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Investigation of the Possibility of Cider Production in Armenia Using Different Dry Active Yeasts

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ABSTRACT

The volume of production and consumption of cider as a low alcoholic and soft drink is increasing year by year. Being a part of the global economy, a similar trend is also noticeable among a smaller scale Armenian producers and consumers. During this research we studied the physicochemical parameters of apple juice and ready-made cider obtained from apple varieties common in Armenia. Fermentation was carried out with four different yeasts intended for cider production and one control sample. The aim was to reveal the effect of each yeast on the quantitative and qualitative characteristics of the final cider.

Introduction

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The climatic conditions of the Republic of Armenia are favorable for growing different types of fruits. Apple cultivation is widespread in Armenia, it is cultivated almost in all regions of the republic. In recent years, an average of 100 000 tons of fresh apples have been produced in Armenia (Statistical Committee of the Republic of Armenia), most of which have been consumed as fresh fruit without processing, and the rest – as raw materials for the production of juices and alcoholic beverages,such as cider and fruit distillates, Calvados.

Cider is known by different names in the world, it can differ in alcohol and sugar content, as well as in production method depending on the country of production (Kosseva, et al., 2017). Being a weak alcoholic and soft drink, the volume of production and consumption of cider is increasing every year (Cider Market Report, 2019-2025). This trend is also noticeable in Armenia, although the volumes are still small.

The purpose of this research was to study the cider production technology from apple varieties common in Armenia, specifically using four different yeasts intended for cider production.

In one of the numerous studies carried out worldwide, non-Saccharomyces yeast strains, combined and uncombined, used for the production of low-alcohol beer, have also shown good results for the production of new types of more aromatic beverages (Madrera, et al., 2021). However, Saccharomyces yeasts strains still occupy a leading position in the fermentation process of the production of various alcoholic beverages due to their efficiency and aromatic characteristics.

To carry out our research, four strains of active dry yeast for the production of cider of the Lesaffre Group French company Fermentis brand were selected. The choice of yeast strains is not accidental, as studies carried out by the producer have shown that the yeast strains can have an important effect not only in terms of the analysis of fermentation dynamics, but also in terms of cider sensory perception. They can be viewed as a powerful tool to diversify the cider offered in the market, and cider makers can produce a distinctive drink using the same raw materials.

Materials and methods

Apple juice from the most common apple varieties grown in Armenia – Golden Delicious, Renet Simirenko and Aydored was used for the research.

For the fermentation, four types of dry active yeasts were used: SAFCIDER™ AB-1, SAFCIDER™ AS-2, SAFCIDER™ AC-4, SAFCIDER™ TF-6. As for cider production in Armenia it is common to use a yeast made for beer production, while SAFALE™ S-04 beer yeast was used as a control sample.

The study of the physicochemical composition of apple juice samples was carried out using the methods accepted in enochemistry, in accordance with the standards developed by the OIV (International Organization of Grapes and Wine) and GOSTs in force in RA, as well as with other experimental methods.

Apple juice provided by Yerevan Beer Factory was poured into glass fermentation containers prepared for five samples. Potassium metabisulphite at 50 mg/l was added to the samples to suppress the possible life activity of undesirable microorganisms. One of the five types of yeasts was added to each of the apple juice sample, according to the manufacturer's recommended dosage, and subjected to fermentation. Fermentation was carried out in glass containers, which were sealed with airlock lids. Fermentation was carried out at a temperature of 18-20 ^{0}C under conditions recommended by the producer. The entire process of fermentation was monitored. After the completion of fermentation, the fermented samples were decanted from the sediment. A study of the chemical composition of one control cider (sample: S-04) and four types of ciders obtained (sample: AB-1, AS-2, AC-4, TF-6) was carried out according to the above mentioned methods. Potassium metabisulphite at 50 mg/l was added to the fermented cider samples, and they were transferred to the refrigerator and stored at +2-+4 ^{0}C temperature conditions. In three months the samples were decanted from the sediment. The samples separated from the sediment were kept for a week at room temperature and then potassium metabisulphite was added at a dose of 50 mg/L. A small part of the samples was degustated on the spot and the rest of the samples was transferred to the refrigerator to be degustated after aging.

The study of the physicochemical indicators of the apple juice and ciders include the following analyses (indicating the appropriate methods of study): total acidity (OIV-MA-AS313-01), pH-active acidity, sugars (measurement by refractometer), total and free sulfur dioxide (OIV-MA-F1-07), fermentable nitrogen YAN (Fracassetti, et al., 2017), total phenols (Zoecklein, et al., 1999), chromatic characteristics (OIV-MA-AS2-07B), Folin Ciocalteu index (OIV-MA-AS2-10) (International Organization of Vine and Wine, 2022). Determination of alcohol content (OIV-MA-AS312-01A), as well as determination of residual sugar, volatile acidity (OIV-MA-AS13-02), aldehydes and acetals, was also performed for cider. Organic acids were measured by HPLC (system: Agilent 1100 Series, detector: Agilent 1260 Infinity) (Schneider, et al., 1987), phenolic compounds – by photocolorimetry (photocoloroimetric) method (Jacobson, 2006).

