. Kenashiebizuru is also closely related to *V. ficifolia*, although its scientific name is given as *V. auskoreana* Hatusima. These grapes are generally included in the *V. ficifolia* group.

Other species (Fig. 4)

As shown in Table 1 and figure 4, in addition to these, many species and varieties are spread in various districts

of Japan. Takeshimayamabudou, a variety of Yamabudou (*V. coignetiae*), was discovered in Hokkaido (around Lake Akan) and Nagano prefecture. It has no hairiness on the lower leaf surface and a thinner leaf compared to Yamabudou. Kumagawabudou has prickly shoots and ovoid leaves; one wild grape (*V. davidii*) with prickly shoots grows naturally in China, however, it differs significantly from Kumagawabudou in its morphological characteristics. Shichituebizuru grows in seaside areas of seven islands of Izu. Moreover, its fruit-set is the highest among all wild grapes native to Japan.

3. The classification of Japanese wild grapes

As genus *Vitis* is mainly classified by morphology, some taxa may be taken as different classifications even if they are the same grape. For example, Shiragabudou has two scientific names, *Vitis shiragai* Makino (Makino, 1918) and *Vitis amurensis* Rupr. (Ohwi, 1953). In this section, wild grapes native to Japan are classified concisely based on chemical, biochemical and genetic knowledge.

Morphological classifications

Classification by leaf structure. Galet (1979) tried to classify genus *Vitis* through ampelographic measurements



Fig. 3 - Geographic distribution of *Vitis ficifolia* Bunge var. *lobata* (Regel) Nakai (Nakagawa *et al.*, 1986).



Fig. 4 - Geographic distribution of wild grapes native to Japan (Nakagawa *et al.*, 1986).

of the leaf (Fig. 5, Table 2). Nakagawa *et al.* (1991) coded the characteristics of grape leaves and the result is presented in Table 3. Code numbers of vein length ratios (ABC) for the five basic leaf shapes are as follows (Galet, 1979):

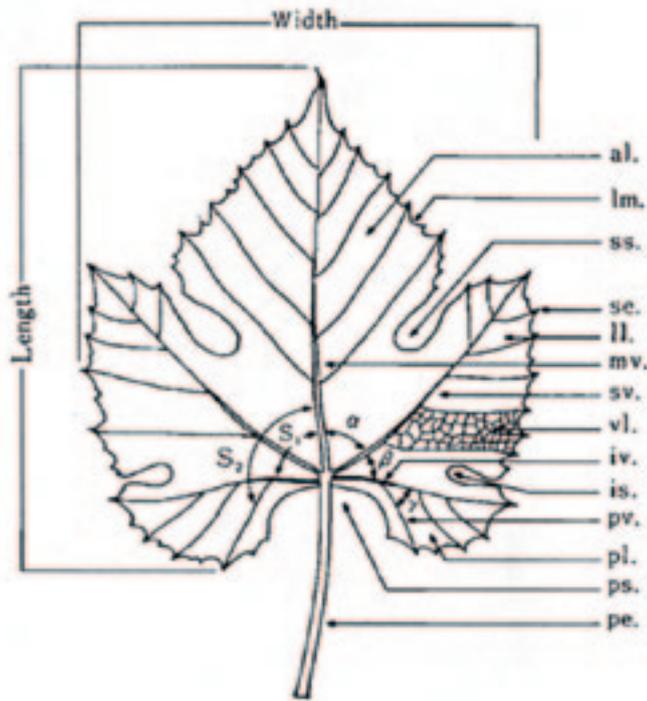


Fig. 5 - The general morphology of a mature grape leaf. al.= Apical lobe; ll.= Lateral lobe; pl.= Proximal lobe; ss.= Superior sinus; is.= inferior sinus; ps. Petiolar sinus; pe.= Petiole; mv.= Midvein; sv.= Superior lateral vein; iv.= Inferior lateral vein; pv.= petiolar vein; vl.= Veinlet; se.= Serration; lm= Leaf margin. (Nakagawa *et al.*, 1991).

Table 2 - The code number of the Galet (1979) ruler for the values of A, B, C, r, S₁ and S₂

Code number	Vales of A, B and C	Vales of r	Vales of S ₁	Values of S ₂
0	0.91~1.00	≤0.80	≤ 70°	≤ 100°
1	0.81~0.90	0.81~0.90	71°~80°	101°~110°
2	0.71~0.80	0.91~1.00	81°~90°	111°~120°
3	0.61~0.70	1.01~1.10	91°~100°	121°~130°
4	0.51~0.60	1.11~1.20	101°~110°	131°~140°
5	0.41~0.50	1.21~1.30	111°~120°	141°~150°
6	0.31~0.40	1.31~1.40	121°~130°	151°~160°
7	0.21~0.30	1.41~1.50	131°~140°	161°~170°
8	0.11~0.20		141°~150°	171°~180°
9	0.00~0.10		≥151°	≥181°

A= L₂ length/L₁ length; B= L₃ length/L₁ length; C= L₄ length/L₁ length, where L₁= Midvein; L₂= Superior lateral vein and L₃= Inferior lateral vein; L₄= Petiolar vein.

r= Leaf length/leaf width.

