



Early cell enlargement by night-time heating of fruit produce watermelon fruit (*Citrullus lanatus* Matsum. et Nakai) with high sucrose content

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ABSTRACT

To investigate the effects of night-time temperature on cell and fruit size, and sugar accumulation in watermelon fruit, fruits were treated with high night-time temperatures in a greenhouse. The minimum night-time ambient temperature of the heating box (18 °C) was approximately 6 °C higher than that of the control. The length, diameter and weight of heat-treated fruit at the end of heating treatment, 16 days after anthesis (DAA), were significantly greater than that of control fruit, but those at harvesting, 42 DAA, were almost the same in both treatments. Mean cell size of the outer portion of heat-treated fruit at 16 DAA was significantly larger than that of the control. Cell size of the fruits at 42 DAA did not differ between heat-treated and control fruits. Sucrose, glucose and fructose content of fruit at 16 DAA did not differ between heat-treated and control fruit. However, sucrose content of the outer portion of heat-treated fruit was 162% of that of control fruit at 42 DAA. Glucose and fructose contents at 42 DAA did not differ between heat-treated and control fruit, except glucose content of outer portion.

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1. Introduction

Watermelon is one of the most important vegetables grown in Ishikawa Prefecture, Japan. In watermelons grown in plastic film greenhouses during spring and shipped in early summer, only the central portion, 10 cm in diameter, is red and sweet; the portion extending from the central portion to the white rind, which is also red, has a lower sugar content. In this type of cultivation (sowing time, March; harvest time, June) night-time temperature in plastic film greenhouses, but not day-time temperature, during early fruit development declines to nearly 10 °C. Glucose and fructose contents in watermelon fruit is almost constant during fruit development, but sucrose content increases rapidly at 30 days after anthesis (DAA) and reaches a maximum at harvest (Elmstrom and Davis, 1981; Kano, 1991). These results show that sucrose content is the most important factor determining watermelon sweetness.

Therefore, we surmise that the reason for the shortage of sweetness in the outer portion of watermelon fruit grown in plastic film greenhouses during spring is the low sucrose content of the outer portion owing to low night-time temperatures during early fruit development stage. On the other hand, rapid cell enlargement at the early fruit development by heating fruit (Kano, 2006) or

treating fruits with auxin (Kano, 2002) followed by active sucrose accumulation in melon fruit shows a preferential accumulation of sucrose in large cells at an early stage. These results suggest that cell enlargement at early stage induces active sucrose accumulation in larger cells, resulting in higher sucrose content. Therefore, watermelon fruits were treated with high night-time temperature in this experiment for the purpose of promotion of cell enlargement and subsequent sucrose accumulation in fruits.

2. Materials and methods

2.1. Plant materials

Watermelon (*Citrullus lanatus* Matsum. et Nakai, cv. Tsukuba no Kaori) was grown in a plastic film greenhouse at Ishikawa Agricultural Research Center, located near Nonoichi Town in Ishikawa Prefecture. Plants were set 40 cm apart in rows spaced 300 cm apart in a 70 m × 10 m vinyl house. Fourteen fruits for both the heated and control treatments were selected from alternate plants on a single bed at the center of the vinyl house. Pinching was done at the 4-true-leaf stage of the primary vines, and 2 secondary vines were kept. On 17 May 2007, flowers set at the 20th node of each vine were pollinated by hand. One fruit was left on each plant. One fruitlet was put into a 30-cm cube wooden box (Fig. 1) from 23 May to 1 June.

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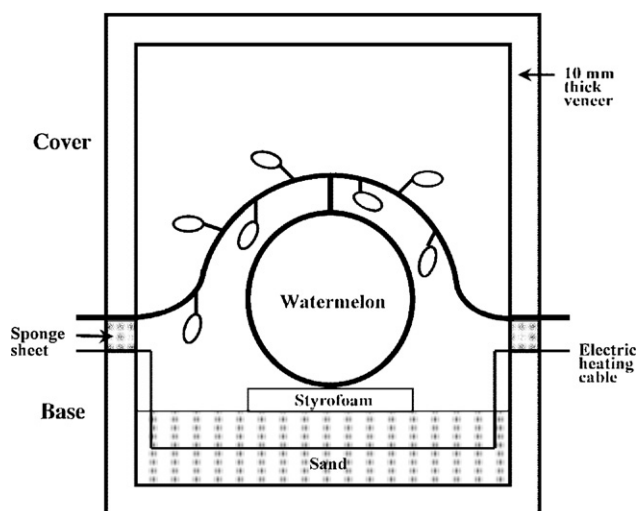


Fig. 1. Schematic diagram of a heating apparatus for watermelon fruit.

2.2. Heating apparatus and temperature measurement

A 30-cm cube box (Fig. 1) was cut 10 cm from the base, and heating elements were set in the lower third for heating the fruit. Sand was poured to a depth of 3 cm in the bottom of the box and an electric cable to create a hotbed (equivalent to 7.7W) was set on the sand and covered with a further 2 cm of sand. As cell division in watermelon fruit ceases at 6 DAA (Kano, 1993), watermelon fruits were heated after 6 DAA to examine the effects of heat treatment on cell enlargement. One fruit was set on the lower third of the box and covered with the upper two-thirds of the box, and the electric current was turned on from 5 p.m. to 7 a.m. each day for 10 days from 23 May (6 DAA) to 1 June (16 DAA) (designated as heat treatment). Control fruits were grown in a plastic film greenhouse without a heating box. Temperatures inside and outside one of the fruit heating boxes, located away from the extremities of the greenhouse, were measured using a thermo recorder equipped with thermistors, called “Ondotori” in Japanese (TR-71S, T and D Inc., Matsumoto, Japan), from 6 DAA (23 May) to 16 DAA (1 June). Temperatures were recorded every hour throughout the whole day during heat treatment.

