Inactivation of polyphenol oxidase from watermelon juice by high pressure carbon dioxide treatment

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Abstract The inactivation of polyphenol oxidase from watermelon juice with high pressure carbon dioxide (HPCD) treatment was investigated. The maximum reduction of polyphenol oxidase (PPO) activity inactivated by HPCD treatment was 95.8% at 30 MPa and 50 °C for 30 min, which was far higher than 50.9% of control treatment at 50 °C for 30 min. The inactivation of PPO was adequately described by a two-fraction model, which indicated that a labile and stable fraction might present in PPO from watermelon juice. The kinetic rate constants k_L and k_S of labile and stable fractions were 1.976 and 0.041 min^{-1} by HPCD treatment of 30 MPa and 50 °C. And the labile fraction was easier to be inactivated by kinetic analysis. HPCD treatment with the combined effects of pressure, temperature, pH reduction, and time was stronger to inactivate PPO from watermelon juice than control treatment at the same temperature.

Keywords Polyphenol oxidase · High pressure carbon dioxide (HPCD) · Watermelon juice · Inactivation · Kinetic analysis

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Watermelon is one of the most popular fruits around the world. Most of the watermelons are eaten freshly and others are processed to juices or fresh-cut products. However, enzymatic browning, which is mainly caused by a polyphenol oxidase (PPO), leads to a deleterious change in sensory properties of watermelon products (Jayaraman et al. 1982; Walker and Ferrar 1998). Traditionally, thermal treatments were used to inactivate PPO (Yemenicioğlu et al. 1997; Matsui et al. 2007; Soysal 2008; Fortea et al. 2009; Bakshi and Masoodi 2010). However, the thermal treatment leads to destruction of heat-sensitive nutrients and quality of food inevitably, such as: texture, color, and flavor (Flora 1976; Hayakawa et al. 1977; Paulus and Saguy 1980; Chawla and Ranote 2009).

High pressure carbon dioxide (HPCD) treatment, a non-thermal technology, attracts more and more interests as the potential alternative or complementary processes to the thermal process in food industry (Ross et al. 2003; Chawla et al. 2011). HPCD has been used to inactivate microorganisms and enzymes effectively (Hong and Pyun 1999; Corwin and Shellhammer 2002; Liao et al. 2007; Liu et al. 2008; Huang et al. 2009; Liu et al. 2010) without destroying the fresh-like sensory, nutrients, and physical properties of liquid foods (Damar and Balaban 2006). For example, HPCD treatment has been used to inactivate the pectinesterase in orange juice (Balaban et al. 1991) and apple juice (Zhi et al. 2008); alkaline protease and lipase in the aqueous solution (Ishikawa et al. 1995); lipoxygenase and peroxidase in sucrose solutions (Tedjo et al. 2000). Specially, PPOs from lobster, brown shrimp, potato, apple juice, and red beet are also inactivated by HPCD treatment, which residual rates are far lower than that of the thermal treatment (Chen et al. 1992; Gui et al. 2007; Liu et al. 2008). Hence, HPCD treatment inactivated PPO more effectively, comparing with the thermal treatment at the same temperature.

PPO has the different composition in different plants. PPO from bean sprouts has two isoenzymes (Nagai and Suzuki 2003); PPO from high L-DOPA containing velvet bean and *Duranta plumieri* has three fractions (Zenin and Park 1978; Roy et al. 2002); PPO in avocado is composed of six isoenzymes (Kahn 1976). The inactivation of PPO by HPCD treatment and the composition of PPO from watermelon juice has not been investigated to our knowledge. Consequently, we try to evaluate the effect of HPCD treatment on inactivation of PPO by kinetic analysis, and the comparison of inactivation capacity between HPCD and the thermal treatment.

Materials and methods

Preparation of watermelon juice The watermelon (Jingling) was harvested maturely in Yanqing farm. The watermelons were washed, peeled, and crushed with a blender (Philips, HR2860, Zhuhai, China). The homogenous solution was filtrated by gauzes. The filtrate was subjected to the following treatments.

