LWT - Food Science and Technology 91 (2018) 70–76

Contents lists available at ScienceDirect

LWT - Food Science and Technology

journal homepage: www.elsevier.com/locate/lwt

Clarification effect of collagen hydrolysate clarifier on chrysanthemum beverage

Qi-Xian Zhang[a], Rui-Jie Fu[a,c], Kai Yao[a], Dong-Ying Jia[a], Qiang He[a,b], Yuan-Long Chi[a,b,c,*]

[a] College of Light Industry, Textile and Food Engineering, Sichuan University, Chengdu 610065, People's Republic of China
[b] Healthy Food Evaluation Research Center, Sichuan University, Chengdu 610065, People's Republic of China
[c] Chengdu Food and Drug Administration Research Institute, Chengdu 610041, People's Republic of China

ARTICLE INFO

Keywords:
Collagen
Clarifier
Clarification
Plant beverage

ABSTRACT

Collagen hydrolysate clarifier (CHC), which was prepared by enzymatic hydrolysis of pigskin shavings (collagen waste) in our previous study, was applied in the clarification of a Chrysanthemum beverage. CHC exhibited an excellent clarification effect on the Chrysanthemum beverage due to the significant improvement of the beverage transmittance from 55.4% to 96.2%. The clarification performance was influenced by CHC dosage, pH, temperature and clarification time. The highest transmittance was obtained at a CHC dosage of 0.8 g/L, pH 5.45, temperature of 29 °C and duration of 3 h. After clarification of CHC, the proteins, polyphenols, sugars and flavonoids in the Chrysanthemum beverage were all slightly reduced, but their retention rates were still more than 80%. The sensory quality and storage stability were improved greatly due to the increased total sensory quality score from 62.7 to 85.6 and the prolonged storage time from 1 day to 12 days. Moreover, the cost was estimated to be as low as $0.93/kg. Therefore, CHC is suitable for application in the clarification of plant beverages.

1. Introduction

Plant beverages are prepared by using plants or plant extracts as raw materials, including herbal beverages, cereal beverages, edible fungus beverages and algae beverages. The main ingredients of plant beverages are polyphenols, sugars, and proteins, and these compounds can combine to generate unstable complexes, causing turbidity and precipitation of the beverage (Cerreti et al., 2017; Lin et al., 2007; Park et al., 2016; Siebert, 2006; Xu et al., 2017). To avoid haze turbidity and sediment in the beverage and to improve its transparency and stability, clarification treatment is necessary. Current clarification treatment methods include natural precipitation, enzymatic hydrolysis and utilization of clarifiers (Bilal, Asgher, Iqbal, Hu, & Zhang, 2017; Feng, Zhang, Ma, & Tian, 2009; Kochergin, Gaudet, & Robert, 2011; Ma, Luo, & Zeng, 2015; Parish, Herbst-Johnstone, Bouda, Klaere, & Fedrizzi, 2017; Sandri, Fontana, Barfknecht, & Silveira, 2011). Natural precipitation involves leaving the beverage at low temperature for a long period, during which the suspended substances settle down slowly without any assistance. This method is time-, energy- and cost-consuming, so it is inefficient. Enzymatic hydrolysis and utilization of clarifiers are popular choices for plant beverage clarification. Enzymatic hydrolysis can reduce the components in the beverage that tend to form precipitates. Utilization of clarifiers is characterized by simple operation, high efficiency and low cost. Commonly used clarifiers include chitosan, modified chitosan, gelatin, activated carbon, and silica; chitosan and gelatin are considered natural and have good biocompatibility (Bilal et al., 2017; Ma et al., 2015; Parish et al., 2017). It is worthwhile to develop new natural clarifiers for plant beverages.

Collagen hydrolysate clarifier (CHC) with a great flocculating property was prepared through pepsin hydrolysis in our previous work (Fu et al., 2017). The raw material of CHC is pigskin shavings (collagen waste) obtained from the slaughterhouse, so it is natural. In addition, CHC prepared by enzymatic hydrolysis has a neutral pH without any chemicals. The pepsin used is of food grade. Therefore, CHC is safe enough to be applied in the food industry. There are many collagen hydrolysate products currently on the market. For example, one oral liquid, developed by American Nature's Bounty Company (New York, USA), is a collagen hydrolysate with a molecular weight lower than 3 kDa and shows good skin care effects. It is also prepared using enzymatic hydrolysis. Our previous work has verified that CHC showed an

* Corresponding author. College of Light Industry, Textile and Food Engineering, Sichuan University, Chengdu 610065, People's Republic of China.
E-mail address: chiyl@scu.edu.cn (Y.-L. Chi).

https://doi.org/10.1016/j.lwt.2018.01.041
Received 14 July 2017; Received in revised form 12 January 2018; Accepted 15 January 2018
Available online 19 January 2018
0023-6438/

excellent flocculation property to kaolin based on charge neutralization and a bridging ability (Fu et al., 2017). Thus, CHC may also be effective in clarification of plant beverages through the same action.

Chrysanthemum beverage is a plant beverage prepared using the flower of a Chrysanthemum (*Dendranthema morifolium* Ramat. Tzvel) as a raw material through the processes of picking, drying, cutting, extraction, filtration and sterilization. It has a bright yellow colour, Chrysanthemum aroma and a slightly bitter taste. Chrysanthemum contains a variety of flavonoids, which impart bioactivity in the beverage (Tao, Duan, Qian, Qian, & Guo, 2016). In this work, a Chrysanthemum beverage was used as a representative plant beverage to investigate the clarification performance of CHC. The effects of CHC dosage, pH, temperature and clarification time on the clarification of Chrysanthemum beverage were investigated and then optimized. The main chemical components, sensory quality and storage stability of Chrysanthemum beverages with and without clarification were analysed. Finally, the production cost of CHC was estimated.

