Detection of Core Rot Symptom of Japanese Pear (*Pyrus pyrifolia* cv. Kosui) by a Nondestructive Resonant Method

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One of the main factors for the lower rating of Japanese pear (*Pyrus pyrifolia*, cv. Kosui) is contamination of core-rotten fruit in the market. An acoustic resonance method originally developed to measure firmness of fruit with the second resonant frequency was applied to the detection of core rot fruit. Second and third resonant frequencies of ‘Kosui’ Japanese pear with different degrees of core rot were nondestructively measured by a resonant method. Resonant frequency of fruit was determined by sandwiching fruit between the vibrator and detector of a resonant device. Obtained vibration voltage signals were analyzed by fast Fourier transformation. An infected pear with core rot was artificially prepared with injections of conidia suspension of *Phomopsis* sp. or *Colletotricum* sp. into the fruit on the tree. Degree of core rot was estimated by the area of the equatorial plane of cut surface. Fruit with less than 500 Hz second resonant frequency had core rot of more than 5% of fruit volume. Discrimination rate was 96.9%. Second resonant frequency more effectively discriminates infected pears with more than 5% core rot from that with less than 5% than the third resonant frequency.

Key Words: core rot, fast Fourier transformation, Japanese pear, nondestructive method, resonant frequency.

Introduction

‘Kosui’ (34.8%) has the largest production of Japanese pear in Japan (MAFF Statistic report, 2011). Contamination of core-rotten fruit of ‘Kosui’ in the market, after consumer complaints, is the main factor for the lower rating of the fruit. Core rot was found to be caused by several fungi, including *Phomopsis* sp. and *Dothiorella* sp. causing more severe rot than other fungi, including *Alternaria* sp., *Fusarium* sp., *Colletotricum* sp., and *Botrytis* sp. (Nitta and Kurihisa, 2006). The core rot symptom in harvested pears is hard to notice from its appearance, especially in the early stage, since mycelium develops initially in the pear core (seed locule) and then spreads from the core into the surrounding cortex of infected fruit. Versatile infra-red spectroscopy can detect such fruit, but this method is not widely used because of its initial high investment (Inoue et al., 2002).

The acoustic resonance vibration method was

developed to detect the firmness of fruit (Taniwaki and Sakurai, 2010). The method is based on the second resonant frequency at which fruit shows resonant vibration in mode ${}_0S_2$ (Terasaki et al., 2001a). Second resonant frequency reflects the internal physical properties of fruit flesh. For example, it relates to its elastic property (Cooke, 1972) and viscous property (Drake, 1962). Measurement of fruit elasticity or firmness by this resonant method evaluates the maturity of fruit such as melon (Taniwaki et al., 2009b, 2010), pear (Taniwaki et al., 2009c), and persimmon (Taniwaki et al., 2009a), since immature fruit generally exhibits hard flesh and overripe fruit has softer flesh; however, the possibility of detecting inner, local deterioration of fruit flesh by this method has not yet been studied. Here we tried to adopt an inexpensive resonant device to detect core rot fruit in ‘Kosui’ Japanese pear.

Materials and Methods

Inoculation with conidial suspensions in pear fruit

Japanese pear fruit (‘Kosui’) of 33-year-old trees in the orchards of the Technical Center of Agriculture of Shimane was used for experiments. Pear fruit with core rot was artificially made by the following treatments

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(Nitta, 2006):

- (1) One mL of conidial suspensions of *Phomopsis* sp. Pho. F strain, a gift from Hiroshima Prefectural Technology Research Institute, Agricultural Technology Center, with a concentration of 10^5 mL⁻¹.
 - (2) One mL of conidial suspensions of *Phomopsis* described above with a concentration of 10^7 mL⁻¹.
 - (3) One mL of conidial suspensions of *Colletotrichum* sp. 99S 198-1 strain gifted from Hiroshima Prefectural Technology Research Institute, Agricultural Technology Center with a concentration of 10^5 mL⁻¹.
 - (4) One mL of sterilized water as a control.
- Phomopsis* sp. is more virulent than *Colletotrichum* sp. (Nitta and Kurihisa, 2006).

