STUDY ON IMPROVEMENT IN COLLOIDAL AND MICROBIOLOGICAL STABILITY OF ARTICHOKE DRINK *(CYNARA SCOLYMUS L.)*

NGHIÊN CỨU CẢI THIỆN ĐỘ BỀN KEO VÀ VI SINH CHO THỨC UỐNG TỪ ARTICHOKE *(CYNARA SCOLYMUS L.)*

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ABSTRACT

The shelf-life of artichoke drink produced on the small scale is limited in several days because of low colloidal and microbiological stability. For overcoming this problem, artichoke drink was firstly treated by bentonite and subsequently filtered through a filter with a pore size of 0.2µm. Technological parameters of the treatment were bentonite content of 0.3%w/w, temperature of 80^{o}C and duration of 5 minutes. The soft drink was then bottled in polypropylene containers, sealed with a polyamide film and pasteurized at 80^{o}C for 10 minutes. The proposed techniques were simple and easy realizing for small scale production. The produced artichoke drink extended the shelf-life up to 6 months when it was stored at ambient temperature.

TÓM TẮT

Thức uống từ artichoke được sản xuất ở quy mô nhỏ có thời gian bảo quản chỉ vài ngày do độ bền keo và vi sinh thấp. Để khắc phục vấn đề này, trước tiên nước artichoke được xử lý với bentonit rồi lọc qua màng lọc với kích thước lỗ lọc xấp xỉ 0.2µm. Các thông số công nghệ của quá trình xử lý nước artichoke với bentonit như sau: lượng bentonit sử dụng là 0.3%w/w, nhiệt độ 80^{o}C và thời gian xử lý là 5 phút. Sau đó, sản phẩm được rót vào bao bì polypropylen, đóng nắp bằng màng polyamit và thanh trùng ở 80^{o}C trong thời gian 10 phút. Khi đó, sản phẩm nước artichoke có thể bảo quản trong thời gian 6 tháng ở điều kiện nhiệt độ phòng.

I. INTRODUCTION

Drinking is one of indispensable demands for human being [1]. Nowadays, soft drinks from medicinal herbs have been considered as highly potential because they not only supply sugars and other nutritive compounds to consumers but also improve the function of some organs in the body. Medicinal herbs abound in our country. They have widely been used in human life and traditional medical treatment [2]. Until present, some soft drinks from medicinal herbs such as artichoke (*Cynara scolymus L.*), corn silk (*Zea mays L.*), dwarf sugar cane (Saccharum sinensis Roxb.), plantain (*Plantago major L.*), ginseng (*Panax ginseng*)… have been produced on the small scale for commercial purpose.

Soft drink industry has a profound impact on the world economy and on our lives in general [1]. Soft drink production with small scale includes various advantages: simple production-line and easy realizing, low cost for equipment investment, high freshness of final product… However, small scale production has certain disadvantages. The soft drink shelf-life is quite short (up to 2-5 days), so the product distribution is limited. In addition, the product must be stored at low temperature (from 4 to 6^{o}C) and this increases the storage cost. Moreover, the colloidal and microbiological stability of medicinal herb drink are quite low [3]. Some precipitates are often appeared in the product and beverage spoilage by contaminated microorganisms is normally observed after several day storage.

This research focused on artichoke drink processing on the small scale. Colloidal stability of this drink concerned with certain colloidal compounds originated from raw material such as pectin, polyphenols, and proteins [4]. These colloids increased drink turbidity and they would precipitate under the change of physico-chemical conditions [3]. Different treatment methods have been proposed to improve colloidal stability of medicinal herb drinks such as precipitation of colloids by high or low temperature, decantation, filtration, addition of chemicals for forming the precipitated complex with colloids,

application of pectinase and protease preparation, use of adsorbents for colloid elimination… [3,5,6]. In this study, a combined method was used: colloidal compounds in artichoke drink were removed by adsorption on bentonite and subsequent filtration.

