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Original Paper

Investigation of Processing Technology for Aged Black Jujube

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Abstract

We investigated how to use jujubes from Xinjiang to make black jujube by temperature-controlled wet high-temperature curing. Single-factor and orthogonal experimental designs $L_9(3)^3$ were used to optimize the color change process of the black jujube. The effect of blackening temperature, blackening time and added water amount on the content of cAMP, 5-HMF, polyphenols, total acids, reducing sugars and moisture content were investigated and optimized. The results showed that the optimum process conditions of red jujube aging and blackening black include blackening temperature of 75 ℃, blackening time of 55 h, and water addition amount of 150 mL per 600 g. Under the best technological conditions, the black jujube was fragrant, sweet, and delicious, and the content of all functional substances, including cAMP (0.0137 g/100g), 5-HMF (0.103 g/100g), polyphenols (2.71 g/100g), total acids (17.09 g/kg), reducing sugars (76.7 g/100g), reached high levels, at a moisture content of 26%.

Keywords

Red jujube, aging, blackening, black jujube, processing technology

1. Introduction

Jujube fruit, also known as big jujube, dry jujube, old Chinese jujube, is the mature fruit of *Ziziphus jujuba* Mill. from the Rhamnaceae family. Jujube is a unique fruit and vegetable resource in China. It is native to China and has been cultivated for over 4000 years (Li, Fan, Ding, & Ding, 2007). With an area and output of more than 90% of the world's total, China is the world's largest producer of jujube fruits (Zhao, Zhang, Liu, Xue, & Pan, 2014). Red jujube is a characteristic fruit of China which integrates nutrition, health care and medicinal functions. Jujube is rich in vitamins, minerals, phytosterols, amino acids, saponins, polyphenols, flavonoids, cAMP, cGMP, and polysaccharides

(Pawlowska et al., 2009). Jujube can enhance immunity, help the body resist inflammation, protect the liver, intestines and stomach, ameliorate insomnia and act as an antioxidant (Chen et al., 2015; Zhang, W. Y., Zhang, L., & Xu, 2016; Almansa, Hernández, Legua, Nicolás-Almansa, & Amorós, 2016; Yue et al., 2015).

At present, there are many red jujube products on the market, such as dry products, fermented products, preserved jujube, red jujube beverages and so on. However, there are few reports on aging black jujube. Ji et al. (2013) have done relevant research on the aged jujube, but they only investigated the changes of composition. There are some conventional jujube products in China, such as Jiaozao, Wuzao, etc., but they are produced differently from the production process of aging black jujube. The processing method of black jujube is similar to that of black garlic. Black jujube is produced by non-enzymatic browning of red jujube at high temperature and high humidity. The color, flavor and nutrition of jujubes are changed after blackening. It has been found that the antioxidant capacity of red jujube is also enhanced after aging and blackening (Park et al., 2012). There is a lack of research on processing technology for black jujube. So, the development of low-sugar aged black jujube and related products has broad market prospects.

2. Materials and Methods

2.1 Plant Material and Reagents

Jujube fruits (moisture content 15-20%) were obtained from Hami (GPS E93°32′N42°49′) Xinjiang Province, China. Chemicals, including zinc acetate, potassium ferrocyanide, NaOH, phenol, concentrated sulfuric acid, and ethanol were purchased from Tianjin Kaitong chemical reagent co., LTD (Tianjin, China); 5-HMF (98%), cAMP (98%) and gallic acid (98%) were purchased from Shanghai Yuanye biotechnology co., LTD (Shanghai, China). HPLC grade methanol and acetonitrile were purchased from Shandong Yuwang industrial co., LTD (Yucheng, Shandong, China).

2.2 The Influence of Blackening Conditions on the Content of Various Index Compounds in Black Jujube

2.2.1 Influence of Blackening Time on the Content of Various Index Compounds in Black Jujube

Dried jujubes were divided into 6 equal portions, 600 g each, and cleaned. According to the proportion of jujube water mass ratio 1:5, the fruits were rehydrate at room temperature for 1 h, removed and drain for 15 min, and then 400 mL distilled water added to the bag and sealed. Then, the red jujube were put in the 80 ℃ oven for blackening. At 36 h, 48 h, 60 h, 72 h, 84 h, and 96 h. Cyclic adenosine monophosphate, 5-hydroxymethylfurfural, polyphenols, total acids, reducing sugars, water content and other indicators of each sample were detected respectively.

