Short Communication

Assessing the supercooling of fresh-cut onions at −5°C using electrical impedance analysis

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Received 6 November 2019; Revised 18 November 2019; Editorial decision 9 December 2019.

Abstract

We supercooled fresh-cut onion at −5°C for 12 h. After supercooling, the electric impedance properties of the samples were evaluated by electrical impedance spectroscopy over the frequency range of 42 Hz − 5 MHz. The time-temperature profiles of samples indicated that the freezing point and supercooling point were −2.3°C ± 0.7°C and −6.9°C ± 1.0°C, respectively. The results indicated that 34 of the 36 supercooled samples exhibited a definite circular arc in the Cole-Cole plot, which suggested that the cell membrane remained intact during supercooling. In the other two samples which did not exhibit a definite circular arc, the cell membrane had sustained serious damage during supercooling. Furthermore, there was large difference in drip loss percentage between supercooled samples exhibited a definite circular arc in the Cole-Cole plot and samples not exhibiting a definite circular arc. Our results suggest that fresh-cut onions can be supercooled at −5°C.

Key words: supercooling; fresh-cut onion; electrical impedance; Cole-Cole plot; supercooling point.

Introduction

The use of low temperature is a common preservation method worldwide for maintaining the freshness of vegetables and fruit because low temperatures reduce the microbial growth, biochemical reactions, respiration, and transpiration of these products. The best temperature for slowing down deterioration is the lowest temperature that can safely be maintained during preservation without freezing the products. Recently, new research into food refrigeration and storage technologies has proposed alternative methods for extending the shelf life of fresh foods by using supercooling (Stonehouse and Evans, 2015), although a supercooled food product is unstable because spontaneous nucleation can occur at any moment (Cox and Moore, 1997).

Previous studies have reported that some vegetables, such as garlic, shallots, and peppers, broccoli, and cauliflower which have been cooled at a slow rate can be stored in a supercooled state at temperatures significantly below their freezing points without freezing occurring (James et al., 2009; James et al., 2011). However, few studies are available on the application of supercooling to fresh-cut vegetables, except for experimental data on fresh-cut cabbage (Koide et al., 2017), leaf lettuce (Quang et al., 2017), and fresh-cut spinach (Koide et al., 2019). In general, fresh-cut vegetables are highly susceptible to microbial spoilage, because of the slicing and peeling operations during their processing (Koseki and Itoh, 2001). Thus, if the supercooling procedure could be applied to fresh-cut vegetables, it would reduce the rate of degradation from microbial sources (spoilage and pathogenic), thereby increasing their shelf life in the food cold chain.

The present study investigated fresh-cut onions, a commonly used cut vegetable, for supercooling tests at a sub-zero temperature. Since our preliminary tests showed that most fresh-cut onions preserved at −5°C exhibited little drip loss and little colour deterioration but significant drip losses and colour deterioration at around a temperature of −7°C after 12 h, we supercooled the fresh-cut onion at −5°C for 12 h. The aim of this study is to measure the freezing point and supercooling point of the sample, and to investigate the supercooled sample using electrical impedance spectroscopy (EIS) to understand the status of the cell membrane of the fresh-cut onions. In addition,
the drip loss percentage of supercooled sample was investigated. To our knowledge, this is the first study to assess the supercooling of fresh-cut vegetables using bulbs vegetables.

Materials and Methods

Materials

Onion bulbs (Allium cepa) were purchased from August to October 2018, at a local supermarket in Morioka, Japan, then stored at room temperature in a dark incubator before the experiment. The experiments on the onion bulbs were performed within a period of 7 days. The onion bulbs were cut longitudinally into 12 equal parts, then the outer and second leaves were removed. The third and fourth leaves were separated by hand then weighed immediately and used as the sample for the supercooling tests. In the present study, samples just cut from the onion bulbs were used as the control. Samples stored in a deep freezer at −80°C for 24 h, followed by thawing at room temperature (dead sample; DS) were used as negative controls. The average mass of the sample was 2.90 ± 0.08 g with an average moisture content of 93.7% ± 0.1% (wet basis), determined using the oven drying method (105°C for 24 h).