S₁= α + β; S₂= α + β + r where α= Angles between L₁ and L₂, β= Angles between L₂ and L₃, and r= Angles between L₃ and L₄.

Cordiform: 357 to 468, Cuneiform: 135 to 247, Truncate: 045 to 247, Orbicular: 015 to 136, Reniform: 014 to 136. According to this method, code numbers are relatively near for close species (e.g. *V. coignetiae* and *V. amurensis*) (Table 3).

Classification by pollen ultrastructure. Mochioka *et al.* (1993) observed ultrastructures of mature pollen grains of wild grapes native to Japan, Korea and China using a scanning electron microscope and reported that the pollen could be classified as one of three types by the lumina forms in muri (Fig. 6). They also reported the

Table 3 - The code number of various wild grapes obtained by using the Galet's method (Nakagawa *et al.*, 1991)

Species	ABC-r-S ₁ S ₂
<i>Vitis coignetiae</i>	146-4-24
<i>Vitis amurensis</i>	146-3-24
<i>Vitis flexuosa</i>	357-7-01
<i>Vitis ficifolia</i> var. <i>lobata</i>	246-3-13
<i>Vitis shiragai</i>	136-3-13
<i>Vitis</i> sp. (Daisankakuzuru)	146-3-12
<i>Vitis ficifolia</i> Bunge var. <i>ganebu</i>	135-3-24
<i>Vitis</i> sp. (Shiohitashibudo)	257-5-02
<i>Vitis ficifolia</i> var. <i>izu-insularis</i>	146-3-01
<i>Vitis kiusiana</i>	368-7-01

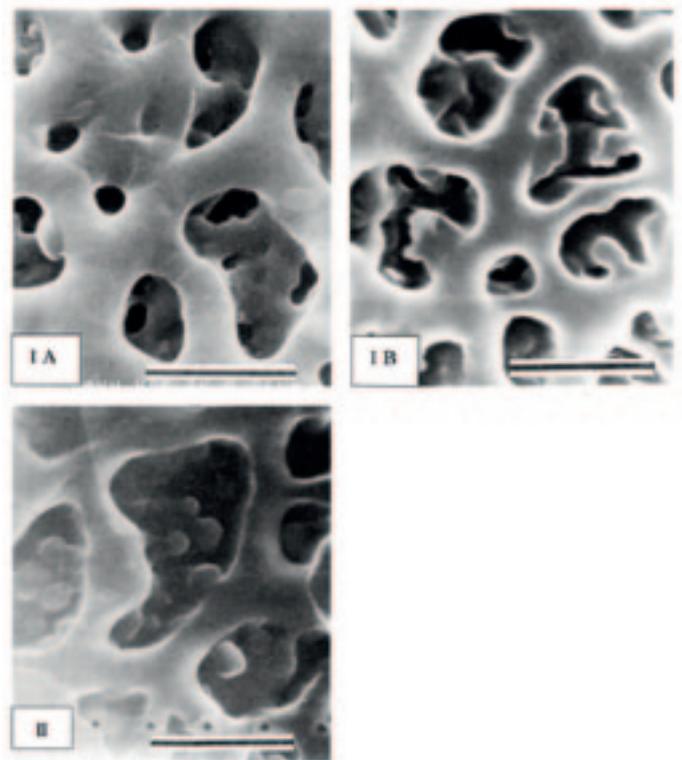


Fig. 6 - Scanning electron microphotographs of grape exine. Scale bars= 1 μm. Type-1 A pollen has perforations in the lumina; type-1 B pollen has perforations and corrugation in the lumina, and type-II pollen has granules in the lumina (Mochioka *et al.*, 1993).

pollen ultrastructures of related species belonged to the same type (Table 4).

Chemotaxonomic classifications

Classification by anthocyanins in grape skin. Mochioka

et al. (1995) analyzed by HPLC anthocyanins in the berry skin of 10 wild grapes (four species, five varieties and one unidentified type) native to Japan. The dendrogram, showing phylogenetic relationships, was drawn from the pairwise comparison of matching coefficients based on anthocyanin

Table 4 - Morphological characteristics of pollen grains of the wild grapes native to Japan, Korea and China (Mochioka *et al.*, 1993)