2.2.2 Influence of Blackening Temperature on the Content of Various Index Compounds in Black Jujube

Dried jujubes were divided into 6 equal portions, 600 g each, then cleaned. According to the proportion of jujube water mass ratio 1:5, rehydrate at room temperature for 1 h, remove and drain for 15 min, and then 400 mL of distilled water was added to the bag and sealed. The portions were separately aged for 60h at 65 ℃, 70 ℃, 75 ℃, 80 ℃, 85 ℃, and 90 ℃. Cyclic adenosine monophosphate (cAMP), 5-hydroxymethylfurfural, polyphenols, total acids, reducing sugars, water content and other indicators of each sample were measured respectively.

2.2.3 Influence of the Amount of Added Water on the Content of Various Index Compounds in Black Jujube

Dried jujubes were divided into 6 equal portions, 600 g each, then cleaned. According to the proportion of jujube water mass ratio of 1:5, the fruits were rehydrated at room temperature for 1 h, removed and drained for 15 min. Then, 100 mL, 200 mL, 300 mL, 400 mL, 500 mL and 600 mL of distilled water was added to individual bags and sealed, followed by aging at 80 ℃ for 60 h. The contents of cAMP, 5-hydroxymethylfurfural, polyphenols, total acids, reducing sugars, water and other indicators of each sample were measured respectively.

2.3 Orthogonal Experimental Design for Optimizing the Blackening Conditions

On the basis of single factor tests, an orthogonal test was carried out by selecting factors such as blackening time, blackening temperature and amount of added water. An orthogonal experiment [$L_9(3)^3$] test design in the blackening mode was used for optimizing the blackening conditions. The key parameters that influenced the contents of key components of black jujube were analyzed, including the time of blackening (A), temperature of blackening (B) and amount of added water (C). Every factor had three levels. Nine extractions were carried out at blackening times 55, 60 and 65 h, blackening temperatures of 75, 80 and 85 ℃, and added water amounts of 150, 200, and 250 mL. The factor levels are shown in Table 1.

Table 1. Orthogonal Design

Level	Factor		
	A, Blackening time/h	B, Blackening temperature (℃)	C, Amount of added water (mL)
1	55	75	150
2	60	80	200
3	65	85	250

2.4 Index Measurement Method

2.4.1 Analysis of Total Acids

The total acid content of black jujube was determined using the GB/T 12456-2008 method. Black samples (20 g) were broken, dissolved in water at 80 ℃, and then placed in a boiling water bath for 30 min (shaken 2-3 times). After the solution was cooled, the volume was set to 250 mL. The sample was filtered, and 25 mL of the filtrate was added to 50 mL of water and titrated with 0.1 mol/L sodium hydroxide solution until the pH was 8.3.

2.4.2 Analysis of Reducing Sugars

The reducing sugar content of black jujube was determined using the GB 5009.7-2016 method. A sample comprising 2.5 g of jujube paste was dissolved in 50 mL of water and transferred to a 250 mL volumetric flask after stirring on a magnetic stirrer for 10 min. Zinc acetate and potassium ferricyanide were added, 5 mL each, to a constant volume, shaken well, and allowed to stand for 30 min. The solution was filtered, the primary filtrate discarded, and the subsequent filtrate saved for later use. Aliquots comprising 5.0 mL alkaline cupric tartrate solution and 5.0 mL alkaline cupric tartrate solution were absorbed, then put in a 150 mL conical flask, 10 mL of water and 2~4 glass beads added. The sample filtrate was used for titration, which was heated to boiling within 2 min, and the titration continued at a rate of 1 drop per 2s until the blue color just faded as the end point.

2.4.3 Analysis of cAMP

A sample comprising 2 g of black jujube paste was added to 80 mL water, stirred with a magnetic stirrer for 10 min, transferred to a 100-mL volumetric flask, and filled to the mark with water. The sample was liquid incubated at 80 ℃ for ultrasonic extraction for 30 min. After cooling, the liquid was passed through a 0.45 μm pore-size membrane and analyzed by HPLC using a sb-c18 column (150 mm x 2.1 mm, 3.5 m; Shimadzu, Japan). The mobile phase was composed methanol and 50 mM potassium dihydrogen phosphate (10:90, V/V). The flow rate was 1 ml/min. The column temperature was 30 ℃. The detection wavelength was 254 nm. The injection volume was 10 μL.

