Liquor Ciders Made from Freeze-Enriched Apple Musts

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Ice cider is a unique apple wine made by the partial fermentation of high-density apple musts enriched by freezing. The chemical and sensory characteristics of this beverage is influenced by different technological factors: the apple cultivars chosen, the freeze-enrichment procedure used to obtain the apple musts, and the yeast strains selected to develop the alcoholic fermentation.

Keywords: yeast strains ; alcoholic fermentation ; volatile compounds ; ice cider ; cider apples

1. History

Ice cider is a special apple wine obtained by the partial fermentation of apple juices enriched by freezing. Inspired by ice wine, this high-end cider was first developed in the early 1990s in Quebec, which cold winter climate allows to naturally increase the concentration of sugars and flavours within the apple. However, it was not until 1998 that the ice cider trademark was officially acknowledged by the Quebec Alcohol Racing and Gaming Board (RACJ) and the Quebec Alcohol Corporation (SAQ). Thereafter, ice cider gained worldwide recognition as a unique beverage, receiving several awards in international contests [1]. The ice cider became an object of identity distinction which favoured the revival of the entire cider industry, resulting in a significant increase in sales of the other alcoholic Quebec´s apple products, such as still and sparkling cider [2].

The Quebec Official Regulation established that ice cider is the cider obtained by the fermentation of apple juices having a sugar content of not less than 30 ºBrix achieved by natural cold, producing a final product with a residual sugar content of 140 g/L and an alcohol by volume (ABV) content between 9 and 13 %. The sensory profile of the ice cider is characterised by its intense sweetness and acidity, smooth mouthfeel and aromas described as fruity, ripe or fresh apple, candy, exotic fruits, and honey [3][4]. The Protected Geographical Indication (PGI) "Cidre de Glace du Québec" was finally achieved in 2014 [3]. Regions like Vermont did consider the making of ice cider a good opportunity for apple growers to obtain higher value for apples in comparison to that obtained from fresh sweet ciders. Similarly to Southern Quebec, Vermont, among other regions in the world, have the ideal climatic characteristic to reliable produce ice cider using natural freezing, as stated by the Canadian regulation [5]. Leading players in the ice cider market are enterprises from Canada and USA, followed by Sweden and Poland [6]. Nevertheless, other countries like France, Spain or Japan are producing ice ciders by means of artificial freezing [2].

The originality of this cider prompted the Asturian cider-making sector to include the ice cider in its product offering and compete in new market niches. Taking into account the novelty of this type of cider, the challenge was to produce high-quality ice ciders preserving, at the same time, some features of the Asturian regional typicity. Today, this beverage is included and defined in the Spanish Legislation [7] and it is being made by seven producers, although on a marginal scale.

The making of ice ciders involves great difficulties. From a technical point of view, the cider apple varieties chosen for the production of the freeze-enriched musts and the yeast strains used to carry out the fermentation are among the most important factors. These factors have been optimised and systematically evaluated for the production of ice ciders by cryo-extraction through two consecutive harvests [8][9][10][11]. Linked to this method, an Exhaustion process has been also evaluated for the obtention of liquor ciders as an option to valorise the juice fractions discarded because of their lower soluble solids content [12]. These two methods are compared on the basis of the chemical characteristics of their respective enriched juices and final ciders.

2. Enrichment and Characteristics of the Apple Juices

2.1 Freeze-enrichment Methods

There are two methods allowed to obtain the ice apple juices [3][7]:

Cryo-concentration or cold-freeze concentration: it consists of pressing the apples and freezing the resulting juices as a single block of ice. After, this block is thawed and several fractions of decreasing soluble solids contents are successively collected. This is the most commonly used method because it allows the producer to get more control of the initial sugar concentration in the must [1].

Cryo-extraction or cold-press extraction, which consists of pressing the frozen apples. The frozen apples are pressed for several hours at a cellar temperature low enough to allow the obtention of the freeze-enriched juices before the water inside the apple tissue thaws [13]. This method requires a careful selection of the apple varieties or apple blends, and a strict control of the apple maturity in order to reach the desired content of sugars and alcoholic degree in the final ciders, and this is not easy to achieve.

