Multivariate statistical analysis of the quality of apple juice to integrate and simplify juice industrial production technologies

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ABSTRACT

Apple juices produced from nine different industrial production technologies were obtained in pilot scale, for simplifying and integrating the production technologies according to the quality comparison of juices. Juice quality characterized by total phenolic content (TPC), total flavonoid content (TFC), total soluble solid (TSS), stability, turbidity, titratable acidity and pH was investigated. The TPCs of traditional pulping combined with belt-style pressing (TP+BSP)-based juices were higher than that of cold pulping combined with horizontal spiral centrifuge separation (CP+HSCS)-based juices, while the CP+HSCS juices showed higher turbidity than the TP+BSP juices. Principal component analysis, linear discriminant analysis and cluster analysis were applied for analyzing juice quality. Nine production technologies were well separated into three categories and that pulping, juicing and stabilizing methods were the main factors affecting the overall juice quality. Our study provided a method to simplify and integrate the current industrial production technologies of juices.

Análisis estadístico multivariado de la calidad de jugo de manzana realizado con el propósito de simplificar e integrar las diversas tecnologías empleadas para la producción industrial de jugo

RESUMEN

Con el fin de simplificar e integrar las distintas tecnologías empleadas para la producción de jugo, se comparó la calidad de jugos de manzana obtenidos en escala piloto empleando nueve diferentes tecnologías de producción industrial. Con este objetivo se investigaron la calidad de los jugos con base en el contenido total de fenólicos (TPC), contenido total de flavonoides (TFC), total de sólidos solubles (TSS), estabilidad, turbiedad, acidez valorable (TA) y pH. El TPC de los jugos elaborados a partir del desulpado tradicional en combinación con prensa de cinta (TP+BSP) fue más elevado que el de los jugos obtenidos por desulpado en frío combinado con la separación mediante centrifugadora horizontal en espiral (CP+HSCS). Asimismo, los jugos obtenidos empleando el método CP+HSCS presentaron mayor turbiedad que aquellos elaborados mediante el TP+BSP. Para analizar la calidad de los jugos obtenidos se aplicaron los siguientes análisis: análisis de componentes principales (PCA), análisis discriminante lineal (LDA) y análisis clúster (CA). Fueron bien separados nueve tecnologías de producción en tres categorías, constatándose que el principal elemento que incide en la calidad del jugo obtenido es el método aplicado: desulpado, extracción, esterilización. Por lo que nuestro estudio proporciona un método que permite simplificar e integrar la tecnología existente actualmente para la producción industrial de jugos.

1. Introduction

Apple (Malus pumila Mill.) is popular all over the world as one of the most widely consumed fruits containing plentiful nutritional phytochemicals for human beings (Wang & Lu, 2014; Yang, Yang, Guo, Jiao, & Zhao, 2013). Apple juice is the most popular and commonly processed fruit juice in the world due to its nutritional value and typical flavor (Hyson, 2011). Unit operations in industrial processing simplify juice producing, while it may also affect the physicochemical properties of juice. Since many production technologies have been designed and different unit operations have been performed during processing, the quality of different technology-based juices was worth to be investigated.

To date, thermal treatment is still an important and most widely used unit operation in food processing industry (Rawson et al., 2011). Phenolic and flavonoid compounds are major bioactive substances present in apple juice (Wlodarska, Pawlak-Lemanska, Khmelinskii, & Sikorska, 2016), and thermal treatment modifies the total phenolic contents (TPCs) and total flavonoid content (TFC) of apple juice. Besides, heating treatment has been reported to increase the total soluble solid (TSS) of fruit pulp (Branco et al., 2016). Physical stability is an important index regarding particle sedimentation and serum separation (Kubo, Augusto, & Cristianini, 2013). Thermal treatment may also affect the juice stability as it has been reported to reduce the...
pectin methyl-esterase activity effectively (Aghajanzadeh, Ziaiifar, Kashaninejad, Maghsoudlou, & Esmailzadeh, 2016). Moreover, heat treatment can vaporize different organic acids, making it change the titratable acidity (TA) and pH values of apple juice (Charles-Rodríguez, Nevárez-Moorillón, Zhang, & Ortega-Rivas, 2007).