2.4.4 Analysis of 5-HMF

A sample comprising 5 g of black jujube paste was suspended in 10 mL methanol, after which a small amount (approx. 5 mL) of water was added. After magnetic stirring for 10 min, the mixture was transferred to a 50 mL brown volumetric flask and filled to the mark with water. Then, the sample solution was ultrasonicated for 30 min and filtered to the sample bottle through a 0.45 μm organics filtering membrane.

A total of 10 μL was injected into the HPLC column. The analytes were separated on a IntertSustain C18 column (250×4.6 mm, 5 μm; Shimadzu, Japan) at 35°C. The mobile phase was a methanol-water mixture (2:98, v/v). The absorbance wavelength for determination was 282 nm.

2.4.5 Analysis of Polyphenols

This analysis was done based on a previous study[10] with some modifications as follows. 1 g of black jujube paste was dissolved in 30 mL 70% ethanol solution and stirred with a magnetic stirrer for 10 min, after which the volume was adjusted to 50 mL with 70% ethanol. The sample was then ultrasonicated at 66 ℃ for 30 min and filtered, after which 0.2 mL of the sample solution was added to 0.5 mL of 100% Folin phenol reagent diluted 1 time, and the mixture was evenly mixed. Then, 1.5 mL of Na_2CO_3 solution with a mass fraction of 10% was added, and the mixture was mixed evenly with distilled water in a constant volume until 10 mL. The mixture was allowed to react at 75 ℃ for 10 min, after which its absorbance at 760 nm was measured.

2.5 Data Processing and Statistical Analysis

All experiments were conducted in triplicate. The experimental results were presented as means±SE. SPSS 20.0 software (IBM Corp., USA) was used for analysis, and Origin 8.0 software (OriginLab Corp., USA)was used for drawing charts.

3. Results and Discussion

3.1 The Influence of Blackening Conditions on the Content of Various Index Compounds in Black Jujube

3.1.1 Effect of Blackening Time on the Biochemical Contents of Black Jujubes

Table 2. Influence of Blackening Time during the Aging Process of Red Jujubes on Important Biochemicals

Index	Blackening time					
	36h	48h	60h	72h	84h	96h
Total acids (g/kg)	19.76±0.00^{b}	18.66±0.16^{a}	19.59±0.00^{b}	22.40±0.00^{c}	23.86±0.00^{e}	23.45±0.00^{d}
Reducing sugars (g/100g)	71.6±0.00^{b}	74.60±0.17^{d}	75.67±0.29^{e}	74.10±0.10^{c}	71.6±0.00^{b}	70.27±0.07^{a}
Polyphenols (g/100g)	2.16±0.01^{a}	2.30±0.09^{ab}	2.32±0.13^{ab}	2.39±0.38^{b}	2.22±0.38^{ab}	2.30±0.09^{ab}
5-HMF (g/100g)	0.143±0.003^{a}	0.177±0.006^{b}	0.203±0.006^{c}	0.290±0.010^{d}	0.323±0.006^{e}	0.353±0.006^{f}
cAMP g/100g	0.029±0.006^{e}	0.026±0.001^{d}	0.023±0.001^{c}	0.021±0.000^{b}	0.015±0.001^{a}	0.014±0.001^{a}

Note. in the same line, numbers marked with the same letters in the upper right corner indicate that there is no significant difference between groups ($p > 0.05$), while those without the same letters indicate that there is significant difference between groups ($p < 0.05$).