Both methods have in common the obtention of high-quality concentrated apple musts, preserving the compositional profile of the apple, and their low extraction yields [10][11][14]. Pando Bedriñana et al. [10] have reported juice extraction yields of less than 10 % w/v in both systems. Recently, an Exhaustion method has been proposed as an option to valorise the apple juice fractions discarded from the cold-pressing method to create new liquor ciders with valuable and diverse aromatic profiles [12]. The Exhaustion process, which is not legally allowed to produce ice ciders, begins by extending the pressing of the apples used for cryo-extraction while these are still frozen. Apple musts are thus obtained with mean total soluble solid contents between 16 and 23 ºBrix, which are higher than those conventionally achieved by pressing unfrozen apples [15]. These juices are subsequently frozen and enriched as in the cold-freeze concentration system. The final juice yield values obtained by the Exhaustion method ranged between 24 and 37% (w/v) depending on the cider apple blends [12].

The ratio between total sugars and total acidity (TS/TA) is paramount as it is related to the perception of sweet taste and smooth sensation, the two key attributes influencing the overall quality scoring of ice ciders by consumers [4]. More than 70 cider apple cultivars are admitted in the Designation of Origin “Cider of Asturias”, most of them are technologically classified as acidic (ranges for total acidity, 90-174 milliequivalents/L; total phenols between 0.70 and 1.45 g tannic acid/L) [16]. The TS/TA ratio together with the aromatic complexity were taken as criteria for the selection of apple varieties suitable for the obtention of ice juices by cryo-extraction among a set of 10 apple varieties widely used for the making of Asturian ciders [8]. Different apple blends were tried on the basis of the best suited varieties in order to select balanced enriched juices with the greater variability in terms of aromatic profiles. This process led to the selection of the cultivar *Durona de Tresali* for producing single-varietal ice ciders, the cultivars *Perico* and *Limón Montés* to produce bi-varietal ciders, and a more complex blend made up with five varieties (*Verdialona, de la Riega, Raxao, Durona de Tresali* and *Regona*), providing the typical mild-sharp profile of Asturian cider [11]. These juices were evaluated for the making of ice and liquor ciders in two consecutive harvests.

2.2. Characteristics of the Apple Juices

The mean contents of most of the components of the freeze-enriched juices, sugars, acids and total phenols, are three-fold those reported for conventional apple musts [17][18][19]. In **Table 1** are summarised the physical-chemical composition of the enriched juices.

The making system significantly influenced the values of density, total soluble solids and total phenols contents. The apple juices made by cryo-extraction exhibited greater variability in density and total soluble solids contents than those obtained by exhaustion. Some of the musts made by the first method did not reach the minimum value of Brix degree necessary to provide ice ciders in accordance with the requirements of the legislation, which illustrates the greater difficulty of the cryo-extraction process [11]. On the contrary, the exhaustion system allowed the final total soluble solids contents of the enriched juices to be standardised at 34 ºBrix [12].

Table 1. Main chemical characteristics of freeze-enriched apple juices according to the making system, juice type and harvest.

				Cryo-extraction				Exhaustion			
	Sy	**JT**	**H**	**Mean**	**SD**	**Max**	**Min**	**Mean**	**SD**	**Max**	**Min**