Species or cultivars		Pollen size (μm)		L/W ratio	No. of colpi	Pollen exine type
		Length (L)	Width (W)			
Japan						
<i>Vitis coignetiae</i> Pulliat	♂	20.0±0.2 ⁽²⁾	19.4±0.2	1.03±0.01	3	I A
	♀	22.0±0.2	20.7±0.2	1.06±0.01	0	I A
<i>Vitis flexuosa</i> Thunb.	♂	20.3±0.5	19.6±0.3	1.04±0.03	3	I A
	♀	20.6±0.2	20.1±0.2	1.03±0.01	0	I B, II
<i>Vitis ficifolia</i> Bunge var. <i>lobata</i> (Regel) Nakai	♂	20.0±0.2	19.2±0.2	1.04±0.01	3	II
	♀	20.9±0.2	20.0±0.2	1.05±0.01	0	II
<i>Vitis ficifolia</i> Bunge var. <i>izu-insularis</i> Hara	♂	20.9±0.3	20.1±0.4	1.04±0.01	3	II
	♀	21.4±0.2	20.2±0.2	1.06±0.01	0	II
<i>Vitis ficifolia</i> Bunge var. <i>ganebu</i> Hatusima	♀	21.4±0.3	20.3±0.2	1.05±0.01	0	I A
<i>Vitis shiragai</i> Makino	♂	21.1±0.2	20.7±0.2	1.02±0.01	3	I A
	♀	22.6±0.2	21.1±0.3	1.08±0.01	0	I A
<i>Vitis kiusiana</i> Momiyama	♀	20.4±0.2	19.5±0.2	1.05±0.01	0	I B
<i>Vitis</i> sp. (provisional name: Shiohitashibudo)	♀	21.3±0.3	20.0±0.3	1.06±0.01	0	II
Korea						
<i>Vitis amurensis</i> Rupr.	♂	20.6±0.3	20.1±0.2	1.03±0.01	3	I A
	♀	23.4±0.2	22.1±0.2	1.06±0.01	0	I B
<i>Vitis</i> sp. (provisional name: Daisankakuzuru)	♀	21.4±0.2	20.3±0.2	1.06±0.01	0	I B
China						
<i>Vitis amurensis</i> Rupr.	♂♀	21.5±0.2	20.7±0.2	1.04±0.01	3	I B
<i>Vitis flexuosa</i> Thunb.	♂	19.8±0.3	19.0±0.3	1.04±0.01	3	I A
<i>Vitis ficifolia</i> Bunge	♂	20.6±0.3	19.0±0.3	1.04±0.01	3	II
<i>Vitis adstricta</i> Hance	♂	21.5±0.2	21.2±0.2	1.02±0.01	3	II
<i>Vitis adstricta</i> Hance var. <i>ternata</i> W.T. Wang	♂	22.3±0.3	21.0±0.4	1.06±0.01	3	II
<i>Vitis bellula</i> (Rehd.) W.T. Wang	♂	19.3±0.3	18.8±0.2	1.03±0.01	3	II
<i>Vitis davidii</i> (Roman.) Foëx	♂	23.1±0.2	22.4±0.2	1.03±0.01	3	I B
	♂♀	20.4±0.2	19.9±0.1	1.03±0.01	3	I B
<i>Vitis pseudoreticulata</i> W.T. Wang	♂	21.3±0.2	20.8±0.2	1.03±0.01	3	I A
<i>Vitis hancockii</i> Hance	♂	20.1±0.2	19.6±0.1	1.03±0.01	3	I B
<i>Vitis chugii</i> Metcalf.	♂	19.9±0.2	19.3±0.2	1.03±0.01	3	I B
<i>Vitis chunganensis</i> Hu	♂	19.9±0.2	19.7±0.2	1.01±0.01	3	I A
Cultivar						
<i>Vitis vinifera</i> L.						
‘Muscat of Alexandria’	♂♀	23.2±0.4	22.4±0.4	1.03±0.01	3	I B
<i>Vitis lambrusca</i> L.						
‘Concord’	♂♀	23.3±0.3	22.9±0.3	1.02±0.01	3	I B
<i>Vitis lambrusca</i> Bailey						
‘Delaware’	♂♀	21.3±0.3	20.1±0.3	1.06±0.01	3	I B
‘Campbell Early’	♂♀	24.6±0.4	23.4±0.4	1.05±0.01	3	II
‘Kyoho’ (tetraploid)	♂♀	28.2±0.3	26.9±0.3	1.05±0.01	3.4	I A

⁽²⁾ Each valure represents the mean of 20 individual measurements ±SE.

components, and agrees well with the morphological taxonomy (Fig. 7, Table 5) (Mochioka *et al.*, 1995).

In this study 19 anthocyanins were identified, and there were more kinds of anthocyanins in the berry skins of wild grapes distributed in southern regions than those of wild grapes distributed in northern regions (Table 5).

Classification by isozyme and DNA analysis. Species-specificity was observed in the alleles dominated by *Gpi-2* and *Pgm-2* gene loci (Fig. 8, Table 6) (Ohmi *et al.*, 1991). The F band of *Gpi-2* and the A and the C bands

of *Pgm-2* existed only in wild grapes native to East Asia (Table 6).

While restriction fragment length polymorphism (RFLP) and random amplified polymorphic DNA (RAPD) analyses were used to analyze the relationships among wild and cultivated grapes, a phenogram of RAPD data obtained showed a clear separation between wild and cultivated grapes (Goto-Yamamoto *et al.*, 1998).

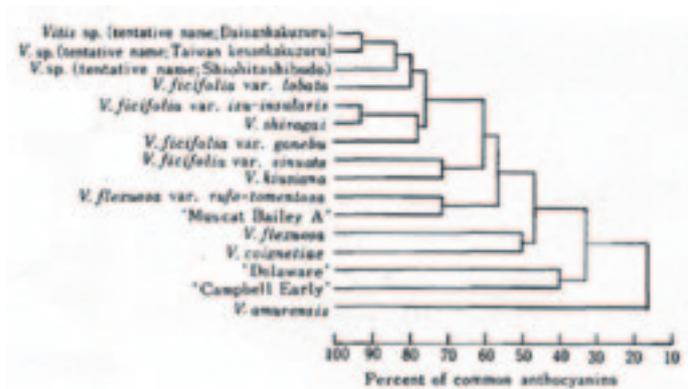


Fig. 7 - Dendrogram of berry skin anthocyanin phenotypes of wild and cultivated varieties (Mochioka *et al.*, 1995).