Homogenization is a commonly used unit operation in juice preparation not only for preservation of foodstuffs but it has also become a valuable tool to promote desirable changes in the physical properties of the products (Augusto, Ibarz, & Cristianini, 2012). It has been reported that homogenization increased the TSS value of strawberry juice (Karacam, Sahin, & Oztok, 2015). Similarly, it also increased the TPC of mango (Guan et al., 2016) and strawberry juices (Karacam et al., 2015). Moreover, homogenization can also affect the turbidity and physical stability of juices (Silva et al., 2010).

Apart from the thermal treatment and homogenization, other common unit operations like pulping and juicing also affect the juice quality (Heinmaa et al., 2016). Although the effect of certain unit operation has been widely studied, there is still much work need to do for comparing quality parameters of apple juices based on different technologies. Until now, many production technologies of apple juices have been designed. However, some of these technologies may provide similar characteristics to the final juices. Besides, since minimal processing is increasingly promoted (Putnik et al., 2017), it will be a tendency to simplify and integrate the current technologies based on the similar product quality. Being an applied statistical data-processing method, multivariate data analysis was widely used in food analyzing and showed advantages in characterizing processing technologies used in food production (Dong, Hu, Chu, Zhao, & Tan, 2017; Kaya, Yıldız, & Ünlütürk, 2015).

Therefore, this study investigated the influence of nine different industrial production technologies on the physicochemical properties of juice including TPC, TFC, TSS, juice stability, turbidity, TA and pH. Multivariate statistical analyses including principal component analysis (PCA), linear discriminant analysis (LDA) and cluster analysis (CA) were performed to investigate the similarities and variations of tested apple juices. This study was intended to provide some references for simplifying and assimilating the current industrial production technologies according to the quality comparison of juices.

2. Materials and methods

2.1. Apple juice processing

Fuji apples (Malus pumila Mill) of high quality were purchased from Shandong province, China, and were transported to pilot test factory in Shaanxi Normal University (SNNU), Xi’an, China. Apples were processed to final apple juices from different nine industrial production technologies (Figure 1). The detailed parameters of unit operations applied in the current study were shown in Table 1. Each red code in Figure 1 represents an individual finished juice product and nine juices were obtained. Among all nine industrial production technologies, unit operations including pulping methods (traditional pulping [TP] and cold pulping [CP]), juicing modes (belt-style pressing [BSP] and horizontal spiral centrifuge separation [HSCS]), homogenization, sterilization methods (conventional thermal pasteurization and ultra-high temperature [UHT]) and two-stage sterilization were designed as factors that may affect the comprehensive quality of different apple juices. The schematic diagram of

![Figure 1. Flow diagram of apple juices based on nine different industrial production technologies. Red parts represent the code names of apple juices.](image)

**Figure 1. Flow diagram of apple juices based on nine different industrial production technologies. Red parts represent the code names of apple juices.**

**Figura 1. Diagrama de flujo de los jugos de manzana obtenidos empleando nueve tecnologías de producción industrial diferentes. Las partes en color rojo corresponden a los nombres codificados de los jugos de manzana.**
CP equipment was shown in Figure 2. After preparation of juices, all samples were stored at 4°C for further measurements.

2.2. Measurements of physicochemical properties

The TPC was measured using the colorimetric Folin–Ciocalteu method (Singleton & Rossi, 1965) with some modifications. Ten milliliter of diluted apple juice was mixed with 40 mL of 70% (v/v) ethanol in a beaker covered with the aluminum foil, incubating at 58°C for 35 min. Then the mixture was centrifuged at 9000 × g for 10 min at 4°C (TGL-16G, Anke Inc., China); 0.5 mL of supernatant was removed and mixed with 7.5 mL of 20% (w/v) sodium carbonate solution and 1 mL of 2 M Folin–Ciocalteu reagent in a 50-mL volumetric flask, filled with distilled water up to the mark and mixed gently. Then mixture was kept at 25°C for 60 min in dark, measuring absorbance at 765 nm (Multiskan Go, Thermo, USA). The amount of TPC was presented as milligram of gallic acid equivalents per liter of sample.

