

Collection of Wild and Cultivated Rare *Malus* Genetic Resources in Northern Kyūshū, Japan

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Summary

We collected two *Malus* species in northern Kyūshū. We explored two native populations of *Malus toringo* (Siebold) Siebold in Ōita and Saga Prefectures and a site where *Malus micromalus* Makino is cultivated in Nagasaki Prefecture. We collected scions from seven trees and tried to proliferate them by grafting. In addition, we collected a living tree of *Malus hupehensis* (Pamp.) Rehder, the Japanese population of which is extinct in nature and now survives only in cultivation, in Kumamoto Prefecture.

KEY WORDS : fruit tree, crabapple, *Malus*, endangered species, Ōita, Saga, Nagasaki, Kumamoto

Introduction

The genus *Malus* Miller is distributed in the Northern Hemisphere, mainly in the cool-temperate zone. *Malus toringo* (Siebold) Siebold, which has the broadest distribution among the four native Japanese *Malus* species, is now most commonly found in northern Japan or at high elevations in central Japan. However, several herbarium specimens were collected in low-elevation areas from either natural or artificial wetlands, such as around ponds, marshes, rivers, and reservoirs. Therefore, at one time this species may also have been distributed more widely in low-elevation areas.

Kyūshū Island is the southern limit of the distribution of *M. toringo* in Japan (Iketani and Ohashi 2001). Only a few specimens from Fukuoka and Saga Prefectures were found in the representative herbaria. At present, it is ranked in the prefectural Red Lists as extinct (Fukuoka Prefectural Government 2011) or critically endangered (Saga Prefectural Government 2010; Ōita Prefectural Government 2011). Despite these circumstances, we found the tree growing in two natural habitats based on our preliminary collection of information from local floras (e.g., Hatusima 2004), herbaria, websites, and conversations with regional botanists.

We also planned to collect germplasms of *Malus micromalus* Makino. This species is one of two traditional Japanese ornamental crabapples (Japanese name kaidō), with the other being *Malus halliana* Koehne. Both species originated in China and were introduced to Japan several hundred years ago. Sources suggest that *M. micromalus* was more common during the Edo Period (Maekawa *et al.* 1961; Kitamura 1985), but today *M. halliana* is the more common species. Although plants of *M. micromalus* are still commercially available, they may be derived from a small stock in nurseries and genetically limited. From our collection of information on *Malus* in this region, we discovered that old trees are cultivated at one site