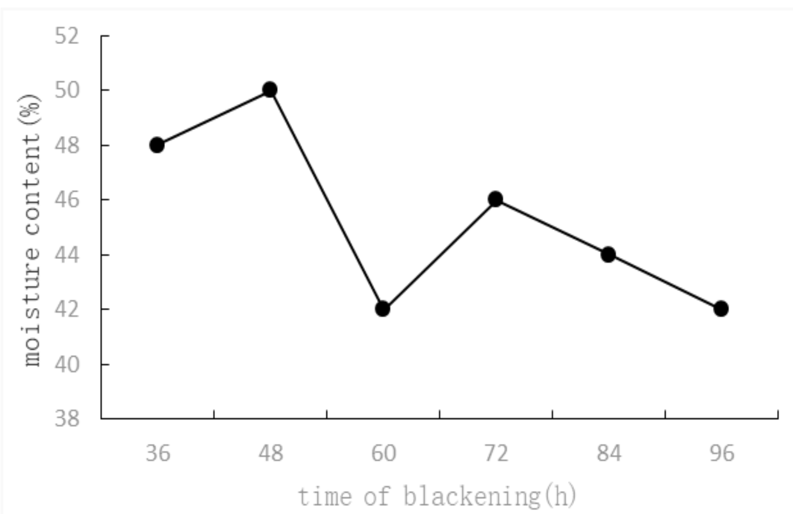

Figure 1. Influence of Blackening Time on the Moisture Content of Aged Black Jujubes

The influence of blackening time on the aging indexes of red jujubes is shown in Table 2 and Figure 1. Blackening time was fixed at 36 h, 48 h, 60 h, 72 h, 84 h and 96 h. It can be seen from Table 2 that the blackening time has a significant effect on the contents of total acids, reducing sugars, furfural, cAMP, flavonoids and water in aged jujubes, but has little effect on the polyphenol content. With the extension of blackening time, the total acid content decreased significantly at 36-48 h, but increased significantly after 48 h. The content of reducing sugars increased significantly between 36 and 60 h, but decreased significantly after 60 h. The content of polyphenols did not change significantly with the extension of blackening time. Furfural content showed a significant increasing trend with the extension of blackening time ($p < 0.05$), the while cAMP content showed a decreasing trend and leveled off after 84 h. As can be seen in figure 1, the water content fluctuated greatly with the extension of blackening time, with the lowest content of 42% observed at 60 h.

During the whole process, the total acid content increased, which may be caused by the fact that the reduced ketones generated by the Maillard reaction can be lysed to produce acids, resulting in the increase of the total acid content (Martins, Jongen, & Van Boekel, 2000; Troise, 2018; Karbasi & Madadlou, 2018). In addition, during the browning reaction, the increase of acids is also related to the production of carboxylic acids. For example, it was reported that carboxylic acids are produced by the oxidation of aldoses (Sang, Cho, Yong, Lee, & Park, 2014). In addition, due to the presence of amino and carbonyl groups, organic acids may change during the Maillard reaction. The increase of reducing sugars may be due to the fact that sucrose was decomposed into monosaccharides or changed into other disaccharides. Our analysis showed that the sucrose content decreased continuously during the aging and blackening of jujube, and finally dropped to 0. Therefore, sucrose may be converted into other reducing sugars during the blackening process. Since reducing sugars are one of the reactants of Maillard reaction, they were continuously consumed and their content decreased with the extension of blackening time. The 5-HMF increased with the extension of black time, but the accumulation rate was higher in the early stage and decreased in the later stages, which may be because 5-HMF is one of the sources of melanoid, the final product of the Maillard reaction. 5-HMF was gradually consumed in the process of increasing melanoid accumulation in the later stages. cAMP was consumed in the non-enzymatic browning reaction during aging.

The reducing sugar content reached the highest point at 75.67 g/100g after 60 h of blackening, whereby the polyphenols and cAMP still remained at a relatively high level, so as to better maintain the nutrients and functional components of jujube. In addition, the moisture content first increased with the increase of blackening time, and then decreased to 42% at 60 h, which was suitable for subsequent processing. However, the total acid index maintained an upward trend, and too much acidity was not conducive to subsequent processing. Furfural has also had a rising trend, but there is still a controversy surrounding the impact of 5-HMF at present (Durling, Busk, & Hellman, 2009), and how it affects the taste of the final product, especially in terms of bitterness. Therefore, blackening jujube for 60 h had the best quality effect.