The TFC was detected using the method reported by Guerrouj, Sánchez-Rubio, Taboada-Rodríguez, Cava-Roda, and Marín-Iniesta (2016). The juice was centrifuged at 3000 rpm for 10 min and 250 µL of supernatant was mixed with 1.25 mL of distilled water. Next, 75 µL of 5% sodium nitrite solution was added and shaken gently. After 6 min, 150 µL of 10% aluminum chloride solution was added. After waiting for further 5 min, 0.5 mL of 1 M sodium hydroxide and 0.275 mL of distilled water were added and mixed well. The absorbance of the mixture (Multiskan Go, Thermo) was measured at 510 nm in triplicate. The results of TFC were expressed as milligram of rutin equivalent per liter of juice.

The TSS, expressing as “Brix, was measured using a refractometer (PAL-1, Atago Inc., Japan). The prism of the instrument was washed with distilled water after each analysis.

The stability was tested using the method reported by Versteeg, Rombouts, Spaansen, and Pilnik (1980) with some modifications. For each sample, before and after centrifugation of 10 min at 320 × g (TGL-16G, Anke Inc.), the absorbance at 660 nm was measured using a spectrophotometer (WFJ 2100, Unico Inc., China). The stability of juice samples was calculated by the following equation:

\[
\text{Stability} \% = \left( \frac{\text{OD}_{660 \text{after centrifugation}}}{\text{OD}_{660 \text{before centrifugation}}} \right) \times 100
\]

The turbidity was determined using a turbidimeter (ET76910, Lovibond Inc., Germany). Prior to measurement, standard samples with turbidity ranged from 0 NTU (nephelometric turbidity units) to 800 NTU were used to calibrate instrument.

Table 1. Parameters of unit operations applied in the juice production.

| Applied unit operations                  | Parameters      |
|-----------------------------------------|-----------------|
| Killing enzymes                         | 98°C, 30 s      |
| Disc separator separation               | 4200 rpm        |
| Horizontal spiral centrifuge separation | 6000 rpm        |
| Homogenization                          | 300 Bar         |
| Thermal pasteurization                   | 98°C, 50 s      |
| Ultra-high temperature                   | 121°C, 15 s     |
| Two-stage sterilization                  | 90°C, 5 min     |

Figure 2. Schematic diagram of cold pulping equipment. The equipment was made by the ministry of China agriculture of modern apple industry technology system (depending on the Shaanxi Normal University) and Xi'an Ding He machinery manufacturing.

Figura 2. Diagrama esquemático del equipo de despulpado en frío. El equipo fue fabricado por el Ministerio de Agricultura chino por el equipo de trabajo de sistemas tecnológicos para la industria de la manzana (dependiente de la Universidad Normal Shaanxi) y de la empresa fabricante de maquinaria Xi’an Ding He.
The TA was determined by titration with 0.1 M solution of NaOH which was standardized before titration. The results were expressed as gram per liter with reference to malic acid. The pH value was measured using a digital pH meter (FE20 Plus, Mettler-Toledo Inc., China) which was calibrated by buffer solutions of pH 4 and pH 7 prior to measurement, and the pH was measured at 25 ± 0.5°C. All analyses were repeated three times for every treatment.

2.3. Statistic and multivariate data analyses

Data obtained in this study were subjected to statistic analysis using SPSS 13.0 (SPSS Inc., USA) and expressed as means ± standard deviations. Results were submitted to the analysis of variance (ANOVA) with significance level of p < 0.05, evaluated by Duncan’s test. For multivariate data analysis, CA and pattern recognition including PCA and LDA were applied according to the physicochemical properties of apple juices.

3. Results and discussion

3.1. Measurement results of physicochemical properties

Physicochemical properties including TPC, TFC, TSS, stability, turbidity, TA, and pH of the nine apple juices were determined and summarized in Table 2. The correlation analyses of all tested variables were shown in Table 3.

For TP+BSP technology-based juices (A1, A2, A3, A4 and A5), apples were pulped by TP and juiced by BSP. However, in case of CP+HSCS technology-based juices (B1, B2, B3 and B4), apples were pulped by CP and juiced by HSCS. As shown in Table 2, it was interesting to see that TPCs of A1, A2, A3 and A4 were significantly higher (p < 0.05) than those of A5, B1, B2, B3 and B4. Due to the characteristics of TP and CP, significant differences of TPC of tested juices were mainly attributed by pulping method. TP crushed full apples and delivered whole pulp to juicing stage. However, as shown in Figure 2, CP crushed full apples but separated peel and seeds from apple pulp before juicing. Thus, the TP+BSP juices were produced from whole apple pulp containing peel and seeds, while the CP+HSCS juices were produced from pure apple flesh pulp without peel and seeds. Since apple peel contains considerably higher contents of total phenols compared to apple flesh (Ahmadi-Azfadi, Nybom, Ekholm, Tahir, & Rumpunen, 2015), it is rational to explain the higher TPC of TP+BSP juices except A5. Although being equally produced by TP+BSP, juice A5 possessed much lower TPC than juices A1, A2, A3 and A4. Due to the fact that juice A5 was sterilized by UHT, the higher temperature treatment inevitably caused deterioration on nutrition (Guan et al., 2016).