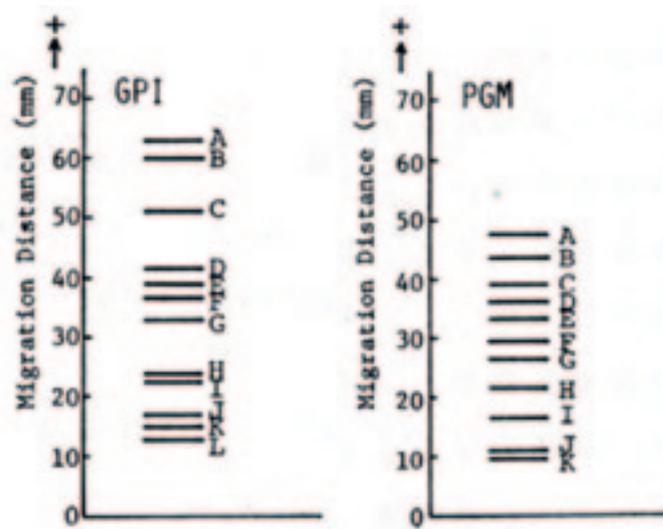


Fig. 8 - GPI and PGM isozymes bands coded by each alleles at *Gpi-2* (left) and *Pgm-2* (right) loci (Ohmi *et al.*, 1991).

Table 5 - Percentage of anthocyanin composition in grape berry skins analyzed by HPLC ⁽²⁾ (Mochioka *et al.*, 1995)

Species or cultivars	Peak No ⁽³⁾																		
	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19
Wild grape																			
<i>Vitis coignetiae</i>			1		4	66			1	4		2			1		18		2
<i>V. amurensis</i>	2		2			81													
<i>V. flexuosa</i>	5		4		4	57			7		4					1	12		2
<i>V. flexuosa</i> var. <i>rufo-tomentosa</i>			2		1	28			3			3	2	1	4	1	38	2	8
<i>V. ficifolia</i> var. <i>lobata</i>	6	2	10		6	44		2	2	1		7	5	10			12		1
<i>V. ficifolia</i> var. <i>izu-insularis</i>	15	3	17		5	27			3	1		3	12	5	2	2	4		
<i>V. ficifolia</i> var. <i>ganebu</i>	8	4	9	8	10	28		2	4			3	4	1	3	2	9		1
<i>V. ficifolia</i> var. <i>sinuata</i>	10	5	39		13	8	9	4	3		6		1		2	1			
<i>V. shiragai</i>	11	7	5	13	15	26			2	1		3	4	2	2	2	4		
<i>V. kiusiana</i>	13	10	28		19	11		1	1		6		3	3	2		1		
<i>V. sp.</i> (tentative name: Shiohitashibudou)	6	3	2	10	15	37		2	6	1	1	4	1	1		2	2		
<i>V. sp.</i> (tentative name: Daisankakuzuru)	5	3	28		17	12	7	7	4	3		2	3	2		2	3		
<i>V. sp.</i> (tentative name: Taiwan kesankakuzuru)	5	2	24		6	36		1	8	3		3	2	2		2	3		
Cultivars																			
'Delaware'					44			26					4	3				9	4
'Muscat Bailey A'			3		21		4	32	23				2	2	1	2	2	1	2
'Campbell Early'		6			6								12	46	6	2	7	6	

⁽²⁾ Absorbance at 520 nm.

⁽³⁾ Peak No. 3= delphinidia 3-monoglucoside, no. 5= cyaniding 3-monoglucoside; no. 9= petunidin 3-monoglucoside; no. 16= malvidin 3-monoglucoside.

Table 6 - Species-specific alleles at 2 loci in grape (Ohmi *et al.*, 1991)

Locus	Allele	Species ⁽²⁾
Gpi-2	A	Vin.
	B	Vin., Amur. (?)
	C	Lab., Rip. (?)
	D	Vin.
	E	Lab., Shir.
	F	Thun., Shir.
	G	Vin., Amur.
	H	Lab., Aest. (?), Vulp (?)
	I	Lab., Rup., Champ., Linc. (?), Coig., Shir.
	J	Bourq. and/or Lab.
	K	Champ.
Pgm-2	A	Shir.
	B	Vin.
	C	Amur., Coig., Thun., Shir.
	D	Vin.
	E	Vin.
	F	Lab. and/or Linc.
	G	Lab., Aest. (?)
	H	Vin., Rup., Champ., Lab., Coig., Aest. (?), Bourq. (?), Rip. (?), Vulp. (?)
	I	Vin.
	J	Rup.
	K	Champ.

