# ナノミストを用いた青果物の品質保持法

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# APPLICATION OF NANOMIST FOR PRESERVING POSTHARVEST QUALITY OF FRESH PRODUCE

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2012



# APPLICATION OF NANOMIST FOR PRESERVING POSTHARVEST QUALITY OF FRESH PRODUCE

By

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A thesis submitted in fulfillment of the requirements for the degree of

## DOCTOR OF PHILOSOPHY

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#### **CHAPTER 1**

#### Introduction

#### **1.1. BACKGORUND OF THE RESEARCH**

Fresh fruit and vegetables, often termed as fresh produce, are considered as commercially important and nutritionally necessary food commodities because of the fact that they provide not only the major dietary source of vitamins, sugars, organic acids and minerals, but also other phytochemicals including dietary fiber and antioxidants with healthbeneficial effects. What is more, fruit and vegetables provide a wide range of features in color, shapes, taste, aroma and texture to refine sensory pleasure in human's diet. There is an increasing demand for fresh produce than processed products due to the raising awareness of people about its health benefit. Fresh produce, unfortunately, are highly perishable in nature and may be unacceptable for consuming if no proper handlings are applied after harvest. In recent years, fresh horticultural products are important items of international commerce following the globalization of trade and free trade agreements. Longer storage, shipments and distribution periods may eventually increase the potential of heavy losses; therefore, the importance of proper cares and techniques for handling fresh fruit and vegetables after harvest has been recognized and emphasized.

All of fresh fruit and vegetables are living tissues consisting of 80-96% water. Due to high moisture content, rich in nutrients and tender nature, they are vulnerable to dehydration, environmental stresses, mechanical injury and pathological breakdown. These characteristics strongly shorten the shelf life of fruit and vegetables and result in

deterioration after harvest. Postharvest losses can occur at any point in the production and marketing change. It is estimated that the magnitude of these losses due to inadequate postharvest handling, storage and transportation in fresh produce is relatively high, between 20-50% in developing countries when compared to 5-25% in developed countries (Kader, 2005). In order to reduce these losses, understanding the causes of deterioration in fresh produce is the fundamental step and followed by using appropriate technology to control weight losses, prevent disease growth and maintain quality of produce.

Temperature is the most effective tool for maintaining the quality and extending the shelf life of fresh produce after harvest, since temperature affects the rate of most biochemical, physiological and microbiological reactions contributing postharvest deterioration (Paull, 1999; Will et al., 2007). Thus, storage at low temperature has been the significant strategy to preserve the harvested horticultural products. In addition, as far as water loss is concerned, a storage environment with high relative humidity (RH) plays an important role in maintaining the quality of produce. Recommended RH level for the storage of fresh fruit and vegetables is commodity specific; levels are generally in the range of 85-95% (Paull, 1999; Rennie et al., 2003; Maguire et al., 2004). However, one of the problems occurring for fresh produce and its package container (corrugated cardboard box) stored under high humidity environment is that there is a risk of wetness on surface of the produce causing developments of postharvest pathogens (Grierson and Wardowski, 1978) and that corrugated cardboards reduce its strength because the boxes become wet during storage. The reduction in strength of cardboards can lead to packaging collapse, thereby causing further mechanical damage to products (Marcondes, 1992).

Nanotechnology is recognised as a revolutionary manufacturing technology of the 21th century involving multidiscipline research issues that rely on the understanding and control of substances at the nanoscale size. This technology encompasses research and development of materials, devices and systems that exhibit novel properties and functions due to their nanoscale dimensions or components. The application of nanotechnology in food and agricultural products, especially for preservation of fruit and vegetables is highly recommended. A nanomist humidifier, a state-of-the-art humidification device was used to generate high humidity with the aim of maintaining the freshness of fresh produce stored in a refrigerated chamber. It is believed that due to ultra fine mist, nanomists could produce some benefits for the field of postharvest. So far there has been no study conducted on the characteristics of nanomist and its advantages on the storage of fresh fruit and vegetables.

#### **1.2. THE AIMS OF THE STUDY**

This research work was undertaken to (a) investigate the particle size distribution of mists produced by humidifiers operating at high RH storage environment, (b) to explore the role of nanomist on the maintenance of strength of corrugated cardboard box under high humidity condition, (c) to discover the effect of nanomist on controlling weight loss of fresh fruit and vegetables, and (d) to find out the postharvest characteristics of fruit following storage under nanomist environment.

#### **1.3 ORGANIZATION OF THE THESIS**

This thesis is organized into six chapters. Chapter 1 introduces the background of the present research work. Chapter 2 looks into the particle size distribution of nanomist and ultrasonic mist under high relative humidity environment. Chapter 3 examines the strength

of corrugated cardboard boxes exposed to nanomist and ultrasonic mist during high RH storage. In chapter 4, controlling the weight loss of fresh produce during postharvest storage under nanomist and ultrasonic mist is investigated. The postharvest characteristics of fruit conditioned with nanomist following storage are also examined in chapter 5. All findings achieved from this study are concluded in chapter 6.

#### **CHAPTER 2**

## Investigation of particle size distributions produced by humidifiers operating in high humidity storage environments

#### **2.1. INTRODUCTION**

Nanotechnology is well known as a revolutionary engineering involving multidiscipline research issues that rely on the understanding and control of substances at the nanoscale size. This technology is not only limited to working with matter at nanoscale, but also encompasses research and development of materials, devices and systems that exhibit novel properties and functions due to their nanoscale dimensions or components. While a variety of definitions of the term "nanoparticles" have been suggested, this study will use the definition suggested by Nickols-Richardson (2007) and Hosokawa et al. (2007) who defined the nanoparticles are generally ultrafine particles whose sizes are in the range from 1 nm to 100 nm. To date, most research into nanotechnology has focused on uses in electronics, medicine and automation (Sozer and Kokini, 2009). The knowledge gained from these sectors could be adapted for the use of nanomists in food and agricultural products in general but for the preservation of fruit and vegetables in particular. A nanomist humidifier (Mayekawa Co., Ltd, Tokyo, Japan), a state-of-the-art humidification device, was used here to generate high humidity with the aim of maintaining the freshness of fresh produce stored in a refrigerated chamber. The mists were generated at nanoscale size, producing a so-called nanomist. The mists were thought to be significantly smaller than those generated by traditional humidifier which is often termed ultrasonic humidifier. It was assumed that nanomists, because of their very fine particle size, would evaporate immediately following atomisation. Therefore, it was also assumed that nanomists could bring a number of benefits to the field of postharvest crop storage. It is assumed that since nanomist humidifiers could provide high humidification without wetting produce, the freshness of produce for a long-term storage could be enhanced. Moreover, it is assumed that when the mists come into contact with the stored produce, and its containers such as corrugated cardboard boxes, wetness does not occur on the surface of those, thereby controlling the deterioration of fresh produce and microorganism growth as well as maintaining the strength of containers during a long storage period.

Particle size distribution is probably the most important physical characteristic of mists. This property influences the evaporation and condensation process of water. However, to date there has been no studies which measure the particle size distribution of nanomist humidifiers under cold storage environments. Therefore, the aim of this chapter was to investigate the characteristics of mist discharged from two types of humidifiers under different storage environments, focusing on those with high relative humidity. In addition, mathematical expressions were used to determine suitable distribution and density functions for describing the experimental data.

#### 2.2. MATERIALS AND METHODS

#### 2.2.1. Operation of the nanomist humidifier

A schematic diagram of the nanomist humidifier used in the experiments is shown in Fig. 2.1. Water is drawn from the water chamber by the centrifugal force of the motor using a screw; subsequently water droplets are generated by the rotating body. Coarse droplets will sediment back into the water chamber and fine drops are dispersed into the mist chamber



Fig. 2.1 Schematic diagram of nanomist humidifier.

where ultrafine mists are dispersed into the external environment by the fan through a specialised filter. The height of the centre of the outlet was 1.7 m.

#### 2.2.2. Experimental setup

The measurements were performed under two environmental conditions, at an ambient air temperature of 22 °C and 70% relative humidity and under a controlled temperature and relative humidity environment. Under ambient conditions, the measurements of particle size were performed at the outlets of humidifiers, at distances of 500 mm and 1000 mm along a straight line extending from the outlet. For controlled environment measurements, particle size was measured under three cold storage environments; one without humidifier, one using the nanomist humidifier and the other with an ultrasonic humidifier. When a humidifier was installed the temperature in the chamber was set to 5 °C and the relative humidity was set to 80% and 90%. Data were recorded at ten-minute intervals throughout the experiment in the container using a humidity and temperature transmitter (model Under controlled environment conditions. HMT337: Vaisala. Helsinki, Finland). measurements were conducted after turning on the humidifiers and refrigerator at time intervals of 2 and 4 h. The measuring position was at the centre point of the container at 1 m above the ground. In order to investigate the difference in particle size distribution with height, measurements were carried out at 700 mm and 1730 mm above the ground after the device had been operating for 18 h. To understand the relationship between particle number concentration and humidifier frequency, the nanomist humidifier was allowed to operate at 40, 50 and 60 Hz. The aerosol sample flow was drawn via a conductive tube 4.5 mm internal diameter and 2100 mm in length. Air velocity was measured at the outlets of the humidifiers using a thermal anemometer (Model ISA-15S, SIBATA Scientific Technology Ltd., Tokyo, Japan).

#### 2.2.3. Instrumentation

Measurements were undertaken inside a container (3060 mm in length, 2130 mm in height and 2320 mm in width) equipped with a nanomist humidifier, cooling system controller and system control panel (Mayekawa Co., Ltd, Tokyo, Japan), and an ultrasonic humidifier (FT -30N-14, UCAN Co., Ltd, Tokyo, Japan).

For particle sizes ranging from 15.1 to 661.2 nm, a scanning mobility particle sizer (SMPS) (SMPS model 3936, TSI Inc, Dylec Corp. Tokyo, Japan) was used to measure particle number concentration and size distribution by separating particles based on their electrical mobility (Fig 2.2). Details of SMPS measurement system were described in previous studies by Choi et al. (2005) and Stipe (2004). The particles of a selected size are detected optically, using a detection technology through which the visibility of fine particles is enhanced by "growing" the particles in condensing butyl alcohol vapour. The device used for particle separation is referred to as a differential mobility size analyser (DMA) (DMA 3081, Dylec Corp. Tokyo, Japan). The particle counter is referred to as a condensation particle counter (CPC) (CPC model 3775, Dylec Corp. Tokyo, Japan). The aerosol sample flow rate was 0.3 1 min<sup>-1</sup>, while the sheath airflow was set to 3.0 1 min<sup>-1</sup>, which allowed the measurement of particles as fine as 15.1 nm diameter. The use of sheath airflow also minimises the diffusion losses of ultrafine particle during sampling. Aerosol instrument manager (AIM) software (version 4.0, TSI Inc, Dylec Corp. Tokyo, Japan) was used for data reduction and analysis of the SMPS output. For each measurement, three SMSP samples were taken, with a scanning time of 100 s.