Results and discussions

The results of the laboratory analyses of the main physicochemical parameters of apple juice and ciders are presented in Table 1.

The acidity of the juice mixtures used for fermentation is important because it helps to control the growth of undesirable microbes. The active acidity index of juice pH partly determines the antibacterial and antioxidant capacity of sulfites to inhibit the growth of wild microflora and yeasts, and as juice pH increases, more quantity of free SO_2 is required (Beech, 1972). Sulfur dioxide inhibits wild yeast growth and allows Sacharromyces strains (which are more resistant to SO_2 than wild yeasts and most potentially spoilage microorganisms) to thrive and dominate fermentation (Jarvis & Lea, 2000). In addition to its antimicrobial effects, SO_2 is a powerful antioxidant that can prevent juice oxidation and browning. However, SO_2 is only effective in its free or unbound form. In our study, despite the relatively high content of sulfur dioxide, the yeasts added to the apple juice were fully functional and they completed the fermentation (Table 1).

The final pH and total acidity of the cider play an important role in the stabilization and shelf life of the bottled product. Low acidity (pH > 3.8) can lead to growth of microorganisms spoilage and bad odors (Lea & Drilleau, 2003). Final acidity also has a big impact on the flavor profile of a cider; high acidity can impart harshness to the cider, which usually requires the addition of some amount of sugar to balance the cider's flavor. As we can see in Table 1, the total acidity of apple juice is 2.75 g/l and the pH is 3.39, in the case of AS-2 yeast, the total acidity increased the most, reaching 3.71 g/l, and the pH – 3.28, respectively. In the case of AC-4 and TF-6 yeasts, total acidity increased by the same amount, reaching 3.58 g/l, and pH – 3.36, in the case of S-04, the amount of total acids increased by a relatively smaller amount, reaching 3.39 g/l, and the pH – 3.38, in the case of AB-1, the amount of total acids remained the same – 2.75 g/l, and the pH, unlike the other samples, increased to 3.53. The indicators of volatile acidity of fermented ciders are within acceptable limits (Table 1). The lowest amount of volatile acidity in cider was obtained with S-04 yeast – 0.19 g/l, then TF-6 – 0.21 g/l, AS-2 – 0.25 g/l, AB-1 – 0.30 g/l, and the highest was in the case of AC-4 – 0.35 g/l.

Apple juice contains a variety of plant secondary metabolites that include an aromatic ring and at least one hydroxyl group and are commonly referred to as phenolics or tannins (Shi, et al., 2003). Polyphenolic compounds in plants act as defense mechanisms against insects, bacteria and fungi. Secondary metabolite concentrations in fruit are influenced by many factors, such as light, temperature and other growth regulators, including terroir. Polyphenolic compounds are broken down by oxidation, which occurs mainly due to contact with air during grinding or crushing of apples, in the presence of the enzyme polyphenoloxidase. Apple pressing and juice fermentation methods for cider can also affect final phenolic concentrations in bottled products (Merwin, et al., 2008). Folin Ciocalteu Index (FCh-I) is a popular international method for measuring the total content of phenolic substances in food. Laboratory analyses performed during our research show that the total content of phenolic substances in apple juice was 696.05 mg/l, and the Folin Ciocalteu index (FCh-I) was 15.82. During fermentation, the amount of the phenolic compounds decreased (Table 1). The total content of phenolic compound is the highest in the S-04 cider sample, 576.71 mg/l, and Folin Ciocalteu index (FCh-I) is 13.10.

Table 1. Results of laboratory analyses of some physicochemical parameters in the studied apple juice and ciders*

Parameters	Unit	Apple juice	Cider S-04	Cider AB-1	Cider AS-2	Cider AC-4	Cider TF-6
Alcohol	Vol.%	-	7.00	7.40	7.20	7.50	7.20
Total acidity	g/l	2.75	3.39	2.75	3.71	3.58	3.58
pH	-	3.39	3.38	3.53	3.28	3.36	3.36
Volatile acidity	g/l	-	0.19	0.30	0.25	0.35	0.21
Sugar	g/l	130	1.47	1.09	1.09	0.93	1.16
Free Sulfur dioxide	mg/l	50.59	3.72	2.79	3.72	3.10	3.41
Total Sulfur dioxide	mg/l	110.01	59.29	55.87	52.46	46.87	63.32
Reductones Sulfur dioxide	mg/l	2.54	2.48	2.48	2.48	2.17	3.10
Yeast Assimilable Nitrogen (YAN)	mg/l	244.72	33.60	39.20	33.60	33.60	30.80
Folin Ciocalteu Index (FCh-I)	-	15.82	13.10	11.82	12.63	12.16	12.69
Total Phenols	mg/l	696.05	576.71	520.22	555.78	535.43	558.55
Aldehydes	mg/l	-	32.56	40.04	57.20	36.08	28.60
Acetals	mg/l	-	17.70	50.74	20.06	12.98	18.88

*Composed by the authors.

