Research Article

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Distinction of Internal Tissue of Raw Ginseng Root Using a Computed Tomography Scanner

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Raw ginseng root of *Panax ginseng* is graded according to its shape and the quality of its internal tissue. A variety of grades are sold with prices according to grade. If an inferior raw ginseng is purchased, the consumer experience an economic loss. This research was conducted in order to explore the possibility of developing a noninvasive method for investigating raw ginseng's internal tissue. It has been determined that computed tomography (CT) scanner images agreed with actual cross-sections of raw ginseng. CT images were obtained to assess the internal portions of raw ginseng, and CT scans of raw ginseng were thoroughly measured using the Hounsfield unit (HU) system, since it allows for a more detailed analysis compared to nuclear magnetic resonance imaging. HU is a measure of attenuation used for CT images, with each pixel being assigned a value using a scale on which air is defined as -1000, water as 0 and compact bone as +1000. It takes about one second to process are slice and produce an image of the raw ginseng by a one channel CT scanner. An image good enough to discriminate the internal tissues can be obtained in 1/24 seconds with a one-channel CT scanner. Using this method, images of raw ginseng can be obtained and the characteristics of the internal tissues can be observed in a short time.

Keywords: Panax ginseng, Internal structure, Korean raw ginseng, Computed tomography scanner

INTRODUCTION

We used a computed tomography scanner in order to noninvasively inspect raw ginseng's internal tissue. A non-destructive imaging process was used to generate a three-dimensional image of the inside of raw ginseng using two-dimensional images [1].

Computed tomography (CT) scanning has begun as a useful aid in medical diagnosis and through the advancement of computer technology and equipment, its use has expanded to high-cost agricultural products, such as ginseng, for the non-destructive study and evaluation of their internal quality. Since the quality of raw ginseng plays a major role in the manufacture of high quality red

ginseng, assessing the internal tissue has become a necessity

Currently, raw ginseng is only graded based on its exterior qualities assessed by the naked eye. Thus, an evaluation method that reflects the interior quality as well as the exterior characteristics is needed. This is particularly important for raw ginseng used to make high quality red ginseng for which the grading process of the internal tissue must be noninvasive. In addition, the results of the inspection of the internal tissue of raw ginseng should be expressed numerically to facilitate an automatic grading system.

Red ginseng is graded as heaven, earth, good and so

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on, according to both its external appearance and the quality of its internal tissue. The prices of red ginseng are greatly affected by the grade determined by both external and internal factors, underscoring the necessity of a grading system that helps in the selection of fine raw ginseng. At the moment, there is no suitable evaluation method. Kim et al. [2] reported on the chemical characteristics of normal and defective red ginseng, and Lee et al. [3] assessed histological characteristics. However, these assessments are not sufficient to facilitate selection of the best raw ginseng.

Until now, the internal quality of raw ginseng has been measured based in the root's specific gravity, but air bubbles found in the rhizome of the root and its rootlets and other variables make a precise evaluation difficult. Additionally, because its measured specific gravity is an average of the whole root, this method neglects the possibility of an inside cavity [4,5]. To address this problem, two-dimensional X-ray photographs were attempted. Three problems occurred in this process. First, the concentration curve's width was changed according to the raw ginseng's thickness. Second, different from industrial products, the shapes of raw ginseng are not a uniformly regular cylinder. Third, entangled roots led to incorrect evaluations. Therefore, the two-dimensional X-ray photograph method was discarded. Nuclear magnetic resonance (NMR) imaging was tried, but it was time-consuming and required that the ginseng should be cut [6,7].

In this study, the cross-section of a raw ginseng specimen was first visually inspected. Using an optical microscope and an electron microscope, the density, presence or absence of starch, cell distribution and the structure of the internal tissue were examined. Then, CT scanner imaging was employed to determine its utility for predicting and judging raw ginseng.

MATERIALS AND METHODS

Materials

The samples used in this research were six-year-old raw ginseng harvested in Jinan, Korea in 2010. Raw ginseng was classified based on tissue characteristics. CT images were acquired for each of the classified ginseng specimens. The images and the actual ginseng cuts were also compared [1].