Supercooling test

Each sample was placed in a 50-ml test tube (2344-050; Iwaki, Sekiyarika Co. Ltd, Tokyo, Japan) then covered with a tube cap for the supercooling test. A refrigerated liquid circulating bath (NCB-3300; Eyela, Tokyo Rikakikai Co. Ltd, Tokyo, Japan) was used for this test. The measurements were made while the test tubes were submerged in ethanol at −5°C. A T-type thermocouple positioned at the geometric centre of the sample was used to measure the temperature. The samples were then stored at −5°C for 12 h. After supercooling, the samples were warmed to 10°C for 12 h then brought to room temperature at 25°C. The samples, except for those used for temperature measurement using a thermocouple, were measured using EIS. The experiments were repeated four times.

Supercooling point and freezing point

The time-temperature profiles of the samples (n = 9) were also measured to determine the freezing point of the fresh samples. This experiment was performed in the same way as the supercooling test but with the following modification. A sample of tissue with a T-type thermocouple positioned at its geometric centre was placed in a 50-ml test tube then cooled to −5°C in the refrigerated liquid circulating bath. When the sample temperature reached −5°C, it was cooled further to −15°C at approximately 0.2°C/min by reducing the temperature of the ethanol in the refrigerated circulating bath. The temperature when the low temperature exothermic occurred in the time-temperature profile was taken as the supercooling point. The freezing point was indicated by the plateau in the time-temperature profiles where the measured temperature is almost constant after rising from that for the supercooled state (Meng et al., 2007).

Electrical impedance measurement

The electrical impedance data from the supercooled, control, and DS samples were measured by EIS as described by Wu et al. (2008). The impedance of each sample was measured using two parallel electrodes spaced 10 mm apart (3532-50, Hioki Corp. E.E., Ueda, Japan). The impedance magnitude, |Z|, and phase angle, θ, of the samples were measured at 50 frequency points (logarithmic frequency intervals) over the frequency range of 42 Hz–5 MHz under a measuring voltage of 1.0 V, and automatically recorded by a computer for analysis. The resistance, R (Ω), and reactance, X (Ω), were calculated using the following equations:

\[ R = |Z| \cos \theta \]

\[ X = |Z| \sin \theta \]

Statistical analysis

The quantitative data are presented as means ± SD.

Results and Discussion

Temperature profile of sample during the supercooling tests

A representative temperature profile for a fresh sample used in the supercooling test is shown in Figure 1. The sample was cooled at 0.5°C/min from 0°C to −2°C. In these measurements, no samples with a thermocouple positioned in the tissue exhibited an exotherm in their temperature profiles under supercooling, thereby indicating that the samples were in a supercooled state.

Freezing point and supercooling point

A representative time-temperature profile for a fresh-cut onion sample for measuring the freezing point and supercooling point is shown in Figure 2. The freezing point of the fresh samples determined from the time-temperature profiles averaged −2.3°C ± 0.7°C, similar to those reported for other vegetables: broccoli (−2.1°C ± 0.3°C), carrots (−1.6°C ± 0.6°C), and shallots (−1.6°C ± 0.2°C) (James et al., 2011), fresh-cut swede (−2.65°C ± 0.35°C) and fresh-cut turnip (−1.97°C ± 0.51°C) (Helland et al., 2016). Next, the supercooling points were obtained in the range from −5.3°C to −8.2°C, with a mean value of −6.9°C ± 1.0°C, which was lower than those reported for other vegetables: broccoli (−4.4°C ± 2.4°C), carrots (−2.7°C ± 0.8°C), and shallots (−5.4°C ± 1.4°C) (James et al., 2011), but similar to those of fresh-cut swede (−6.26°C ± 2.45°C), and fresh-cut turnip (−6.37°C ± 1.59°C) (Helland et al., 2016). The present results demonstrate that fresh-cut onion can be preserved at a temperature of −5°C. However, more information on the effects
of speed of cooling, sample size, preservation time in relation to ice formation are needed for future investigations.

