

Morphological characteristics and ancestry of Japanese garlic clones – An overview

Yoshihiko Shiga, Sakio Tsutsui and Tetsuo Mikami*

Hokkaido Agricultural Laboratory for Business Development, Eniwa, 061-1405, Japan. *E-mail: t-mikami@imb.me-h.ne.jp

Abstract

Garlic (*Allium sativum* L.) is an ancient and clonally propagated crop that has been under rather continuous selection since antiquity. In Japan, each garlic-growing region had its own local cultivars (clones) at one time. This is still the case in some regions, but with expanding transport networks and the requirements for high yield of commercial quality, only a few clones dominate the garlic production in the country. Here we describe the morphological characteristics of Japanese garlic clones. The review also focusses on the possible ancestry of Japanese garlics inferred from molecular genetic analysis.

Key words: *Allium sativum* L., garlic, genetic diversity, germplasm, morphological characteristics

Introduction

Garlic is an old horticultural crop which loses its fertility during domestication, and needs to be propagated vegetatively either by cloves derived from the compound bulbs or by inflorescence bulbils (topsets) (Simon and Jenderek, 2003; Stavěliková, 2008). Despite the exclusive vegetative propagation, cultivated garlic genotypes exhibit great phenotypic diversity (Pooler and Simon, 1993; García-Lampasona *et al.*, 2003; Volk *et al.*, 2004; Panthee *et al.*, 2006). Such diversity presumably results from cross pollination between wild ancestors when this plant species still had the ability of sexual reproduction (Pooler and Simon, 1993; Panthee *et al.*, 2006). Variation may also have its origin in the accumulation of mutations throughout the history of garlic cultivation. Furthermore, it has been proposed that variation in plant performance of clonal species may be due in part to microbial infection, which is passed from generation to generation by asexual propagation (Silander, 1985).

The genetic variability found in garlic has allowed adapted types to be selected for different environments, and selection continues for new types to extend garlic culture into both colder and warmer regions. In Japan, the first reliable documentation of garlic cultivation came from the 10th century (<http://www.wakunaga.co.jp/garlic/history/japan.html>). A number of locally adapted garlic clones (we hereafter use the term 'clone' to denote garlic cultivars or genotypes that are exclusively clonally propagated) have been cultivated in this country (Etoh, 1985; Ishibashi *et al.*, 1987), yet only a few clones are at present cultivated on a commercial scale. This may lead to a decline in genetic diversity. In addition, there has been only fragmented information available on the Japanese garlic germplasm (Etoh, 1985). With such a situation in mind, we herein present an overview of Japanese garlic clones with special reference to their characteristics and ancestry.

Garlic production in Japan: The bulb is economically the most important part of the garlic plant and is mainly used for flavouring meat, fish, sauces and salads. Apart from bulbs, the young floral stems are also consumed by Japanese people. Additionally, garlic is much valued as a medicinal crop. In a 5-year average from 2009

to 2013, garlic was harvested from *ca.* 2,200 ha with a total yield of *ca.* 20,000 t annually in Japan (Table 1). The import volume of this crop is roughly equal to its home production (Table 1). Today, China is the dominant source of imported garlic in Japan. Other main suppliers of garlic to this country are the United States, Spain and Argentina. Most of the home production consists of two clones: 'Fukuchi White' and 'Shanghai Wase'.

Table 1. Production and import of garlic in Japan (MAFF Statistical Yearbook, 2014)

	2009	2010	2011	2012	2013
Cropping acreage (ha)	2,140	2,240	2,240	2,300	2,340
Total harvest (t)	19,800	19,700	20,600	20,100	20,900
Import volume (t)	19,842	18,557	19,248	19,862	18,527

is the most popular and economically important clone.

Japanese garlic clones: Garlic clones are primarily divided into three groups: complete-bolting (hard neck) garlic with a floral stem or scape; nonbolting (soft neck) garlic without a scape; and incomplete- or partial-bolting garlic with partially exerted and swollen inflorescences, bulbils within the scape, or incompletely elongated scapes (Takagi, 1990). The bolting habit and phenotypic characteristics of major Japanese garlic clones are described below. Since garlic phenotypes are known to vary according to environmental conditions, the descriptions for these clones are only relevant in the conditions in their main growing regions.

Fukuchi White: This clone stemmed from Japanese landraces of unknown origin and is most often found on supermarket shelves. It grows well in cold northern climates and is classified as an incomplete-bolting and late-maturity genotype (Table 2). The plants have generally flat and broad leaves. 'Fukuchi White' produces large, round and symmetrical bulbs. Each bulb typically contains six or so cloves (Table 2) that are normally plump with glossy white skin, and thus the clone is the most attractive looking garlic in Japan. Cloves have mild odour and flavour, and they also store well.