One of the above four treatment suspensions was injected into the pear fruit core from its petal side by a syringe. The dates of inoculation were July 31 (n=34, harvested on August 5), August 14 (n=152, harvested on August 19), August 21 (n=83, harvested on August 25), and August 28 (n=91, harvested September 1). Infection periods were four to five days.

The number of fruit was 96 for (1), 100 for (2), 105 for (3) and 59 for (4) treatment. Total number of tested fruit was 360. Average and standard deviation of fruit weight (g) were 352 and 75.9, respectively. Minimum and maximum fruit weights (g) were 173 and 612, respectively.

Measurement of resonant frequency

One day after harvest, fruit weight and diameter were measured. The fruit was sandwiched between the vibrator and detector of a firmness vibration device (model NSP-1; Applied Vibro-Acoustics Inc., Hiroshima, Japan). The vibrator (25 mm in diameter) emitted vibrations with a frequency from 100 to 1000 Hz sine wave and vibrated the fruit. The PC and detector (15 mm in diameter) with a piezoelectric sensor detected the vibration of pear fruit. The vibration was converted to voltage signals by the sensor and introduced into PC. Resonant frequency was calculated by fast Fourier transformation by custommade software installed on the PC.

Elastic index (EI) was calculated using fruit mass and second (f_2) or third resonant frequency (f_3) as follows,

$$\begin{aligned} \text{Em}_2 &= m^{2/3} \times f_2^2 \\ \text{Em}_3 &= m^{2/3} \times f_3^2 \end{aligned}$$

where m is fruit mass and Em_2 and Em_3 are elastic indices calculated with fruit mass and f_2 and f_3 , respectively (Terasaki et al., 2001a, b). First resonant frequency (f_1) is related only to the limited area of fruit flesh at which the vibrator was attached. (Blahovec et al., 2006). Another elastic index (Ed_2) was calculated with diameter d and f_2 by

$$\text{Ed}_2 = d^2 \times f_2^2$$

Estimation of core rot volume

After measurement by the device, pear fruit was cut equatorially and the cut surface was photographed by digital camera. The area of core rot in the photograph was quantitatively determined by an automatic area meter (model AAC-410; Hayashi Denko, Tokyo, Japan). Ratio of the volume of infected core rot was calculated by three-second power of the ratio of the area of core rot observed in the equatorial plane of the photograph. Relationship between the ratio of the volume of infected core rot and resonant frequency or the elastic index was examined, since the resonant measurement affords the physical properties of the three-dimensional spherical body.

Results and Discussion

Figure 1 shows the degree of the area of core rot of 'Kosui' Japanese pear. The rotten part becomes dark or black. Pathogenic fungi invade the core from the bottom of the fruit without any trace of invasion. Volume of seed locule was approximately 5% for pear 'Kosui'. The percentage in the picture shows the core rot volume calculated based on the colored (brown or black) area. Fruit with less than 5% core rot may be sold without customer complaints, but fruit with brown or black flesh may not.

Figure 2 shows the relationship of elastic index Ed_2 with Em_2 for each pear fruit. It demonstrates that the correlation coefficient 0.996 was high enough to be nearly 1 and the scattering of data was distributed linearly, therefore, elastic index Ed_2 can be used instead of Em_2 . Em_2 needs each fruit to be weighed before measurement, which is not practical in the orchard.

Figure 3 shows the relationship between second resonant frequency and the ratio (%) of core rot volume to total fruit volume. Above 500 Hz second resonant frequency, the core rot volume was always less than 5% and was confined only to the seed locule region. Below 500 Hz, the ratio increased as second resonant frequency decreased, suggesting that the degree of core rot relates to the second resonant frequency. The decrease in second resonant frequency of fruit with core rot suggests that core rot causes the softening of flesh or the region of core rot became softer than the other flesh region, since there are a number of reports that the second resonant frequency or calculated elastic index decreased as fruit ripened in kiwifruit (Terasaki et al., 2001b), apple (Motomura et al., 2004; Osanai et al., 2003), pear (Murayama et al., 2006; Taniwaki et al., 2009c), melon (Kuroki et al., 2006; Taniwaki et al., 2009b), persimmon (Taniwaki et al., 2009a), and grape (Takahashi et al., 2010). There seems to be a threshold resonant frequency of around 500 Hz to segregate fruit with more than 5% core rot from fruit with less than 5% core rot.