Methods

Computed tomography scanner

Raw ginseng with a diameter of 0 to 40 mm was mea-

sured using a MD-CT scanner system (24 channels, 600 mm, SOMATOM Sensation 4; Siemens, Forchheim, Germany) at Sun Hospital (Daejeon, Korea). Images were of the transverse cross-section and 1,000×1,000 pixels in size. The field of view was 250×250 mm, and the slice thickness was set between 1 and 2 mm (varied for specific objectives). The other parameters were also altered for specific objectives.

Scanning electron microscopy and optical microscopy

To investigate each sample, slices were cut vertically and horizontally from designated parts of the root, attached to a stage using carbon double-sided tape, gold-coated with high vacuum vapor deposition (Polaron SC502 sputter coater; West Sussex, UK), and then observed using a scanning electron microscope (SEM; DSM960A, Zeiss, Oberkochen, Germany) as previously described methods [3,8].

Preparation of samples for the SEM was quickly frozen and critical-dried using CO₂ gas. The dried samples were gold-coated for SEM and then observed. Paraffin embedding for optical microscope observation was as follows. Fresh ginseng was soaked at room temperature for 48 h in every step into 70%-, 80%-, 90%-, 95%-EtOH and finally 100%-xylene to dehydrate water in ginseng. It was soaked at 60°C for 72 h into the mixture of xylene and paraffin in the order of the ratio of 2:1, 1:1, and 1:2, and finally into 100% paraffin. Paraffinsaturated ginseng was placed in an aluminum mold to pour the liquid paraffin over it and then followed by cooling on ice. Paraffin-embedded ginseng was cut at a thickness of 10 µm using a microtome and replaced on a slide glass to melt the paraffin at 60°C for 2 h with a pyrostat. The tissue of ginseng was stained for 2 h with Harris' hematoxylin and eosin stain. A few drops of the solution of permount:xylene=1:1 were dropped on the stained sample onto a slide glass and sealed with a cover glass to solidify it at room temperature for one hour. The tissue was observed with a microscope to take pictures and count cells.

Reagent was made by dissolving 0.5g of iodine and 0.5 g of potassium iodine into a small quantity of water and then adding water to make 1 L. Five grams of fresh ginseng was placed with the reagent in a test tube and gently shaken to facilitate color development. The stained sample was washed once with water and observed on eye.

The oven volatile method was used to measure the water content.

RESULTS AND DISCUSSION

CT scanners were the first imaging devices used for detailed three-dimensional visualization of the internal anatomy of living creatures and were initially only viewed as tomographic reconstructions of slice views or sections. Since the early 1990s, with advances in computer technology and scanners using spiral CT technology, internal three-dimensional anatomy was viewable from multiple perspectives on computer monitors using three-dimensional software reconstructions. By comparison, conventional X-ray images are two-dimensional projections of the true three-dimensional anatomy, i.e., radiodensity shadows.

CT scanning was originally established by Sir Godfrey Newbold Hounsfield, one of the principal engineers and developers of computed axial tomography [1]. CT scanners present density in Hounsfield units (HUs), which range from -1,000 to 1,000. Hard bone or plaster equals +1,000 HUs, while air is -1,000 and distilled water is defined as 0. The density of raw ginseng with compact tissue measures between -200 and 100 HUs, with the core density registering the lowest. This is thought to be the result of ginseng repeating a cycle of taking from storage and re-storing it as the root grows. On Fig. 1C, the CT

density analysis of normal ginseng shows annual rings.

NMR imaging depends on moisture, which is easily influenced by temperature, and irregular moisture distribution makes it even trickier to produce an acceptable image. On the other hand, CT images depend on the density of the ginseng's neutrons and protons, rather than moisture content, which makes a quick discriminate analysis of the internal tissue possible.

As seen in the picture below (Fig. 1A), raw ginseng's two-dimensional X-ray images do not allow a specific evaluation of the internal tissue. This is especially true when a bundle of raw ginseng is held by a technician. Fig. 2 presents a view of a cross-section of raw ginseng showing its various tissues using a CT scanner and compares the cross-section with the HU values assigned to each part.