Density (g/mL)	*	ns	ns	1.14085	0.0091	1.15031	1.12377	1.14833	0.00042	1.14897	1.14782
Total Soluble Solids (ºBrix)	*	ns	ns	32.87	1.89	34.81	29.32	34.41	0.08	34.54	34.32
pH	ns	**	ns	3.31	0.06	3.40	3.23	3.31	0.09	3.43	3.22
Total Acidity (g sulphuric acid/L)	ns	**	ns	12.41	1.46	14.28	10.98	13.73	1.53	15.59	12.07
Total Phenols (tannic acid/L)	***	ns	ns	3.4	0.2	3.8	3.2	4.2	0.2	4.42	3.79
Sucrose (g/L)	ns	**	ns	68.2	13.2	89.3	53.8	71.1	14.7	89.5	49.9
Glucose (g/L)	ns	*	ns	56.8	8.2	64.7	44.3	57.3	7.3	67.3	46.0
Fructose (g/L)	ns	*	**	172.9	28.8	200.6	122.6	177.0	20.7	201.9	148.2
Total Sugars (g/L)	ns	ns	**	297.9	33.8	331.4	243.4	305.5	19.5	329.3	282.6
Sorbitol (g/L)	ns	**	ns	22.3	3.9	28.4	17.5	23.1	3.3	26.8	18.1
Malic acid (g/L)	ns	ns	**	19.2	2.6	22.4	15.9	20.4	2.6	23.2	17.4

Sy: making system; JT: juice type; H: harvest; Results of the Kruskal Wallis test: (***): significant at $p < 0.01$; (**): significant at $p < 0.05$; (*): significant at $p < 0.10$; ns: not significant.

The content of total phenols was significantly higher in the juices made by exhaustion. The juice type or apple blends and harvest influence pH, total acidity, and the contents of sugars.

3. Fermentation of the Freeze-Enriched Apple Juices

The fermentation of the enriched juices is a technological challenge due to the large hyperosmotic stress that such environment produces in the yeasts, which often leads to altered metabolism -decreased yeast growth and sugar consumption rate, production of high contents of glycerol and acetic acid- and difficulties in reaching the required alcoholic degree [20][21][22]. Although different indigenous yeasts species are present in the apple musts, such as *Metschnikowia pulcherrima* and *Saccharomyces* sp. they may not be sufficient to develop the fermentation of juices thus, a selected yeast strain is routinely inoculated [10][23].

The use of autochthonous yeast strains as starters of fermentation should ensure the capacity to overcome the above cited stress conditions providing ciders with their typical characteristics. *Saccharomyces bayanus* is a cryo-tolerant species widely spread in fermenting cider habitats [15].

The main criteria of yeast strain selection are its ability to ferment sugars in high-density must to a desired alcohol content and its tolerance to ethanol. Bearing this in mind, a series of tests on synthetic media was carried out to select suitable starters among a collection of 74 autochthonous *Saccharomyces bayanus* yeast strains. The fermentation performance of ten pre-selected yeast strains was evaluated in small scale experiments by using a 32 ºBrix-apple juice obtained by cryo-extraction, the final selection based on their capacity to provide ciders with good sensory quality characteristics [9]. Three of those yeast strains were subsequently evaluated for producing ice ciders by cryo-extraction in two consecutive harvests [11].

Once the starter culture has been selected it is important to inoculate it in a proper amount, in a stepwise acclimatisation method to condition the yeast to the high-density medium. It is also advisable to provide yeasts with assimilable nitrogen and micronutrients to ensure their viability and fermentation performance [20]. Following this methodology, the *Saccharomyces bayanus* strains inoculated in the ice apple juices reached maximum rates of ethanol production ranging between 3.46 and 4.75 g ethanol/L/day, achieving in three months a final alcoholic degree greater than 8 (% *v/v*) [11]. The final ciders presented glycerol contents ranging between 6.2 and 10.1 g/L and low values of volatile acidity (0.12 and 0.52 g acetic acid/L), which can be associated with the cryo-tolerant character of this species [24].

Saccharomyces cerevisiae together with *Saccharomyces bayanus* strains are routinely used for producing ice wines and ice ciders although some non-*Saccharomyces* have been proposed in mixed fermentations to improve aroma complexity of ice wines [23][25]. Some strains of *Torulaspora delbrueckii* can tolerate ethanol contents of up to 8% ABV, so they can be used for individual fermentations of ice wine and liquor ciders [[12], [26]].