Fig. 2.2 Measurement systems of scanning mobility particle sizer (SMPS) and light scattering aerosol spectrometer (Welas).

For particle sizes ranging from 0.2 to 10  $\mu$ m diameter, a light scattering aerosol spectrometer (Welas 2000, Dylec Corp. Tokyo, Japan) was used to determine particle concentration and particle size distribution. The detailed operation of this instrument was described in the study of Luu et al., (2010). It has a wide concentration range from 1 to 10<sup>5</sup> particles/cm<sup>3</sup>. Data analysis was performed using PDControl 1.0 software (Dylec Corp. Tokyo, Japan).

The complete size distribution for droplet diameters was represented using the method described by Ziegr *et al.* (2010). The particle size distribution between 15.1 nm and 10  $\mu$ m was obtained by combining the light scattering data at diameters above 661.2 nm with the SMPS data at diameters below 661.2 nm. A little difference in diameter between the two instruments was obtained at the merging point, even though the SMPS measures a mobility diameter and light scattering measures an optical diameter. The particle size distribution was presented according to the method used by Babinsky (2002).

#### 2.3. MATHEMATICAL FORMULATION OF DROPLET SIZE DISTRIBUTION

#### 2.3.1. Distribution functions for particle size distribution

Various mathematical distribution functions have been used to fit particle size experimental data. In this paper, the equations of Rosin-Rammler (Macías-García *et al.*, 2004) and Nukiyama-Tanasawa (Tanasawa, 1963; González-Tello *et al.*, 2008) were applied to obtain the size distribution of droplets from the measured data.

Nukiyama-Tanasawa equation can be expressed as

$$f(x) = \frac{1}{N_T} \frac{dN}{dx} = Ax^{\alpha} \exp(-Bx^{\beta})$$
(2-1)

where x is the particle diameter, f(x) is the probability density function of the number distribution (nm<sup>-1</sup>),  $N_T$  is total number of particles, N is number particle and A, B,  $\alpha$ , and  $\beta$ are adjustable parameters. Li and Tankin (1987) assume that  $\alpha$ =2. Microsoft Excel Solver was used to fit the data series. The program finds the values of the fitting parameters that give the best fit between the model and the data.

The Rosin-Rammler model can be stated as

$$F(x) = 1 - \exp\left[-\left(\frac{x}{l}\right)^m\right]$$
(2-2)

where x is the particle diameter, F(x) is the probability cumulative density distribution (dimensionless), *m* and *l* are adjustable parameters characteristic of the distribution. These parameters were done by using Microsoft Excel Solver. Usually, Rosin-Rammler equation is used for weight distribution, but in this paper we use it for number distribution because the weight is represented as a function of number as described below .

$$dw = \frac{\pi \bar{x}^3}{6} dN \tag{2-3}$$

Where w is weight (kg),  $\rho$  is density (kg m<sup>-3</sup>),  $\bar{x}$  is mean particle diameter in frequency interval (m) and N is number of particles. The number frequency distribution can be obtained by taking the derivative of the cumulative distribution,

$$f(x) = \frac{1}{N_T} \frac{dN}{dx} = \frac{m}{l^m} x^{m-1} \exp\left[-\left(\frac{x}{l}\right)^m\right]$$
(2-4)

In order to compare the accuracy of predicted data between Eq. (2.1) and Eq. (2.4), root mean square error (RMSE) was used.

$$RMSE = \sqrt{\frac{\sum \left\{ f(x) - f(x_p) \right\}^2}{n}}$$
(2-5)

where f(x) is the measured value,  $f(x_p)$  is the predicted value and n is the number of observations.

#### 2.3.2. Representative diameters

The Sauter mean diameter ( $x_{vs}$ ) represents the size of a droplet with the volume to surface area ratio of the entire spray and is known to characterise the heat and mass transfer of the droplets better than, for example, the arithmetic mean diameter (Semião *et al.*, 1996). Sauter mean diameter is expressed as

$$x_{VS} = \frac{\int_{0}^{\infty} x^{3} f_{N}(x) dx}{\int_{0}^{\infty} x^{2} f_{N}(x) dx}$$
(2-6)

Arithmetic mean diameter (AMD) indicates droplet diameter (Bhanarkar *et al.*, 2008) and is defined as

$$AMD = \frac{\int_0^\infty x f_N(x) dx}{\int_0^\infty f_N(x) dx}$$
(2-7)

#### 2.4. RESULTS AND DISCUSSION

#### 2.4.1. Particle size distribution at ambient environment

The temperature and relative humidity of ambient air were measured at 22 °C and 70% RH. Fig. 2.3 presents particle number concentration and size distribution measured at the outlet generated by different operating frequencies of the nanomist humidifier. It can be seen that the observed size distributions were unimodal and a peak value occurred at around 40 nm. There was a good correlation between the particle number concentration and frequency of the generator. The particle number concentration increased with increasing generator frequency. Adiga (2005) observed that the droplet size and particle concentration depended on nanomist generator frequency. However, here changes in size distribution were not observed when frequency was varied.

The difference in particle size distribution between nanomist and ultrasonic mist at distances of 500 mm and 1000 mm on the straight line extending from the outlet is described in Fig. 2.4. Again, the nanomist size distribution shown was unimodal. The peak value, and corresponding number concentration, measured at the outlets of the nanomist and ultrasonic mist humidifiers were about 40 nm and 120 (particle/cm<sup>3</sup>/nm), and 60 nm and 6985 (particle/cm<sup>3</sup>/nm), respectively. With regard to the nanomist humidifier, the size distribution did not vary with distance, but the particle number decreased with increasing distance. Both size distribution and particle concentration of the ultrasonic mist varied with distance from 0 m to 500 mm (Fig. 2.4b) although air velocity measured at the outlet of ultrasonic humidifier was 8.3 m s<sup>-1</sup>; only slightly stronger than the 7.8 m s<sup>-1</sup> measured with distance



Fig. 2.3 Particle size distribution generated by different frequencies of nanomist humidifier at ambient air.



Fig. 2.4 Changes in particle size distribution generated by 50 Hz nanomist (a) and ultrasonic (b) humidifiers at ambient air from different positions.

from the outlet to a distance of 500 mm; it slowly reduced from 500 mm to 1000 mm. The particle size distribution became coarser with distances > 500 mm; the peak number mode was approximately 216 nm for both distances of 500 and 1000 mm. The reduced particle number concentration may be attributed to the evaporation of mists or smaller particle coagulating into coarser particles (Wang, 1982; Shi *et al.*, 1999; Zhang *et al.*, 2001). The lower reduction in nanomist concentration can be attributed to the low particle concentration; perhaps the frequency of collision was low and coagulation did not occur.

#### 2.4.2. Particle size distribution under controlled temperature and relative humidity

Fig. 2.5 presents the difference of particle size distribution observed under three cold storage environments. Relative humidity recorded in the chamber without the humidifier, in chambers with nanomist and ultrasonic humidifiers was 60.2%, 92.0% and 91.6% respectively, whilst temperature was maintained at around 5 °C for all chambers. It is clear that the concentration of mist and particle size observed in the chamber without humidifier was very low compared to measurements with humidifiers. The mode value produced by the nanomist at 50 Hz was 61.5 nm, about 3 times smaller than that of ultrasonic humidifier whilst the nanomist concentration was 210 (particle/cm<sup>3</sup>/nm), about 4 times lower than that of ultrasonic mist, although the discharge rate of both humidifiers was similar (490 g h<sup>-1</sup> for nanomist and 417 g h<sup>-1</sup> for ultrasonic mist). Barrow and Pope (2007) showed that evaporation rate is greater with fine droplets because they have greater surface area relative to their volume (surface area to mass ratio) compared with coarse droplets. In this experiment, although the instruments were able to measure the particle size up to 10  $\mu$ m, just few particles with between 1  $\mu$ m to 2  $\mu$ m diameter were observed with the ultrasonic



Fig. 2.5 Particle size distribution measured under various cold storage

environments at 5 °C.

mist. This size range lies in with data range provided by UCAN Co., Ltd, Tokyo, Japan (personal communication).

Fig. 2.6 shows the particle size distribution obtained from the nanomist and ultrasonic mist chambers at temperature of 5 °C and relative humidity of 80% and 90% at intervals of 2 and 4 h. As can be seen from the graph, the particle size distribution observed at 2 and 4 h was similar for both types of mists at relative humidity of 80% and 90%. On the other hand, when relative humidity was increased to 90%, a corresponding increase in particle number concentration was observed. This result is in agreement with the findings of Zhang (2001) which showed that the particle number concentration correlated positively with relative humidity. Changes in relative humidity could decrease particle number concentration due to evaporation but would not change particle size distribution.

The particle size distribution of the nano- and ultrasonic mists at a temperature of 5 °C and 90% relative humidity after 18 h is shown in Fig. 2.7. As with other measurements, the peak value of the nano- and ultrasonic mists were 64 nm and 210 nm, respectively. Interestingly, the particle number concentration of the nanomist measured at 700 mm above from the ground was lower than that measured at 1730 mm above the ground (Fig. 2.7a) whilst the particle number concentration of ultrasonic mist was almost the same at two locations. The lower nanomist concentration observed at the lower height may be explained that since nanomist particle size is fine, the mist tends to move with the airflow direction and is carried to the upper site by the air stream. It had similar evaporation as shown in Fig 2.4.



Fig. 2.6 Particle size distribution generated by 50 Hz nanomist (a) and ultrasonic (b) humidifiers at 5 °C, 80% and 90% relative humidity (RH) during four hours.



Fig. 2.7 Particle size distribution generated by 50 Hz nanomist (a) and ultrasonic (b) humidifiers at 5 °C and 90% relative humidity (RH) after 18 hours at two points

#### 2.4.3. Fitting experimental data to mathematical models

Table 2.1 shows the representative diameters and the parameters *A*, *B*,  $\alpha$ ,  $\beta$  and *m*, *l* values obtained when the Rosin-Rammler and Nukiyama-Tanasawa equations were used. As can be seen from the table that mode diameter of the nanomist did not depend on the frequency of the generator. The AMD of nanomist became finer with increased frequency, but the SMD was not affected by generator frequency. It is noticeable that the SMD of nanomist was finer than that of ultrasonic mist, so it is clear that nanomist produces a mist that is easier to evaporate than ultrasonic mist. It can be also seen that there was a connection between parameter *B* in Nukiyama-Tanasawa equation and SMD; the smaller the parameter *B*, the higher the SMD. Tanasawa (1963) reported that parameter *B* is a function of SMD. As a result, if parameter *B* is identified, then the SMD of particles can be understood. Based on experimental results, the particle size of nanomist was about 2-3 times smaller than that of ultrasonic mist depending on the generator frequencies used.