In Fig. 3, CT scanner images clearly reveal a cavity in the ginseng, as is seen in a cross-section of raw ginseng. On Fig. 3B is a comparison of normal ginseng's CT images and an actual cross-section of raw ginseng. The CT cross-section images reveal its density, which cannot be seen clearly with the naked eye. On Fig. 3A, the cavity filled with air on the CT scan is assigned a value of -1,000 HU. Compact areas without a cavity are measured between -200 and 50 HU. The cortex of raw ginseng

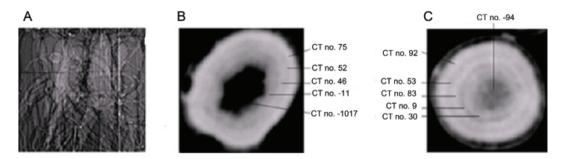


Fig. 1. Comparison between raw ginseng's two-dimensional X-ray (A), the cavitated raw ginseng's computed tomography (CT) density analysis (B), and the normal raw ginseng'CT density analysis (C).

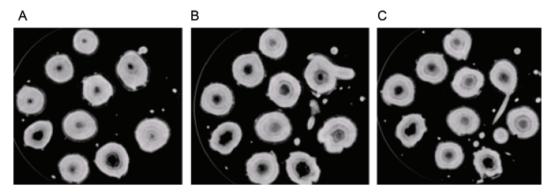


Fig. 2. Cross-sections of parts of the body of raw ginseng containing various internal tissues visualized using a computed tomography scanner. Three images of (A), (B), and (C) adjoined one another.

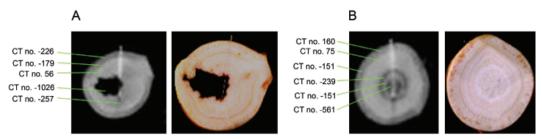
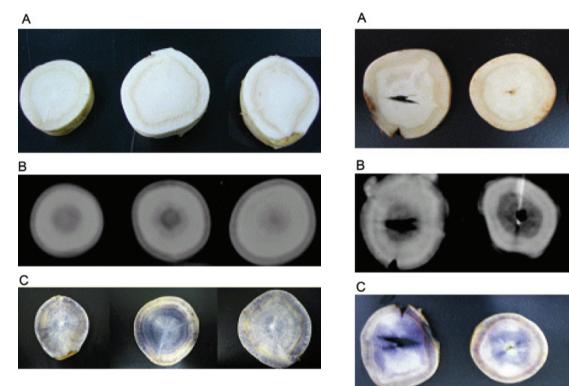


Fig. 3. Comparison between scanned images and actual cross-sections of a raw ginseng root with an internal cavity (A) and one without (B).



 $Fig.\ 4.\ (A)$ Visual inspections, (B) noninvasive computed tomography scan images, and (C) iodine-stained images of raw ginseng without an internal cavity.

with a cavity measured -179 to -233 HU, while that of raw ginseng without a cavity measured -181 to 75 HU less than the specimen with a cavity. The HU value at the center of the xylem was as low -220 HU in both cases. Cavities measured approximately -1,000 HU, making them distinguishable on the CT images.

In Figs. 4 and 5, the results of a visual inspection and a CT scan of high density ginseng without a cavity are compared. Cracks and the presence of a cavity in manufactured red ginseng are thought to be related to the starch content in the inner tissue of raw ginseng. To prove this relationship, starch in the tissue was iodine-stained and observed. However, neither the naked eye without the aid of staining nor the iodine stain was able to confirm the pattern and the staining results were erratic. From the mi-

Fig. 5. (A) Visual inspections, (B) noninvasive computed tomography images, and (C) iodine-stained images of raw ginseng with an internal cavity.

croscopic observations, it could be inferred that the tissue around a cavity should an irregular structure, while the tissue without a cavity should the opposite [9].