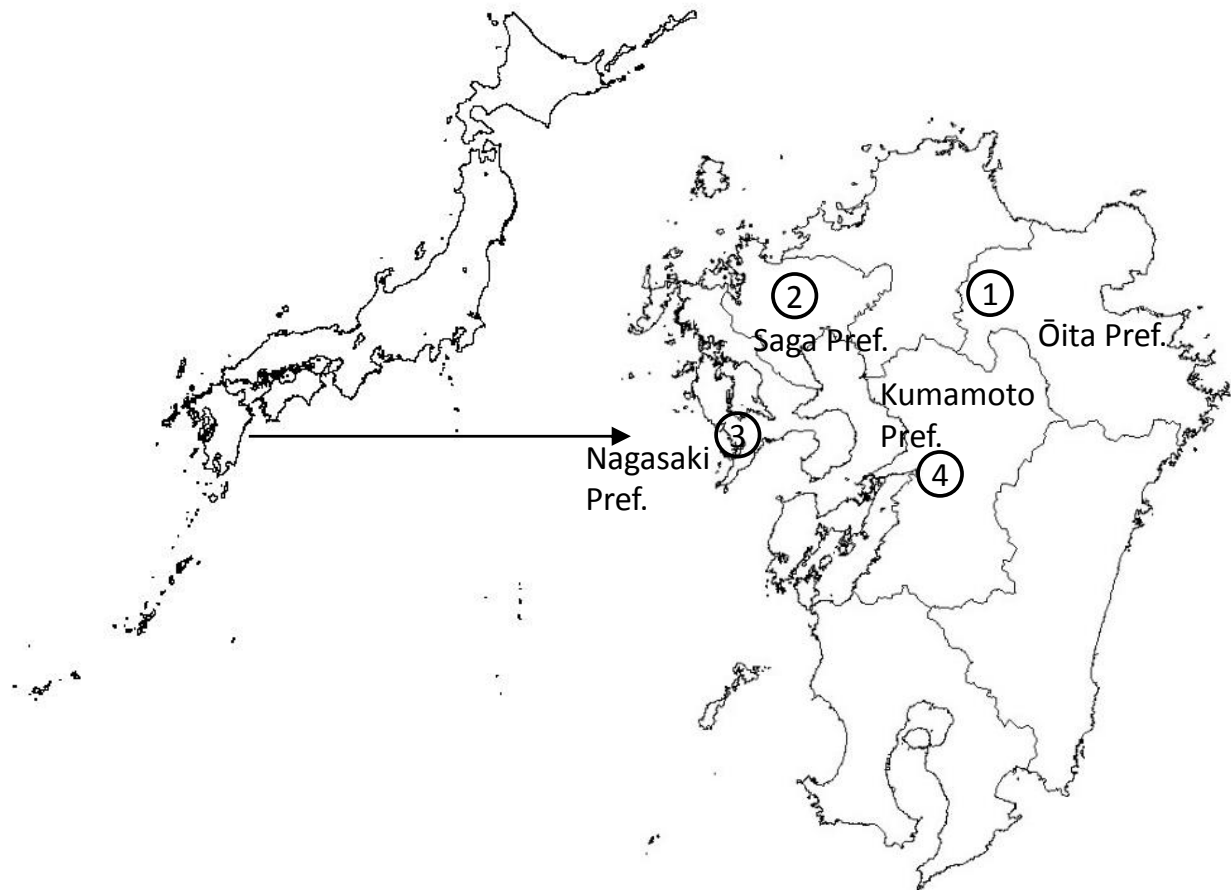


Fig. 1. Map of study sites, produced using the KenMap software (Kamada 2013).

in Nagasaki-shi.

Finally, we also aimed to collect germplasms of *Malus hupehensis* (Pamp.) Rehder, which was found in the wild in Kumamoto Prefecture in the 1920s. Although the information on its natural habitat was once missing, it was rediscovered in 1970 (Omori and Yoshizumi 1973). However, that site was destroyed during the expansion of an agricultural reservoir. We found that some trees transplanted from the natural habitat as well as their seedling offspring have been cultivated at the adjacent settlement.

Methods

In addition to the information from local floras, herbaria, and websites, we obtained detailed information on the natural habitat of *M. torino* from two specialists on the local flora of this region, Mr. M. Arakane (information on Ōita Prefecture) and Mr. H. Kamiaka (Saga Prefecture). We also obtained information on *M. hupehensis* from Mr. A. Kawakami.

Based on our preliminary research, we located two natural habitats of *M. torino*. One is situated at Fushiki, Hita-shi, Ōita Prefecture. The site has been designated as a natural monument of Hita City and is cared for by the Hita City Office and the Fushiki settlement. We obtained permission to investigate and collect plant materials from both bodies. The other natural habitat is situated at Tairano, Kyūragi, Karatsu-shi. This site is owned and cared for by the Kyūragi Branch of the Karatsu City Office and Saga Prefectural Kyūragi High School. We also obtained permission from both bodies to collect plant materials.

Malus micromalus trees planted in the precincts of the Catholic Nakamachi Church in Nagasaki-shi

were investigated and materials were collected with permission from the church.

We searched around Nishiyama settlement, which is adjacent to the former natural habitat of *M. hupehensis*, to discover cultivated trees. Two field trips were carried out from 12 to 13 October 2013 and 23 to 27 March 2014, with the aim of investigating the state of the local population. We then made a third field trip from 3 to 6 November 2014, when we collected scions and fruits for the propagation of the germplasm.

Results and Discussion

The state of the populations and collection of germplasm

Basic data on the study sites are compiled in Table 1, and their locations are mapped in Figure 1. Both natural populations of *M. toringo* grow at wet sites. The Fushiki population is at the upstream edge of an agricultural reservoir, where there is a small coastal wetland (Photos 1, 2). Although such habitats in human settlements were once common for this species, beginning in the 1960s most such agricultural reservoirs were rebuilt and these small wetlands were destroyed. The Fushiki population is perhaps an exceptional case due to the retention of the old reservoir.

The Tairano population is located in a natural wetland (Tairano-shitsugen Marsh) in a mountainous area (Photo 3; about 670 m a.s.l.). Such montane natural wetlands are very rare on Kyūshū because almost all of them have been converted to paddy fields. Therefore, this population is also an exceptional survivor. Unfortunately, we found only three wild individuals. In addition, we found two planted trees in an artificially developed but deserted land adjacent to the marsh. Apparently, tourism development of the marsh was planned about a decade ago and several seedling trees were planted, but this plan fell through.

We found four cultivated *M. micromalus* trees in the precincts of the Catholic Nakamachi Church in Nagasaki-shi (Photos 4, 5). Three trees are at the north of the church building and one is at the south. The maximum trunk diameter is only about 10 cm, indicating that these trunks are not so old. However, because we observed many root sprouts at the bases of the trees, the trunks may have regenerated several times. Though the exact origin of these trees is not recorded, they may have been cultivated for several decades.

We found several cultivated trees of *M. hupehensis* in private gardens and adjacent forest at the Nishiyama settlement in Misato-machi (Photo 6), and we received a living tree from one of the owners.

In total, we collected seven scion germplasms from two *M. toringo* wild populations and *M. micromalus* cultivated trees and a living tree of *M. hupehensis* (Table 2). In addition, we collected fruits from *M. toringo* and *M. micromalus*.