3.1.2 Effect of Blackening Temperature on the Biochemical Composition of Black Jujube

Table 3. Influence of Blackening Temperature on the Aging Process of Red Jujubes

Index	Blackening temperature					
	65 ℃	70 ℃	75 ℃	80 ℃	85 ℃	90 ℃
Total acids (g/kg)	13.81±0.01[a]	15.03±0.01[b]	17.59±0.02[c]	20.34±0.01[d]	21.72±0.17[e]	25.29±0.15[f]
Reducing sugars (g/100g)	75.59±0.03[e]	74.41±0.05[d]	67.40±0.03[b]	77.54±0.03[f]	71.61±0.03[c]	63.06±0.00[a]
Polyphenols (g/100g)	3.64±0.11[a]	3.50±0.06[a]	3.43±0.32[a]	5.47±0.10[c]	4.46±0.07[b]	4.25±0.04[b]
5-HMF (g/100g)	0.043±0.001[a]	0.097±0.003[b]	0.171±0.006[c]	0.256±0.012[d]	0.418±0.005[e]	0.698±0.027[f]
cAMP g/100g	0.034±0.001[e]	0.027±0.002[d]	0.021±0.002[bc]	0.023±0.001[c]	0.018±0.001[b]	0.014±0.000[a]

Note. In the same line, the numbers marked with the same letters in the upper right corner indicate that there is no significant difference between groups ($p > 0.05$), while those without the same letters indicate that there is significant difference between groups ($p < 0.05$).

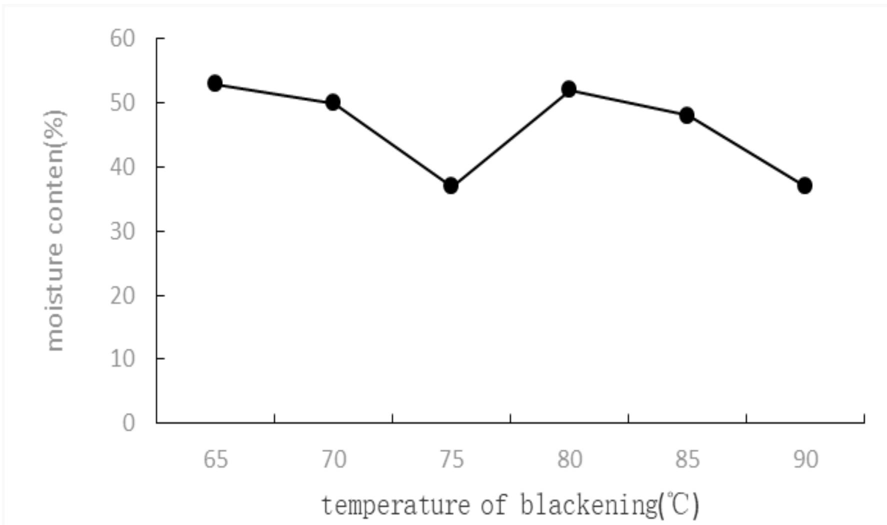

Figure 2. Influence of Blackening Temperature on the Moisture Content of Aged Black Jujubes

The influence of different blackening temperatures on cAMP, furfural, total acid, reducing sugar, polyphenol and moisture indexes was studied, as shown in Table 3 and Figure 2. The blackening temperature was fixed at 65 ℃, 70 ℃, 75 ℃, 80 ℃, 85 ℃ and 90 ℃. As shown in Table 3, with the increase of blackening temperature, the content of total acids and furfural showed a significant increasing trend. It is generally believed that 5-HMF is a product of dehydration and decomposition of fructose or glucose under acidic conditions, and it is a common intermediate product of the Maillard reaction, ascorbic acid oxidation reaction and caramelization. Studies have pointed out that the accumulation of 5-HMF is strongly correlated with the browning rate. However, 5-HMF is produced at high temperatures and is the most important pollutant among heat-induced products, especially in baked goods (Capuano & Fogliano, 2011). The reducing sugar content increased significantly with the increase of blackening temperature. At 80 ℃ the content was 77.54 g /100g. The increase of the reducing sugar content may be associated with the decomposition of other sugars. Studies have shown that the reducing sugar content is directly proportional to the browning rate in the Maillard reaction. In the later stages, with the increase of blackening temperature, the Maillard reaction was accelerated, melanoid-like substances accumulated more, and the color of the jujube was deepened (Majid, Mehdi, & Moein, 2019). For the polyphenol content, the rise of temperature was within the range 65-75 ℃, but the maximum of 5.47 g/100g reached at 80 ℃ was significantly lower. At temperatures higher than 85 ℃ the content stabilized. With the increase of temperature, the increase of polyphenol content may be caused by the formation of Maillard reaction products or by the cleavage of the esterified and glycosylated compounds (Kavita et al., 2015). Overall, cAMP displayed a reducing trend, but increased somewhat at 80 ℃, in spite of generally faster decomposition at higher temperatures. As can be seen in Figure 2, with the increase of blackening temperature, water content reduced first reduced and then rose again, but was significantly lower at 75 ℃. This may be because the maillard reaction rate was faster in the early stage and the water consumption was faster. When the temperature exceeded 80°C, the maillard reaction was weakened by the temperature, and the evaporation of water increased with the temperature in the later stage, resulting in the water content of jujube decreasing. Therefore, the jujubes blackened at 80 ℃ were optimal, with the functional material content maintaining a high level.

