Inheritance and Softening of the Stony Hard Peach

Introduction

Based on the flesh softening process, peaches (Prunus persica Batsch) were classified into melting and non-melting types. Most of the table peaches had a melting texture. The melting-type mature fruit soften rapidly, whereupon they become prone to damage. To enhance their durability and the postharvest life span of fresh market peaches, enhancing preservability is one of the key objectives in table peach breeding. Non-melting type peaches are characterized by their slow softening rate and no notable decrease in flesh firmness, even when overripe. This texture has been also referred to as rubbery. Most peaches used for commercial canning are of the non-melting type, because their flesh can be boiled while maintaining the texture integrity. In 1976, a third type of flesh texture, stony hard, was found by Yoshida. He reported that the stony hard peach had crisp flesh and hardly softened on the tree and after harvest, but melted like melting cultivars when heated. Since non-melting and stony hard peach fruit have high preservability, these traits may be useful to significantly improve the preservability of table peaches. However, the physiological and genetic characteristics of the stony hard peach have remained unclear to date.

Relationship between Softening and Endogenous Ethylene in the Stony Hard Peach

Ethylene is a plant hormone that controls fruit ripening. The ripening of fruit can be classified as climacteric and non-climacteric types. In the former, ethylene is known to play key roles in the fruit maturation process. Indeed, the level of ethylene production has been regarded as key in determining varietal differences in the storage life of melons and apples. In the tomato for example, it has been shown that inhibiting ethylene production...
using a gene recombinant is a novel method to enhance fruit preservability. Although peaches classed as climacteric fruit, the differences in ethylene production ability among melting, non-melting and stony hard genotypes remain unclear.

Haji et al. reported that both melting and non-melting mature peaches showed increased endogenous ethylene production and softer flesh, although the degrees differed, while the stony hard peach produced little or no ethylene and remained firm after harvest. They also showed that during the growth, maturation and senescence of the fruit, the flesh of the ‘Yumyeong’ stony hard cultivar was hard and showed neither ethylene production nor postharvest softening, but the increase in fruit size, change in the fruit skin ground color increased sugar content and decreased titratable acid normally. In Japan, soft peaches are usually preferred, and traits of hard flesh and non-softening are undesirable. Since fruit that have their ethylene production and softening repressed, such as kiwi, are usually softened by ethylene treatment, Haji et al. examined the effect of exogenous ethylene on postharvest softening of the stony hard peach. They demonstrated that the fruit of the stony hard cultivar ‘Manami’, ‘Odoroki’ and ‘Yumyeong’ did not soften in an ethylene-free atmosphere, but rapidly softened with continuous ethylene treatment, showing a pattern similar to melting cultivars and became mealy.

The difference in softening between melting and non-melting cultivars is attributed to the presence of both endo- and exo-polygalacturonase (PG) in melting cultivars whereas non-melting cultivars have only exo-polygalacturonase. Non-melting is thus attributable to a mutation in the polygalacturonase gene. Conversely, stony hard is distinguished from non-melting as a mutant, the softening of which is inhibited by suppressing ethylene production in ripening fruit, and anticipated as a new genetic source to improve the preservability of the table peach.

The involvement of ethylene and 1-aminocyclopropane-1-carboxylic acid in softening of the stony hard peach

In higher plants, ethylene is biosynthesized from S-adenosyl-L-methionine (SAM) via 1-aminocyclopropane-1-carboxylic acid (ACC). The first step is catalyzed by ACC synthase and the second by ACC oxidase. The addition of ACC, which is the immediate precursor to ethylene, significantly promotes the synthesis of ethylene in tissues that otherwise produce almost none of the substance. Haji et al. investigated the effects of exogenous ethylene treatment and ACC application on ethylene synthesis and softening of the stony hard peach fruit. Ethylene treatment of ‘Yumyeong’ fruit with 1,000 ppm for three consecutive days at 25°C effectively softens them. When this treatment was interrupted, the ‘Yumyeong’ fruit stopped softening and no endogenous ethylene was produced. Treating the ‘Yumyeong’ fruit with 100 nmol/gFW ACC at 25°C stimulated it to synthesize ethylene and soften. These results indicate that in the stony hard peach, ACC oxidase activity and ethylene sensing are normal, and endogenous ethylene production is inhibited due to the inability to synthesize ACC. To clarify the suppression mechanism of ethylene production in the stony hard peaches, Tatsuki et al. examined the expression levels of ACC synthases (Pp-ACS1, Pp-ACS2, and Pp-ACS3) and ACC oxidase (Pp-ACO1) in melting and stony hard cultivars. They concluded that inhibition of endogenous ethylene production in the stony hard peach fruit was due to the suppressed expression of Pp-ACS1 mRNA. Hayama et al. studied the expression of genes encoding cell-wall modification enzymes in the stony hard peach with and without ethylene treatment, and identified three genes as related to softening in the
peach\textsuperscript{12}. These reports show that the stony hard variety is an ideal mutant for studying the relationship between fruit softening and ethylene.