As shown in Table 2, compared to TPC of juices A1, A3, B1 and B3, TPC of A2, A4, B2 and B4 increased significantly (p < 0.05), respectively. According to Figure 1, this phenomenon indicated that the two-stage sterilization increased the TPC of juices. Previous studies had shown that the effect of thermal treatments on TPC is variable (Azofeifa, Quesada, Pérez, Vaillant, & Michel, 2015). Many studies reported that thermal treatment caused significant loss in TPC (Aguilar-Rosas, Ballinas-Casarrubias, Nevarez-Moorillon, Martin-Bellos, & Ortega-Rivas, 2007; Sanhíjarsegaram, Razali, & Somasundram, 2013), while a number of researches provided different findings (Hager, Howard, & Prior, 2010; Mena et al., 2013). Branco et al. (2016) found that TPC and TFC increased after pasteurization (about 7.2% and 16.4%, respectively). Since this variability is related to many complex factors including the severity of the heat process, the characteristic of the food matrix and the sensitivity of different phytochemicals to the temperature (Rawson et al., 2011), further investigations need to be conducted in this regard. In the current study, the juices were the not from concentrate (NFC) cloudy apple juices. Due to the preference of consumers, NFC cloudy juice is expected to be full of dispersed flesh particles which are mainly formed by the cellular tissues of fruits. Although the heat treatment degraded the TPC of juices (De Paepe et al., 2014), but it greatly ruptured a large amount of cells at the same time, allowing relatively more phenolic compounds to leak out and increasing the TPC of final juice.

The mean values of TFC showed the same tendency of variation as TPC showed, while almost no significant differences (p > 0.05) of TFC were observed within TP+BSP juices and CP+HSCS juices, respectively. The correlation analysis

|   | TPC (mg/L) | TFC (mg/L) | TSS (% Brix) | Stability | Turbidity (NTU) | TA (g/L) | pH |
|---|------------|------------|--------------|-----------|----------------|----------|----|
| A1 | 239.2 ± 4.3c | 63.0 ± 2.7ab | 12.7 ± 0.1a  | 57.2 ± 6.1ab | 175.7 ± 9.7d | 3.35 ± 0.02cde | 3.79 ± 0.10a |
| A2 | 257.7 ± 7.5ab | 64.1 ± 2.5a | 12.7 ± 0.2a | 59.2 ± 5.0ab | 204.3 ± 9.3c | 3.29 ± 0.08cde | 3.88 ± 0.12a |
| A3 | 246.3 ± 7.2bc | 61.3 ± 1.8abc | 12.6 ± 0.1a | 53.5 ± 3.1bc | 169.7 ± 5.7c | 3.39 ± 0.03cd | 3.81 ± 0.09a |
| A4 | 261.9 ± 8.8a | 62.8 ± 2.6ab | 12.6 ± 0.1a | 57.6 ± 5.2ab | 183.7 ± 10.6d | 3.35 ± 0.01cde | 3.85 ± 0.12a |
| A5 | 197.4 ± 3.6d | 58.4 ± 3.4bcd | 12.7 ± 0.1a | 44.4 ± 6.3c | 180.7 ± 9.0d | 3.27 ± 0.07de | 3.84 ± 0.01a |
| B1 | 150.7 ± 5.7f | 56.4 ± 2.3cd | 12.7 ± 0.1a | 64.2 ± 6.7ab | 312.7 ± 11.1b | 3.78 ± 0.08a | 3.80 ± 0.04a |
| B2 | 187.2 ± 2.9de | 58.6 ± 2.0abcd | 12.6 ± 0.1a | 67.8 ± 4.8a | 336.7 ± 11.8a | 3.41 ± 0.04c | 3.83 ± 0.07a |
| B3 | 146.2 ± 7.5f | 53.8 ± 1.8d | 12.7 ± 0.1a | 62.7 ± 5.0ab | 307.7 ± 6.9b | 3.63 ± 0.04b | 3.81 ± 0.14a |
| B4 | 178.7 ± 7.2e | 57.4 ± 2.6bcd | 12.7 ± 0.2a | 66.2 ± 5.3ab | 324.7 ± 8.3b | 3.24 ± 0.05e | 3.82 ± 0.11a |

