World First¹ in Practical Application of Technology

Success in Propagating a Sapling Using Tissue Culture from a "Sacred Plum Tree" in front of the Kitano Tenmangu Main Shrine

Sumitomo Forestry Co., Ltd. has conducted research and development in relation to the breeding of saplings for the purpose of protecting and preserving the plum trees in front of the Kitano Tenmangu Honden (Main Shrine) in Kyoto that are worshipped as sacred trees. On this occasion, Sumitomo Forestry succeeded in using tissue culture, a biotechnological technique, to propagate saplings that will ensure this valuable plum tree is passed down to future generations.

The "sacred plum tree" in front of the Honden (Main Shrine) is estimated to be more than 300 years old. This successful propagation from old plum trees and the research and development envisaging practical applications such as seedling production are world firsts.¹

The Kitano Tenmangu shrine was built in 947 as the grand head shrine for about 12,000 Tenmangu and Tenjinja shrines nationwide which deify Sugawara no Michizane. As a place remembered in connection with Sugawara no Michizane, who dearly loved *ume* (plums), approximately 1,500 plum trees of 50 varieties have been planted here, making it a place well known for beautiful plum blossoms. Michizane has also long been widely embraced as a god of learning.

The plum tree designated as a "sacred tree" in front of the Main Shrine is said to be the tree in the *Tobi-Ume* ("Flying Plum") legend, in which a plum tree yearned so much for Sugawara no Michizane after he had been transferred to Dazaifu (present-day Dazaifu Tenmangu) that it flew all the way to Dazaifu before the night was out. In addition to carrying on the history and culture of Kitano Tenmangu and of Tenmangu and Tenjinja shrines nationwide, this successful propagation should also contribute greatly to preserving and further promoting the Tenjin faith spanning more than 1,000 years.

Kitano Tenmangu has named the successfully propagated sapling "*Beniwa Kombai*: the Sacred Tree in the *Tobi-Ume* Legend," and plans to immortalize it for future generations.

1. Search of sites providing access to academic materials: Web of Science / Google Scholar / J Dream III

Background to propagation by tissue culture, and earlier developments

In recent years, the "plum pox virus" (PPV), which broadly infects the rose family including plums and peaches, has become prevalent in various regions around the world. In 2009, Japan's first case of PPV infection was confirmed in plum trees in Ome City, Tokyo. Plants infected by PPV sustain such damage as developing chlorosis spots or rings on their leaves, or spots on the skin of their fruit, reducing their commercial value. Given that no treatment or preventive chemicals have been found at present, PPV has resulted in more than 400,000 plum trees nationwide being felled and incinerated so far.

Emergency control measures have been implemented by both the Ministry of Agriculture, Forestry and Fisheries (MAFF) and by individual prefectures, and the Kitano Tenmangu shrine has also beefed up its preventive measures stronger than ever before. Nevertheless, in order to prepare for the unexpected and to pass down the historically valuable "sacred plum tree" to future generations, Kitano Tenmangu submitted a request for technical cooperation to Sumitomo Forestry with its successful track record in various arbor technologies concerning tissue culture and seedling production.

With regard to the tissue culture method, seedlings propagated by the shoot-tip culture method (a technique using the meristematic tissue at the tip of the bud, called the "shoot apex") are supposedly likely to be virus-free, unlikely to inherit tree diseases and so on. In addition, they stand a good chance of experiencing "rejuvenation" (blastogenesis) relative to the age of the subject trees. From the perspectives of protecting and preserving the "sacred plum tree" and handing it down to future generations, both sides confirmed that the tissue culture method was the most appropriate technique, and subsequently launched the research project in 2009.

Sumitomo Forestry's Tsukuba Research Institute, which was involved in the research and development, initially undertook research on cultivating the "sacred plum tree" based on literature and on its previous achievements in propagating cherry trees and other plants. The research, however, confirmed that *ume* (plum) trees are often crossbred with other plants, such as *sumomo* (Japanese plum) and *anzu* (apricot) trees, and that culture conditions differ for each variety. After five years of repeated experiments, the research ended in success.

• Future initiatives: Using tissue culture for DNA identification of different varieties, and protecting the sights of Kitano for future generations

Besides protecting and preserving the "sacred plum tree" of Kitano Tenmangu, it is hoped that these seedlings propagated using tissue culture will contribute significantly to maintaining the Kyoto landscape and carrying on its culture. In addition to conducting studies of plant varieties through DNA identification, the project team will also continue to carry out surveys and research for the enjoyment of all, promoting the use of tissue culture technologies to protect and preserve other plum trees at the shrine, and protecting the Kitano Tenmangu landscape for posterity.

Sumitomo Forestry also plans to begin providing advice on the propagation of historic plum trees and old plum trees nationwide based on the technologies developed in this initiative. For further details, please contact the division in charge: Forest and Landscape Research Center (http://sfc.jp/flrc/).

Past activities of the Heisei no Tobi-Ume Project

| 2009 | April | Japan's first case of PPV infection confirmed in plum trees in Ome City, |
|--------------|----------|---|
| 2009 | December | Tokyo Work begins on the project for research and development relating to the Kitano |
| 2009 | December | Tenmangu "sacred plum tree" |
| | | Survey and research policy determined, trees to be protected and preserved |
| | | selected |
| 2010 to 2011 | | Preliminary research and development begins on tissue culture technologies |
| 2012 | January | Collection of material from the Kitano Tenmangu "sacred plum tree" begins |
| 2015 | February | Success achieved in propagating sacred plum trees using tissue culture |

 Overview of propagation technology using tissue culture [workflow]

- (1) Collect winter buds, and from them use a microscope to extract only the meristematic tissue of the shoots (shoot apex).
- (2) Transfer the shoot apexes to test tubes, pour in a culture solution developed for the sacred plum tree, and cultivate to produce a large quantity of shoots (multiple shoots). <u>Photo 1</u>
- (3) Grow long shoots from the multiple shoots by cultivating the multiple shoots in a solid medium. Photo 2
- (4) Divide the large quantity of long shoots into individual shoots, and transplant them into artificial compost with an added culture solution to encourage the shoots to take root. The shoots will take root in about 4 weeks, reproducing a whole plant (seedling). The steps to this point are conducted under sterile conditions. Photo 3
- (5) Cultivate the seedlings in a greenhouse to acclimatize them to outside conditions (acclimatization process). Photo 4



Photo 1 Multiple shoots (3rd month of culture)



Photo 2 Multiple shoots (6th month of culture)



Photo 3 Seedlings taken root in artificial soil (8th month of culture)



Photo 4 Sapling growing in ordinary soil

About Sumitomo Forestry

Founded in 1691, Sumitomo Forestry Co., Ltd. and its Group companies have broadened business activities focused on wood. Based on its corporate philosophy—"utilize timber as a renewable, healthy and environmentally friendly natural resource, and contributes to a prosperous society through all types of housing-related services"— and with its approximately 250,000 hectares of owned and managed forest, the global network that spans more than 20 countries and expertise and technology in housing-related businesses, Sumitomo Forestry Group is developing the Forestry and Environment Business, the Timber and Building Materials Business, the Housing Business, the Overseas Business, the Lifestyle Service Business and other businesses both in Japan and abroad. Adding such businesses as wooden biomass power generation and Timber Solution, it will continue to pursue the potential of timber.

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