3.1.3 Effect of Added Water Amount on the Biochemical Contents of Black Jujube

Table 4. Influence of Added Water Amount on the Aging Process of Red Jujubes

Index	Added water amount					
	100mL	200mL	300mL	400mL	500mL	600mL
Total acids (g/kg)	21.69±0.12^{e}	21.94±0.01^{f}	21.15±0.00^{d}	18.77±0.01^{c}	19.26±0.01^{b}	17.15±0.08^{a}
Reducing sugars (g/100g)	63.78±0.02^{a}	70.47±0.09^{c}	66.92±0.11^{b}	72.68±1.66^{d}	76.10±0.16^{e}	77.27±0.13^{e}
Polyphenols (g/100g)	3.45±0.11^{b}	4.37±0.04^{e}	3.07±0.05^{a}	3.61±0.03^{bc}	4.15±0.06^{d}	3.63±0.03^{c}
5-HMF (g/100g)	0.337±0.010^{e}	0.314±0.006^{d}	0.281±0.002^{c}	0.264±0.002^{b}	0.260±0.005^{b}	0.245±0.000^{a}
cAMP g/100g	0.017±0.002^{a}	0.018±0.001^{a}	0.019±0.001^{a}	0.020±0.001^{a}	0.027±0.002^{b}	0.026±0.002^{b}

Note. In the same line, the numbers marked with the same letters in the upper right corner indicate that there is no significant difference between groups ($p > 0.05$), while those without the same letters indicate that there is significant difference between groups ($p < 0.05$).

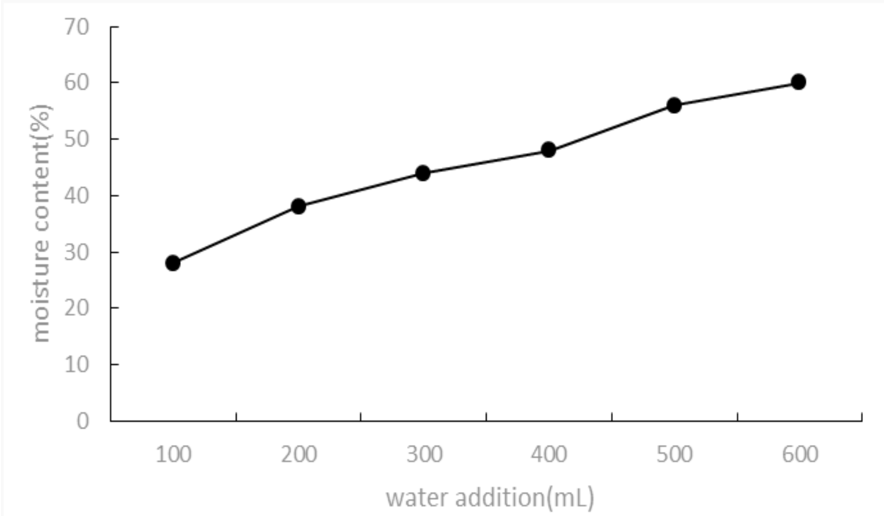

Figure 3. Influence of Water Addition on the Moisture Content of Aged Black Jujubes

The effects of different amounts of added water on cAMP, furfural, total acid, reducing sugar, polyphenols and water indexes in the blackening process were studied, as shown in Table 4 and Figure 3. The amount of added water was fixed at 100 mL, 200 mL, 300 mL, 400 mL, 500 mL and 600 mL. As shown in Table 4, with the increase of water content, the total acids and furfural showed a significant downward trend, i.e., the higher the humidity, the lower the content. There was a significant difference of reducing sugars between 100 mL and 400 mL of water added, but it tended to be stable after 500 mL. The polyphenol content fluctuated greatly, and the increase was the most obvious at 200 mL, reaching 4.37 g/100g. There was no significant difference in the content of cAMP at 100-400 mL and 500-600 mL. As can be seen from figure 3, with the increase of total water content, the water content of the jujube also showed an upward trend. When the amount of added water exceeded 400 mL, the black jujube not only had a large water content, but also had a large amount of residual water in the blackening container, resulting in an incomplete appearance and soft collapse of the black jujube. When 100 mL of water was added, the water content of black jujubes was 28%, and the black jujubes were hard and bitter. To sum up, when the water content was 200 mL, the water content of jujube was appropriate, and cyclic adenosine monophosphate, reducing sugars and polyphenol were all at high levels. Therefore, the water content of jujube was the most appropriate with the addition of 200 mL of additional water.