All data were presented as means ± standard deviations. Values followed by different letters within the same column are significantly different (Duncan’s test, p < 0.05). Total phenolic content, total flavonoid content and titratable acidity were expressed as gallic acid equivalent, rutin equivalent and malic acid equivalent, respectively.

TPC: total phenolic content; TFC: total flavonoid content; TSS: total soluble solid; TA: titratable acidity.

Todos los datos se presentan ± las desviaciones estándares. Los valores que exhiben letras distintas al interior de la misma columna son significativamente diferentes (prueba de Duncan, p<0,05). El contenido fenólico total, el contenido de flavonoides total y la acidez valorable se expresan como equivalentes de ácido gálico, de rutina y de ácido málico, respectivamente. TPC, contenido fenólico total; TFC, contenido de flavonoides total; TSS, total de sólidos solubles; TA, acidez valorable.
Table 3. Correlations analysis of the physicochemical properties.

| TPC     | TFC  | TSS   | Stability | Turbidity | TA     |
|---------|------|-------|-----------|-----------|--------|
| TFC     | 0.75** |
| TSS     | -0.08 | 0.02  |
| Stability | -0.32 | -0.46 | -0.13   |
| Turbidity | -0.80** | -0.58 | 0.05 | 0.66* |
| TA      | -0.59 | -0.46 | 0.06 | 0.25 | 0.41   |
| pH      | 0.15 | -0.20 | 0.30 | 0.14 | -0.10 | -0.16 |

* and ** mean the correlations are significant at $p < 0.05$ and $p < 0.01$ levels, respectively.

Also demonstrated that the TPC was significantly and positively correlated with TFC ($R^2 = 0.75$, Table 3). Being an important part of total phenols, flavonoids are a group of about 4000 naturally polyphenolic compounds, found universally in plant origin (Liu, Ponnumasmy, Lee, Jong, & Chen, 2017; Sharma & Jammeda, 2017). As shown in Table 2, TFC of the juices changed variably but showed no sharp fluctuations. The ANOVA indicated that the TFCs of juices were relatively stable than TPC. Being frequently modified with glucose and rhamnose residues, aglyconic flavonoids generally exist in the form of flavonoid glycosides (Beck & Stengel, 2016). Due to the stable structure of aglyconic flavonoids, the TFC is much less strongly affected by the processing operations, accounting for the stability of TFC (Capanoglu, De Vos, Hall, Boyacioglu, & Beekwilder, 2013).

TSS is a measure of inorganic and organic contents in a liquid in molecular, ionized or microgranular suspended form, which is predominantly sugar in case of apple juice. It can be seen that no significant difference ($p > 0.05$) was detected among TSS of the nine apple juices, indicating that the TSS of apple juice was much less strongly affected by processing.

For stability, no significant difference ($p > 0.05$) was detected among most juice samples except A5 which showed the lowest stability. The result indicated that apple juice sterilized by UHT exhibited relatively worse physical stability than samples treated by conventional thermal pasteurization or conventional thermal pasteurization combined with two-stage sterilization. The same phenomenon was detected in comparison between pasteurized milk and UHT-heated milk (Heilig, Celik, & Hinrichs, 2008). In the current study, the stability values of juices were measured using centrifugal process. According to the Stokes’ law, it is easier for the large-size particles to precipitate, whereas the smaller particles tend to remain in the supernatant after centrifugation (Lv, Kong, Mou, & Fu, 2017). Although no statistically significant difference was observed, the mean values of stability of CP+HSCS juices were higher than those of TP+BSP juices (Table 2), indicating the existence of smaller size particles in CP+HSCS juices. Moreover, turbidity, which is also called as serum cloudiness, is another important indicator to reflect the juice stability. As shown in Table 3, the turbidity was positively correlated with the stability values of juices ($R^2 = 0.66$). The results of ANOVA showed that the CP+HSCS juices possessed higher turbidity (about 75%) compared to TP+BSP juices. The lower values of turbidity reflected the unstable characteristics of juices (Lv et al., 2017), demonstrating better stability of CP+HSCS juices. Besides, it should be concerned that turbidities of samples within TP+BSP juices and CP+HSCS juices showed the same tendency. It can be seen that juices treated with two-stage sterilization (A2, A4, B2 and B4) possessed higher turbidity than juices only treated by thermal pasteurization (A1, A3, B1 and B3). Since heating greatly ruptures cell structure which allows pectin to leak out, the higher concentration of colloidal pectin in juice attributes the increased turbidity (Santhirasegaram, Razali, George, & Somasundram, 2015). Among all tested juices, A2 and B2 held the highest turbidity within their groups, respectively, which indicated that the combination of homogenization and two-stage sterilization increased turbidity significantly ($p < 0.05$).