Inheritance and Softening of the Stony Hard Peach

Almost all table peaches in Japan are thought to be derived from offspring or related cultivars of ‘Hakuto’ \textsuperscript{21} and this cultivar is a heterozygote of the stony hard gene\textsuperscript{22}. The stony hard gene is thus expected to be distributed among the table peach cultivars in Japan. The melting and non-melting characters are controlled by a single allele ($M/m$) and the latter is recessive\textsuperscript{3}. In 1976, Yoshida reported that the stony hard was a recessive trait described as one of melting flesh/stony hard flesh\textsuperscript{22}. However, the genetic separation of flesh texture characteristics in the offspring of non-melting and stony hard genotypes remained unclear.

Since melting and non-melting genotype can be distinguished by differences in softening at ripening\textsuperscript{8,11}, these genotypes could not be determined in the stony hard peach; the softening process of which is blocked by the absence of ethylene production\textsuperscript{9}. In 2005, Haji et al.\textsuperscript{11} demonstrated that melting and non-melting genotypes in the stony hard peach fruit could be distinguished by differences in softening following exogenous ethylene treatment. Furthermore, they examined the genetic segregation of flesh texture characteristics in $F_1$ and $F_2$ seedlings by crossing of the ‘Nishiki’ non-melting cultivar with the ‘Yumyeong’ stony hard cultivar. In this exercise, 40 plants were obtained as $F_1$ seedlings and 72 $F_2$ seedlings were provided. All fruit from the $F_1$ seedling were judged as normal, namely in which ethylene was produced from mature fruit, and melting. In the $F_2$ population, however,

**Fig. 2.** Changes in flesh firmness and ethylene production of the ‘Yumyeong’ stony hard peach cultivar treated with 1,000 ppm ethylene at 25°C for one day (24 h) and eight days
Vertical bars represent SE ($n = 3$ to 4). Reproduced from Haji, T. et al. (2003)\textsuperscript{9} with the permission of the Japanese Society for Horticultural Science.

**Fig. 3.** Changes in ethylene production and flesh firmness of the ‘Yumyeong’ stony hard peach cultivar treated with 100 nmol/gFW ACC at 25°C
Vertical bars represent SE ($n = 3$ to 4). Reproduced from Haji, T. et al. (2003)\textsuperscript{9} with the permission of the Japanese Society for Horticultural Science.

**Inheritance of melting, non-melting and stony hard texture in peaches**

Since melting and non-melting genotype can be distinguished by differences in softening at ripening\textsuperscript{8,11}, these genotypes could not be determined in the stony hard peach; the softening process of which is blocked by the absence of ethylene production\textsuperscript{9}. In 2005, Haji et al.\textsuperscript{11} demonstrated that melting and non-melting genotypes in the stony hard peach fruit could be distinguished by differences in softening following exogenous ethylene treatment. Furthermore, they examined the genetic segregation of flesh texture characteristics in $F_1$ and $F_2$ seedlings by crossing of the ‘Nishiki’ non-melting cultivar with the ‘Yumyeong’ stony hard cultivar. In this exercise, 40 plants were obtained as $F_1$ seedlings and 72 $F_2$ seedlings were provided. All fruit from the $F_1$ seedling were judged as normal, namely in which ethylene was produced from mature fruit, and melting. In the $F_2$ population, however,
40 plants were judged to be normal melting, 12 plants normal non-melting, 15 plants stony hard melting, and 4 plants stony hard non-melting respectively. The distribution of normal melting, normal non-melting, stony hard melting and stony hard non-melting in the F₂ population was not inconsistent with the ratio 9:3:3:1 (Fig. 4). These results indicated that the normal/stony hard (Hd/hd) trait was inherited independently of the melting/non-melting (M/m) trait.

**Conclusion**

Stony hard peaches have a unique texture, which is characterized by the absence of both ethylene production and softening in mature fruit (Fig. 5). The suppression of fruit softening in the stony hard peach is due to the inhibition of ethylene production. Stony hard peach fruit are subjected to ethylene-induced softening, while melting and non-melting genotypes in the stony hard peach fruit are distinguished by ethylene treatment. Normal/stony hard (Hd/hd) traits are inherited independently of melting/non-melting (M/m) traits. In conclusion, the stony hard peach is expected to be used as a genetic source for breeding new table peaches, the flesh firmness of which can be controlled by ethylene treatment.

**References**