Electrical impedance

EIS is often used to analyse the physical state of cell membranes in biological tissues (Zhang and Willison, 1992; Wu et al., 2008; Ando et al., 2016; Watanabe et al., 2016). The structural properties of the cell appear in the range of frequencies from approximately 100 Hz to 10 MHz (Ando et al., 2014). The impedance of the fresh-cut onion as the control, the supercooled sample, and DS as the negative control were plotted with respect to frequency (Figure 3). Focusing on the control, there was a clear dependence of the impedance on frequency: the impedance decreased as the frequency increased, especially between 1 and 100 kHz. This dependence of impedance on frequency showed that the cell tissue structure was composed of an aggregation of closed cell spaces separated by cell membrane (Ohnishi et al., 2003). While the impedance of DS at a lower frequency was very low compared with the control, it was almost independent of the frequency between 1 kHz and 5 MHz, showing that the cell membrane had been so seriously damaged that it could no longer retain the cell contents (Wu et al., 2008). The Cole-Cole plots for the control, supercooled, and DS samples are shown in Figure 4. The right side of the circular arc describes a low frequency area, and the left side a high frequency area. It has been reported that the Cole-Cole plot for fresh vegetables describes a circular arc (Wu et al., 2008; Watanabe et al., 2016), but no arcs were produced for the frozen-thawed samples. Similar results have been reported for several frozen-thawed vegetables: carrots (Ohnishi et al., 2003), eggplant (Wu et al., 2008), potato (Ohnishi et al., 2003; Ando 2014), fresh-cut cabbage (Koide et al., 2017), and fresh-cut spinach (Koide et al., 2019).

Regarding the supercooled samples, the impedance of 34 of the 36 samples (SC-1: supercooled samples with a high impedance at 1 kHz, the same as that for the fresh sample) depended on frequency (Figure 3), and there were definite circular arcs in the Cole-Cole plots (Figure 4), suggesting that the cell membranes had remained intact after supercooling. However, 2 of the 36 samples (SC-2: supercooled samples with a low impedance at 1 kHz) had a very low impedance similar to that for DS (Figure 3), and there were no definite circular arcs in the Cole-Cole plots (Figure 4), almost the same trend as for DS.

The average values of drip losses, expressed as the loss of weight of the sample after supercooling as a percentage of the initial weight, were 2.0% for SC-1 and 13.6% for SC-2. While, the average drip loss percentage of DS was 18.6%. The large difference in drip loss percentage between SC-1 and SC-2 may be due to the state of cell membrane.
membranes before and after supercooling. Taking the results of EIS, it can be suggested that the physiological structure of the cell membranes of SC-2 had been disrupted by the ice crystals formed during the supercooled state of the test sample.

The temperature profile of the sample during supercooling and the evaluation of the supercooled sample by EIS indicated that fresh-cut onions could be preserved by supercooling at −5°C. This would contribute to reducing the amount of product wastage and the negative indices of marketability such as drip loss, visual alterations, and colour deterioration (Ohnishi et al., 2003; Stonehouse and Evans, 2015; Jha et al., 2019). Further investigations into the supercooling preservation of fresh-cut vegetables regarding temperatures and durations of treatment to achieve stable supercooling conditions are underway.

Conclusions

In this study, fresh-cut onions were preserved by supercooling at −5°C for 12 h, with the following conclusions:

1. The time-temperature profiles of the samples indicated that the freezing and supercooling point were −2.3°C ± 0.7°C and −6.9°C ± 1.0°C, respectively.
2. The dependence of impedance on frequency leading to the Cole-Cole plots would be an effective method for assessing the supercooling of fresh-cut onions.
3. Electrical impedance spectroscopy (EIS) indicated that the cell membranes of most of the supercooled samples remained intact.

Our study suggests that fresh-cut onions can be supercooled at −5°C.

Funding

This work was supported by JSPS KAKENHI, grant number JP16H05001 [Grant-in-Aid for Scientific Research (B)] and JP16K15010 [Grant-in-Aid for Exploratory Research].

Author contributions

SK coordinates the study, AY contributed to the measurement and analysis of all data, TU contributed to the discussion of the electrical impedance analysis, and MU contributed to the discussion of supercooling state from viewpoints of cryobiology.