⁽²⁾ Abbreviations: Amur.= *Vitis amurensis*; Aest.= *V. aestivalis*; Bourq.= *V. aestivalis* var. *bourquiana*; Champ.= *V. champini*; Lab.= *V. labrusca*; Linc.= *V. lincecumii*; Rip.= *V. riparia*; Rup.= *V. rupestris*; Shir.= *V. shiragai*; Tun.= *V. thunbergii* (= *V. ficifolia* var. *lobata*); Vin.= *V. vinifera*; Vulp.= *V. vulpina*.

Classification by general judgments

Since the past horticultural plant classification was qualitatively performed considering a small number of characteristics, a different result for some researchers might be found (e.g. Shiragabudou). Therefore, using plural classification methods is desirable because just one method might induce the wrong result.

What follows is a brief discussion of judgments about some Japanese wild grapes with questionable taxonomic points.

Shiragabudou. Shiragabudou was discovered in Okayama prefecture, Honshu and was first named by Makino (1918). As the leaf shape of this wild grape resembles that of *V. amurensis* Rupr. Ohwi (1953) changed its scientific name to *Vitis amurensis* Rupr. The leaf morphology and pollen ultrastructures of these two wild grapes are in the same group (Table 3, 4), but anthocyanin composition in berry skins (Table 5) and species-specific alleles at 2 loci (Table 6) are apparently different.

Furthermore, ecological differences exist between Shiragabudou and *V. amurensis*. Shiragabudou is distributed over the warm lowland from 20 to 240 m above sea level in Okayama while *V. amurensis* has a growth area in the cold districts at 40 to 50° N latitude. The cross section morphology of Shiragabudou shoots is hexagonal, while that of *V. amurensis* is circular.

These differences show that Shiragabudou and *V. amurensis* are not the same species, thus *Vitis shiragai* Makino should be used as the scientific name for Shiragabudou.

Ebizuru and its varieties. Ebizuru is distributed widely in Japan. There are a number of varieties and ecotypes in the Ebizuru group, and morphological differences are various. Even if they are the same species, there are several synonyms for this group. Even now, *Vitis ficifolia*, *V. ficifolia* var. *lobata*, and *V. thunbergii* are used as scientific names for Ebizuru. An isotype of *V. ficifolia* is the wild grape native to China. There are definitely differences in leaf morphology and bearing habit between Chinese *ficifolia* and Japanese Ebizuru.

Natural hybrids. Different *Vitis* species can be hybridized easily with each other, so there are many natural hybrids. *V. yokogurana* is supposed to be a hybrid of *V. flexuosa* and *V. saccharifera* (Makino, 1918); *V. flexuosa* var. *tukubana* is supposed to be a hybrid of *V. flexuosa* and *V. ficifolia* var. *lobata* (Murata, 1971).

Yamabudou. Yamabudou (*V. coignetiae*) must be a species related to *V. amurensis*, but there is no report that both these two species are simultaneously distributed over the same regions in Japan or other countries.

Shiohitashibudou. (tentative name): Shiohitashibudou is an unidentified species, and it was discovered in Kagoshima prefecture, Kyushu (Nakagawa *et al.*, 1991). Its leaf morphology is definitely different from that of other Japanese wild grapes. Shiohitashibudou might be a related species or natural hybrid of Ebizuru because its flowering habit is ever-bearing and the pollen ultrastructure is type II. Its bud endodormancy is deeper, and the soluble solid content of its juice is higher than other Japanese wild grapes (Mochioka, 1996).

Ryuukyuganebu. Ryuukyuganebu is distributed over Amami, Ryukyu and Yaeyama Islands, and is supposed to be a variety of *V. ficifolia*. However its leaf shape is different from *V. ficifolia* var. *lobata* (Fig. 9, Table 3), and its pollen ultrastructure is also different (Table 4). Ryuukyuganebu is ever-green in its habitat.

4. Physiological and ecological traits of wild grapes native to Japan

Four species [*V. coignetiae*, *V. flexuosa*, *V. shiragai*, and Shiohitashibudou (tentative name. *Vitis* sp.)] and two varieties (*V. ficifolia* var. *lobata* and *V. ficifolia* var. *ganebu*) of wild grapes native to Japan, and two species [Chosen Yamabudou (*V. amurensis*) and Daisankakuzuru (tentative name. *V. sp.*)] grown in Korea, and 'Delaware' (*V. labrus-*