Fig. 2.8 shows the fit of the Rosin-Rammler and Nukiyama-Tanasawa equations to the experimental data obtained from nanomist (50Hz) and ultrasonic humidifiers operated at 5 <sup>o</sup>C and 90% relative humidity after 2 h. Data shows that both the Nukiyama-Tanasawa (Eq. 2-1) and Rosin-Rammler (Eq. 2-4) equations fit the experimental data acceptably. However, the Nukiyama-Tanasawa equation had a lower RMSE (0.00257 and 0.051 for nanomist and ultrasonic) compared to the RMSE for the Rosin-Rammler equation (3.079 and 0.081 for nanomist and ultrasonic). Thus, the curve obtained from Nukiyama-Tanasawa equation fitted the experimental data better than that obtained using the Rosin-Rammler equation. Thus, the Nukiyama-Tanasawa equation could be used to estimate the particle size distributions produced by nano- and ultrasonic mists.

## Table 2.1 Parameters in distribution equations and representative diameters

Humidifier	Parameters				Representative diameters (nm)				
	Nukiyama-Tanasawa			Rosin-I	Rammler	Mode	AMD	SMD	
	α	β	A	B	m	l			
Nano-40 Hz	2.0	1.45	5.84E-06	2.56E-03	2.18	110.6	61.5	104	333
Nano-50 Hz	2.0	1.35	1.18E-05	5.63E-03	2.16	91.5	61.5	86	235
Nano-60 Hz	2.0	1.38	1.35E-05	5.24E-03	2.17	86.7	65.5	82	314
Ultrasonic	2.0	2.66	2.37E-07	5.81E-07	2.66	247.6	210	234	591

### at 5 °C and 90% RH

Mode is diameter associated with the maximum number of particles in a distribution; AMD is Arithmetic mean diameter; SMD is Sauter mean diameter



Fig. 2.8 Fitting of the Rosin-Rammler and Nukiyama-Tanasawa equations to experimental data generated by 50 Hz nanomist (a) and ultrasonic (b) humidifiers at 5 °C and 90% relative humidity (RH) after 2 hours.

#### **2.5. CONCLUSIONS**

This study investigated the characteristics of mists formed by two types of humidifiers in storage environments with high humidity. The results showed that the number mode diameter of nanomist humidifier at 60 Hz operating at 5 °C and 90% RH was 65.5 nm, about 3 times finer than that produced by the ultrasonic mist. Moreover, particle number concentration of the nanomist was 4 times lower than that of the ultrasonic mist. It was also shown that the mist number concentration was very much dependent on generator frequency and the relative humidity. From these results, it can be concluded that nanomist evaporates more easily than that of ultrasonic mist. Therefore, the nanomist could be useful for raising relative humidity without wetting the produce. This will serve as the basis for future studies and also supports the hypothesis that the nanomist could bring several benefits to the postharvest storage of fresh produce. Further investigations into the effects of mist particle size on the strength of corrugated cardboard and the quality of fresh produce during a long storage period are recommended.
# **CHAPTER 3**

# Examination of the strength of corrugated cardboard exposure to nanomist and ultrasonic mist under high humidity condition

# **3.1. INTRODUCTION**

Corrugated cardboard is the most widely used type of package for the packaging and distribution of a wide variety of commodities ranging from fruits and vegetables, consumer products, to industrial items. It is equally suitable for all the different modes of storage and transport such as shipping by sea or by air. The most important feature of containers made from corrugated cardboards is to protect the packaged commodities against damage during storage and transport. Therefore, the maintenance of strength of cardboard during storing, marketing and distribution of horticultural commodities is needed to take into consideration.

The storage of produce is usually maintained under low temperature environment. As far as commodity's transpiration is concerned, however, high relative humidity of the storage environment plays an important role in maintaining the quality of produce. Recently, low temperature and high humidity storage has attracted the attention of horticultural industries in Japan (Tanaka, 2000). Recommended humidity level for the storage of fresh fruit and vegetables is commodity specific; levels are generally in the range of 85% to 95% (Paull, 1999; Rennie *et al.*, 2003; Maguire *et al.*, 2004). One of the problems occurring for corrugated cardboard stored under high humidity environment is that corrugated cardboards reduce its strength because the boxes become wet during storage. The reduction in strength of cardboards can lead to packaging collapse, thereby causing further mechanical damage

to products (Marcondes, 1992). Furthermore, wetting may result in biodeterioration of cardboard (Koivula and Hanninen, 1999).

Various studies (Ackerman, 1997; Twede and Selke, 2005; Modzelewska, 2006; Allaoui *et al.*, 2009a; Allaoui *et al.*, 2009b) have been performed on the corrugated cardboard and its compressive strength. They found that corrugated cardboard is very sensitive material to the environmental conditions, especially high relative humidity. Once wet, the corrugated cardboards lose their rigidity. Modzelewska (2006) indicated that when humidity of ambient air was about 100%, boxes made from corrugated cardboards practically disintegrated. For this reason, the idea of using mists with smaller droplets to raise high humidity for storage environments is considered to apply. It is reported by Barrow and Pope (2007) the evaporation rate is greater with smaller droplets because small droplets have greater surface area relative to their volume (surface area to mass ratio) as compared with larger droplets. The rapid evaporation of droplets results in the more dryness on the surface of the corrugated cardboard; in that way, strength of board is preserved.

At present, the large size or conventional mists are being employed to raise relative humidity in the storage environment. These mists are mostly generated by ultrasonic humidifier. Our recent study showed that ultrasonic humidifier emitted the mists with average mode diameter of 210 nm (Hung *et al.*, 2010b). The large size mists may easily damp the corrugated cardboards, thus their strength are lost. Nanomists, which are defined as particles in the range from 1 nm to 100 nm in diameter (Nickols-Richardson, 2007), are assumed to evaporate immediately after atomization and are considered not to damp the corrugated boxes in comparison with the larger size ultrasonic (Hung *et al.*, 2009). It is the goal of this study which proposes to investigate the effect of nanomist and ultrasonic-mist

on the moisture content and compressive strength of corrugated cardboard under high relative humidity condition.

# **3.2. MATERIALS AND METHODS**

### 3.2.1. Sample preparation

The experiments were conducted on three types of commercially made corrugated cardboard boxes used for the package of strawberry, broccoli and mizuna. The cardboard used in this study was single wall type. Table 3.1 lists the name of corrugated cardboard boxes tested, along with their key specifications. The moisture content and strength tests were performed on both test specimens and empty boxes.

Specimen tests of selected corrugated cardboards were prepared according to JIS Z0403-2 (JSA, 1999) (Fig. 3.1). For every measurement of moisture content, a sample of 5 specimens was tested for each type of cardboard box. The edge of specimens was wrapped with aluminum tape to prevent water vapor and mist penetration from the edge. With regard to mechanical test, a sample of 12 pieces was used to measure the relationship between the moisture adsorption and compressive strength. In order to investigate the differences in characteristics of moisture adsorption of specimens, the samples were horizontally and vertically placed on the stand at the center of chambers. For corrugated cardboard box test, a sample of 5 strawberry and broccoli boxes was tested. The samples were placed at ambient temperature for 24 h and then put near the outlet of humidifiers and at the center of storage chambers at temperature about 6  $^{\circ}$ C and relative humidity of 94%.

Box Type	Basi	s weight (g/	$cm^2$ )	Electer terms	Numbers	Dimension (mm) (H x L x W)	
	Outer liner	Medium	Inner liner	Flute type	of flaps		
Strawberry	180	125	180	С	0	60 x 305 x 220	
Mizuna	210	200	170	А	4	440 x 390 x 210	
Broccoli	220	160	220	А	4	430 x 330 x 250	

# Table 3.1 Properties of the tested corrugated cardboard boxes.

H: height, L: length, W: width

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Fig. 3.1 Geometry of corrugated cardboard specimen.

For the test of moisture content, the specimens were conditioned in a thermo-hygrostat under conventional condition (23 °C and 50% RH) for over 24 h (JSA, 1998). Once the equilibrium is reached, specimens were weighed and these measurements were used as dry basis. The samples were then placed in the storage environments at temperature about 6  $^{\circ}C$ and relative humidity of 94% where nanomist and ultrasonic-mist were sprayed to raise humidity. Nanomist is generated by nanomist humidifier (Test model, Mayekawa Co., Ltd, Tokyo, Japan) while ultrasonic-mist is emitted by ultrasonic one (FT -30N-14, UCAN Co., Ltd, Japan). The amount of water absorbed was measured by the change in mass of the cardboards as a function of time. The change in mass of samples was first measured at intervals of 6, 12 and 24 hours, and then daily over 7 days. For mechanical test, specimens were also conditioned as above-mentioned and stored at the same temperature and RH condition as described previously. The samples were tested several times during storage. Data of temperature and RH were recorded at ten-minute intervals in the containers using humidity and temperature transmitter (model HMT337, Vaisala, Helsinki, Finland). This device can measure the accuracy at temperature  $\pm 0.2$  °C at range from -70 °C to + 180 °C and RH  $\pm$  1.7% at range from 90% to 100%.

#### 3.2.2. Compression test

The strength test of piece samples was performed by using a tensile and compression testing instrument (Tensilon UTM-7, Toyo Measuring Instruments Co. Ltd., Japan) at crosshead speed of 3 mm/min based on a column crush test (JIS Z0403-2), whereas the top-to-bottom compression test of empty boxes was conducted by using compression test machine (AG-100 kNE, Shimadzu, Japan) at crosshead speed of 12 mm/min (Fig 3.2).



Fig. 3.2 Compression testing instruments of Tensilon UTM-7 and Autograph AG-100 kNE.

Two metal plates with smooth surfaces are attached to the upper and the lower compression jaws of the machine to evenly distribute the compression load on the corrugated cardboards.

### 3.2.3. Statistical analysis

Data analysis was performed using analysis of variance (GenStat Discovery Edition 3, VSN Internaional Ltd., UK). Significant differences were further examined with Tukey's test at  $P \le 0.05$ .

### **3.3. RESULTS AND DISCUSSION**

#### 3.3.1. Specimen test

The moisture content of specimens exposed to nanomist and ultrasonic-mist after 7 days is shown in Table 3.2. The average temperature and relative humidity measured during the experiment were  $5.9 \pm 0.37$  °C and  $94.2 \pm 1.46\%$  for nanomist and  $6 \pm 0.38$  °C and  $94.3 \pm$ 1.35% for ultrasonic-mist. As can be seen from the table that type of cardboard did not affect the moisture content (P = 0.423), but type of mist and position of placing specimens significantly influenced on the absorption of water vapor (P<0.05). Moisture content of specimens exposed to nanomist after 7 days was about 10% lower than that exposed to ultrasonic-mist regardless of position. Moreover, moisture content of specimens stored in the nanomist container was not affected by the position while moisture content of vertically-positioned specimens in the ultrasonic container was significantly lower than that of the ones placed horizontally. The main reason causing the differences in moisture content between specimens conditioned with nanomist and ultrasonic-mist is attributed to the difference in size and particle concentration emitted by two different humidifiers. The previous data showed that mode-based diameter of nanomist is about 65.5 nm, such mist Table 3.2 Moisture content (% d.b.) of specimens exposed to nanomist and ultrasonicmist at ca. 6  $^{\circ}$ C and 94% RH after 7 days .