2. Materials and methods

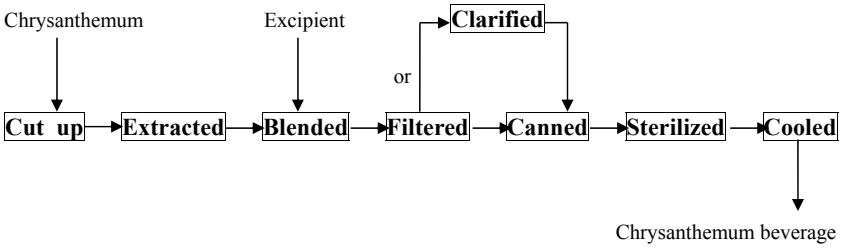
2.1. Materials, reagents and instruments

CHC was prepared based on our previous work (Fu et al., 2017). A Chrysanthemum beverage was prepared as described in section 2.2. Chrysanthemum and sugar powder were purchased from a local market in Chengdu, China. Bovine serum albumin, Coomassie brilliant blue G-250, Folin-Ciocalteu phenol reagent, gallic acid, glucose, and rutin were of analytical grade and purchased from Sigma Co, Ltd. in Shanghai, China. Ethanol and phosphoric acid were of analytical grade and purchased from Kelong Chemical Reagent Company in Chengdu, China. Trehalose was of food grade and purchased from Kelong Chemical Reagent Company in Chengdu, China. The Coomassie brilliant blue G-250 solution was made by mixing 10 mg of Coomassie brilliant blue G-250, 5 mL of 90% (v/v) ethanol and 10 mL of 85% (v/v) phosphoric acid and diluting to 100 mL by deionized water.

The instruments included a UV–Vis spectrophotometer (UV-1100, Shanghai Mapada Instrument Co., Ltd., China), a magnetic stirrer with a temperature control system (90-3, Shanghai Huxi Analysis Instrument Factory Co., Ltd., China), a pH metre (PHS-3C, Shanghai INESA Instrument Limited by Share Ltd., China), a waterproof incubator (9080, Shanghai Yiheng Technical Co., Ltd., China), a spectrophotometer (CM-5, Konica Minolta Co., Ltd., Japan), and a handheld refractometer (WY-020R, Chengdu Wanchen Optical Instrument Factory, China).

2.2. Preparation of chrysanthemum beverage

The main production processes of a Chrysanthemum beverage are listed below.



(1) **Cutting:** Chrysanthemum was cut into small strips with 0.3 cm width and 1 cm length after cleaning and drying at 45 °C for 2 h and at 65 °C for another 2 h.
(2) **Extraction:** 10 g Chrysanthemum strips were packaged and added to 400 mL water. Extraction was conducted for 30 min at 100 °C. The extracted liquid was poured out and collected. Another 300 mL water was added, and the second extraction was conducted for 20 min at 100 °C. The extracted liquid from the second extraction was collected and mixed with that from the first extraction.
(3) **Blended:** 16 g trehalose and 16 g sugar powder (100 mesh) were fully dissolved into 96 mL water and added to the extracted liquid.
(4) **Sterilized:** after canning, the beverage was sterilized for 20 min at 121 °C.

2.3. Influence factors on clarification

2.3.1. CHC dosage

A total of 10 mL of Chrysanthemum beverage was added to a 10-mL cylinder with a plug at its natural pH of 5.0. Then, 2 mg, 4 mg, 6 mg, 8 mg and 10 mg of CHC were added to obtain a Chrysanthemum beverage with CHC dosages of 0.2 g/L, 0.4 g/L, 0.6 g/L, 0.8 g/L and 1.0 g/L, respectively. The beverage without CHC added was used as a control. The mixtures were set at room temperature (30 °C) and left to stand for 4 h. A sample was obtained at 7-mL line of the cylinder to detect the transmittance (T, %) using the UV–Vis spectrophotometer at 700 nm. A higher transmittance indicates a better clarification effect (Cerreti et al., 2017; Jing et al., 2011).

2.3.2. pH

A total of 10 mL of Chrysanthemum beverage was adjusted to pH 4.0, 4.5, 5.0, 5.5, 6.0, 6.5 and 7.0 by using a 1 mol/L HCl or a 1 mol/L NaOH solution. CHC was added, and the dosage in all mixtures was 0.6 g/L. The mixtures were set to room temperature (30 °C) and left to sit for 4 h. A sample was obtained at 7-mL line of the cylinder to detect the transmittance (T, %) by UV–Vis spectrophotometer at 700 nm.

2.3.3. Temperature

A total of 10 mL of Chrysanthemum beverage with a CHC dosage of 0.6 g/L at its natural pH of 5.0 was adjusted to 5 °C, 10 °C, 15 °C, 20 °C, 25 °C, 30 °C, 35 °C and 40 °C and left to stand for 4 h. A sample was obtained at 7-mL line of the cylinder to detect the transmittance (T, %) by UV–Vis spectrophotometer at 700 nm.

2.3.4. Clarification time

A total of 10 mL of Chrysanthemum beverage with a CHC dosage of 0.6 g/L at its natural pH of 5.0 was set to room temperature (30 °C) and left to stand for 6 h. A sample was obtained at 7-mL line of the cylinder at certain intervals to detect the transmittance (T, %) by UV–Vis spectrophotometer at 700 nm.

2.4. Optimization of clarification conditions

Based on the single factor experiment results and the Box-Behnken experimental design principle (Cerreti et al., 2017), three factors—CHC dosage (A), pH (B) and temperature (C)—all of which had a stronger