Inspection of ginseng with an SEM and an optical microscope revealed fissures in the xylem tissue that had not been seen in the macroscopic observations. Partial deformation of cells was also observed. A portion of raw ginseng had a relatively compact tissue that was observed in the CT images (Fig. 6). In addition, cell structures were observed for each part of raw ginseng (Fig. 6A; C, X1, and X2). Fig. 7 shows raw ginseng with relatively non-compact tissue due to a cavity, as observed with a CT scanner. Cell structures were observed in parts C, X1,

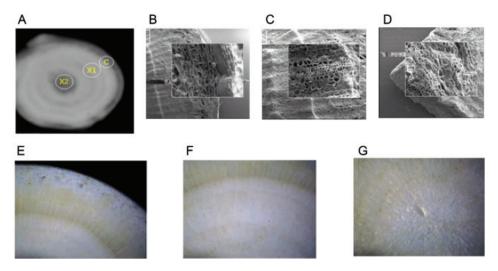
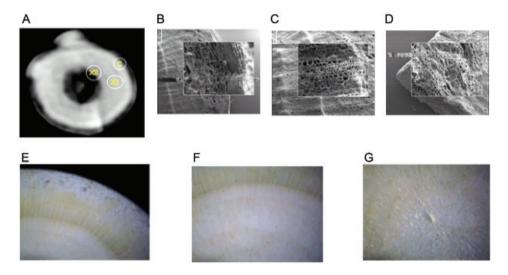


Fig. 6. Tissues taken from a raw ginseng root without a cavity visualized under an electron microscope and an optical microscope. (A) Computed tomography scanning; (B) cortex, 100 times; (C) xylem, 100 times; (D) xylem pith, 100 times (electron microscope); (E) cortex vidual, 30 times; (F) xylem, 30 times; (G) xylem pith, 30 times (electron microscope).



 ${f Fig. 7.}$ Tissues taken from the root of raw ginseng with a cavity visualized under an electron microscope and an optical microscope. (A) Computed tomography scanning; (B) cortex, 100 times; (C) xylem, 100 times; (D) xylem pith, 100 times; (E) cortex vidual, 30 times; (F) xylem, 30 times; (G) xylem pith, 30 times.

and X2 in Fig. 7A under an electron microscope and an optical microscope.

The SEM observations showed that the cell distribution was regular and dense. On the other hand, if a cavity was present in the body, the cell distribution was irregular, illustrating the presence of destroyed cells. Closer to the cavity the tissue showed more damage, irregularity in the size and shape of cells, and fewer cells were visible. The tissue with a cavity showed less regularity in its cellular arrangement compared to the tissue without a cavity (Fig. 6). Also, small irregular cavities were found inter-

mittently in the center of the tissue.

Fig. 8 shows a starch-containing cell (S) and a non-starch cell (SN). Fig. 8B shows SN magnified by 2,000 times, and Fig. 8C shows S magnified by 1,000 times, respectively. Fig. 8D shows a cell and a non-starch cell. In the tissue with a cavity, the number of stored starch particles was either low or there were none (Fig. 7). In Fig. 6, compact tissues of raw ginseng indicate a directly proportionate relationship between the assigned HU values and the amount of stored starch [10,11]. Fig. 9 presents the correlation between tissue patterns in raw

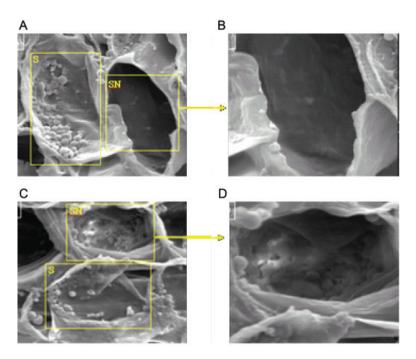


Fig. 8. Cells comprising the internal tissue of raw ginseng visualized using scanning electron microscope. (A) Cell with starch (S), 1,000 times; (B) cell without starch (SN), 2,000 times; (C) SN, 1,000 times; (D) S, 2,000 times.