Doy Type	Nanc	omist	Ultrasonic-mist			
Box Type	Vertical	Horizontal	Vertical	Horizontal		
Strawberry	$19.5 \pm 0.97$ a	$17.9 \pm 0.74$ a	$26.8\pm0.94~b$	$30.7 \pm 1.48$ c		
Mizuna	$18.7 \pm 0.57$ a	$18.5 \pm 0.87$ a	$29.0\pm1.03~\text{b}$	$30.4 \pm 1.07 \text{ c}$		
Broccoli	$18.3 \pm 0.45$ a	$18.0 \pm 0.67$ a	$28.1 \pm 1.37$ b	$30.5 \pm 1.1 \text{ c}$		

A different letter showed significant difference at P < 0.05 according to Tukey's test. Data are accompanied by the standard deviations of the means (n = 5). Vertical; vertically-positioned specimens and horizontal; horizontally-positioned specimens.

evaporates easily compared with ultrasonic-mist which has a diameter of around 210 nm (Hung *et al.*, 2010b). The effect of position on moisture content of specimens exposed to ultrasonic-mist can be explained that large size mist directly falls down on the surface of specimens without evaporating and it wet the board, thus water is adsorbed into the cardboard.

In order to analyze the adsorption kinetics of water vapor on the corrugated board, the pseudo first- order kinetics model was used to analyze the experimental data. One of the purposes of using the numerical analysis is to predict the results without doing experiments. This equation has been used for dye adsorption by cotton (Chairat *et al.*, 2008) and water adsorption by brown rice (Genkawa *et al.*, 2008). A simple kinetic analysis of adsorption is the Lagergren equation. A pseudo first-order equation describes the kinetics of the adsorption process as follows:

$$\frac{dM}{dt} = k_1 \left( M_e - M \right) \tag{3-1}$$

where  $k_1$  is the rate constant of pseudo first-order adsorption (day<sup>-1</sup>), and  $M_e$  and M are the moisture content (% d.b.) at equilibrium and at time *t*. After definite integration by applying the initial conditions ( $t = 0, M = M_0$ ), Eq. (3.1) becomes

$$M = M_{e} + (M_{0} - M_{e})\exp(-k_{1}t)$$
(3-2)

where  $M_0$  is the initial moisture content.  $k_1$  and  $M_e$  were obtained by nonlinear least square method. These parameters were shown in Table 3.3.

The result in Fig. 3.3 presents moisture content of strawberry specimens exposed to nanomist and ultrasonic-mist. The absorption of water vapor increased with time until it reached the saturation. The data represented by a solid line were obtained from the pseudo



Fig. 3.3 Moisture content of strawberry specimens exposed to nanomist and ultrasonic mist at ca. 6 °C and 94% RH. Solid lines are fitted values. Bars represent standard deviations of the means (n = 5).

Table 3.3 Parameters for prediction of moisture content of specimens and corrugatedcardboard boxes.

	Nanomist				Ultrasonic-mist			
Parameters	Specimen		Box		Specimen		Box	
	Vertical	Horizontal	Outlet	Center	Vertical	Horizontal	Outlet	Center
M <sub>o</sub>	7.7	7.77	8.18	8.08	9.73	9.08	8.18	8.49
$M_{e}$	18.74	17.49	24.22	29.6	19.63	21.68	29.47	21.92
<i>k</i> 1	3.5	2.69	4.32	5.69	1.97	2.06	1.09	1.65

first-order equation (3-2). The fitted values agreed very well with the experimental data. This result suggested that Eq. (3-2) is available for analyzing water adsorption of corrugated cardboard.

The compressive strength of corrugated cardboard was calculated according to JIS Z 0403-2 which is defined as  $P_m = 0.02F$ , where  $P_m$  is compressive strength of cardboard (kN/m), *F* is the maximum compressive load (N) and 0.02 is the reciprocal number of width (50 mm) of test piece (1/mm). Compressive load of the corrugated cardboard with 8.71 % d.b. of moisture content was proportional to strain in the middle of test after which the increasing rate of the load gradually decayed; finally the cardboard bent as shown in other material tests (Fig. 3.4).

Fig. 3.5 shows the relationship between the moisture content and compressive strength of specimens. Relationship between moisture content (M) and compressive strength ( $P_m$ ) was represented by using an exponential Eq. (3-3) and parameters were shown in Table 3.4.

$$P_m = a M^b \tag{3-3}$$

The coefficient of determination calculated from data of strawberry and mizuna specimens was 0.926 and 0.956, respectively. It shows that compressive strength of board exponentially decreased with the moisture content. It has been well documented that the yield stress decreased with the increase in the moisture content (Ackerman, 1997; Twede and Selke, 2005; Allaoui *et al.*, 2009a). In addition, compressive strength of cardboard can be predicted by using Eq. (3-4) which was obtained from Eq. (3-2) and Eq. (3-3), and parameters were presented in Table 3 and 4.

$$P_m = a[M_e + (M_0 - M_e)\exp(-k_1 t)]^b$$
(3-4)



Fig. 3.4 Compressive curve of the corrugated cardboard with 8.71 % d.b. of moisture content.



Moisture content *M* (% d.b.)

Fig. 3.5 Relationship between moisture content and compressive strength of the specimens. Solid curves are fitted values (n = 12).

Box Type	S	pecimen tes	st		Box test	
	а	b	$r^2$	С	d	$r^2$
Strawberry	17.4	-0.87	0.926	252.7	-1.81	0.963
Mizuna	45.5	-0.97	0.956	-	-	-
Broccoli	-	-	-	1489.8	-2.62	0.887

# Table 3.4 Parameters for prediction of compressive strength of specimens andmaximum compressive load of corrugated cardboard boxes.

*a, b, c, d* : parameters in Eq. (3), (5);  $r^2$  : coefficient of determination

Fig. 3.6 compares the compressive strength of vertically-positioned specimens exposed to nanomist and ultrasonic mist at about 6 °C and 94.3% relative humidity after 7 days. The data shown at day 0 were experimental data whereas those shown at day 7 were predicted values. In general, it can be easily seen that compressive strength of specimens conditioned with nanomist was significantly different with that of those exposed to ultrasonic-mist (P < 0.05). After 7 days the strength of specimens exposed to nanomist remained at 43.4% to 55.6% while that of those exposed to ultrasonic-mist stayed at 29.9% to 33.4% depending on types of cardboards. It is also noted that compressive strength of cardboard was affected by type of cardboard. Strawberry cardboard box, which is made from corrugated cardboard type of C with thickness of 3 mm, has a lower initial strength than the others with thickness of 5 mm; but once stored under nanomist environment its strength remained at 55.6% at the end of experiment. According to the study of Modzelewska (2006), the effect of moisture content on the behavior of corrugated cardboard boxes is attributed to the material employed for its manufacture rather than to the structure (layers, flutes...). For the cardboard boxes made from the same material, the flute height influences the properties of the corrugated cardboard. The higher the flute, the better the rigidity of cardboard, but the product will have greater absorption property and consequently the strength was weaker. From this result, it can be concluded that nanomist has a great effect on controlling strength of corrugated cardboard.



Fig. 3.6 Compressive strength of vertically-positioned specimens exposed to nanomist and ultrasonic mist ca. 6 °C and 94% RH after 7 days. Bars represent standard deviations of the means (n = 5).

## 3.3.2. Corrugated cardboard box test

Given in Fig. 3.7 is the change in moisture content of strawberry cardboard box exposed to nanomist and ultrasonic-mist over a period of 7 days at 6 °C and 94% RH. The data represented by a solid line were obtained from the pseudo first-order Eq. (3-2) using parameters in Table 3.3. It is evident that moisture content of cardboard exposed to nanomist was significantly lesser than that of those exposed to ultrasonic-mist. Additionally, the position of placing boxes in the chambers affected the moisture content. The boxes placed close to the outlet of ultrasonic humidifier had a moisture content of 30.8% d.b. whilst the ones put in the center had a moisture content of 24.8% d.b. On the contrary, the boxes located near the outlet of nanomist had a lower moisture content that the ones in the center. The result obtained from broccoli box was similar to the strawberry's (data not shown). The larger size ultrasonic mists emitted near the outlet are ascribed to be responsible for the wetting of corrugated. The results obtained from the experiments of cardboard boxes again confirmed that the moisture content of the boxes exposed to nanomist was about 10% lower than that of those exposed to conventional mist.

Fig. 3.8 shows the relationship between the moisture content and maximum compressive load of strawberry and broccoli cardboard boxes. In this study, we could not determine cross sectional area of the box, thus a unit of F was used instead of  $P_m$ . Relationship between moisture content (M) and maximum compressive load (F) was represented by using an exponential Eq. (3-5) and parameters were shown in Table 3.4.

$$F = c M^d \tag{3-5}$$



Fig. 3.7 Moisture content of strawberry corrugated cardboard box exposed to nanomist and ultrasonic-mist at ca. 6 °C and 94% RH. Solid lines are fitted values.

Bars represent standard deviations of the means (n = 5).



Fig. 3. 8 Relationship between moisture content and maximum compressive load of strawberry and broccoli cardboard boxes. Solid curves are fitted values (n = 5).

It can be seen from the graph that maximum compressive load of corrugated board was strongly dependent upon the moisture content. The coefficient of determination calculated from data of strawberry and broccoli was 0.963 and 0.887, respectively. The maximum compressive load of corrugated cardboard box in Fig. 3.9 was predicted using Eq. (3-6) which was obtained from Eq. (3-2) and Eq. (3-5), and parameters in Table 3.3 and 3.4.

$$F = c[M_e + (M_0 - M_e)\exp(-k_1 t)]^d$$
(3-6)

Data indicated that maximum compressive load of corrugated cardboard boxes exposed to nanomist and ultrasonic-mist decreased gradually over the time. The boxes exposed to nanomist maintained its maximum compressive load at 28.1%, whereas those exposed to ultrasonic-mist remained at 14% after 7 days (Fig. 3.9). Twede and Selke (2005) reported that high humidity storage conditions can severely degrade strength of the box in a matter of hours. At 85% RH, a box loses about 40% of its compression strength. As relative humidity increases to 90%, the moisture content of the board increases to 20%. This increase in moisture content lowers the compression strength by nearly 50%. An evidence was shown from our experiments that even the cardboard exposed to 95% humidity of nanomist, the moisture content of the cardboard remained approximately 20% d.b. and maintained its maximum compressive load at 28%, while the boxes stored in the same humidity of ultrasonic-mist absorbed 30% d.b. moisture content and its maximum compressive load reduced to 14% after 7 days.



Fig. 3.9 Change in maximum compressive load of strawberry corrugated cardboard box exposed to nanomist and ultrasonic-mist at ca. 6 °C and 94% RH.

Data were calculated using Eq. (3-6).

# **3.4. CONCLUSIONS**

In this investigation, the aim was to evaluate the effects of relative humidity and particle size on the moisture content and compression strength of corrugated cardboard under high relative humidity condition. The results of present research show that relative humidity affected moisture content of corrugated cardboard. One of the more significant findings to merge from this study is that the ultrasonic mist raised a moisture content of corrugated cardboard as compared with nanomist at the same humidity. Furthermore, there was a good correlation between moisture content and compressive strength of corrugated cardboard and the cardboard was weakened with the increase in the moisture content. From these results, it can be concluded that nanomist is useful for raising humidity during cold storage of fresh fruit and vegetables since it helps preserve the strength of corrugated cardboard.