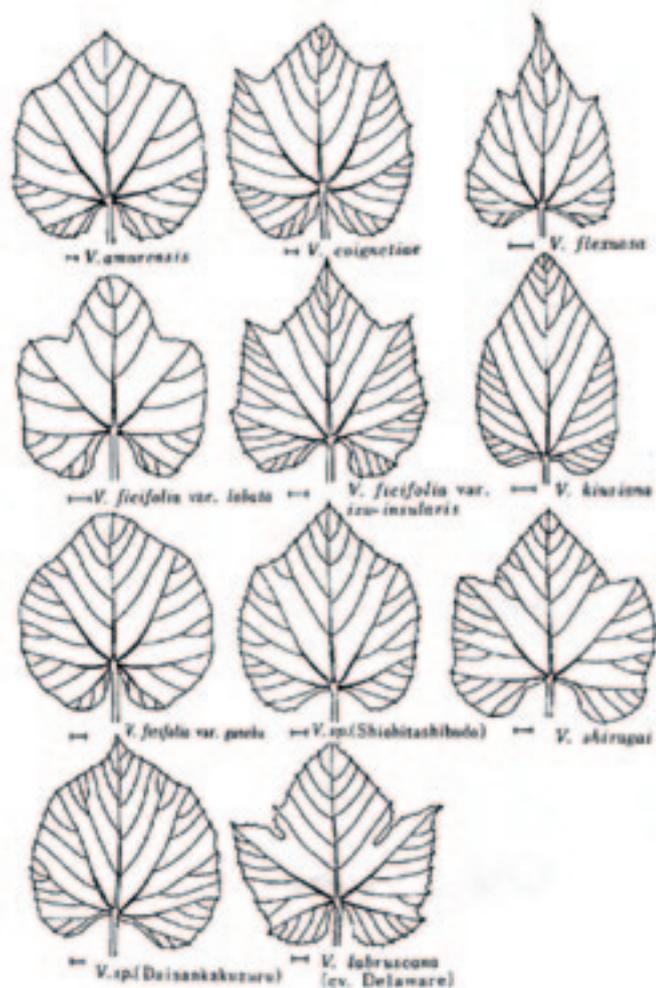


Fig. 9 - Standard design of mature leaf of various wild grapes (—| 1 cm) (Nakagawa *et al.*, 1991).

cana Bailey) (Table 7) were cultivated in the Horticultural Experiment Field at Osaka Prefecture University (Sakai, Osaka) and their physiological and ecological traits were compared (Nakagawa *et al.*, 1986). Here, Chosen Yamabudou is a wild grape grown naturally in the northern and northeastern regions of China, northern region of Korea and southeastern region of the former Soviet Union. The results are illustrated below.

Growth cycle

Bud burst. Bud burst of Yamabudou and Chosen Yamabudou occurred earlier among the wild grapes, followed by Sankakuzuru, Ebizuru, and Daisankakuzuru. Shiohitashibudou showed the latest bud burst in this study (Table 7). 'Delaware' broke bud later than all wild grapes except for Shiohitashibudou.

Full bloom (50% cap off). In grape cultivation, early bud burst does not always mean early bloom. Indeed, the orders of bloom date differed from those of bud burst (from early to late bloom): Yamabudou, Sankakuzuru Ebizuru/Shiragabudou, Daisankakuzuru, Shiohitashibudou, and Ryuukyuuganebu. Here, Ebizuru blooms in the same period as Shiragabudou, and the bloom date of Sankakuzuru was comparable to that of 'Delaware' (Table 7).

Veraison (berry coloring begins). The skin color of Yamabudou changed earliest among the wild grapes. Changes in skin color in Chosen Yamabudou and 'Delaware' occurred on the same date. Skin color change occurred the latest in Daisankakuzuru, Shiohitashibudou, and Ryuukyuuganebu; and Sankakuzuru, Shiragabudou and Ebizuru were the next to latest (Table 7).

Maturity. Maturity is denoted when berry weights and soluble solids attain maximum maturation. 'Delaware' matured in mid-August, which was earlier than the studied wild grapes. Sankakuzuru and Ryuukyuuganebu matured in mid-September and Daisankakuzuru in early-October (Table 7). Thus, wild grapes tend to have medium to late maturation in Osaka.

Defoliation. Defoliation indicates the date when all leaves (from basal to tenth leaf) fall completely. Defoliation in Sankakuzuru and Daisankakuzuru took place in early-November, Yamabudou and Chosen Yamabudou shed their leaves in mid-November, and Shiohitashibudou, Ebizuru, and Shiragabudou in late-November. Interestingly, Ryuukyuuganebu, a subtropical grape, showed extremely late defoliation; in some cases, leaves did not fall until January.

Characteristics of organ

Shoot. Observations of shoot growth in summer enabled us to classify the wild grapes into three types: continuous, subcontinuous, and discontinuous. The continu-

Table 7 - Growth cycle of wild grapes native to Japan at Sakai Osaka (Nakagawa *et al.*, 1986)

Species and varieties	Bud burst	Full bloom	Veraison	Harvest	Leaf fall
Chosen yamabudou (<i>V. amurensis</i> Rupr.)	3/27	5/14	8/1	9/29	11/14
Yamabudou (<i>V. coignetiae</i> Pulliat)	3/27	5/12	7/26	9/29	11/13
Sankakuzuru (<i>V. flexuosa</i> Thunb.)	3/29	5/26	8/6	9/14	11/7
Ebizuru [<i>V. ficifolia</i> Bunge var. <i>lobata</i> (Regel) Nakai]	3/29	6/4	8/11	9/11	11/27
Shiragabudou (<i>V. shiragai</i> Makino)	3/29	6/4	8/6	9/15	11/23
Daisankakuzuru (tentative <i>Vitis</i> sp.)	3/29	6/10	9/4	10/6	11/4
Shiohitashibudou (tentative <i>Vitis</i> sp.)	4/10	6/13	9/4	9/22	11/25
Ryuukyuuganebu (<i>V. ficifolia</i> Bunge var. <i>ganebu</i> Hatusima)	4/3	6/19	8/31	9/20	after end of Dec.
Delaware (<i>V. labruscana</i> Bailey)	4/6	5/27	7/30	8/20	11/8

ous group had constant growth of some shoots in summer; Shiohitashibudou and Daisankakuzuru were included in this type. The subcontinuous type exhibited slight growth of some shoots in summer; Ryuukyuganebu, Sankakuzuru, Ebizuru, and Shiragabudou corresponded to this type. Finally, the discontinuous group stopped shoot growth in summer, for example Yamabudou and Chosen Yamabudou in this study. Tendril placement of all *Vitis* species and varieties native to Japan is intermittent.