For TA of the nine juices, it can be seen that, although varied to different degree, no significant difference ($p > 0.05$) was detected among TP+BSP-based juices. For CP+HSCS-based juices, TA in apple juices decreased significantly after two-stage sterilization since thermal treatment caused evaporation of organic acids (Charles-Rodriguez et al., 2007). Similar to the case of TSS, no significant difference was detected in pH among the nine apple juices (Table 2). Theoretically, TA is much related to the pH value as total acidity affects the pH directly (Cairns, Watson, Creanor, & Foye, 2002). However, the correlation coefficient between pH and TA in the current study was only −0.16 (Table 3). Fruit juice is one of the most common natural buffer systems which contains a significant amount of weak acids, providing juice a resistance to change in the pH. As in the current study the variations in TA values (from 3.24 to 3.78 g/L, Table 2) were very small, consequently the pH values were remained comparatively stable due to the existence of buffer system leading to an inapparent correlation between pH and TA. Since juices produced by different technologies exhibited varied physicochemical properties and showed different benefits in some certain characteristics among the seven tested parameters, it was hard to determine which technology proved better for the production of apple juice and whether some technologies provoked similar effects to the overall quality of juices. Thus, multivariate data analyses including PCA, LDA and CA were applied to treat the original data in Table 2 for discriminating the commonality and distinction of apple juice samples.

3.2. Multivariate data analysis

Since the results of physicochemical measurements were accurate and reliable, physicochemical data of the nine apple juices and three parallel subsamples for each were treated. Multivariate data analyses were conducted by PCA, LDA and CA. It was shown that the sum of the first three dimensions of PCA (Figure 3) and that of the first two dimensions of LDA (Figure 4) were 79.15% and 95.9%, respectively. This indicated that the PCA and LDA can explain most original data and give reliable results to characterize the quality of studied juices. Besides, the clusters of CA (Figure 5) exhibited the same result that verified the results of PCA and LDA once again.

3.2.1. Principal component analysis

Being an unsupervised pattern recognition method (Visnevski-Necrasov et al., 2015), PCA is a way of identifying patterns in data and expressing the data in a way as to emphasize their similarities and differences (Shin, Craft, Pegg, Phillips, & Eitenmiller, 2010). It can compress data set...
and reduce the number of dimensions without losing much information of original data set. To observe the general effects of the different industrial production technologies on the physicochemical properties of final apple juices, a PCA procedure was performed according to the measured variables summarized in Table 2 (27 samples × 7 physicochemical properties). During this procedure, the correlation matrix was used for extraction analysis and the rotation was obtained using varimax.

Table 4 showed the total variance explained in PCA procedure. The first principal component (PC1) had the highest eigenvalue of 3.16 and accounted for 45.08% of the variability in the data set. The second and the third PCs (PC2 and PC3) had eigenvalues of 1.34 and 1.05 and accounted for 19.14% and 14.93% of the variance in the data set, respectively. Since PC4 to PC7 yielded progressively smaller eigenvalues, which were less than 1.0, the first three PCs were used for further study according to Kaiser’s rule. Thus, the original seven-dimension data matrix was compressed and reduced to a three-dimension one and presented in a three-dimension score plot (Figure 3). It can be seen that the first three dimensions explained 79.15% of the variances of all measured physicochemical properties. Generally speaking, when the PCs have more than 70% cumulated reliability of the original data set, these PCs can be used to replace the original one, and the corresponding results could be competent. As most of the total variance was explained, Figure 3 was proved to be impactful for representing the original data set of seven tested variables.