3.2 Optimization of the Blackening Process of Red Jujube Using an Orthogonal Experiment

Table 5. The Orthogonal Experimental Design for Assessing Different Blackening Conditions

Level		A	B	C	5-HMF (g/100g)	cAMP (g/100g)	Total acids (g/kg)	Reducing sugars (g/100g)	Polyphenols (g/100g)
1		1	1	1	0.103	0.0137	17.09	76.7	2.71
2		1	2	3	0.234	0.0116	22.48	72.2	3.605
3		1	3	2	0.391	0.0082	24.69	66.3	3.122
4		2	1	2	0.135	0.0150	19.44	72.5	3.42
5		2	2	1	0.277	0.0124	23.22	72.3	3.302
6		2	3	3	0.395	0.0092	24.59	61.3	3.296
7		3	1	3	0.157	0.0158	19.91	76.7	2.799
8		3	2	2	0.325	0.0100	25.13	70.2	3.509
9		3	3	1	0.475	0.0217	24.99	66.2	3.525
5-HMF	K1	0.728	0.395	0.855					
	K2	0.807	0.836	0.851					
	K3	0.957	1.261	0.786					
	R	0.199	0.866	0.069					
cAMP	K1	0.0335	0.0445	0.0478					
	K2	0.0366	0.034	0.0332					
	K3	0.0475	0.0391	0.0366					
	R	0.014	0.0105	0.0146					
Total acids	K1	64.26	56.44	65.3					
	K2	67.25	70.83	69.26					
	K3	70.03	74.27	66.98					
	R	5.77	17.83	3.96					
Reducing sugars	K1	215.2	225.9	215.2					
	K2	206.1	214.7	209					
	K3	213.1	193.8	210.2					
	R	9.1	32.1	6.2					
Polyphenols	K1	9.437	8.929	9.537					
	K2	10.018	10.416	10.051					
	K3	9.833	9.943	9.7					
	R	0.581	1.487	0.514					

Table 6. Variance Analysis Table of Furfural for Orthogonal Test Design

Factor	Sum of squares	Freedoms	Mean square error	F	Sig.
Correction model	0.135[a]	6	0.023	502.583	0.002
Intercept	0.690	1	0.690	15409.588	0.000
A	0.009	2	0.005	100.722	0.010
B	0.125	2	0.063	1395.859	0.001
C	0.001	2	0.001	11.169	0.082
Error	8.956E-005	2	4.478E-005		
Total	0.825	9			
Corrected total	0.135	8			

According to the range and variance analysis shown in Tables 5 and 6, the primary and secondary order of each influencing factor is: B blackening temperature > A blackening time > C water addition amount. The blackening temperature and blackening time had a significant effect on furfural, while the addition of water had no significant effect. With furfural as index, an orthogonal test was done to determine the best technology of blackening for $A_1B_1C_2$, which were found to encompass a blackening time of 55 h, blackening temperature of 75 ℃, and added water of 200 mL.

Table 7. Anova Table of Cyclic Adenosine Monophosphate for Orthogonal Test Design

Factor	Sum of squares	**Freedoms**	Mean square error	F	Sig.
Correction model	9.333E-005[a]	6	1.556E-005	0.716	0.682
Intercept	0.002	1	0.002	70.737	0.014
A	3.605E-005	2	1.802E-005	0.830	0.547
B	1.838E-005	2	9.190E-006	0.423	0.703
C	3.891E-005	2	1.945E-005	0.896	0.528
Error	4.345E-005	2	2.172E-005		
Total	0.002	9			
Corrected total	0.000	8			

According to the range and variance analysis shown in Tables 5 and 7, the primary and secondary order of each influencing factor was: C water addition amount > A blackening time >B blackening temperature. The effect of blackening temperature, blackening time and water content on cAMP was not significant. With cAMP as index, an orthogonal test was conducted to determine the best technology of blackening for $A_3B_1C_1$, which was found to encompass a blackening time of 65 h, blackening temperature of 75 ℃, and added water of 150 mL.