Remarks and future prospects

Malus micromalus is an important find because the existing genetic resources of this species in Japan are limited. In addition, the two surveyed populations of *M. toringo* are perhaps the last remaining wild populations on Kyūshū. They are very isolated from other populations on nearby islands. The nearest one is perhaps in the mountainous area of Yamaguchi Prefecture, about 100 km away. Therefore, genetic diversity of two populations may already be low. While trees of the Tairano population bore no or only a few fruits, trees of the Fushiki population bore many fruits (Photo 2). From these findings, we suspect that the former and latter populations are diploid with sexual reproduction and polyploid with apomictic reproduction, respectively. We will examine this issue using clonally propagated germplasms and seedlings from the collected fruits.

The last species, *M. hupehensis*, is especially interesting. Natural populations of this species were

previously discovered in Japan in only two places. Both discoveries were reported about 90 years ago, and the species has not been observed again at Nakatsu-shi in Ōita Prefecture (the site not surveyed here). Therefore, cultivated trees in Nishiyama settlement represent the only true remaining population. For a long time the examinable materials were only a few sheets of herbarium specimens, and this species was thought to be a misidentification of *M. torino* (Kitamura 1979; Ohashi 1989; Iketani and Ohashi 2001). However, the morphological characters of the remaining trees are clearly identifiable as *M. hupehensis* (data not shown). In our future research, we will compare this and other materials to investigate whether Japanese trees are genetically diversified from the continental ones, as well as the ploidy level and reproductive system.

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和文摘要

北九州地域において、自生種のズミ (*M. toringo* (Siebold) Siebold), 近世以前に中国から導入された栽培種のカイドウ (*M. micromalus* Makino), 及び野生絶滅したツクシカイドウ (*M. hupehensis* (Pamp.) Rehder) の3種のリンゴ属植物の探索を行った。ズミは、現存する2箇所の自生個体群を調査し、それぞれから2点の遺伝資源を採取した。カイドウは長崎市で栽培個体2点を採取した。ツクシカイドウは、絶滅前の野生地に近い集落で栽培個体を発見し、1点の遺伝資源を採取した。

Table 1. Basic data of the study sites

Location No.	Species	Place	Latitude	Longitude	Altitude (m)	Status	Number of Individual
1	<i>Malus toringo</i>	Fushiki, Hita-shi, Ōita Pref.	33° 22′ 37″ – 39″	131° 0′ 00″ – 02″	480	Wild on a coastal marsh of a reservoir	ca. 20
2	<i>Malus toringo</i>	Tairano-shitsugen Marsh, Kyūragi-chō Amakawa, Karatsu-shi, Saga Pref.	33° 22′ 28″ – 40″	130° 8′ 8″ – 11″	670	Wild at a riverside marsh and adjacent forest, and cultivated	5
3	<i>Malus micromalus</i>	Catholic Nakamachi Church, Nakamachi, Nagasaki-shi, Nagasaki Pref.	32° 45′ 5″ – 6″	129° 52′ 26″ – 27″	8	Cultivated	4
4	<i>Malus hupehensis</i>	Nishiyama, Misato-machi, Kumamoto Pref.	32° 39′ 14″ – 30″	130° 46′ 43″ – 59″	70–100	Cultivated	ca. 10

Table 2. List of collected germplasms

Species	Locality No. ^{a)}	sample	Designation ^{b)}	JP No.	Field No.
<i>Malus toringo</i>	1	Scion	COL/OOITA/2014/NIAS/001	252892	OiHtFk-t009
<i>Malus toringo</i>	1	Scion	COL/OOITA/2014/NIAS/002	252893	OiHtFk-t015
<i>Malus toringo</i>	2	Scion	COL/SAGA/2014/NIAS/001	252894	SgKr-t003
<i>Malus toringo</i>	2	Scion	COL/SAGA/2014/NIAS/002	252895	SgKr-t006
<i>Malus toringo</i>	2	Scion	COL/SAGA/2014/NIAS/008	252896	SgKr-t008
<i>Malus micromalus</i>	3	Scion	COL/NAGASAKI/2014/NIAS/001	252897	NgkNgk-m001
<i>Malus micromalus</i>	3	Scion	COL/NAGASAKI/2014/NIAS/002	252898	NgkNgk-m002
<i>Malus hupehensis</i>	4	Living tree	COL/KUMAMOTO/2014/NIAS/001	252899	KmMyNs-h101

a) correspond to numbers in Table 1

b) Designation using in the NIAS Genebank



Photo 1. *Malus toringo* wild population at Fushiki, Hita-shi, Ōita Prefecture (location no. 1; 12 October 2013).



Photo 2. Infructescences of *Malus toringo* (location no. 1; 12 October 2013).



Photo 3. A wild individual of *Malus toringo* at Tairano-shitsugen Marsh, Kyūragi-chō Amakawa, Karatsu-shi, Saga Prefecture (location no. 2; 26 March 2014).



Photo 4. Cultivated trees of *Malus micromalus* (location no. 3; 27 March 2014).



Photo 5. Mature fruits of *Malus micromalus* (location no. 3; 6 November 2014).



Photo 6. Branches of *Malus hupehensis* with immature flowers and the previous year's fruits (location no. 4; 23 March 2013).