According to Vietnam standard TCVN 7041:2002, the number of total aerobic microorganisms in beverage must be less than 10^2cfu/mL [7]. For improving the microbiological safety and the shelf-life of artichoke drink, pasteurization process was then examined.

II. MATERIALS AND METHODS

Materials

- Artichoke (*Cynara scolymus L.*): dried artichoke flower originated from Khoi nguyen Co., Ltd. (Lam dong) was used in the present study. The moisture of this raw material was 11%.

- Sucrose was purchased from Bien hoa sugar Co., Ltd. The moisture and sucrose content were 0.03% and 99.8% of dry mass, respectively.

- Citric acid (food grade) was originated from Hunan Dongting Citric Acid Chemicals Co., Ltd. (China). The moisture and citric acid content were 7.5% and 99.5% of dry mass, respectively.

- Potable water was used for preparation of artichoke extract and sucrose syrup.

- Bentonite was originated from Tianjin Bentonite Minchem Co., Ltd (China). The moisture was approximately 9%. The diameter of bentonite grains was less than 0.075mm.

Methods

- Production line schema for artichoke drink processing:

Artichoke → Extraction with hot water → Filtration with cotton filter for eliminating solid residue → Mixing artichoke extract with sucrose syrup to the sugar content of 100g/L → Adjustment to pH of 3.9 by citric acid → Treatment by bentonite → Filtration (filter with pore size of 0.2μm) → Cooling to ambient temperature → Bottling and sealing → Pasteurization → Dating → Artichoke drink

The technological parameters of the artichoke extract and sucrose syrup preparation were previously described elsewhere [2,8]. In this study, artichoke drink was bottled in 200mL containers made by polypropylene. The products were sealed with a polyamide film.

- Analytical methods:

- Total solutes in the artichoke extract and drink were determined by refractometer Atago (Japan) and expressed in ^oBx
- pH was measured by pH-meter Mettler Toledo (EU)
- Essential oil, carotenoids, alkaloids, flavonoids, organic acids, reducing compounds, and tannins in artichoke extract and drink were qualitatively determined by a classical method described elsewhere [9]
- Total and reducing sugars were quantified by spectrophotometric method, using phenol and dinitrosalycylic acid reagents, respectively [10]
- Turbidity of artichoke drink was measured by Turbidity LAB-IR ISO7027 (Germany). The result was expressed in Number of Turbidity Units (NTU) [10]
- Aerobic microorganisms, yeasts and molds, Coliforms, Streptococci faecal, *E. coli*, and *C. perfringens* were quantified by plate count agar method [11]

- Statistical analysis:

The presented results in this research were the average of three independent experiments. The obtained data were subjected to analysis of variance (ANOVA), $p<0.05$ using Startgraphics plus, version 3.2

III. RESULTS AND DISCUSSION

Improvement in colloidal quality of artichoke drink by bentonite treatment

In juice processing, bentonite has been considered as potential adsorbent for eliminating colloidal compounds and improving the drink transparence [3]. In this study, bentonite was added to hot water (90^oC) for

preparing 20%w/w bentonite solution. This solution was stored for 12 hours at ambient temperature and then added to the artichoke drink. The bentonite content used in the treatment was expressed by the ratio of weight of bentonite in dry matters to weight of artichoke drink (%w/w).

In the first experiment, different contents of bentonite were added to the artichoke drink for adsorption of colloidal compounds. The treatment was realized at 90^{o}C for 10 minutes. The artichoke drink was then filtered for removing solid residue and bentonite grains. The collected filtrates were used to determine the turbidity. The treatment was not carried for control sample. The results are given in Table 1. It can be affirmed that artichoke drink treatment by bentonite ameliorated significantly the product transparence. This phenomenon was also observed in juice processing [3], and winemaking [5,6]. The turbidity of artichoke drink treated by bentonite was 2.9 – 24.3 times lower than that of the control sample. Increase in bentonite content from 0.1 to 0.3%w/w reduced remarkably the turbidity of the artichoke beverage. However, treatment with higher bentonite content did not make significant differences in product turbidity. The suitable of bentonite content used in the treatment was therefore 0.3% w/w.