# **CHAPTER 4**

# Weight loss and quality attributes of fresh produce following postharvest storage under nanomist and ultrasonic mist environments

# **4.1. INTRODUCTION**

Fresh fruit and vegetables are living tissues that continue to lose water after harvest, but, unlike growing crops, they can no longer replace lost water from the soil and must rely on the water content present at harvest. The loss of water from fresh produce following harvest is a serious problem because it causes shrinkage and weight loss. Most commodities become unsalable as fresh produce after losing 3% to 10% of their weight (Ben-Yehoshua and Rodov, 2003).

The quality and storage life of fresh produce are highly dependent upon the vapor pressure gradient between the produce and the storage atmosphere. Therefore, when the produce and the storage environment are maintained at the same temperature (assuming factors such as air velocity are held uniform), the transpiration rate is highly correlated with the relative humidity (RH) during storage time (Grierson and Wardowski, 1978; Sharkey and Peggie, 1984). Transpiration through the actions of the stomata, lenticels, cuticles and epidermal cells (Ben-Yehoshua and Rodov, 2003) is considered the major cause of postharvest weight loss and poor quality in leafy vegetables. Transpiration in produce is a mass transfer process in which water vapor moves from the surface of plant organs to the surrounding air. This process occurs when there is a gradient of water vapor pressure between the tissue and the surrounding air.

Produce is usually stored under a low-temperature environment. For the commodity's transpiration, however, a high RH storage environment plays an important role in maintaining the quality of produce. Recommended RH levels for the storage of fresh fruit and vegetables are commodity specific, with levels generally in the range of 85% to 95% (Paull, 1999; Rennie et al., 2003; Maguire et al., 2004).

At present, large mists are used to raise RH in the storage environment. These mists are occasionally generated by ultrasonic humidifiers, and their average particle mode diameters vary between 2.9 µm (Rodes et al., 1990) and 210 nm (Hung et al., 2010b). The large mists can easily wet the surface of produce, which is covered by a laminar film of water. The RH of the laminar film is considered to be nearly 100%. Wetting on the surface of the produce causes the stomata to open (Lange et al., 1971), resulting in water loss. Our previous study showed that nanomists, generated by nanomist humidifiers, have average particle diameters of 65.5 nm (Hung et al., 2009). Nanomist humidifiers are thought to provide improved capability for generating ultrafine mists for high humidification. Nanomists, because of their very fine particle size, presumably evaporate immediately after atomization. The fine mists, which are present on the surface of the produce, also easily evaporate and therefore do not wet the produce in comparison with the larger ultrasonic mists. Nanomists could bring a number of benefits to the field of postharvest crop storage. Hung et al. (2010a) have demonstrated that the strength of corrugated cardboard is well-preserved under a nanomist environment with high relative humidity. The aim of this chapter is to investigate the effect of nano- and ultrasonic mists on the control of weight loss and several postharvest quality attributes of three types of horticultural produce stored under high relative humidity environment.

# 4.2. MATERIALS AND METHODS

### 4.2.1. Source of materials

Eggplant fruit (*Solanum melongena* L., cv Chikuyo), mizuna (*Brassica rapa* L. *var. Japonica* cv Kyomizore) and fig fruit (*Ficus carica* L., cv Toyomitsu-Hime) were purchased from a Fukuoka wholesale market. The produce was carefully handled as they were transported to the laboratory and the experiment. Eggplant fruits possessing uniform color and weight between 145 and 200 g each were chosen for the experiment. Mizuna plants weighing approximately 200 g were packed in polyethylene bags with the leafy portion of the plant left open. The figs were sorted to eliminate any that had defects, and those with a uniform color and a weight of approximately 80 grams each were chosen. The experiments were repeated twice in October 2009 and May 2010 for the mizuna and eggplant and in October and November 2010 for the fig fruit.

### 4.2.2. Storage condition

The samples were placed in storage environments at 5.5 °C for mizuna and eggplant and 7 °C for fig fruit. A nanomist humidifier (test model, Mayekawa Co., Ltd, Tokyo, Japan) and an ultrasonic humidifier (FT-30N-14, UCAN Co., Ltd, Japan) were used to maintain the humidity at approximately 95%. Forty-five eggplant fruits and 15 mizuna bags were used for each storage condition for the experiment performed in October 2009, while 10 eggplant fruits and 8 mizuna bags were used in May 2010. Forty figs were used in both experiments for each storage condition. To investigate the effect of particle size on the quality of produce, in addition to the effect of RH, the eggplant and fig fruits were placed on trays and were directly exposed to the mists, whereas the mizuna bags were vertically

placed into plastic trays with an opening on the top of each bag. All samples were assessed for weight loss, chilling injury and color throughout the experiment. Determinations were made at the beginning of storage and at 3 and 6 days for mizuna, 4 and 8 days for fig and 5 and 10 days for eggplant.

The temperature and RH were recorded at 5-minute intervals in the containers using a humidity and temperature transmitter (model HMT337, Vaisala, Helsinki, Finland). This device can measure the accuracy of the temperature to  $\pm 0.2$  °C and the RH to  $\pm 1.7\%$  at a range of 90% to 100%.

## 4.2.3. Methods for parameter assessment

#### 4.2.3.1. Weight loss

Weight loss was measured as a reduction in the weight of the produce stored and was expressed as the percentage of weight change compared with initial weight, namely, the weight loss rate.

# 4.2.3.2. Chilling injury (CI) index in eggplant fruit

On the day of sampling, the external CI symptoms were visually observed on a subjective scale. The level of CI severity was calculated according to the following scale, similar to that proposed by Concellon *et al.* (2007) and Lederman *et al.* (1997): 1 = no damage, 2 = low damage, 3 = regular damage, 4 = moderate damage and 5 = severe damage. The CI index (*CI*) was expressed as follows:

$$CI = \frac{\sum_{i=1}^{5} (L_i N_i)}{N_{Total}}$$
(4-1)

where  $L_i$  is the chilling injury level,  $N_i$  is number of fruits on the level and  $N_{total}$  is the total number of fruits in the treatment.

### 4.2.3.3. Determination of color

The color of eggplant skins and mizuna leaves was measured using a chromameter (Minolta CR 200, Japan). The measurements were expressed as  $L^*$  values (dark to light), and parameters  $a^*$  and  $b^*$  represent redness to greenness and yellowness to blueness, respectively. Three readings were taken from the lower, central and upper sections of each eggplant fruit, and the average of the values for each fruit was calculated. Ten fruits were used for each measurement. For the mizuna leaf, the determination of color was made from three leaves per pack and eight packs were employed for each storage condition.

To evaluate the browning of the pulp tissue of the eggplant, only the lightness of the pulp tissue indicated by parameter  $L^*$  was used. Two slices (thickness = 1 cm) were cut at the central section of each fruit, and the reading was taken from one side of each slice immediately after cutting. The results were expressed as  $L^*_{0}$ , which was calculated as the mean of five fruits per storage time and condition.

# 4.2.3.4. Examination of stomatal aperture

The stomatal data were collected following the method described by Hirose *et al.* (1992). A glass slide with a small drop of instant adhesive was attached to the epidermal surface of the minuza leaf and the skins of the figs for approximately 30 seconds. After removing the leaves and skins, the glass slide was placed on a microscopic base to observe the stomatal image through a confocal laser scanning system (CLSM) (Olympus Fluoview FV-300, Tokyo, Japan). The collection of stomatal impressions using instant adhesive was performed inside a storage environment under low light on six glass slides from six

leaves/fruits per storage time and condition. From each glass slide, two images were taken at 20-fold magnification with a size of 1024 x 1024 pixels. The stomatal area was measured using ImageJ 1.42q (Wayne Rasband, National Institute of Health, USA). Twenty stomata for minuza and six stomata for fig were randomly selected from each image to study the stomatal area, and a total 240 and 72 stomata were used to calculate stomatal the area for mizuna and fig, respectively, in each treatment.

### 4.2.4. Statistical analysis

The experimental data are presented as the mean  $\pm$  SE of two independent experiments. An analysis of variance was performed using the statistical software GenStat (Discovery Version 3, VSN International Ltd., UK). Differences between the means of attributes were compared by a least significant difference (LSD) test at a significance level of  $P \le 0.05$ .

### **4.3. RESULTS AND DISCUSSION**

The temperature and RH monitored in all experiments during postharvest storage are shown in Table 4.1. From the table, it can be seen that the average temperature recorded from the two storage environments was nearly the same in each experiment. The average RH measured from the nanomist was equal to or smaller than those measured from ultrasonic storage except for fig in experiment 1. For instance, in the second experiments for mizuna and eggplant, the RH recorded from the nanomist was 1.2% and 1.4% smaller than those in the ultrasonic chamber, respectively. Fig. 4.1 presents the changes in temperature and RH during fig storage as an example. The graphs show various patterns of RH readings obtained from the two chambers. There were several points with an RH over 100%. There are two possible reasons explaining this phenomenon. Firstly, this may reflect supersaturation, which occurs when water is present in the form of tiny droplets (Rodov *et al.*, 2010). Secondly, this could be an error of hygrometer (Vaisala) because this device is designed with a warmed probe head which covers around a humidity sensor to avoid condensation. The warmed probe head is heated continuously so that its temperature is always higher than in environment. When relative humidity reaches 100% at a certain temperature, there is water adsorption adhered to the inner surface of the case of humidity sensor. Due to heating of warmed probe, water attached to the inner case evaporates to internal environment, resulting in more water vapor near the sensor. Thus, relative humidity calculated by the device is higher than actual relative humidity in environment.

Produce	Experiment	Nanomist				Ultrasonic-mist			
		Temperature (°C)	RH (%)	Cumulative VPD (Pa)	Weight loss rate (%)	Temperature (°C)	RH (%)	Cumulative VPD (Pa)	Weight loss rate (%)
Minuza	1	$5.9 \pm 1.2$	96.9 ± 3.8	49886	$3.1\pm0.6$	$5.8\pm0.9$	97.3 ± 5.1	45766	$8.4\pm0.5$
	2	$5.5 \pm 0.7$	94.4 ± 5.7	93925	$4.2\pm0.2$	$5.5\pm0.8$	95.8 ± 7.1	89440	$6.2\pm0.6$
Eggplant	1	5.9 ± 1.1	97.4 ± 3.8	66448	$4.4 \pm 0.1$	$5.8\pm0.8$	97.5 ± 4.9	64231	$7.3 \pm 0.3$
	2	$5.5\pm0.7$	$94.4\pm5.7$	119987	$6.2\pm0.1$	$5.6\pm0.8$	95.6 ± 7.1	118419	$9.7 \pm 1.1$
Fig	1	$7.1 \pm 0.5$	$94.3 \pm 4.1$	117216	$7.8\pm0.2$	$7.0 \pm 0.5$	$94.0 \pm 5.7$	133326	$13.3\pm0.3$
	2	$7.0\pm0.4$	93.3 ± 3.9	143102	$9.9\pm0.2$	$7.0\pm 0.5$	93.6 ± 5.9	139137	$16.2\pm0.2$

Table 4.1 Temperature, RH, cumulative VPD and weight loss of produce measuredfrom all experiments.