HPCD process system The diagram of the HPCD system was described by Liao et al. (2007), as shown in Fig. 1. The stainless steel pressure vessel with a volume of 850 ml was designed to withstand a pressure of 50 MPa. The vessel temperature was maintained by a THYS-15 thermostatic bath (Ningbo Tianheng Instrument Factory, Zhejiang, China). An XMTA-7512 temperature controller (Yuyao Temperature Meter Factory, Zhejiang, China) was used to monitor the temperature with two thermocouples. One thermocouple was fixed in the vessel lid to monitor the CO₂ temperature in the upper part of the vessel and the other was placed at the middle wall of the vessel to monitor

the temperature of watermelon juice in the vessel. A 2TD plunger pump (Huaan Supercritical Fluids Extraction Co. Ltd., Jiangsu, China) with a maximum pressure of 50 MPa and a maximum flow rate 50 l/h was used to pressurize the vessel. A DBY-300 pressure transducer (Shanxi Qingming Electronic Group Corporation, Shanxi, China) was fixed in the vessel lid to monitor the vessel pressure. All the data of temperature and pressure were displayed on a control panel. All parts of the system exposed to high pressure were made of stainless steel. The vessel had gas-tight connections to the gas inlet and outlet. The vessel lid could be sealed by screws during HPCD processing. Commercially available CO₂ of 99.5% purity was purchased from Beijing JingCheng Co. (Beijing, China), and was passed through an active carbon filter before entering the pressure vessel.

Control treatment of watermelon juice The watermelon juice (20 mL) in 50 mL plastic tube was subjected to water bath of 30, 40, and 50 °C for 2.5, 5, 10, 20, and 30 min at atmospheric condition, respectively. The heating time was recorded after the temperature of watermelon juice reached the desired temperatures with a short come-up time. After the treatment, the juice was cooled down in an ice-water bath for further analysis.

HPCD treatment of watermelon juice The watermelon juice (20 mL) in 50 mL plastic tube was placed in the vessel which had been preheated to the experimental temperature, and then pressurized by CO₂. The sample was held at constant pressure and temperature during the treatment. At the end of treatment, the vessel was rapidly depressurized. After treatment, the watermelon juice was removed and immediately cooled in an ice bath. The residual activity and pH value were determined after equilibrating to ambient temperature.

Fig. 1 Diagram of Dense Phase Carbon Dioxide Processing Equipment 1 CO₂ cylinder; 2 CO₂ filter; 3 Pressure gauge; 4 Cooling unit; 5 Plunger pump; 6 Pressure transducer; 7 Biohazard clean safety facility for aseptic operation; 8 High pressure vessel; 9 Thermocouples; 10 Thermostatic bath; 11 Vacuum pump; 12 Displaying panel

Four levels of the pressure (8, 15, 22, and 30 MPa) were evaluated at 50 °C and 3 levels of temperature (30, 40, 50 °C) evaluated at 30 MPa. For each pressure and temperature, the juice was treated for 2.5, 5, 10, 20, and 30 min, respectively.

Determination of PPO activity The watermelon juice was stirred with 1% polyvinyl polypyrrolidone (Sigma-Aldrich Co., Shanghai, China) and 1% Triton X-100 at 4 °C for 1 h, and centrifuged at 10,000×*g* (Sigma, 3–18 sartorius, Germany) and 4 °C for 20 min. The supernates were prepared for PPO assay. PPO activity was assayed according to the method described by Duangmal et al. (1999) with minor modifications. Aliquots of the supernate (0.6 mL) were mixed with 2.4 mL substrate solution [100 mM catechol in 100 mM citrate buffer (pH 5.4)] and measured at 420 nm (Unico, UV-3802, Shanghai, China). The initial velocity of each reaction was calculated from the linear portion of the absorbance–time curve and taken as the enzymatic activity. One unit of PPO activity was defined as the change in absorbance of 0.001 min^{−1} at 420 nm and per milliliter of watermelon juice. The residual activity of PPO was calculated according to Eq. 1.

$$\text{Residual activity} = \frac{\text{PPO activity treated with HPCD} \times 100\%}{\text{PPO activity before HPCD}} \quad (1)$$

Kinetics analysis Inactivation kinetics of PPO from the watermelon juice was fitted by a 2-fraction model (Castro et al. 2006; Nunes et al. 2006). This model took into account the existence of two isoenzymes, labile and stable fraction, and the stable fraction was more resistant to temperature and/or pressure than the labile one. Both fractions were considered to be inactivated according to a first-order model, being independent of each other. There was a fast inactivation period followed by a decelerated decay, the total activity decay was described by Eq. 2.

$$A = A_L \exp(-k_L t) + A_S \exp(-k_S t) \quad (2)$$

where A_L and A_S refer to the activity of the labile and stable fractions in relation to total enzyme inactivation, respectively; k_L and k_S are the inactivation rate constants of the labile and stable fraction (min^{−1}), respectively.