In the next experiment, treatment temperature was varied from 70 to 90^{o}C. The bentonite content and treatment time were fixed at 0.3%w/w and 10 minutes. Table 2 presents the results. Increase in temperature decreased the turbidity of the final product. However, high temperature could damage some thermo-sensitive compounds and affect negatively the sensory properties of the soft drink. From Table 2, 80^{o}C was selected as appropriate temperature for the treatment in artichoke drink processing. In comparison with winemaking, the treatment temperature by bentonite was approximate 15^{o}C. However, the treatment time was prolonged to 10 days [6]. In artichoke processing, the treatment could not be carried out at ambient or lower temperature and for a long duration because of preventing the drink spoilage by contaminated microorganisms.

In the following experiment, treatment time was changed from 5 to 15 minutes. The bentonite content and treatment temperature were maintained at 0.3%w/w and 80^{o}C. The obtained results are shown in Table 3. It can be noted that increase in treatment time more than 10 minutes did not decreased the product turbidity. Hence, the suitable treatment time was 10 minutes.

Improvement in microbiological quality of artichoke drink by pasteurization

The pH value of artichoke drink was lower than 4.6, hence the treatment temperature for inactivation of the contaminated microorganisms would be lower than 100^{o}C [12].

Firstly, the pasteurization temperature was varied from 75 to 90^{o}C. The treatment time was 15 minutes. A control sample was also realized without pasteurization. All samples were than incubated in the acceleration conditions of 37^{o}C and 99% relative humidity for 3 weeks. The total aerobic microorganisms in each sample during the incubation are seen in Table 4. After 1 week incubation, the number of total aerobic microorganisms in the control sample exceeded the value of 10^2cfu/mL approved by Vietnam standard TCVN 7041:2002 [7]. On the contrary, the result of all pasteurized samples matched the national standard. However, for ensuring the soft drink with high microbiological quality, 80^{o}C was chosen as suitable pasteurization temperature.

Table 1. Effect of bentonite content on NTU of artichoke drink

Bentonite content (%w/w)	0.1	0.2	0.3	0.4	0.5	Control sample
NTU of artichoke drink	22.57^b	5.59^c	2.71^d	2.50^d	2.18^d	65.76^a

Different letters mean significant difference (P<0.05)

Table 2. Effect of treatment temperature by bentonite on NTU of artichoke drink

Temperature (^oC)	70	80	90
NTU of artichoke drink	2.97^a	2.77^b	2.77^b

Different letters mean significant difference (P<0.05)

Table 3. Effect of treatment time by bentonite on NTU of artichoke drink

Time (min)	5	10	15
NTU of artichoke drink	2.99^a	2.77^b	2.73^b

Different letters mean significant difference (P<0.05)

Table 4. Effect of pasteurization temperature on total aerobic microorganisms of artichoke drink (After pasteurization, all samples were incubated in the acceleration conditions of 37^{o}C and 99% relative humidity)

Pasteurization temperature (^oC)	Total aerobic microorganisms (cfu/mL)			
	After 1hour pasteurization	After 1 week incubation	After 2 week incubation	After 3 week incubation
75	$< 1*10^{-1\ b}$	$< 1*10^{-1\ b}$	33^d	63^e
80	$< 1*10^{-1\ b}$	$< 1*10^{-1\ b}$	$< 1*10^{-1\ b}$	$< 1*10^{-1\ b}$
85	$< 1*10^{-1\ b}$	$< 1*10^{-1\ b}$	$< 1*10^{-1\ b}$	$< 1*10^{-1\ b}$
90	$< 1*10^{-1\ b}$	$< 1*10^{-1\ b}$	$< 1*10^{-1\ b}$	$< 1*10^{-1\ b}$
Unpasteurized sample (control)	36^a	$1,1*10^{2\ c}$	-	-

Different letters mean significant difference (P<0.05)

Table 5. Effect of pasteurization time on total aerobic microorganisms of artichoke drink (After pasteurization, all samples were incubated in the acceleration conditions of 37^{o}C and 99% relative humidity)