Data except for cumultive VPD are the mean and accompanied by the standard error of the means. RH: relative humidity, VPD: vapour pressure deficit.



Fig. 4.1 Changes in temperature (T) and relative humidity (RH) under two environments of nanomist (a) and ultrasonic (b) during postharvest storage of fig.

More readings with an RH over 100% were observed in the ultrasonic chamber than in the nanomist chamber. This difference could be one reason that the number density of mists produced by the ultrasonic humidifier was four times greater than the nanomist humidifier (Hung *et al.*, 2010b).

The results shown in Table 4.1 indicate that the weight loss rates of the samples stored in the nanomist were 3.7%, 5.3% and 8.8% for mizuna, eggplant and fig, respectively, while those stored in the ultrasonic mist were 7.3%, 8.5% and 14.7%, respectively. From these data, it can be observed that storing fresh fruit and vegetables in the nanomist chamber reduced the weight loss rates (3.6%, 3.2% and 5.9% for mizuna, eggplant and fig, respectively) compared with those stored under the ultrasonic mists. The differences in the weight losses of the produce between the two treatments can be explained using Fick's law of diffusion. According to Ben-Yehoshua (2003), the flow rate of water vapor through the surface of fruit or vegetables is proportional to the difference between the humidity of the internal atmosphere of the produce and the humidity of the storage atmosphere, behaving according to Fick's law of gas diffusion. This model has been applied to estimate the transpiration rate of mushrooms (Mahajan *et al.*, 2008). Therefore, the weight loss of fresh produce in a chamber in which the RH changes with time can be calculated based on Fick's law and is expressed as follows:

$$J = k \left( \frac{P_s - P}{R_d T_A} \right) \tag{4-2}$$

where *J* is the mass flux of water vapor; *k* is the mass transfer coefficient;  $R_d$  is the gas constant of water vapor per mass unit;  $T_A$  is the absolute temperature and  $(P_s-P)$  is the vapor pressure deficit (VPD).

VPD also can be expressed as

$$VPD = P_s(100-RH)/100$$
(4-3)

with  $\varphi$  (decimal)=*RH*/100 and VPD =*P<sub>s</sub>*(1- $\varphi$ ), where  $\varphi$  is actual relative humidity (RH %) and *P<sub>s</sub>* is the saturated vapor pressure in pascals (Pa) and is calculated using Tetens's equation, as presented by Rodov et al. (2010):

$$P_s = f\left(a \exp\left\{\frac{bT}{c+T}\right\}\right) \tag{4-4}$$

 $a = 611.21, b = 17.502, c = 240.97 \text{ and } f = 1.0007 + 3.46 \text{ x } 10^{-8} P$ 

where T = temperature in degrees Celsius (°C) and P = atmospheric pressure in pascals,

Eq. (4.2) can be rewritten as

$$\frac{1}{A}\frac{dm}{dt} = KP_s(\varphi_s - \varphi) \tag{4-5}$$

where *m* is the mass of water vapor and *t* is the time.

$$K = \frac{k}{R_d T_A} \tag{4-6}$$

As the rate of change of absolute temperature during the experiment is extremely small,  $T_A$  can be considered constant.

To find the change in mass, namely, water loss  $\Delta m$ , we integrate Eq. (5) as follows:

$$\Delta m = \int_{i}^{f} dm = KA \int_{i}^{f} P_{s}(\varphi_{s} - \varphi) dt$$
(4-7)

where *i* is the onset of experiment and *f* is the end of experiment. By discretizing Eq. (7), we obtain Eq. (8)

$$\Delta m = KA\Delta t \sum_{i=1}^{n} P_{s_i} \left( \varphi_s - \varphi_i \right)$$
(4-8)

where  $\Delta m$  is the mass of water transpirated from the produce (kg),  $\Delta t = t_i - t_{i-1}$  is partial storage time (s),  $\varphi_s$  is the equilibrium of the RH in the tissue of the produce (intercellular RH is described below),  $\varphi_i$  (decimal) is the RH of the storage environment at time  $t_i$ , A is the surface of the produce through which water vapor passes (m<sup>2</sup>) and K is a constant related to the mass transfer coefficient, as expressed by Eq. (4-6)(s m<sup>-1</sup>).

The expression  $P_{s_i}(\varphi_s - \varphi_i)$  in Eq. (4-8) is the vapor pressure deficit (VPD), which shows the difference in the partial pressure of water vapor between the storage environment and the produce. The rate of transpiration from fresh produce is proportional to the VPD of the surrounding atmosphere at a given temperature if *K* and *A* are constant. The higher the VPD, the greater the water loss.

In both empirical and theoretical studies, it has been well demonstrated that intercellular water vapor in the tissues of fresh produce is very close to saturation with a  $\varphi_s$  of approximately 0.995 (Nobel, 1974; Ben-Yehoshua and Rodov, 2003). We can calculate the cumulative VPD using Eq. (4-9) based on the RH and temperature data from each experiment. The values of  $\varphi_i$  more than or equal to 0.995 were not calculated because, at this level of  $\varphi_i$ , the transpiration process does not occur.

$$CumulativeVPD = \sum_{i=1}^{n} P_{s_i} (\varphi_s - \varphi_i)$$
(4-9)

The data on cumulative VPD calculated from each experiment are shown in Table 4.1. If we hypothesize that the parameters A and K in Eq. (4-8) are the same in both storage
environments, the weight loss of the produce then depends on the cumulative VPD. We can see that the cumulative VPDs obtained from the experiments exposed to the nanomists were higher than those treated with the ultrasonic mists, except for fig in experiment 1. Although the cumulative VPDs of the experiments stored with nanomists were higher than those of the ultrasonic mists, the weight loss rates of the nanomist-exposed produce were always smaller than those under the ultrasonic mists. Furthermore, the mean RH recorded in the ultrasonic mists of experiment 2 was 1.2% and 1.4% higher than that in the nanomists for mizuna and eggplant, respectively; the weight loss rate of the produce stored in the ultrasonic mists would have been lower than those in the nanomist. There was one case in which the cumulative VPD of the nanomist experiment was smaller than the ultrasonic, but the ratio of cumulative VPD for ultrasonic to nanomist (1.1) was very different, with a weight loss rate ratio of ultrasonic to nanomist of 1.7. Therefore, it can be concluded that one of the two above-mentioned parameters (K, A) in both storages was not the same. It is also possible that the weight losses of the produce may be influenced by the particle size, resulting in the partial closing of the stomata pores which is relative to the A parameter in Eq. (4-8), as indicated below.

Fig. 4.2 shows the stomatal opening of the mizuna leaves and the fig fruit stored in the nanomist (a) and the ultrasonic mist (b). The stomatal pore is presented in Table 4.2. Significant differences were found in stomatal area between the samples stored in the nanomist and ultrasonic conditions. The average value of the stomatal area measured prior to storage was 99.2  $\mu$ m<sup>2</sup> and 172.5  $\mu$ m<sup>2</sup> for mizuna and fig, respectively. At that time, the recorded temperature and RH were 23 °C and 61%, respectively, under light. Nevertheless, the stomata of the samples partially closed during storage, especially the stomatal pores of the samples stored in the nanomist chamber, which closed by 34.7  $\mu$ m<sup>2</sup> and 51.5  $\mu$ m<sup>2</sup> for



Mizuna



Fig

Fig. 4.2 Stomatal opening of mizuna leaves and fig fruit stored in the nanomist (a)

and in the ultrasonic mist (b).

Products	Treatments	Storage time	Stomatal pore $(\mu m^2)$	Color			CI index	Pulp color $(L *_0)$
		(days)		$L^*$	<i>a</i> *	$b^*$		
Mizuna	Nanomist	0	$99.2 \pm 5a$	$45.9\pm0.3a$	$-18.4\pm0.2a$	$25.4 \pm 0.3a$		
		3	$69.4\pm3b$	$46.0\pm0.2a$	$-18.2 \pm 0.2a$	$25.1 \pm 0.4a$		
		6	$64.5\pm2b$	$46.2\pm0.4a$	$-18.4 \pm 0.2a$	$25.5\pm0.4a$		
	Ultrasonic-mist	0	99.2 ± 5a	45.9 ± 0.3a	-18.4 ± 0.2a	25.4 ± 0.3a		
		3	$83.4\pm3.7a$	$45.5\pm0.3ab$	$-17.1 \pm 0.2b$	$23.7\pm0.4b$		
		6	$87.6\pm2.2a$	$45.0\pm0.3b$	$-17.2 \pm 0.2b$	$23.0\pm0.4b$		
Eggplant	Nanomist							
		0		$27.0\pm0.2a$	$2.2 \pm 0.3a$	$-0.9 \pm 0.1a$		$86.4 \pm 0.3$ ab
		5		$25.8\pm0.2b$	$2.7 \pm 0.4a$	$-0.8 \pm 0.1a$	$2.6\pm0.2c$	$86.7\pm0.2ab$
		10		$24.5\pm0.4c$	$2.6 \pm 0.4a$	-0.7 ± 0.1a	$3.2\pm0.2b$	$87.1\pm0.3a$
	Ultrasonic-mist	0		27.2 . 0.2	22.02	0.0 . 0.1		064.02
		0		$27.3 \pm 0.2a$	$2.2 \pm 0.3a$	$-0.9 \pm 0.1a$	2.1 0.11	86.4 ± 0.3a
		5		$25.6\pm0.2b$	$2.6 \pm 0.5a$	$-0.7 \pm 0.1a$		$85.9 \pm 0.3$ ab
		10		$23.6\pm0.2d$	$2.2 \pm 0.4a$	$-0.7 \pm 0.1a$	4.4 ± 0.2a	$85.7 \pm 0.3b$
Fig	Nanomist	0	172.5 ± 10.6a					
		4	$113.9 \pm 4.9c$					
		8	$121.3 \pm 4.4c$					
	Ultrasonic-mist	0	172.5 ± 10.0	50				
		4	$172.3 \pm 10.0a$ 147.0 ± 6.3b					
		8	$170.5 \pm 6.8a$	1				

# Table 4.2 Several quality attributes of three commodities preserved under two environments of nanomist and ultrasonic mist during postharvest storage.

Data are the mean of two experiments (No 1 and No 2 shown in Table 1) except for pulp color of eggplant and stomatal pore of fig and accompanied by the standard error of the means. Pulp color of eggplant and stomatal pore of fig were obtained from experiment No 2. Different letters in one column separated by products show significant difference by statistical programme at P < 0.05.

mizuna and fig, respectively, compared to their initial openings, while in the ultrasonic chamber, they were closed by 15.8  $\mu$ m<sup>2</sup> and 25.5  $\mu$ m<sup>2</sup>, respectively.