The decimal reduction time (D value) defined as the treatment time needed for 90% inactivation of initial activity of PPO at a given condition was obtained.

$$D = \frac{2.303}{k} \quad (3)$$

The pressure and temperature increase needed for a 90% reduction of D value are reflected by Z_P (MPa) (pressure

sensitivity parameter) and Z_T (°C) (temperature sensitivity parameter), respectively, evaluated by Eqs. 4 and 5:

$$\log \left[\frac{D_1}{D_2} \right] = \frac{P_2 - P_1}{Z_P} \quad (4)$$

$$\log \left[\frac{D_1}{D_2} \right] = \frac{T_2 - T_1}{Z_T} \quad (5)$$

The pressure- and temperature-dependence of k can be expressed by the activation volume (V_a , cm³/mol) and activation energy (E_a , kJ/mol), evaluated by Eqs. 6 and 7.

$$\ln \left[\frac{k_1}{k_2} \right] = \frac{V_a}{RT} [P_2 - P_1] \quad (6)$$

$$\ln \left[\frac{k_1}{k_2} \right] = \frac{E_a}{R} \left[\frac{1}{T_2} - \frac{1}{T_1} \right] \quad (7)$$

where P_2 and P_1 , T_2 and T_1 are pressures and temperatures corresponding to the decimal reduction times D_1 and D_2 or constant k_1 and k_2 , respectively. R is the gas constant; T is the absolute temperature (K). The values of Z_P and Z_T are obtained as the negative reciprocal slope of the regression line representing $\log D$ versus P and T relationship, respectively. The activation energy and V_a are estimated from linear regression of $\ln k$ versus $(1/T)$ and P , respectively.

Determination of pH The pH value of the sample was measured at 25 °C with a Thermo Orion 868 pH meter (Thermo Fisher Scientific, Inc., MA, U.S.A.), which was calibrated with pH 4.0 and 7.0 buffer.

Statistical analysis Analysis of variance (ANOVA) and Duncan's multiple range tests were carried out by using the software SAS (SAS Institute Inc., Cary, NC, U.S.A.). The ANOVA test was performed for all experimental run, to determine the significance at 95% confidence interval. All experiments were performed in triplicates.

Results and discussion

Effect of control treatment on PPO residual activity The influence of control treatment on PPO residual activity of the watermelon juice is shown in Table 1. In general, PPO residual activity decreased with the treatment time increasing at the same temperature. Moreover, it decreased with the temperature increasing at the same treatment time. The minimum of PPO residual activity was 49.1% at 50 °C for 30 min by control treatment.

Table 1 Effect of control treatment on polyphenol oxidase (PPO) residual activity (%) from the watermelon juice

Treatment time (min)	PPO residual activity (%)		
	30 °C	40 °C	50 °C
2.5	79.8±4.62 ^b	69.7±3.48 ^b	60.1±0.76 ^b
5	77.6±1.32 ^b	58.8±3.80 ^c	59.7±0.76 ^b
10	71.9±3.04 ^c	62.3±1.52 ^c	56.6±1.32 ^b
20	67.6±2.01 ^c	59.2±1.32 ^c	52.6±2.63 ^c
30	68.0±2.01 ^c	51.8±2.01 ^d	49.1±0.76 ^d

All data represent means±SD, $n=3$.

Within the same column, the different letters mean the values are not significantly different at $P<0.05$.

The initial PPO activity is 100.0±3.48 a (%)

For most enzymes studies, thermal inactivation could be described by a first-order reaction (Gui et al. 2007; Liu et al. 2008; Yu et al. 2010). Dimick et al. (1951) reported that the inactivation of PPO from purees of Bartlett pears by heating confirmed a first-order reaction. Gui et al. (2007) studied that a first-order reaction could well describe the inactivation of PPO in the cloudy apple juice by thermal treatment under atmospheric conditions. Soysal et al. (2008) also showed that thermal inactivation of apple PPO at 45, 55, 65 and 75 °C with 4-methyl catechol as substrate was fitted by a first-order reaction model. However, inactivation of PPO of watermelon juice by control treatment was well fitted by a two-fraction model with the regression coefficient higher than 0.857 (Table 2). The kinetic parameters of inactivation by control treatment were shown in Table 2. The similar inactivation effect is also shown in PPO residual rate of kiwifruit (Park and Luh 1985) by the thermal treatment. PPO residual activity and treatment time presented a nonlinear relationship. Discrepancy was due to that enzymes of different sources had different characteristics.