In AS-2 and TF-6 cider samples, the indicators are almost the same: 555.78 mg/l, FCh-I: 12.63 and 558.55 mg/l, FCh-I: 12.69. AC-4 cider sample: 535.43 mg/l, FCh-I: 12.16, and AB-1 cider sample: 520.22 mg/l, FCh-I: 11.82.

It is desirable that the sugar content of the apple juice intended for fermentation is not higher than 140 g/l to make cider with alcohol content close to 7.00 vol. % as a result (Berry & Slaughter, 2003). The sugar content of the apple juice of our study was 130 g/l (Table 1), and as a result of its fermentation, the highest amount of alcohol was obtained in the case of AC-4 yeast – 7.5 vol. %, and the lowest amount in the case of S-04 – 7.00 vol. %. In the case of AB-1 yeast, the amount of alcohol was 7.40 vol. %, in the case of AS-2 and TF-6 – 7.20 vol. % (Table 1). The content of residual sugar did not exceed 1.5 g/l in any sample (Table 1).

The amount of yeast assimilable nitrogen (YAN) in apple juice can vary greatly depending on the apple variety (Technical Overview on Cider Production, 2018). The content of YAN in the apple juice of our study was 244.72 mg/l (Table 1), and as the YAN (mg/l)/Sugar (g/l) ratio is equal to 1.88, that is, it is higher than the instructions provided by the company that produces the yeast, no additional nutrients were given during fermentation. During fermentation nitrogen consumptions were in full accordance with the manufacturer's instructions. TF-6 consumed the most, leaving a residue of 30.80 mg/l, samples S-04, AS-2 and AC-4 had the same consumption of fermentable nitrogen, leaving a residue of 33.60 mg/l, and sample AB-1 had the least consumption, leaving 39.20 mg/l residue.

Aldehydes are produced as a result of the non-enzymatic process of oxidative-deamination of amino acids (alanine) and in the Krebs cycle. In small concentrations, they have a pleasant smell and participate in the formation of special aromas (Kazumyan, et al., 2022). The quantity of aldehydes in our examined samples was: S-04 – 32.56 mg/l, AB-1 – 40.04 mg/l, AS-2 – 57.20 mg/l, AC-4 – 36.08 mg/l, TF-6 – 28.60 mg/l (Table 1). Acetals have a direct, positively significant correlation with methanol, ethers and aromatic acids. The addition of acetals during fermentation increases their presence in the wine. The quantity of acetals in our samples was 17.70 mg/l in S-04, 50.74 mg/l in AB-1, 20.06 mg/l in AS-2, 12.98 mg/l in AC-4 and in TF-6 – 18.88 mg/dm^3 (Table 1).

The color characteristics of the samples were measured with a UNICO 2802 UV/VIS photoelectric colorimeter at 420, 520, and 620 nm (International Organization of Vine and Wine, 2021). As a result, the color intensity of the apple juice was 1.72, and the color intensity of the cider samples in descending order was: AS-2 – 0.39, TF-6 – 0.34, S-04 – 0.34, AB-1 – 0.32, AC- 4 – 0.32. In apple juice, the color shade is 2.22, and in the cider samples, the highest color shade is as follows: AB-1 – 3.84, AC-4 – 3.55, AS-2 – 3.31, TF-6 – 3.16, S-04 – 3.13.

Color characteristics are made up by combining three colors measured respectively – at 420 nm: yellow, 520 nm: red, and 620 nm: blue (Table 2). The percentage ratio of color characteristics in the investigated apple juice and cider samples is presented in Diagram.

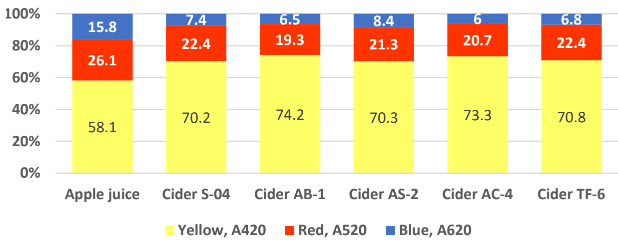

Diagram. Color characteristics of the studied apple juice and ciders, % *(composed by the authors).*

Table 2. Color characteristics of the studied apple juice and ciders*

Parameters	Apple juice	Cider S-04	Cider AB-1	Cider AS-2	Cider AC-4	Cider TF-6
Yellow, A420	0.9984	0.2390	0.2375	0.2757	0.2355	0.2410
Red, A520	0.4498	0.0764	0.0618	0.0834	0.0663	0.0763
Blue, A620	0.2722	0.0249	0.0206	0.0330	0.0194	0.0230
Color intensity	1.72	0.34	0.32	0.39	0.32	0.34
Color shade	2.22	3.13	3.84	3.31	3.55	3.16

*Composed by the authors.