In this sense, the fermentation performance of a selected *Saccharomyces bayanus* strain referred to as C6 was compared with that of two commercial yeasts (*Saccharomyces cerevisiae* and *Torulaspora delbrueckii*) in juices enriched by exhaustion. All the yeast strains provided ciders with alcoholic degrees greater than 10 % (*v/v*) but at different rates. The ciders made by exhaustion exhibited volatile acidity values between 0.41 and 0.81 g acetic/L, the *Saccharomyces cerevisiae* strain being the main producer of acetic acid [12].

In **Table 2** are summarised and compared the oenological characteristics of all the ciders obtained by fermentation with the *Saccharomyces bayanus*. Among these samples are included ciders made from three types of apple juices in two consecutive harvests [11][12].

The year of harvest together with the type of juice significantly influenced most of the components analysed. The sugar content present in the starting musts conditioned the fermentation activity of the yeast and thus, the final composition of the ciders. The ciders made by exhaustion presented less variability in their residual sugar content than those produced by cryo-extraction. Moreover, the exhaustion system provided ciders with values for total and volatile acidity significantly higher than those observed in the corresponding ciders made by cryo-extraction.

Table 2. Oenological characteristics of ciders made by Cryo-extraction and Exhaustion obtained by fermentation with the *Saccharomyces bayanus* C6.

				Cryo-extraction				**Exhaustion**			
	Sy	**JT**	**H**	**Mean**	**SD**	**Max**	**Min**	**Mean**	**SD**	**Max**	**Min**
Density (g/mL)	ns	ns	***	1.06510	0.01552	1.08273	1.03708	1.07091	0.00590	1.07704	1.06201
pH	ns	***	*	3.59	0.10	3.74	3.47	3.59	0.09	3.73	3.49
Volatile acidity (g acetic acid/L)	***	ns	***	0.34	0.14	0.53	0.13	0.54	0.10	0.67	0.43
Total acidity (g sulphuric/L)	***	***	ns	11.11	1.42	13.11	9.59	12.54	1.17	14.10	11.09

Total phenols (g tannic acid/L)	**	**	ns	3.1	0.5	3.8	2.3	3.6	0.2	4.1	3.3
Alcoholic degre (% v/v)	ns	**	***	9.85	0.91	11.23	8.68	10.02	0.74	11.17	9.17
Total sugars (g/L)	ns	ns	**	126.8	31.1	161.3	71.0	128.4	10.6	143.5	114.1
Glycerol (g/L)	ns	ns	***	8.0	1.0	9.5	6.4	7.9	1.0	9.6	6.5
Sorbitol (g/L)	ns	**	***	21.6	4.1	29.0	16.3	21.8	3.8	26.9	16.7
Malic acid (g/L)	ns	***	**	15.5	2.1	19.0	12.5	16.8	1.9	19.9	14.6

Sy: making system; JT: juice type; H: harvest; Results of the Kruskal Wallis test: (***): significant at $p < 0.01$; (**): significant at $p< 0.05$; (*): significant at $p<0.10$; ns: not significant.

4. Volatile Compounds of Ciders

The analysis of the volatile profiles of the ciders led to the quantification of 38 components. An optimised Stir Bar Sorptive Extraction (SBSE) was used to determine the minor volatile compounds [11][12]. In **Table 3** are summarised the volatile profiles of all the ciders obtained by fermentation with the *Saccharomyces bayanus.*

Alcohols were quantitatively the main group of flavour compounds, amyl alcohols and 2-phenylethanol being major components. The group of esters was qualitatively the most numerous, the ethyl acetate being the main ester present in ciders. Despite the chemical and processing singularities of the ice ciders, the ranges of concentration of all of these components were similar to the values observed in other types of cider [27][28].

Table 3. Volatile profiles of ciders made by Cryo-extraction and Exhaustion obtained by fermentation with the *Saccharomyces bayanus* C6.