Inflorescence. Wild grapes native to Japan (seven species and eight varieties) are dioecious, meaning that they contain imperfect individual male and female plants. Three types of fruiting habits were found (Fig. 10). “A” type: as is the case of Yamabudou, Chosen Yamabudou, Sankakuzuru, Daisankakuzuru, Shiragabudou, and Kumagawabudou,

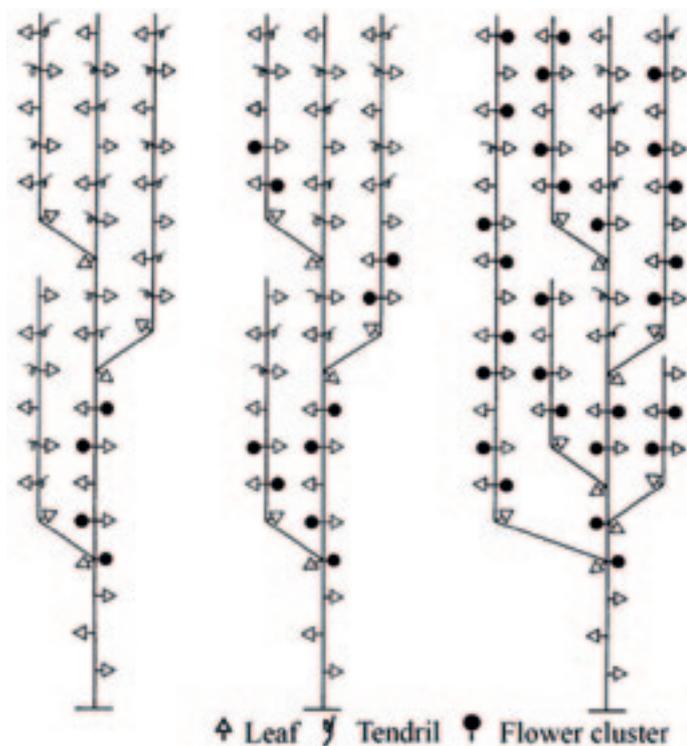


Fig. 10 - Fruiting habit of wild grapes native to Japan.

with two to four inflorescences at the basal part on each shoot without any inflorescences on lateral shoots. “B” type: as seen in Shiohitashibudou, Shichitoubizuru, and some Shiragabudou, there are two to four inflorescences at the basal part on each shoot with some inflorescences on lateral shoots. “C” type: Ebizuru and Ryuukyuganebu belong to this type, with two to six inflorescences at the basal part and the upper part of each shoot, having contiguous inflorescences from the base to the top of lateral shoots.

Grape comparison and fruit quality (Table 8)

Sugars. Shiohitashibudou attained a sugar concentration of 17.7%, which is the highest among the other species (measured as 12-14%) except Ebizuru and Ryuukyuganebu, which had sugar concentrations of around 8%. Almost all studied species have high glucose content, especially Kumagawabudou, Shiohitashibudou, and Daisankakuzuru, which contained two to three fold more glucose than fructose.

Acids. Kumagawabudou and Daisankakuzuru contained about 0.7 and 0.8% of organic acids, while almost all other species contained about 0.5%; Shiragabudou showed the lowest level of organic acids (0.36%) (Table 8).

Amino acids. The concentration of amino acids varied widely from 50 mg% (Ebizuru) to 294 g% (Shiohitashibudou).

Anthocyanins. Northern species such as Yamabudou and Chosen Yamabudou showed small amounts of anthocyanin, while the southern species, Ryuukyuganebu, contained a larger amount of this component (Table 8).

Dormancy and cold hardiness

Dormancy. Although all species have a dormancy trait, its intensity depends on the species. Ryuukyuganebu showed a short dormancy period, while it was generally longer in Kumagawabudou and Shiohitashibudou (Nakagawa *et al.*, 1986; Nakagawa, 1989).