Shanghai Wase: 'Shanghai Wase', which is a bolting-type

Table 2. Phenotypic characteristics of two garlic clones ‘Hokkaido Zairai (Tokoro-kei)’ and ‘Fukuchi White’ (Y. Shiga and K. Nakayasu, unpublished)

Trait	Hokkaido Zairai (Tokoro-kei)	Fukuchi White
Plant height (cm)	70	63
Number of leaves	5.7	7.5
Scape length (cm)	61	5
Bulb fresh weight (g)	68	64
Number of cloves per bulb	6.7	6.3
Bulb wrapper colour	White to light yellow	White to light yellow
Clove skin colour	Pinkish purple	White

Garlic was grown in 2010/2011 at Urausu-chou (43°26'N, 141°49' E, ca. 100 m a.s.l.), Hokkaido, Japan. Fifty cloves per accession were planted in rows 120 cm apart and 15 cm distant within row. The harvest time was the end of July. Plant height, number of leaves and scape length were recorded in mid-June of harvest year.

clone introduced originally from China, is well-adapted to warm or mild southern climates. It has round bulbs, each with 12 or so comparatively small, white cloves. This genotype is also characterised by its very vigorous growth and early maturation date. Scape removal is recommended to increase bulb yield.

Isshu Wase: The clone is considered to have come originally from Korea and grows under the same climate conditions as ‘Shanghai Wase’. It normally yields a solid scape, resulting in the classification of complete-bolting type. Leaves are generally medium in width and deep green in colour. Total cloves per bulb range from 10 to 12, white or light brown cloves being usually smaller than those produced on ‘Fukuchi White’.

Okinawa Wase: ‘Okinawa Wase’ is grown in the southernmost part of Japan. This clone has practically no dormancy and is ready to harvest earlier than any of other Japanese garlics. It produces oval bulbs, each with more than 12, small pale-pink cloves. Leaves are usually narrow and pale green.

Furano: This origin-unknown clone is classified as an incomplete-bolting and late-maturity genotype. It is suitable for cold northern climates. ‘Furano’ typically has six or so reddish-brown cloves per bulb in a circular configuration around a central scape. Leaves are generally long and dark green.

Hokkaido Zairai (Tokoro-kei): A local clone ‘Hokkaido Zairai (Tokoro-kei)’, just like ‘Fukuchi White’, is better suited to cold climates. When ‘Hokkaido Zairai (Tokoro-kei)’ and ‘Fukuchi White’ were grown in the experimental field of northern Japan, the two clones were observed to be similar in their plant height, number of leaves and bulb fresh weight (Y. Shiga and K. Nakayasu, unpublished, Table 2). However, the two clones differ from each other in the extent of bolting and colour of the cloves: ‘Hokkaido Zairai (Tokoro-kei)’ is categorized as a complete bolting type and has six or so pinkish-purple cloves per bulb (Table 2, Fig. 1). The scapes usually need to be removed. This clone is also characterised by its clove traits: the cloves often have a rich, spicy flavour, and are hot and pungent when eaten raw. Recent study indicated that the content of alliin, which is one of the organosulphur compounds and confers garlic’s flavour (Block, 1985; Ovesná *et al.*, 2007; Robledo-Paz *et al.*, 2012), was 1.5 times higher in ‘Hokkaido Zairai (Tokoro-kei)’ than in ‘Fukuchi White’ (Sarengaowa *et al.*, 2011). Moreover, the thiosulphinates (a health benefit compound) content in ‘Hokkaido Zairai (Tokoro-

kei)’ was found to be 1.7 times as high as that in ‘Fukuchi White’ (Sarengaowa *et al.*, 2011).

Ancestry of Japanese garlics: The origin and cultivation history of most Japanese garlic clones are not certain. The primary centre of origin for garlic is believed to be Central Asia, from where the plant expanded in two directions, towards India and China and towards Europe and Russia (Maaß and Klaas, 1995; Lallemand *et al.*, 1997; Etoh and Simon, 2002). Molecular markers such as isozymes, RAPD (Random Amplified Polymorphic DNA), AFLP (Amplified Fragment Length Polymorphism) and SSR (Simple Sequence Repeats) have been employed to examine the genetic diversity and phylogenetic relationships among garlic germplasm accessions collected from different regions of the world. Using RAPD and AFLP analyses, several research groups were able to distinguish bolting, incomplete-bolting and nonbolting garlic accessions from one another by clustering them into separate groups (Maaß and Klaas, 1995; Al-Zahim *et al.*, 1997; Ipek *et al.*, 2003).