				Cryo-extraction				**Exhaustion**			
	Sy	**JT**	**H**	**Mean**	**SD**	**Max**	**min**	**Mean**	**SD**	**Max**	**min**
Major volatiles (mg/L)											
Ethyl acetate	ns	ns	**	49	4	56	43	50	6	60	42
Methanol	ns	**	**	41	10	61	29	44	8	55	30
Propanol	ns	*	***	19	3	22	10	21	4	25	16
Iso-Butanol	ns	ns	***	34	5	44	29	37	8	49	26
1-Butanol	ns	***	*	5	2	8	2	4	1	6	3
Amyl alcohols	ns	ns	***	290	33	370	253	268	44	317	216

2-Phenylethanol	***	**	ns	111	31	177	81	78	34	155	55
Minor Volatiles (μg IS/L)											
Esters (sum of 23 compounds)	***	ns	ns	962.6	254.8	1645.2	654.7	394.3	115.8	562.2	174.3
Alcohols (sum of 2 compounds)	***	ns	ns	1.2	0.1	1.4	1.0	2.4	0.8	3.6	1.2
Volatile phenols (sum of 2 compounds)	**	***	ns	4.2	2.9	9.2	1.1	3.4	1.8	4.6	2.2
Fatty Acids (sum of 4 compounds)	**	ns	***	118.3	24.6	144.4	75.8	71.6	37.7	123.2	27.3

Sy: making system; JT: juice type; H: harvest; Results of the Kruskal Wallis test: (***): significant at $p < 0.01$; (**): significant at $p < 0.05$; (*): significant at $p < 0.10$; ns: not significant.

As seen in **Table 3**, the making system had a significant effect on the contents of 2-phenylethanol, present in higher concentration in the ciders made by cryo-extraction. Among the minor components, the general trend observed was that the contents of all the odorants were lower in the ciders obtained by exhaustion. This feature was remarkable in the case of esters such as ethyl tetradecanoate and isoamyl acetate, whose contents in the ciders made by exhaustion were, respectively, 10 and 4.8 times lower than those observed in the ice ciders [12]. Specific characteristics of the fermentation medium –sugar and lipids contents, dissolved oxygen, precursor concentrations– could explain the differences in the volatile profiles of the two types of ciders [29].

Gas Chromatography-Olfactometry is a powerful technique to characterise the aroma of ciders. The olfactometric profiles of a set of ice ciders made by cryo-extraction presented 58 peaks including many components that have been already found in other types of cider together with 23 new odorants. Moreover, a set of high-boiling point compounds with fruity, floral, sweet or spicy characters was observed; this latter profile differed from that found in still Spanish ciders [4][27].

5. Conclusions and Perspectives

The great sensory originality of ice cider has attracted the attention of makers and consumers around the world since its creation in the late 1990s. It is a unique high-end beverage, whose production process is labour-demanding and very exigent with respect to the selection of apple varieties and their maturation state. In addition, the legal requirements that regulate the production and characteristics of this product open up various technological challenges.

First, the prestige of this cider makes it necessary to develop reliable analytical techniques to control its authenticity. Fourier Transform Infrared together with fluorescence spectroscopies allowed to distinguish sugar and phenolic profiles of ice ciders from those of conventional still and sparkling ciders with a high classification percentage [30]. The contents of minor volatiles have shown their suitability to differentiate among ice ciders obtained by cryo-extraction and liquor ciders made by exhaustion. The application of rapid chromatographic analysis techniques together with appropriate multivariate methods allow discriminating among ciders of different making-processes and origins [12][31].

The olfactometric technique could also be used for authentication purposes. As said, the ice ciders showed different olfactometric profiles compared to still conventional ciders. Linked to this question, Lan et al. [32] reported that some odorants with smoky and sweet characters were perceived with higher intensity in the ice wines from the Beibinghong cultivar in comparison to the corresponding dry wines.

Second, the juice extraction yield of the cryo-extraction process could be improved by applying Pulsed Electric Fields (PEF). As reported elsewhere, the treatment of apples by PEF previous freezing increased the juice yields during pressing, giving apple juices with higher contents in sugars than the untreated apples [13]. Besides, the application of PEF, either in the apples or in their juices, could help prevent the establishment of spoilage microorganisms, thus positively influencing the development of fermentation. Some issues related to the viscosity and the inhomogeneous particle size distribution must be addressed to implement this technique in an industrial-scale process [33].

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