2.3. Measurements of cell size in the fruit

Seven fruits were harvested at 16 DAA (1 June) and 42 DAA (27 June), respectively. After calculation of mean fruit weight at 16 DAA and 42 DAA, fruits were used for cell and sugar analysis. Two disks 20 mm thick were cut from 7 fruits for each treatment. One disk was taken from the maximum transverse diameter toward the calyx end for cell analysis and the other from the maximum transverse diameter toward the peduncle end for sugar analysis. Using a sharp table knife, 1 strip, a sample measuring approximately 10 mm × 10 mm, was removed from the disk, centered on the diameter line of each disk (Fig. 2). Rectangular parallelepipeds (RPs), each measuring approximately 15 mm, were serially sampled across the diameter of the disk using the same sharp table knife. The 3rd and 4th RPs from the left and right ends, and 2 central RPs of the fruits 16 DAA and 42 DAA, were used for cell analysis. The RPs were dehydrated using a graded ethanol series (70%, 80%, 90% and 100%), and were embedded in paraffin. Seven sections 10 μm thick were prepared from these paraffin blocks, and the clearest section from each sample for each treatment was examined under a microscope. Approximately 49 and 38 cells per sample of 16- and 42-day fruits were investigated, respectively. The maximum diam-

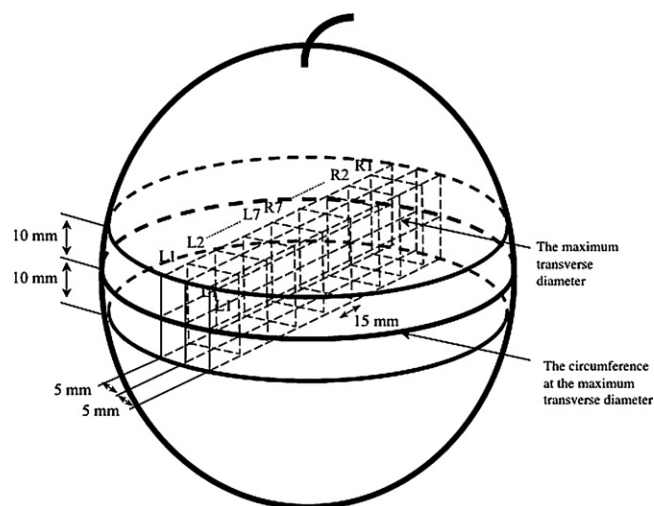


Fig. 2. Illustration of the collection of rectangular parallelepipeds (L1–R1) for the determination of cell size and sugar content in watermelon fruit. This is an example of untreated fruit at 42 days after anthesis.

eter (designated as cell size, shown by arrow lines) of individual cells from the maximum transverse diameter in each sample was measured (Figs. 2 and 3). Mean cell size of all the portions (L3, L6, R5, R3 of control and L3, L7, R7, R3 heated at 16 DAA; L4, L8, R8, R4 of control and heated at 42 DAA; refer to Fig. 2) was determined.

2.4. Sugar analysis

The other strips, each measuring approximately 15 mm, were serially sampled across the diameter of the disk using the same sharp table knife. All of the RPs except outermost 4 RPs were wrapped in cheesecloth and squeezed using pincers to extract the juice. The juice was diluted 10 times with distilled water. The solution was centrifuged at 8000 × g for 15 min before being filtered through a 0.45 μm PTFE hydrophilic filter (Millipore, Bedford, MA, USA). Twenty microliters of the filtrate was injected into an HPLC apparatus (LC-10ADvp, Shimadzu Inc., Kyoto, Japan) fitted with a refractive index detector (RID-10A, Shimadzu Inc.) and a column (Shim-pack SCR-101C, Shimadzu Inc.) at 0.8 ml min⁻¹ and 80 °C. To determine the concentration of each sugar, 20 μl of a standard solution of sucrose, glucose and fructose (each at 20 g l⁻¹) were injected into the HPLC apparatus before injection of the filtrates. Mean sugar content at the outer portion (L3, L4, R3, R4 of control and heated at 16 DAA and 42 DAA; refer to Fig. 2) and the central portion (L6, R5 of control, and L7, R7 of heated at 16 DAA, and L8, R8 at 42 DAA; refer to Fig. 2) was determined.

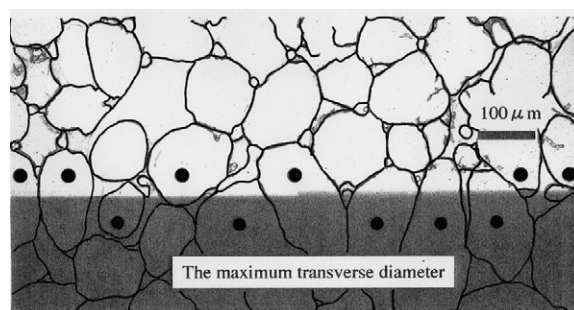


Fig. 3. Illustration of the measurement of cell size of watermelon fruits. Black dots indicate the actual cells measured.

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