The variation in stomatal area between the two treatments could be the reason for the differences in weight loss. Open or partially closed stomata will affect parameter *A*, thereby changing the water loss, as shown in Eq. (4-8). The partial closure of the stomata observed in the nanomist chamber could be due to the following reasons. Lange *et al.* (1971) found that the opening of the stomatal aperture depends on the wetness of the surface of the plant, which suggests that less water on the outer surface of the epidermis results in a closing of the stomata, while air that is nearly saturated leads to maximal opening of the stomata. Furthermore, the structure and function of stomata are described as follows. Stomata are composed of two guard cells. These cells have walls that are thicker on the inner side than the outer side. This unequal thickening of the paired guard cells causes the stomata to open when they take up water and close when they lose water. The opening and closing of stomata is governed by increases or decreases of water in the guard cells, which cause them to take up or lose water, respectively (Masuda, 1989; Taiz and Zeiger, 2003).

In our study, we observed that the stomata of the produce stored in the nanomist closed more than those stored in the ultrasonic mist. The reason may be that under the nanomist environment, the number of nanomist particles on the surface of the produce is smaller than that of the ultrasonic mist. Moreover, the very fine mists present on the surface of the produce are believed to evaporate quickly, therefore not wetting the produce and thus making the stomata close more than those stored in the ultrasonic, where the large mists and their number, which directly drop on the produce and do not easily evaporate, are thought to wet the surface of the samples. The CI index of the eggplant fruits increased by day 5 (Table 4.2). The CI of the fruits stored in the nanomist chamber was lower than that of the ultrasonic mist, and the CI symptoms increased with storage time. Concellon *et al.* (2004) reported that a high CI index ( $CI_i = 4$ ) of eggplant fruits was observed on day 12 at 5 °C, similar to the CI of the eggplant fruits stored in the ultrasonic chamber. Moisture loss has been implicated as the causal factor in the CI of grapefruit (Purvis, 1984) and eggplant fruit (Fallik *et al.*, 1995). Severe CI may induce water loss by the destruction of cuticles and the softening of the tissue. Because the CI was not severe in our experiment, the reduction of CI in the eggplant fruit stored under nanomist is probably due to the reduced water loss from the fruit.

Changes in the color parameters of mizuna, eggplant fruit and the pulp of eggplant during the storage period are shown in Table 4.2. For mizuna, the  $L^*$  value was different between the two storage conditions at the end of the experiment. Moreover, the  $a^*$  and  $b^*$  values showed a significant difference between the samples stored in the two environments. A more negative  $a^*$  (greenness index) value was recorded for the samples stored in the nanomist, which means that the leaves were greener than those stored in the ultrasonic chamber. For eggplant, the lightness of skin ( $L^*$ ) was reduced slightly over the storage period and was not significantly different at day 5, though it was significantly different between the two treatments during storage at the end of experiment. Parameters  $a^*$  and  $b^*$ were not different between the two treatments and were constant throughout storage. The variations in parameter  $L^*$  between the two treatments were attributed to differences in surface CI symptoms. Concellon *et al.* (2007) suggested that CI symptoms are related to variations in the color parameter. The lightness of eggplant pulp ( $L^*_0$ ) was a good indication of the browning evolution of the fruit tissue during cold storage. As shown in Table 4.2, the  $L^*_0$  readings of the fruits stored under nanomist were constant over storage time and were similar to the initial value, which was around 87. However, a significantly lower value was observed in the fruit stored under ultrasonic mist, indicating the browning of seed and pulp. The difference in the lightness of eggplant pulp may be attributed to the difference in chilling injury.

#### **4.4. CONCLUSION**

In this study, the purpose was to investigate the effects of nanomist and ultrasonic mist on the postharvest attributes of mizuna, eggplant and fig fruit preserved under high relative humidity condition. The results of present research show that storing selected vegetables under nanomist environment reduced the weight loss rates (3.6%, 3.2% and 5.9% for mizuna, eggplant and fig, respectively) compared with those stored under the ultrasonic mists. Colour of mizuna leaves stored in the nanomist showed greener than those stored in ultrasonic. Furthermore, the eggplant fruit stored in the nanomist chamber had a lower index of chilling injury than those stored in the ultrasonic. A significant finding of this study is that the stomatal pores of the samples exposed to the nanomists closed much more than those stored in ultrasomic mist. This results in the difference in transpiration of fresh produce stored in two environments. From the results obtained in this study, it is possible to conclude that nanomists are useful for raising humidity to control the weight loss of fresh fruit and vegetables during cold storage or transportation. Further research regarding the impact of nanomists on postharvest diseases of fruit is highly recommended.

# **CHAPTER 5**

# Investigation of postharvest quality of fig (*Ficus carica* L.) fruit stored under nanomist and ultrasonic mist condition with high relative humidity

#### **5.1. INTRODUCTION**

Figs (*Ficus carica* L.) are a nutritious fruit rich in fiber, potassium, calcium, and iron with higher level than other fruit such as apples, grapes and strawberries (Michailides, 2003). Additionally, figs are an important source of vitamins, amino acids and antioxidants (Solomon *et al.*, 2006; Crisosto *et al.*, 2010). Fresh fig fruit is known as one of the most perishable horticultural commodities. The particular structure of the fruit as described by presence of an apical pore (ostiole) makes fig fruit very susceptible to a number of diseases caused by fungi and bacteria. The high metabolic activity and the easiness of pathogen development are the main causes of deterioration (Doster *et al.*, 1996; Piga *et al.*, 1998). The shelf life of fresh fig harvested at fully ripe stage is limited to 2 days under the ambient temperature of the harvest season at the end of summer (Hamanaka *et al.*, 2010).

The quality and storage life of fresh produce are highly dependent upon the difference in vapor pressure between the produce and the storage atmosphere. Hence, when both produce and storage are maintained at the same temperature (assuming factors such as air velocity are held constant), transpiration rate is related to the relative humidity (RH) during storage (Grierson and Wardowski, 1978; Sharkey and Peggie, 1984). Transpiration through the actions of the stoma, lenticels, cuticles and epidermal cells is considered to be the main cause of postharvest weight loss and poor quality in fresh produce (Ben-Yehoshua and Rodov, 2003).

Several studies have been conducted on postharvest storage of fresh fig fruit under controlled atmosphere (Colelli et al., 1991; Kaynak et al., 1998; D'Aquino et al., 2005). The RH used in these studies ranges from 80% to 90%. Taking commodity's transpiration into consideration, high relative humidity up to 95% (Paull, 1999) of the storage environment plays an important role in maintaining the quality of produce. However, one of the problems occurring for the produce stored under high humidity environment is that there is a risk of wetness on the produce surface causing developments of postharvest pathogens (Grierson and Wardowski, 1978). Germination of fungal spores is promoted by the wetness; therefore, presence of condensed water on the fruit surface favorably affects the growth of postharvest diseases (Rodov et al., 2010). Previous studies have demonstrated the techniques of controlling postharvest decay of figs using sodium carbonate (Molinu et al., 2006) and chlorine dioxides (Karabulut et al., 2009). Although these methods were found to be effective in preventing microorganisms' growth, the easy absorption of chemicals by the tissue and their penetration through ostiole has raised public concern about contamination of perishables with chemical residues. Therefore, the proper choice of storage conditions and environmentally friendly technology need to be applied to improve postharvest life of horticultural produce in general and shelf life of fig fruit in particular.

Nanomist humidifier (Mayekawa Co., Ltd, Tokyo, Japan), a state-of-the-art humidification device, is used to produce high humidity in a refrigerated chamber. The mists were generated at nano-scale size, producing a so-called nanomist with average particle mode diameter of 65.5 nm (Hung *et al.*, 2010b). These mists were thought to be significantly smaller than those generated by traditional humidifier which is often termed ultrasonic

mists; their average particle diameters vary between 210 nm (Hung et al., 2010b) and 2.9 µm (Rodes et al., 1990), and much larger than mists produced by tabor atomizer system with droplet size of 10 µm (Afek et al., 2000). Nanomists, because of their very fine particle size, presumably would evaporate immediately following atomisation. This characteristic could bring a number of benefits to the field of postharvest crop storage comparing to the ultrasonic. For example, when nanomists drop on the surface of the produce, they easily evaporate and thus do not wet the produce surface, thereby controlling the deterioration of fresh produce and microorganism development. Hung et al. (2010a) have demonstrated that the strength corrugated cardboard was well-maintained under nanomist environment with high relative humidity in comparison with ultrasonic mist condition. Furthermore, Hung et al. (2011) have suggested that postharvest weight loses of vegetables kept in nanomist condition after a week reduced between 3.2% and 5.9% depending upon commodities as compared to those stored under ultrasonic mists with the same temperature and RH. The objective of present work was to compare the effect of nanomists and ultrasonic mists on postharvest quality of fresh fig fruit stored under high relative humidity environment.

## **5.2. MATERIALS AND METHODS**

#### 5.2.1. Source of fruit

Fig fruit (*Ficus carica* L., cv Toyomitsu-Hime) were purchased from the Asakura agricultural cooperative. A careful handling of the produce was taken during transport to the laboratory and the experiment. The figs were sorted to eliminate any that had defects, and those with a uniform color and a weight of approximately 80 grams each were chosen. The experiments were repeated twice in October and November 2010.

#### 5.2.2. Storage condition

The figs were kept in storage environments at 7 °C and RH at approximately 94% where the nanomist humidifier (Test model, Mayekawa Co., Ltd, Tokyo, Japan) and the ultrasonic humidifier (FT-30N-14, UCAN Co., Ltd, Japan) were used to raise humidity. Forty figs were used for each storage condition. The fruits were placed on cardboard and were directly exposed to the mists in order to investigate the effect of particle size on the quality of produce. A sample of 25 figs was used for assessing color, decay incidence and visual overall quality throughout the experiment. The remainder of samples was used for measuring flesh firmness, total soluble solids and titratable acidity. Determinations were made on day 0, 4 and 8.

The temperature and RH inside the chambers were recorded at 5-minute intervals in the containers using a humidity and temperature transmitter (model HMT337, Vaisala, Helsinki, Finland). This device can measure the accuracy at the temperature to  $\pm 0.2$  °C and the RH to  $\pm 1.7$  % at a range of 90 % to 100%.

### 5.2.3. Methods for parameter assessment

#### Firmness

Flesh firmness determination was measured at the equatorial region of the fruit using a rheometer (RE-3305 Rheoner, Yamaden, Tokyo, Japan) with a 7.9 mm cylindrical probe. The fruit was cut in half. Immediately after removal of the skin, each half was placed on a stationary plate and was punctured to a depth of 10 mm. The probe descended toward the sample at crosshead speed of 1 mm/s and the maximum value of force was expressed in

Newton (N). The mean of the two measurements was considered as one replicate. Six fruit per treatment and date were evaluated.

#### Decay incidence

The fruits with visible mold development were expressed as percentage of fruit showing decay symptoms and decay severity was assessed according to a 1-5 scale, where 1 = no visible mould, 2 = one lesion less than 1 cm in diameter, 3 = one lesion between a diameter of 1-2 cm, 4 = one lesion larger than 2 cm but smaller than 3 cm or two lesion each larger than 1 but smaller than 2 cm, and 5 = one lesion larger than 3 cm or more than three lesions. Results were expressed as decay incidence index.