The loading variables of PCA in the first three PCs, representing the correlation between tested physicochemical properties and components, were shown in Table 5. Thus, PC1 was negatively correlated to TPC and positively correlated to TA and turbidity. PC2 was positively correlated to stability, and PC3 was positively correlated to TSS and pH. As shown in Figure 3, the score plot defined by the first three PCs, a clear separation of juices according to the measured physicochemical properties, was obtained. All data was located in three separated regions. A1, A2, A3 and A4 were located in an isolate group, and B1, B2, B3 and B4 were scattered in another independent region; while only A5 lied in a separate zone singly. Samples lying close together in the score plot were characterized by a high similarity in juice quality. For the CP+HSCS technology-based juices (B1, B2, B3 and B4), the data scattered positively in PC1 and PC2, indicating higher TA and turbidity, lower TPC, as well as higher stability of these juices. Apart from A5, the TP+BSP technology-based juices (A1, A2, A3 and A4) were located negatively in PC1, exhibiting that these four juices possessed higher TPC, as well as lower TA and turbidity. Equally being produced by TP+BSP technology but treated by UHT, A5 was separated singly. Being scattered negatively in PC2, A5 correspondingly showed relatively worse stability compared to other juices. All these information derived from Figure 3 well accounted for the original data and were in agreement with the results summarized in Table 2.

Although apple juice samples were separated into three groups and there was no overlapping area in PCA plot, the total variance explained in the first three PCs still not sufficient. Thus, a further analysis needs to be performed for confirming the PCA result.

3.2.2. Liner discriminant analysis

Compared to PCA, LDA is a supervised pattern recognition method that can maximize the variance between categories and minimize the variance within categories to separate the different samples completely (Qiu, Wang, & Gao, 2015). It only looks for a sensible rule to discriminate between them by forming linear functions of the data maximizing the ratio of the between-group sum of squares to the within-group sum of squares (Zhao, Wang, Lu, & Jiang, 2010).

To achieve a clear separation among studied juice samples, LDA was subsequently performed on the autoscaled data matrix composed of 27 juice samples. The different juice technologies were set as grouping variables. All the measured physicochemical properties (Table 2) were selected as markers through stepwise procedure and the canonical discriminant analysis resulted in seven discriminant functions. A satisfactory differentiation according to the juices was achieved with a recognition ability.
of 100.0%. However, for the prediction ability, 92.6% of original grouped cases were correctly classified, while only 59.3% of cross-validated grouped cases were correctly classified, evaluated by using the leave-one-out cross-validation (supplementary Table 1). The scores of the first two functions for each sample were plotted in Figure 4. As the two-dimensional score plot of LDA explained 95.9% of the total variance, with LD1 and LD2 representing 88.5% and 7.4% of the variance, respectively, LDA provided a high reliability to differentiate apple juice samples from each other.

The studied apple juice samples produced from the nine industrial production technologies were distinguished into three isolated regions, being space bounded by three circles in Figure 4. Expectantly, the classification result of LDA was corresponding to that of PCA. For both PCA and LDA, TP+BSP juices except A5 were located in an isolate group, while A5 was separated singly. CP+HSCS juices were scattered in another independent region. Juices located in the same area possess similar overall physicochemical properties. It should be concerned that the differences of production technologies of A1, A2, A3 and A4 were homogenization and two-stage sterilization and the same to CP+HSCS juices (B1, B2, B3 and B4). Although the two-stage sterilization significantly increased TPC of apple juices and the homogenization affected the juice stability and turbidity (Table 2), the results of both PCA and LDA indicated that the overall quality of studied apple juices was less strongly affected by two-stage sterilization and homogenization.

3.2.3. Cluster analysis

According to PCA and LDA, apple juices produced from the nine industrial production technologies were discriminated into three categories and the separations of both methods were the same. To further verify the results of PCA and LDA, data set was treated by CA method, a technique that attempts to separate data into specific groups based on similarities or distances among observations (Huang, Guo, Qiu, & Chen, 2007). It can combine the closest data more flexibly, ignoring the category of sample.