There are also some reports illustrating that the molecular grouping correlates with the geographical distribution of the groups. For example, mitochondrial DNA polymorphism revealed two major types of garlic clones: East Asian and European types. Japanese (*e.g.*, ‘Fukuchi White’, ‘Isshu Wase’), Chinese and Taiwanese clones formed together a well-defined type (*viz.*, East Asian type). European type was further divided into four subtypes: Russian, the former Yugoslavian, and two European sub-



Fig. 1. Phenotypes of Japanese local garlic clone ‘Hokkaido Zairai (Tokoro-kei)’. Scape development (A) and bulbs consisting of pinkish-purple cloves (B: Photo: S. Nakoshi)

types (Tsuneyoshi *et al.*, 1992). The analysis also revealed that East Asian, Russian and European type garlics were grown in Central Asia (Tsuneyoshi *et al.*, 1992). Japanese garlics were subsequently reported to be separated from Korean and Chinese garlics by an SSR-based study, though it is unclear whether 'Fukuchi White' and 'Isshu Wase' were included in the assay (Jo *et al.*, 2012).

Most recently, Yoshida *et al.* (2012) have determined the nucleotide sequences of the P1 protein gene and its flanking untranslated region of leek yellow stripe virus (LYSV) isolated from different sources of garlic germplasm accessions to understand the phylogenetic relationships between the LYSV isolates. As might have been expected, garlic clones grown in southern Japan were shown to harbor LYSVs similar to those in Chinese garlics, implying that these garlics have close ancestry. By contrast, it is interesting to note that the LYSV isolates from garlic samples (*e.g.*, 'Fukuchi White' and 'Hokkaido Zairai (Tokoro-kei)') in northern Japan were different from the Chinese isolates, and are probably not of Asian origin, but rather of European origin. The northern garlic clones are known to be phenotypically more similar to some European garlics than Chinese garlics (Yoshida *et al.*, 2012). These results collectively suggest that the current Japanese garlics have mixed ancestry, though the definite proof remains to be given.

An additional comment seems to be appropriate. The demand for high-quality garlics continues to increase. Consumers are also attracted to pigmented, unique garlic types for different culinary purposes (Simon and Jenderek, 2003; Volk and Stern, 2009). In this context, 'Hokkaido Zairai (Tokoro-kei)' is of interest because it produces pinkish-purple cloves (Fig. 1) and contains high amount of health benefit compounds. The clone can thus be considered as an important source for breeding economically valuable garlics. Finally, the number of accessions of cultivated garlics in Japan is very small, which is likely to handicap future genetic improvement. More collection of garlic germplasm accessions and the evaluation of the accessions *via* morphological, physiological and molecular characterisation will help the conservation of genetic diversity and development of new elite garlic cultivars.

Acknowledgements

We thank Mr. Makoto Nakamura (Managing Director, Hokkaido Agricultural Laboratory for Business Development) for support throughout this work. Thanks are also due to Dr. Yasuhiro Kuramoto (Wakunaga Pharmaceutical Co., Ltd.), Dr. Fumihide Inoue (Nihon Pharmaceutical Co., Ltd.), and Mr. Shin-ichi Nakoshi (Japan Agricultural Cooperatives, Tokoro) for valuable comments.