## Overall quality

All fruit were evaluated for overall quality on a 1-5 scale as described by Colelli et al. (1991) with minor modifications, where 1 = unacceptable, 2 = fair (limit of usability), 3 = acceptable (limit of marketability), 4 = good, and 5 = excellent. Results were expressed as an overall quality index.

#### Color

The color of fig fruit was measured using a chromameter (Minolta CR 200, Japan). The measurements were expressed as lightness ( $L^*$  darkness to lightness, on a scale of 0–100), chroma (C, indicating intensity or saturation of the color), and hue (H, angle that indicates the pure spectrum color). Measurements were taken from two spots located on opposite sides of the equatorial region of each fruit and the average of the values for each fruit was calculated.

#### Total soluble solids (TSS) and titratable acidity (TA)

The fruit were wrapped in soft gauze No 3 (Hayashi Eizai Co. Ltd., Fukuoka, Japan), squeezed by hand, and the expressed juice was used for measurements of TSS and TA. TSS was determined at room temperature using a digital refractometer PR-101 $\alpha$  (Atago Co. Ltd., Tokyo, Japan) and expressed as <sup>o</sup>Brix. Titratable acidity was measured using titration method. One mL fruit juice was added to 9 mL distilled water plus a drop of phenolphthalein and titrated with 0.1N NaOH. The results were expressed as the percentage of citric acid.

#### 5.2.4. Statistical analysis

The experimental data are presented as the mean  $\pm$  SE of two independent experiments. An analysis of variance was performed using the statistical software GenStat (Discovery Version 3, VSN International Ltd., UK). Differences between the means of attributes were compared by a least significant difference (LSD) test at a significant level of 0.05.

#### **5.3 RESULTS AND DISCUSION**

#### 5.3.1. Temperature and relative humidity

The average temperature and relative humidity monitored from the nanomist and the ultrasonic chambers were nearly the same and being  $7.1 \pm 0.5$  °C and  $93.8 \pm 4\%$ , and  $7.1 \pm 0.5$  °C and  $93.8 \pm 5.8\%$ , respectively.

#### 5.3.2. Color and firmness

Fig. 5.1 shows changes in relative values of color parameters of fig fruit during a period of storage. In general, there was a reduction in the value of color parameters over time. Significant differences were observed in lightness, chroma and hue angle of fruit stored under the nanomist and the ultrasonic mist at the end of experiment. Color of fruit stored under the ultrasonic mist became darker, as reflected by a decrease in  $L^*$  value and showed less intensive (lower *C* value) and more red (smaller *H*) during the storage period. The flesh firmness is shown in Fig. 5.2. Significant difference in firmness of fruit between two conditions was detected after 8 days of storage. Fruit stored under ultrasonic mist became softer than those under nanomist.

### 5.3.3. Total soluble solids (TSS) and titratable acidity (TA)

Total soluble solids of figs at the harvest time were around 16 <sup>o</sup>Brix (Fig.5.3). No significant difference in TSS between these two treatments was found at day 4, but significant difference was detected at day 8. Previous study demonstrated that after 8 days of cold storage, weight loss rate of the figs stored in the nanomist was 6% lower than those stored in the ultrasonic mist. The difference in weight loss rate between two treatments may be the reason causing difference in TSS.



Fig. 5.1 Changes in relative values of the lightness, chroma and hue angle of figs stored at 7  $^{\circ}$ C and 94% RH under two environments during postharvest storage. Different letters show significant difference by statistical program at *P*<0.05



Fig. 5.2 Firmness of figs stored at 7  $^{\circ}$ C and 94% RH under two environments during postharvest storage. Different letters show significant difference by statistical program at *P*<0.05



Fig. 5.3 Total soluble solids of figs stored at 7 °C and 94% RH under two environments during postharvest storage. Different letters show significant difference by statistical program at *P*<0.05

The titratable acidity (TA) calculated as citric acid is presented in Fig. 5.4. The initial TA value was 0.34%. There was a slight increase in the acidity throughout storage, but it increased substantially in the fruit stored under ultrasonic mist at the end of experiment. No significant difference was found at day 4, but significant difference between two conditions was detected at day 8. An increase in TA was influenced by the weight loss which favored the concentration. Similar results were reported in the study on "Craxiou de porcu" figs by D'Aquino *et al.* (1998).

#### 5.3.4. Decay incidence and visual quality

The molds usually originated in the ostiole and skin cracks and then started to spread over the entire fruit. The number of figs showing visible mould development under the nanomist was 18% and 32% at days 4 and 8, respectively while it was 46% and 66% under the ultrasonic mist during the same period (Fig 5.5a). With regard to mold severity, decay index was found to be significant between two treatments at day 8 (Fig. 5.5b). Decay severity of the fruit stored under the ultrasonic mist occurred more severely than those stored under the nanomist. The reason could be related to the mist size. Due to very fine particle size, when the nanomists are present on the surface of the fruit, fine mists are believed to rapidly evaporate and therefore do not wet the fruit surface, thereby preventing microorganism development. On the other hand, with larger mists and their higher number, the ultrasonic mists are assumed to directly drop on the fruit and do not easily evaporate and thus bring about wetness on the fruit surface. The wetness in the ostiole may induce germination of pathogen spore (Karabulut *et al.*, 2009). The findings of present study are in agreement with that of Afek et al. (2000) who investigated that small droplet size can generate 96% RH without depositing free water on the produce surface while with larger droplets once the RH increases to 94%, free water starts to accumulate on the produce, making them susceptible to diseases.

From a marketing perspective, visual appearance is a critical feature of fig quality. As can be seen in Fig. 5.6, figs stored under the nanomist showed a better overall appearance than those kept in the ultrasonic mist for the entire duration of experiment. The major causes resulting in reduction of visual quality of the fruit were water loss and the onset of fungal decay.

#### **5.4. CONCLUSIONS**

In this study, the aim was to compare the effects of nanomists and ultrasonic mists on the postharvest quality of fig fruit preserved under high relative humidity condition. The results of present work show that in comparison with ultrasonic mists, preserving fresh fig fruit under a nanomist environment reduced fruit softening and decay incidence, thereby improving overall visual quality. In addition, the postharvest quality attributes such as color, total soluble solids and titratable acidity were better maintained by nanomists during postharvest storage. From these results, it is possible to conclude that nanomist humidifier is a useful tool to generate mists for raising humidity during cold storage or transport of fresh fruit.



Fig. 5.4 Titratable acidity of figs stored at 7 °C and 94% RH under two environments during postharvest storage. Different letters show significant difference by statistical program at P<0.05



Fig. 5.5 The percentage of infected fruit (a) and decay index of figs (b) stored at 7  $^{\circ}$ C and 94% RH under two environments during postharvest storage. Different letters show significant difference by statistical program at *P*<0.05



Fig. 5.6 Overall quality index of figs stored at 7  $^{\circ}$ C and 94% RH under two environments during postharvest storage. Different letters show significant difference by statistical program at *P*<0.05

# **CHAPTER 6**

# **Summary and conclusions**

The studies presented in this thesis were performed to explore the particle size distribution of mists produced by nanomist and ultrasonic humidifiers operating at high relative humidity storage environment and investigate the effects of nanomist on the strength of corrugated cardboard boxes and on the preservation of postharvest quality of fresh produce stored under high relative humidity condition. The summary and conclusion for each chapter are addressed below.

In chapter 2, the investigation of characteristics of mists formed by nanomist and ultrasonic humidifiers in storage environments with high humidity was made. The results showed that the number mode diameter of nanomist humidifier at 60 Hz was 65.5 nm, about 3 times finer than that produced by the ultrasonic mist. Moreover, particle number concentration of the nanomist was 4 times lower than that of the ultrasonic mist. It was also shown that the mist number concentration was very much dependent on generator frequency and the relative humidity. From these results, it can be concluded that nanomist evaporates more easily than ultrasonic mist. In addition, the Nukiyama-Tanasawa equation estimated the size distribution better than the Rosin-Rammler equation.

In chapter 3, the effects of particle size on the moisture content and compression strength of corrugated cardboard under high relative humidity condition were examined. The results of present research show that relative humidity affected moisture content of corrugated cardboard. The moisture content of both specimen and cardboard box tests exposed to the nanomist and ultrasonic-mist at the end of experiments was 19.9% d.b. and 30.4% d.b.,

respectively (dry basis: g-water in material/ g-dry weight). One of the more significant findings from this chapter is that the ultrasonic mist raised a moisture content of corrugated cardboard as compared with nanomist at the same humidity. Furthermore, there was a good correlation between moisture content and compressive strength of corrugated cardboard and the cardboard was weakened with the increase in the moisture content. From these results, it can be concluded that nanomist is able to preserve the strength of corrugated cardboard during long cold storage of fresh fruit and vegetables.

In chapter 4, the purpose of the research was to investigate the effects of nano- and ultrasonic-mists on the postharvest attributes of fresh fruit preserved under high relative humidity condition. The postharvest quality of three types of horticultural produce, eggplant fruit (Solanum melongena), mizuna (Brassica rapa) and fig fruit (Ficus carica), was investigated under storage environments of two kinds of fine mists producing relative humidity as high as 95% at 5.5 °C and 7 °C for 10, 6 and 8 days, respectively. The results of present research showed that storing fresh fruit and vegetables in the nanomist chamber reduced the weight loss rates (3.6%, 3.2% and 5.9% for mizuna, eggplant and fig, respectively) compared with those stored under the ultrasonic mists. Furthermore, the eggplant fruit stored in the nanomist chamber had a lower index of chilling injury than those stored in the ultrasonic. The stomatal pores of the samples exposed to the nanomists closed by 34.7  $\mu$ m<sup>2</sup> and 51.5  $\mu$ m<sup>2</sup> for mizuna and fig, respectively, compared with their initial openings, while in the ultrasonic mists, they closed by 15.8  $\mu$ m<sup>2</sup> and 25.5  $\mu$ m<sup>2</sup>, respectively. This induces the difference in transpiration of fruit stored in nanomist and ultrasonic mist. The colour of mizuna leaves stored in the nanomist was greener than those placed in the ultrasonic mist during the postharvest storage. From these results, it is possible to conclude that nanomists are useful for preserving postharvest quality of fresh fruit and vegetables during storage and distribution.

In chapter 5, the aim of this work was to compare the effects of nanomists and ultrasonic mists on the postharvest quality of fig fruit preserved under high relative humidity condition. The two experiments were performed to store fig at 7 °C and 94% RH for 8 days. The results of present work show that in comparison with ultrasonic mists, preserving fresh fig fruit under a nanomist environment significantly reduced fruit softening and decay incidence, thereby maintaining overall visual quality. In addition, the postharvest quality attributes such as color, total soluble solids and titratable acidity were better preserved by nanomists during cold storage. Based on the obtained results, it is likely to state that nanomist humidifier is a useful tool that can be used to preserve the quality of fresh fruit during postharvest storage.

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