Pasteurization time (min)	Total aerobic microorganisms (cfu/mL)			
	After 1 hour pasteurization	After 1 week incubation	After 2 week incubation	After 3 week incubation
5	$< 1*10^{-1\ b}$	$< 1*10^{-1\ b}$	27^c	73^d
10	$< 1*10^{-1\ b}$	$< 1*10^{-1\ b}$	$< 1*10^{-1\ a}$	$< 1*10^{-1\ a}$
15	$< 1*10^{-1\ b}$	$< 1*10^{-1\ b}$	$< 1*10^{-1\ a}$	$< 1*10^{-1\ a}$
20	$< 1*10^{-1\ b}$	$< 1*10^{-1\ b}$	$< 1*10^{-1\ a}$	$< 1*10^{-1\ a}$
Unpasteurized sample (control)	43^a	$1,2*10^{2\ b}$	-	-

Different letters mean significant difference (P<0.05)

Table 6. Some physico-chemical characteristics of artichoke drink

Characteristics	Before pasteurization	After pasteurization
Content of total solutes (^{o}Bx)	11.0[a]	11.1[a]
Reducing sugar (g/L)	19.6[b]	21.3[c]
Total sugar (g/L)	103.3[d]	106.4[e]
pH	3.9[f]	3.9[f]

Different letters in each row mean significant difference (P<0.05)

Table 7. Chemical compounds with bioactivity in the artichoke drink

Chemical compounds	Artichoke extract	Artichoke drink before pasteurization	Artichoke drink after pasteurization
Essential oil	+	+	+
Fats	+	+	+
Carotenoids	+	+	+
Alkaloids	+	+	-
Flavonoids	+	+	+
Organic acids	+	+	+
Reducing compounds	+	+	+
Tannins	+	+	+
Saponins	-	-	-

(+): *positive result,* **(-):** *negative result*

Then the pasteurization time was changed from 5 to 20 minutes. The pasteurization temperature was fixed at 80^{o}C. The obtained results in Table 5 showed that the number of total aerobic microorganisms in all samples matched the national standard after 3 week incubation in the acceleration conditions, except the control sample that was spoiled after 1 week incubation in the same conditions. 10 minutes was therefore selected as appropriate pasteurization time to guarantee high safety for artichoke drink. Similar results were also observed by Liang Y.R. (2006) in the study on pasteurization of soft drink from green tea. This author found that optimal treatment temperature and time were 85^{o}C and 10 minutes, respectively [13].

From the experimental datum, it can be concluded that if the pasteurized artichoke drink was stored at ambient temperature, the product shelf-life would not lower than 6 months.

Evaluation of product quality

Table 6 presents some physico-chemical characteristics of the artichoke drink before and after pasteurization. Low pH value of 3.9 and high pasteurization temperature of 80^{o}C facilitated sucrose hydrolysis and this led to an increase in total and reducing sugar contents. The microbiological test on total aerobic microorganisms, yeasts and molds, coliforms, streptococcii faecal, *E. coli* and *C. perfringens* showed that no colony was appeared in the selected agar media used in the plate count agar method.

Table 7 showed the qualitative results of the presence of some chemical compounds with bioactivity in the artichoke extract, and in the artichoke drink before and after pasteurization. It can be confirmed that artichoke contained different valuable compounds, except saponins [4]. The beverage treatment by bentonite at 80^{o}C

and for 5 minutes did not change qualitative composition of the artichoke extract. However, alkaloids were lost during the product pasteurization. For preventing this phenomenon, thermal treatment with ultra-high temperature/ short time and aseptic packaging must be used. Nevertheless, these techniques were costly options and they were not suitable for application to small scale production.

IV. CONCLUSIONS

Application of the treatment by bentonite and pasteurization to artichoke drink processing could improve significantly the colloidal and microbiological stability of the final product. The proposed techniques were simple and easy realizing. In addition, the cost of new equipment investment for small enterprises was quite low. These techniques were therefore potential for practical application.

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