Cold hardiness. Cold hardiness varied markedly among the wild grapes. For example, Ryuukyuganebu and Kumagawabudou were very susceptible to severe damage at -3°C. On the contrary, Yamabudou, Ebizuru, Shiragabu-

Table 8 - Berry composition of wild grapes native to Japan (Nakagawa *et al.*, 1986)

Species and varieties	Reducing sugars (%)	Glucose (%)	Fructose (%)	Glucose/ Fructose	Organic acid (%)	Amino acid (mg %)	Anthocuanin (OD 537 nm)
Chosen yamabudou (<i>V. amurensis</i> Rupr.)	12.7	9.2	3.5	2.6	0.50	191.2	0.12
Yamabudou (<i>V. coignetiae</i> Pulliat)	12.3	5.2	7.1	0.7	0.50	155.3	0.21
Sankakuzuru (<i>V. flexuosa</i> Thunb.)	14.2	8.0	6.2	1.3	0.52	214.0	0.30
Ebizuru [<i>V. ficifolia</i> Bunge var. <i>lobata</i> (Regel) Nakai]	8.1	4.1	4.0	1.0	0.51	50.7	0.30
Shiragabudou (<i>V. shiragai</i> Makino)	12.0	6.7	5.3	1.3	0.36	222.6	0.45
Daisankakuzuru (tentative <i>Vitis</i> sp.)	12.3	8.1	3.9	2.2	0.81	250.8	0.42
Kumagawabudou (<i>V. kiusiana</i> Momiyama)	12.0	9.2	2.8	3.3	0.72	180.5	0.49
Shiohitashibudou (tentative <i>Vitis</i> sp.)	17.7	12.7	5.0	2.5	0.48	294.5	0.41
Ryuukyuganebu (<i>V. ficifolia</i> Bunge var. <i>ganebu</i> Hatusima)	7.8	3.7	4.1	0.8	0.51	138.0	0.70
Delaware (<i>V. labruscana</i> Bailey)	16.8	7.5	9.3	0.8	0.77	220.8	0.05

dou, Shichitoubizuru, and Chosen Yamabudou showed moderate cold hardiness; their survival has even been reported at -10°C (Nakagawa, 1989).

5. Value and use of wild grape germplasms in Japan

Wine

Between the 1960s and 1980s, Yamabudou was successfully cultivated in commercial vineyards for wine-making in the town of Ikeda (Hokkaido); its cultivation has attracted attention as a means to revitalize towns in Japan. In China, *V. quinquangularis* is processed into an excellent wine (Li *et al.*, 1992). Kumagawabudou, which is thought to be the same species as *V. quinquangularis* (Li *et al.*, 1991), therefore, may be an important resource for wine making.

Breeding

The major cultivars, bred using wild grapes, are 'Sawanobori Waingurando' and 'Yama Sauvignon' in Japan, both of which are used for wine production. 'Sawanobori Waingurando' is a cross seedling of *V. amurensis* × (Seibel 13053 × Nakajima No.1, a strain of Yamabudou) and it was released in 1998. By contrast, 'Yama Sauvignon' is a progeny of Yamabudou × 'Cabernet Sauvignon', which was released in 1990 by Dr. Yoshihide Yamakawa at the University of Yamanashi. 'Yama Sauvignon' has the following superior characteristics: 1) no cracking of berry; 2) resistance to ripe rot, downy mildew, and gray mold; 3) adaptability to the prevailing weather conditions in Japan; 4) high productivity; and 5) high quality wine with typical aroma and taste (Yamakawa *et al.*, 1989).

Considering the potential use of wild grapes as breeding material, these grapes have the following notable characteristics: drought resistance, cold hardiness, salt tolerance, water logging tolerance, heat tolerance, disease resistance, high concentration of important substances, ever bearing, and short dormancy. Four characteristics are especially promising: i) ever bearing, ii) short dormancy, iii) salt tolerance, and iv) heat tolerance. Therefore, we explain the usefulness of these characteristics for grape breeding programs in more detail.

i) Ever bearing

Strains of Ryuukyuganebu and Ebizuru bloom and fruit as long as the growth of axillary buds continues. This trait makes it possible to carry out year-round culture and/or culture using a factory system, like some vegetables, using artificial light and controlled irrigation. However, the major gene related to this trait has not yet been identified.

ii) Short dormancy

Ryuukyuganebu can be released from dormancy after being subjected to low temperature for extremely short periods. This trait may be profoundly related to ever bearing. Thus, it may be possible to force culture inside greenhouses due to the low cost, for year-round culture and/or culture using a factory system.

iii) Salt tolerance

Salt accumulation through the use of chemical fertilizers causes serious problems which sometimes result in the loss of plants. Shiohitashibudou is excellent in its resistance to salts and Ryuukyuganebu grows naturally along the seashore. Although the mechanism underlying salt tolerance of this grape has not yet been fully elucidated, this trait could be useful not only for scion but also for root stock.

iv) Heat tolerance

As global warming progresses, fruit skin, including grape, shows poor coloration, which leads to a defective appearance and reduced commercial value. Poor coloration also affects wine production. Interestingly, the coloration of wild grapes native to Japan is very high, even under high temperature conditions during their ripening season. In grape, since MYB is involved in red skin coloration, it is valuable to compare MYB genes between wild grapes and primary Japanese cultivars such as 'Kyoho' and 'Aki Queen'. Thus, this trait could be useful for sustainable grape production with high quality and high adaptability in the production area.

6. Conclusions

Wild grapes native to Japan have been actively studied over a long period, but in recent years attention has declined. However, wild grapes native to Japan can offer many useful characteristics, such as short dormancy, ever bearing, heat and salt tolerance. These traits are very attractive, not only for their use as rootstock, but also in terms of breeding material or for genetic studies. We have recently begun a breeding study using Ryuukyuganebu. It is expected that some novel grapes will be bred to withstand increasing global temperatures or for use in grape cultivation factories.

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