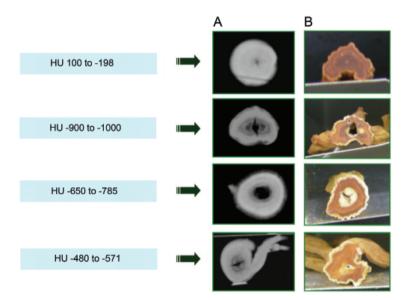


Fig. 9. Quality comparison between (A) cross-sectional computed tomography images of raw ginseng and (B) actual cross-sections of the resulting red ginseng. HU, Hounsfield unit.

ginseng and the quality of the red ginseng made from it. Raw ginseng with a density ranging from 100 to -198 HU was used to create compact red ginseng tissue. Raw ginseng with a cavity that measured -900 to -1,000 HU (similar to that of air) resulted in red ginseng with a cavity. Raw ginseng that measured -650 to -785 HU at the center resulted in an un-reddened area inside; the inside was white. Raw ginseng with a small cavity in the center

averaging -480 to -571 HUs resulted in an uncolored area and a small cavity in the red ginseng.

The raw ginseng was paraffin-embedded for observation. The internal tissues from raw ginseng were observed using an SEM or iodine staining, and then with an optical microscope. The paraffin embedding process involved dehydration, paraffin-soaking and cutting ultrathin sections using a microtome. Table 1 and Fig. 10

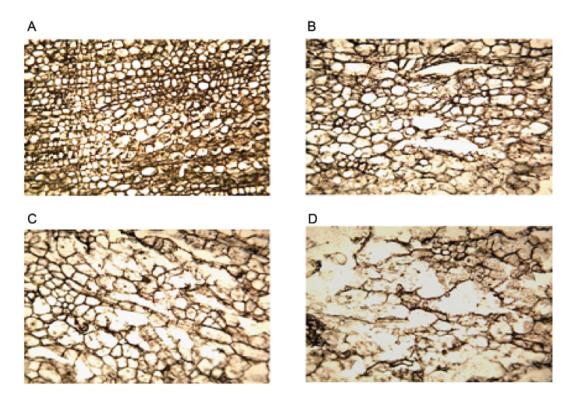
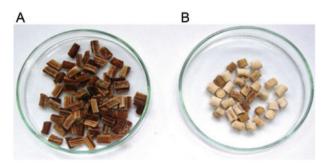


Fig. 10. Comparison between Hounsfield unit (HU) values and the number of cells in the internal tissue of raw ginseng. (A) Cortex tissue shows HU value of -17 to 45. (B) Xylem tissue shows HU value of 30 to 89. (C) Xylem tissue shows HU value of -59 to -320. (D) Xylem tissue shows HU value of -351 to -669.



 $Fig.\ 11.$ Comparison between the tissue with a Hounsfield unit (HU) value greater than 50 (A) and the tissue with a HU value less than -250 (B) in terms of color after being processed and dried.

detail the number of cells in the internal tissues. At the center of the xylem, ginseng without a cavity (-59 to -320 HUs) had 1.43 times more cells in a given area than the tissue around a cavity. The number of cells in the tissue around a cavity dropped significantly and was directly proportional to the HU value assigned to that area. Variations in the size and shape of cells as well as changes in the number of cells were seen, which was likely due to the effects of cracks and cavities that occurred during the red ginseng production process.

A piece of tissue measuring greater than 50 HU and an-

 $Table\ 1.$ Comparison between HU values and the number of cells in the raw ginseng

Tissue	Hounsfield no. (HU)	No. of cells in an equivalent square
Cortex	-17 to 45	2,115 to 2,332
Xylem	30 to 89	1,284 to 1,394
	-59 to -320	1,036
	-351 to -669	738

HU, Hounsfield unit.

other measuring less than -250 HU was cut, steamed, dried, and then compared for color differences (Fig. 11). The sample that originally measured 50 HU matched the color and histological characteristics of normal red ginseng, while the sample that measured less than -250 HU exhibited a white area and lacked normal red ginseng characteristics.

The specific gravities, dehydrated specific gravities and water contents of the two tissues samples were compared in Table 2. The specific gravity and dehydrated specific gravity of the tissue greater than 50 HU were both greater than those of the less than -250 HU sample. However, unexpectedly, the moisture content was higher in the loose tissue.

Table 2. Comparison of specific gravities and water content from two raw ginseng samples measured before and after being dried

Variables	Greater than 50 HU	Less than -250 HU
Specific gravity of the raw ginseng	1.011	0.777
Specific gravity of the raw ginseng after dried	0.2604	0.181
Water content (%)	74.35	78.26

HU, Hounsfield unit.

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