Effect of HPCD treatment on PPO residual activity The effect of HPCD treatment on PPO residual activity is shown in Figs. 2 and 3. PPO residual activity decreased with the

treatment time at the same temperature and pressure. Furthermore, PPO residual activity decreased with the temperature at the same pressure.

PPO residual activity of watermelon juice by HPCD treatment was well fitted by a two-fraction model with the regression coefficient higher than 0.970 (Table 3). Consequently, PPO of the watermelon juice consisted of the labile and stable fractions. The similar inactivation effect was shown in PPOs of lobster, shrimp and potato (Chen et al. 1992) by HPCD treatment. However, the different phenomena presented in some other investigations. The inactivation of PPO in cloudy apple juice exposed to HPCD treatment followed first-order reaction kinetics (Gui et al. 2007). The HPCD inactivation process of PPO in red beet extract followed first-order kinetics (Liu et al. 2008). And heat inactivation of Crimson seedless grape PPO also was well fitted by first-order kinetics in the reaction medium contained 10 mM sodium acetate buffer (pH 3.0), 2.5 mM TBC and 2.6 µg/mL (Fortea et al. 2009). The discrepancy among these investigations might result from two aspects. On one hand, the different sources of PPOs led to the different change trends of residual activities. On the other hand, different reaction medium was the other reason to affect the inactivation. The reaction medium in this study and Gui et al. (2007) were different juice system, and it was water system in the study of Liu et al. (2008), while the inactivation of Crimson seedless grape PPO was carried out in buffer. The deduce was in accordance with the study of Saraiva et al. (1996), which reported that pH, enzyme concentration, and ions concentration affected the kinetic patterns.

The inactivation rate constants and decimal values of PPO from the watermelon juice were described as k_L , D_L (labile fraction) and k_S , D_S (stable fraction), respectively (Tables 2 and 3). D_L of control treatment was 15.7, 14.6, and 12.7 min at 30, 40, and 50 °C, and D_S was 1785.3, 719.7, and 531.9 min, respectively. Similarly, D_L was smaller than its corresponding D_S at each temperature and pressure after HPCD treatment, suggesting that the labile fraction could be inactivated easily. Moreover, D_L and D_S decreased after HPCD treatment at the same temperature compared to control treatment, which was in

Table 2 Kinetic parameters for inactivation of polyphenol oxidase (PPO) from the watermelon juice by control treatment based on the two-fraction model

T (°C)	k_L (min ⁻¹)	k_S (min ⁻¹)	D_L (min)	D_S (min)	R^2 ($P<0.05$)	$Z_{T,L}$ (°C)	$Z_{T,S}$ (°C)	$E_{a,L}$ (kJ/mol)	$E_{a,S}$ (kJ/mol)
30	0.147	0.001	15.7	1785.3	0.857				
40	0.158	0.003	14.6	719.7	0.983	215.1	38.0	8.7	49.6
50	0.182	0.004	12.7	531.9	0.957	$R^2=0.964$	$R^2=0.923$	$R^2=0.960$	$R^2=0.932$