Table 3. Results of laboratory analyses of organic acids in the studied apple juice and ciders*

Parameters	Unit	Apple juice	Cider S-04	Cider AB-1	Cider AS-2	Cider AC-4	Cider TF-6
Malic acid	g/l	4.11	3.38	2.69	3.77	3.87	3.51
Lactic acid	g/l	0.05	0.43	0.17	0.17	0.2	0.31
Tartaric acid	g/l	-	-	-	-	-	-
Formic acid	g/l	0.14	0.31	0.26	0.17	0.22	0.31
Ascorbic acid	g/l	-	-	-	-	-	-
Shicimic acid	mg/l	8.60	14.10	9.60	11.40	11.60	20.00
Acetic acid	g/l	-	0.26	0.34	0.31	0.31	0.33
Citric acid	g/l	0.30	0.61	0.43	0.74	0.83	0.64
Succenic acid	g/l	0.16	0.57	0.45	0.38	0.48	0.55
Fumaric acid	mg/l	1.10	3.00	7.20	10.80	6.10	2.40

*Composed by the authors.

Apart from water, the main components of cider are organic acids, sugars, alcohols and polyphenolic compounds. The main organic acid present in apples is malic acid (Merwin, et al., 2008). The amount of malic acid in our apple juice sample was 4.82 mg/l. Depending on the characteristics of the yeast, malic acid decreased in different amounts during fermentation, and in some cases, new formations in the form of other organic acids appeared (Table 3).

Organic acid concentrations were tested in apple juice and all cider samples. The result shows the presence of malic acid in the juice – 4.11 g/l and in all samples in the range of 2.69 g/l – 3.87 g/l. From the results we can conclude that malolactic fermentation has occurred in all the samples. Acetic acid concentration is equivalent within the range of volatile acid values. The citric acid content in the juice is 0.30g/l, but in all cider samples it has increased in the range of 0.43-0.83 g/l. The highest concentration of citric acid was identified in the AC-4 sample – 0.83 g /l, it has been increased 2.7 times compared with the juice. In the Krebs Cycle, citric acid is formed through several derivatives, which are converted into succinic acid. Succinic acid was detected in all studied samples. The highest concentration of succinic acid was identified in S-04 sample – 0.57 g /l. The Krebs Cycle ends with dehydrogenation to fumaric acid. Fumaric acid was detected only in a very little amount in the range of 2.40 – 10.80 mg/l. Shikimic acid is an indispensable intermediate product in the biosynthesis of aromatic compounds (Kazumyan, et al., 2022). Shikimic acid was detected in all the samples in the range of 9.60-20.00 mg/l. The highest concentration of shikimic acid was identified in TF-6 sample – 20 mg/l. The role of the organic acids is important for the freshness in the mouth, balance of cider taste and aroma bouquets.

Conclusion

The results of this research allow us to conclude that the selected four different yeasts are favorable and effective for apple juice fermentation and cider production. The choice of yeasts depends on the initial chemical content of the apple juice and the expected final flavor profile of the cider. According to the results of the research, it is advisable to use S-04 yeast for cider production with a relatively low alcohol and acidity content, and to use AS-2 yeast for cider with medium alcohol content and relatively high acidity. Compared with the control sample, ciders produced with all the other yeasts increased content of total acidity, except for AB-1, which didn't have any impact on the acidity of the cider. Although the indicators of volatile acidity of fermented ciders are within acceptable limits, their concentration in all the other samples was slightly higher compared with the control sample. The content of phenolic substances was slightly inferior to that of the control sample, but the content of aldehydes was predominant in the AB-1 sample and the content of acetals was predominant in the AS-2 sample. In terms of color characteristics, the content of yellow color prevailed in all samples compared to apple juice. The sample with the highest color intensity was AS-2, and the most expressive color shade was observed in the AB-1 sample. All yeasts contributed to the formation of organic acids during their metabolism, but they were manifested in different ways in terms of the amounts of the produced organic acids. It is planned to carry out a sensory evaluation with the samples matured on the above-mentioned cider yeast sediment, with the method of characterization through tasting. Based on the tasting results, aroma wheels will be created for each sample yeast to help characterize the yeast and understand which yeast produces the best sensory performance.

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