Table 8. Total Acid Variance Analysis Table for Orthogonal Test Design

Factor	Sum of squares	Freedoms	Mean square error	F	Sig.
Correction model	67.831[a]	6	11.305	109.511	0.009
Intercept	4513.152	1	4513.152	43717.976	0.000
A	5.551	2	2.776	26.887	0.036
B	59.646	2	29.823	288.890	0.003
C	2.634	2	1.317	12.756	0.073
Error	0.206	2	0.103		
Total	4581.190	9			
Corrected total	68.037	8			

According to the range and variance analysis shown in Tables 5 and 8, the primary and secondary order of each influencing factor is: B blackening temperature > A blackening time > C water addition. The blackening temperature and blackening time had a significant effect on the total acid content, while the addition of water had no significant effect. With total acids as index, an orthogonal test was conducted to determine the best technology of blackening for $A_1B_1C_1$, which was found to encompass a blackening time of 55 h, a blackening temperature 75 ℃, and added water of 150 mL.

Table 9. Anova Table of Reducing Sugars for Orthogonal Test Design

Factor	Sum of squares	Freedoms	Mean square error	F	Sig.
Correction model	199.307[a]	6	33.218	7.759	0.119
Intercept	44718.151	1	44718.151	10445.454	0.000
A	15.136	2	7.568	1.768	0.361
B	176.962	2	88.481	20.668	0.046
C	7.209	2	3.604	0.842	0.543
Error	8.562	2	4.281		
Total	44926.020	9			
Corrected total	207.869	8			

According to the range and variance analysis shown in Tables 5 and 9, the primary and secondary order of each influencing factor is: B blackening temperature > A blackening time > C water addition. The time of blackening and the amount of water added had no significant effect on reducing sugars, while the temperature of blackening had a significant effect. Using separate raw sugars as an index, an orthogonal test was conducted to determine the best technology of blackening for $A_1B_1C_1$, which was found to encompass a blackening time of 55 h, blackening temperature of 75 ℃, and water addition of 150 mL.

Table 10. Anova Table of Orthogonal Test Design for Polyphenols

Factor	Sum of squares	Freedoms	Mean square error	F	Sig.
Correction model	0.490[a]	6	0.082	0.503	0.782
Intercept	95.310	1	95.310	588.045	0.002
A	0.059	2	0.029	0.181	0.847
B	0.385	2	0.192	1.187	0.457
C	0.046	2	0.023	0.142	0.876
Error	0.324	2	0.162		
Total	96.123	9			
Corrected total	0.814	8			

According to the range and variance analysis shown in Tables 5 and 10, the primary and secondary order of each influencing factor is: B blackening temperature > A blackening time > C water addition. The effect of blackening time, blackening temperature and water content on polyphenols was not significant. With the polyphenol content as index, an orthogonal test was conducted to determine the best technology of blackening for $A_2B_2C_2$, which encompassed a blackening time of 60 h, blackening temperature of 80 ℃, and water addition of 200 mL.

In conclusion, the three factors of blackening time, blackening temperature and water addition had no significant influence on the cAMP and polyphenol indexes. Therefore, furfural, total acid, reducing sugar indices were used as the basis to determine the optimal processing parameters for $A_1B_1C_1$, which were a blackening time of 55 h, blackening temperature of 75 ℃, and water addition of 150 mL.

4. Conclusion

Red jujube is widely planted in China, but there are few reports on the production technology of black jujube. In this paper, an orthogonal $L_9(3)^3$ test was used to optimize the production process of black jujube. The results revealed optimum processing parameters as follows: aging blackening time 55 h, blackening temperature 75 ℃, added water 150 mL. The black jujube produced under the optimal technological conditions had a strong fragrance, sweet and sour taste, and contents of various functional substances reaching (dry matter meter): cAMP 0.0137 g/100g, 5-HMF 0.103 g/100g, polyphenols 2.71 g/100g, total acids 17.09 g/kg, reducing sugars 76.7 g/100g, and water content 26%.

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