Figure 4 shows the relationship between third resonant frequency and the ratio of core rot. The tendency of the relation is similar to that of Figure 3, but the threshold

Fig. 1. Photographs of core rot in 'Kosui' Japanese pear. Core rot was artificially made by inoculation of conidia of *Phomopsis* sp. at a concentration of 10^5 or 10^7 mL $^{-1}$. 1, intact equatorial plane of pear fruit; 2–6, those of infected fruit with core rot. The core rot volume was shown as % in parentheses. Volume of seed locule of 'Kosui' is approximately 5%.

Fig. 2. Relationship between elastic indices Ed_2 and Em_2 . Ed_2 was obtained with fruit diameter and its second resonant frequency and Em_2 with fruit mass and its second resonant frequency.

resonant frequency is more unclear. A number of fruit with core rot volume more than 5% showed third resonant frequency of more than 800 Hz. Consequently, segregation of core rot fruit by resonant frequency is more effective in Figure 3 than in Figure 4, suggesting that second resonant frequency reflects the mechanical property of the central part of fruit more than the third.

Figure 5 shows the relationship between the ratio of core rot and elastic indices (Em_2) calculated from the second resonant frequency and fruit mass. The graph is

Fig. 3. Relationship between core rot volume of pear 'Kosui' and second resonant frequency obtained by a resonant measurement. Vertical line in the graph shows 500 Hz threshold frequency.

similar to Figure 3. There is also a threshold Em_2 around 1.2×10^7 to segregate core rot fruit from intact fruit. Apparently core rot segregation by second resonant frequency is better than that by third resonant frequency, probably because smaller fruit with higher resonant frequency may be more resistant to the invasion of pathogens than larger fruit with lower resonant frequency.

Figure 6 shows the relationship between the ratio of

Fig. 4. Relationship between core rot volume of pear 'Kosui' and elastic index (Em_2) calculated with fruit mass and second resonant frequency. Vertical line in the graphs shows 1.2×10^7 threshold Em_2 .

Fig. 5. Relationship between core rot volume of pear 'Kosui' and third resonant frequency obtained by a resonant measurement.

core rot and elastic indices (Em_3) calculated from third resonant frequency and fruit mass. Scattering of the data with core rot volume of more than 5% is smaller for Figure 6 than for Figure 4. Third resonant frequency and Em_3 may not be a better index for removal of infected pear fruit than second resonant frequency or Em_2 .

Using the data shown in Figure 3, the discrimination of fruit with core rot was simulated using 500 Hz as a threshold frequency (Table 1). Second resonant frequency successfully evaluated 173 sound fruits out of 184, and 176 core rot fruits out of 176. The resultant

Fig. 6. Relationship between core rot volume of pear 'Kosui' and elastic index (Em_3) calculated with fruit mass and third resonant frequency.

Table 1. Discrimination of core rot fruit of pear by second resonant frequency.

	Below 500 Hz	Above 500 Hz	Total
Normal ^z	11	173	184
Core rot fruit	176	0	176
Total	187	173	360

^z Included fruit with rot confined only to the seed locule.

discrimination rate was 96.9% (= 349/360). The data clearly demonstrated that the criterion of 500 Hz second resonant frequency effectively indicates pear fruit with core rot. More precisely, Em_2 should be used for discrimination, but measuring the mass or diameter of fruit is not practical when working in an orchard. Table 1 confirms the use of second resonant frequency for practical segregation work in the orchard. A segregation test of pear spontaneously infected with core rot in orchards should be performed in a future study.

Core rot and mouldy core disease are widely known in apples (Serdani et al., 2002; Sørensen et al., 2009; van der Walt et al., 2010). Apple core rot is more important than its mouldy core, in that core rot symptoms penetrate the fruit flesh and lead to economic losses (Li et al., 2011). If the core rot region in infected apples is softer than the other peripheral flesh, the same resonant method can be applied to discriminate infected apples from sound apples nondestructively.

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