A hierarchical CA method was applied to treat 27 apple juice samples based on the measured physicochemical properties. Prior to the treatment, each variable was standardized. Moreover, a heat map (Heml 1.0.3.3, Heatmap Illustrator, China) was applied to display how the nine juices varied in physicochemical properties. For cluster membership, single solution was chosen. The Ward’s linkage method provided a high reliability to differentiate apple juice samples from each other.

Table 4: Total variance explained in PCA.

| Component | Initial eigenvalues | % of Variance | Cumulative (%) |
|-----------|---------------------|--------------|---------------|
| 1         | 3.16                | 45.08        | 45.08         |
| 2         | 1.34                | 19.14        | 64.22         |
| 3         | 1.05                | 14.93        | 79.15         |
| 4         | 0.67                | 9.57         | 88.72         |
| 5         | 0.53                | 7.55         | 96.27         |
| 6         | 0.22                | 3.11         | 99.37         |
| 7         | 0.04                | 0.63         | 100.00        |

Extraction method: principal component analysis.
Método de extracción: Análisis de componentes principales.

Table 5: Loadings of the features in the first three principal components for apple juice.

| Component  | Component 1 | Component 2 | Component 3 |
|------------|-------------|-------------|-------------|
| TPC        | −0.901      | −0.271      | −0.011      |
| TA         | 0.793       | 0.021       | −0.021      |
| Turbidity  | 0.699       | 0.547       | −0.053      |
| TFC        | −0.601      | −0.588      | −0.146      |
| Stability  | 0.216       | 0.854       | −0.088      |
| TSS        | 0.217       | −0.289      | 0.847       |
| pH         | −0.341      | 0.442       | 0.750       |

Extraction method: principal component analysis. Rotation method: Varimax with Kaiser normalization.
Método de extracción: Análisis de componentes principales. Método de rotación: Varimax con normalización de Kaiser.
was taken as the clustering algorithm and the squared Euclidean distance was taken as a measure of interval as shorter distances showed more similarity. The heatmap combined with dendrogram of CA was shown in Figure 5.

Regarding the CA, all juice samples were grouped into three clusters being space bounded by red frames, in accordance with the results of PCA (Figure 3) and LDA (Figure 4). The clusters demonstrated that A1, A2, A3 and A4 were grouped closely possessing higher contents of total phenols and total flavonoids, while A5 formed a single group in the dendrogram. Besides, the CP+HSCS technology-based juices (B1, B2, B3 and B4) were clustered into the same group for their relatively better stability, higher turbidity and TA.

In case of the clustering of CP+HSCS juices, it should be concerned that B1 showed high similarity to B3 for they were grouped into the same cluster, whereas B2 and B4 were in high similarity. According to the production technologies (Figure 1), B2 and B4 were additionally performed by the two-stage sterilization compared to B1 and B3, respectively. This indicated that, via CA, the two-stage sterilization affected juice quality to some extent. Although differences were observed within CP+HSCS juices through CA, the two clusters were combined and grouped into the same cluster in the next merge process. For three clusters presented in the dendrogram, the memberships in each cluster were in high coincidence with the results of PCA and LDA. Overall, the high coincidence of the results based on PCA, LDA and CA indicated the reliability for characterizing the apple juices produced from different production technologies.

4. Conclusion

The physicochemical properties of juices produced from nine different production technologies were investigated, and the multivariate data analyses including PCA, LDA and CA were performed for characterizing the qualities of juices. Juices of nine technologies were well grouped into three categories according to the determined variables. Pulping methods (TP and CP), juicing modes (BSP and HSCS) and sterilization methods (conventional thermal pasteurization and UHT) were proved to be the three main factors affecting the general quality of apple juices. As the juices grouped into the same category exhibited similar quality score in our study, their production technologies can be integrated or simplified. Our outcomes revealed that quality index of final juices was actually grouped into three categories instead of nine different production technologies. Thus, some of the current production technologies can be integrated in order to simplify the production procedures. Moreover, as minimal processing is increasingly encouraged, our study may provide a new perspective and reference to simplify current industrial production technologies.

Acknowledgment

This research was supported by an earmarked fund from China Agriculture Research System (CARS-28).

Disclosure statement

No potential conflict of interest was reported by the authors.

Funding

This research was supported by an earmarked fund from China Agriculture Research System (CARS-28).

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