Effect of pressure of HPCD treatment on PPO Z_p and V_a were estimated by Eqs. 4 and 6. Z_p represents the pressure increase needed for a 90% reduction of D value, and V_a means the pressure-dependence of k . The pressure sensitivity parameters and activation volume of the labile and stable fractions of PPO from watermelon juice were described as $Z_{p,L}$, $Z_{p,S}$ and $V_{a,L}$, $V_{a,S}$, respectively (Table 3). The labile fraction exhibited higher $Z_{p,L}$ (22.8 MPa) and lower absolute value of $V_{a,L}$ ($-271.7 \text{ cm}^3/\text{mol}$) than those ($Z_{p,S}=20.1 \text{ MPa}$ and $V_{a,S} = -307.9 \text{ cm}^3/\text{mol}$) of the stable fraction. Consequently, the stable fraction of PPO was more susceptible to pressure than the labile one. In previous studies, Z_p of PPO in red beet and apple juice is 125.0 and 66.7 MPa after HPCD treatment at 55 °C (Liu et al. 2008; Gui et al. 2007). PPO from the watermelon juice was easier to be inactivated than that of apple juice (Gui et al. 2007), because the treatment conditions and juice system were similar. The absolute value of V_a of PPO from the watermelon juice was higher than that of previous studies. For example, V_a of the labile fraction of purified pepper pectin methylesterase (20 mM citrate buffer, pH 5.6) is $-27.4 \text{ cm}^3/\text{mol}$ at 50 °C after ultrahigh pressure treatment (300–800 MPa) (Castro et al. 2006). V_a of avocado PPO is from $-34.1 \text{ cm}^3/\text{mol}$ to $-20.4 \text{ cm}^3/\text{mol}$ in buffers with different pH (Weemaes et al. 1998). V_a of red beet extract is $-35.5 \text{ cm}^3/\text{mol}$ for POD and $-50.2 \text{ cm}^3/\text{mol}$ for PPO above 7.5 MPa by HPCD treatment (Liu et al. 2008), respectively. The differences resulted from the different treatment methods, treatment conditions, and food systems of these investigations.

Effect of temperature of HPCD treatment on PPO E_a and Z_T are two important parameters for inactivation of PPO as a function of the temperature, which were listed in Table 2 and 3. Z_T expresses the temperature increase needed for a 90% reduction of D value, and E_a indicates the temperature-dependence of k . In general, the high necessarily E_a means that the rate of this process is strongly-temperature dependent in the same treatment condition (Forte et al. 2009). Compared to control treatment, $E_{a,L}$

increased from 8.7 to 43.2 kJ/mol and $Z_{T,L}$ decreased from 215.1 to 44.5 °C after HPCD treatment, respectively. Conversely, the $E_{a,S}$ decreased from 49.6 to 36.9 kJ/mol and $Z_{T,S}$ increased from 38.0 to 52.6 °C after HPCD treatment. Hence, the high pressure would increase thermal sensitivity of the labile fraction of PPO and lowered that of the stable one of watermelon juice. The explanation for such result is that inactivation occurs through several mechanisms, each with its own temperature dependence. The inactivation with thermal treatment is generally the result of a process of protein unfolding. However, with HPCD treatment, the inactivation attributes to some other process with lowering E_a , such as the loss of some functional group or the metal ion (i.e., Cu^{2+} in PPO) (Forte et al. 2009). Hence, the combination of these mechanisms contributed to the E_a increasing and Z_T decreasing with the pressure increasing. This result of the stable fraction was in agreement with the previous investigations. Gui et al. (2007) reported that E_a was decreased from 72.0 to 18.0 kJ/mol and Z_T increased from 27.0 to 108.0 °C after HPCD treatment.

In addition, $Z_{T,L}$ (44.5 °C) was lower than $Z_{T,S}$ (52.6 °C) and $E_{a,L}$ (43.2 kJ/mol) was higher than $E_{a,S}$ (36.9 kJ/mol) after HPCD treatment (Table 3). It presented that the labile fraction of PPO from watermelon juice was more susceptible to temperature than the stable one, which was opposite from the susceptibility to pressure. Contrary to this study, the stable fraction of pectin methylesterase in apple juice was more susceptible to temperature than the labile one after HPCD treatment (Zhi et al. 2008). The results showed the different characteristic of two fractions of the enzymes.

Effect of pH of HPCD treatment on PPO The effect of HPCD treatment on the pH of watermelon juice is shown in Table 4. The initial pH of watermelon juice was 5.8. After HPCD treatment, the pH of watermelon juice decreased significantly ($P<0.05$) which decreased from 5.8 to 5.5~5.7. This result was in agreement with the investigations of carrot juice, apple juice, and horseradish peroxide

Table 4 Effect of high pressure carbon dioxide (HPCD) treatment on pH of watermelon juice at different conditions