Conflict of Interest Statement

None declared.

References

Ando, Y., Maeda, Y., Mizutani, K., Hagiwara, S. W., Nabetani, H. (2016). Impact of blanching and freeze-thaw pretreatment on drying rate of carrot roots in relation to changes in cell membrane function and cell wall structure. IWT - Food Science and Technology, 71: 40–46.
Ando, Y., Mizutani, K., Wakatsuki, N. (2014). Electrical impedance analysis of potato tissues during drying. Journal of Food Engineering, 121: 24–31.
Cox, D., Moore, S. (1997). A Process for Supercooling. Patent Application No. WO97/18879. World Intellectual Property Organisation.
Helland, H. S., Leufvén, A., Bengtsson, G. B., Pettersen, M. K., Lea, P., Wold, A. B. (2016). Storage of fresh-cut swede and turnips: effect of temperature, including sub-zero temperature, and packaging material on sensory attributes, sugars and glucosinolates. Postharvest Biology and Technology, 111: 370–379.
James, C., Hansen, P., James, S. J. (2011). Super-cooling phenomena in fruits, vegetables and seafoods. In: Proceedings of 11th International Congress on Engineering and Food (ICEF 2011), Athens, Greece, pp. 22–26.
James, C., Seignemartin, V., James, S. J. (2009). The freezing and supercooling of garlic (Allium sativum L.). International Journal of Refrigeration, 32: 253–260.
Jha, P. K., Xanthakis, E., Chevallier, S., Jury, V., Le-Bail, A. (2019). Assessment of freeze damage in fruits and vegetables. Food Research International (Ottawa, Ont.), 121: 479–496.
Koide, S., Kumada, R., Hayakawa, K., Kawakami, I., Orikasa, T., Katabira, M., Uemura, M. (2017). Survival of cut cabbage subjected to subzero temperatures. In: Proceedings of VI Postharvest Unlimited: ISH International Conference, 17–20 October 2017, Madrid, Spain. http://postharvest2017.sicongresos.com/posters/23.jpg. Accessed 6 November 2019.
Koide, S., Obusga, R., Orikasa, T., Uemura, M. (2019). Evaluation of electrical and physiological properties of supercooled fresh cut spinach. Journal of the Japanese Society for Food Science and Technology - Nippon Shokuhin Kagaku Kogaku Kaishi, 66: 335–340. (In Japanese with English abstract).
Koseki, S., Itoh, K. (2001). Prediction of macrobiol growth in fresh-cut vegetables treated with acidic electrolyzed water during storage under various temperature conditions. Journal of Food Protection, 64: 1935–1942.
Meng, Q. R., Liang, Y. Q., Wang, W. F., Du, S. H., Li, Y. H., Yang, J. M. (2007). Study on supercooling point and freezing point in floral organs of apricot. Agricultural Sciences in China, 6: 1330–1335.
Ohnishi, S., Fujii, T., Miyawaki, O. (2003). Freezing injury and rheological properties of agricultural products. Food Science and Technology Research, 9: 367–371.
Quang, T. N., Iwamura, K., Shrestha, R., Sugimura, N. (2017). A study on supercooled storage of leaf lettuces produced in plant factory. Japan Journal of Food Engineering, 18: 25–32.
Stonehouse, G. G., Evans, J. A. (2015). The use of supercooling for fresh foods: a review. Journal of Food Engineering, 148: 74–79.
Watanabe, T., Orikasa, T., Shono, H., Koide, S., Ando, Y., Shina, T., Tagawa, A. (2016). The influence of inhibit avoid water defect responses by heat pretreatment on hot air drying rate of spinach. Journal of Food Engineering, 168: 113–118.
Wu, L., Ogawa, Y., Tagawa, A. (2008). Electrical impedance spectroscopy analysis of eggplant pulp and effects of drying and freezing-thawing treatments on its impedance characteristics. Journal of Food Engineering, 87: 274–280.
Zhang, M. I. N., Willson, J. H. M. (1992). Electrical impedance analysis in plant tissues: the effect of freeze-thaw injury on the electrical properties of potato. Canadian Journal of Plant Science, 72: 345–353.