References

- Al-Zahim, M., H.J. Newbury and B.V. Ford-Lloyd, 1997. Classification of genetic variation in garlic (*Allium sativum* L.) revealed by RAPD. *HortScience*, 32: 1102-1104.
- Block, E. 1985. The chemistry of garlic and onion. *Scientific Amer.*, 252: 114-119.
- Etoh, T. 1985. Studies on the sterility in garlic, *Allium sativum* L. *Mem. Fac. Agr. Kagoshima Univ.*, 21: 77-132.
- Etoh, T. and P.W. Simon, 2002. Diversity, fertility and seed production of garlic. In: *Allium Crop Science: Recent Advances*, H. Rabinowitch and L. Currah (eds.). CABI Publishing, New York. p.101-117.
- García-Lampasona, S., L. Martínez and J.L. Burba, 2003. Genetic diversity among selected Argentinean garlic clones (*Allium sativum* L.) using AFLP (Amplified Fragment Length Polymorphism). *Euphytica*, 132: 115-119.
- Ipek, M., A. Ipek and P.W. Simon, 2003. Comparison of AFLPs, RAPD markers, and isozymes for diversity assessment of garlic and detection of putative duplicates in germplasm collections. *J. Amer. Soc. Hort. Sci.*, 128: 246-252.
- Ishibashi, Y., T. Ogawa and N. Matubara, 1987. Ecological and morphological classification of garlic cultivars. *Bul. Nagasaki Agr. For. Expt. Sta.*, 15: 95-111.
- Jo, M.H., I.K. Ham, K.T. Moe, S.W. Kwon, F.H. Lu, Y.J. Park, W.S. Kim, M.K. Won, T.I. Kim and E.M. Lee, 2012. Classification of genetic variation in garlic (*Allium sativum* L.) using SSR markers. *Austral. J. Crop Sci.*, 6: 625-631.
- Lallemand, J., C.M. Messiaen, F. Briard and T. Etoh, 1997. Delimitation of varietal groups in garlic (*Allium sativum* L.) by morphological, physiological and biochemical characters. *Acta Hort.*, 433: 123-132.
- Maaß, H.I. and M. Klaas, 1995. Intraspecific differentiation of garlic (*Allium sativum* L.) by isozyme and RAPD markers. *Theor. Appl. Genet.*, 91: 89-97.
- MAFF (Ministry of Agriculture, Forestry and Fisheries, Japan) Statistical Yearbook, 2014, <<http://www.maff.go.jp/j/tokei>>
- Ovesná, J., L. Kučera, J. Králová, L. Leišová, H. Stavěliková and J. Velišek, 2007. Genetic diversity among garlic clones as revealed by AFLP, phenotypic descriptors and S-amino acids level. *Veg. Crops Res. Res. Bul.*, 66: 105-116.
- Panthee, D.R., R.B. Kc, H.N. Regmi, P.P. Subedi, S. Bhattarai and J. Dhakal, 2006. Diversity analysis of garlic (*Allium sativum* L.) germplasms available in Nepal based on morphological characters. *Genet. Resour. Crop Evol.*, 53: 205-212.
- Pooler, M.R. and P.W. Simon, 1993. Characterization and classification of isozyme and morphological variation in a diverse collection of garlic clones. *Euphytica*, 68: 121-130.
- Robledo-Paz, A. and H.M. Tovar-Soto, 2012. Biotechnological tools for garlic propagation and improvement. In: *Innovations in Biotechnology*, E.C. Agbo (ed.). DOI: 10.5772/30636. <<http://www.intechopen.com/books/innovations-in-biotechnology/biotechnological-tools-for-garlic-propagation-and-improvement>>
- Sarengaowa, T., Yamagishi and F. Inoue, 2011. Chemical quality evaluation of the garlic. *The 58th Annual Meeting of the Japanese Society of Pharmacognosy* (Abstr. P354).
- Silander, J.A. 1985. Microevolution in clonal plants. In: *Population Biology and Evolution of Clonal Organisms*, J.B.C. Jackson, L.W. Buss and R.E. Cook (eds.). Yale University Press, New Haven. p.107-152.
- Simon, P.W. and M.M. Jenderek, 2003. Flowering, seed production, and the genesis of garlic breeding. *Plant Breeding Rev.*, 23: 211-244.
- Stavěliková, H. 2008. Morphological characteristics of garlic (*Allium sativum* L.) genetic resources collection - Information. *Hort. Sci.*, 35: 130-135.
- Takagi, H. 1990. Garlic *Allium sativum* L.. In: *Onion and Allied Crops*, Volume III: Biochemistry, Food Science, and Minor Crops, J.L. Brewster and H.D. Rabinowitch (eds.). CRC Press, Inc. Boca Raton, Florida. p.109-146.
- Tsuneyoshi, T., A.V. Nosov, Y. Kajimura, S. Sumi and T. Etoh, 1992. RFLP analysis of the mtDNA in garlic cultivars. *Jpn. J. Breeding*, 42 (Supplement 2): 164-165.
- Volk, G.M., A.D. Henk and C.M. Richards, 2004. Genetic diversity among U.S. garlic clones as detected using AFLP methods. *J. Amer. Soc. Hort. Sci.*, 129: 559-569.
- Volk, G.M. and D. Stern, 2009. Phenotypic characteristics of ten garlic cultivars grown at different North American locations. *HortScience*, 44: 1238-1247.
- Yoshida, N., H. Shimura, K. Yamashita, M. Suzuki and C. Masuta, 2012. Variability in the P1 gene helps to refine phylogenetic relationships among leek yellow stripe virus isolates from garlic. *Arch. Virol.*, 157: 147-153.