Treatment time (min)	30 MPa 30 °C	30 MPa 40 °C	30 MPa 50 °C	22 MPa 50 °C	15 MPa 50 °C	8 MPa 50 °C
5	5.5±0.07 ^b	5.4±0.02 ^b	5.5±0.10 ^b	5.5±0.04 ^c	5.5±0.04 ^b	5.6±0.01 ^b
10	5.6±0.09 ^b	5.4±0.08 ^b	5.5±0.07 ^b	5.6±0.02 ^b	5.6±0.05 ^b	5.6±0.03 ^b
20	5.5±0.09 ^b	5.4±0.04 ^b	5.5±0.03 ^b	5.6±0.04 ^b	5.5±0.08 ^b	5.6±0.04 ^b
30	5.4±0.10 ^b	5.4±0.05 ^b	5.5±0.04 ^b	5.5±0.04 ^c	5.5±0.08 ^b	5.7±0.02 ^b

All data represent means±SD, $n=3$.

Within the same column, the different letters mean the values are not significantly different at $P<0.05$.

The initial pH is 5.8±0.02^a

solution (Zhou et al. 2009; Gui et al. 2006, 2007). The decrease of pH was caused by CO₂ dissolving into juices or solutions, which further dissociated to bicarbonate, carbonate and H⁺ ions. Hence, there was no change in the pH was observed in muscadine grape juice, after the removal of CO₂ by vacuum during the last stage of dense phase CO₂ processing (Pozo-insfran et al. 2007). However, pH did not change in orange juice after HPCD treatment (Kincal et al. 2006). It was because that the dissociation constants of carbonic acid and bicarbonate were pK_a=6.57 and pK_a=10.62 (Damar and Balaban 2006), the low pH in the original orange juice resulted in the carbonic acid formed by CO₂ dissolving into the juice difficultly being dissociated into H⁺ ions.

Moreover, pH was an important factor to affect the enzyme activities. Zhi et al. (2008) studied that the residual activity of apple PME not subjected to HPCD lost about 21.2% of its original activity after pH adjustment from 7.5 to 5.0. Alternatively, Denès et al. (2000) reported that the activity of purified PME (citrate-phosphate buffer, 6 mM) which optimum pH was 7.5 from apples (*Golden Delicious*) at pH 4.0 remained only 1%. Hence, pH might play some part in PPO inactivation of juice.

Effect of CO₂ of HPCD treatment on PPO Through analyzing, pressure of HPCD treatment was a factor to affect PPO activity of watermelon juice. However, during high pressure with heat treatment, PPO activities reduced only if pressure reached to 100 MPa (Gomes and Ledward 1996; Weemaes et al. 1998; Anese et al. 1995). Hence, CO₂ was another factor to result in PPO activity decreasing sharply by HPCD treatment below 30 MPa in our study.

A large amount of CO₂ dissolved into the watermelon juice during HPCD treatment. Enzyme-bound arginine can easily interact with CO₂ to form a bicarbonate-protein complex under a lower pH environment, which resulted in the loss of enzyme activity (Weder 1984; Weder and Bokor 1992). In addition, Chen et al. (1992) revealed that CO₂ treatment resulted in changes in the secondary structures of the PPO by spectropolarimetric analysis. Gui et al. (2006) also obtained that the reduction of horseradish peroxidase activity caused by supercritical CO₂ treatment was related to the changes in the secondary and tertiary structures. Hence, CO₂ might be another factor to reduce PPO activity in watermelon juice by changing protein structure.

Conclusion

PPO residual activity from watermelon juice decreased drastically to 4.2% at 50 °C, 30 MPa for 30 min under HPCD treatment, which was far lower than 49.1% of control treatment at 50 °C for 30 min. The kinetic analysis

indicated that PPO from watermelon juice was composed of the labile and stable fraction. The labile was more susceptible to temperature and less susceptible to pressure under HPCD treatment. In general, the labile fraction of PPO was inactivated more easily than the stable one. The lowering of pH might be a factor inducing the inactivation of PPO by changing the structures of enzyme protein. Hence, combined effects of pressure, temperature, treatment time, pH reduction, and CO₂ were suggested as the cause of PPO inactivation. HPCD treatment might be more effective method to inactivate PPO from watermelon juice than control treatment at the same temperature.

In addition, the low temperature can save energy and the price of CO₂ is cheap, so the production costs of commercial watermelon juice can be reduced. Moreover, continuous HPCD system had been fabricated and the continuous-process could be achieved. The production efficiency can meet the demand of commercial process. In conclusion, HPCD-treated watermelon juice has large market potential.

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