Sveriges lantbruksuniversitet
Swedish University of Agricultural Sciences

Faculty of Natural Resources and Agricultural Sciences
Department of Food Science

Ice cider product development

– Effects of concentration, yeast strains and processing conditions on biochemical and sensory quality traits

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Abstract

Ice cider is produced by fermenting apple juice that has been concentrated by freezing (cryoconcentration or cryoextraction). Ice cider is more a sweet wine than a cider, with an intense apple flavour and sweetness, and acidity to balance the flavours. It originates from Canada, where specifications includes a pre-fermentation sugar content of not less than 30 °Brix, and a finished product with a residual sugar content of not less than 130 g/l, containing 7-13 % alcohol. This project aims to investigate and document some of the aspects of ice cider production process for Swedish conditions. The ambition is to start building experience and knowledge useful for ice cider production in Sweden. The scientific documentation on ice cider fermentation and biochemical properties of cryoconcentrated apple juice is very limited. In this project factors important for the quality of cider and ice wine were reviewed, and methods for producing ice cider through cryoconcentration and subsequent fermentation were evaluated. It was demonstrated in this project that concentrating the apple juice by cold has an equally concentrating effect on sugars, total acids and total phenolics. It was also demonstrated that the level of concentration of the juice highly influences the fermentation kinetics and output. Higher sugar level slowed down the rate of fermentation, and the amount of ethanol produced was lower. In order to produce a good ice cider, the juice needs to be sufficiently concentrated to produce the alcohol content and residual sugar level that defines the product, while not be too concentrated and place excessive hyperosmotic stress upon the yeast cells, resulting in slow and potentially stuck fermentations. The juice needs to hold a minimum of 32 °Brix, while a juice of above 42°Brix will be very difficult to ferment. It was further demonstrated in this project that the selection of yeast strain has an effect on level of ethanol produced, and acid development during fermentation of ice cider. The level of phenolics was found to remain fairly stable across different yeast strains during fermentation. The yeast strain was also demonstrated to have a small impact on the flavour of the ice cider.

Table of contents

List of tables

List of figures

Introduction

Ice cider is produced by fermenting apple juice that has been concentrated by freezing. Ice cider is more a sweet wine than a cider, with an intense apple flavour and sweetness, and acidity to balance the flavours. Ice cider is typically served chilled in a small port or sherry glass, preferably accompanied by cheese, charkuteries or buttery-creamy desserts. Inspired by the ice wine industry, ice cider (or cidre de glace) was developed in Quebec, Canada in the early 1990'ies. The sweet, concentrated juice is produced either by cryoextraction (pressing of frozen apples) or by cryoconcentration (freezing and extraction from apple juice). In Canada the ice cider industry is developing rapidly since its dawn in 1990ies. Today there are over 50 producers in Canada, and in 2011-2012 the net sales was estimated to 12.8 M CAD (approximately 88 M SEK) or 800 000 bottles (www, CARTV, 2015). Ice cider is also exported from Canada to many parts of the world. It has only very recently, in small scale, been made in Sweden with Swedish apples.

Sweden has a long tradition of apple production. In recent years the interest for Swedish apples for consumption has increased, and the industry is planting to meet future demands (Jordbruksverket, 2013) of consumers, year round. The tradition of making fermented beverages from apples in Sweden has in large been limited to large-scale production of beverages from fermented apple juice diluted with water and additions of sugars and artificial flavours. In Sweden a "cider" needs to contain at least 15 % of fruit juice, while in France the denomination cider is strictly for fermented beverages containing 100 % fruit juice, no additions allowed (Riktig Cider, 2015). In the origin of ice cider, Canada, there is equally a strict definition of what is to be called an ice cider.

Ice cider, being a fairly new product, has very little scientific documentation regarding its properties or production practices. Based on fermented apple juice, it will certainly share some similarities with apple cider in its biochemical composition and production demands. On the other hand, the apple juice is extensively concentrated, and this results in a very different environment for the yeast cells during the fermentation. The osmotic pressure on the yeast cells form the sugar level is extremely high and thus the fermentation conditions will be much different from those of cider. Similarly high levels of sugar can be found in ice wine production, where freeze-concentrated grape juice is fermented to a sweet ice wine. Thus, even if the starting material is different, many parallels can be drawn between the making of ice wine and ice cider. However, the composition of apple juice is not identical to grape juice, and the findings on ice wine production does not necessarily hold true for ice cider. The specific nature and requirements for ice cider and its production remains to be described.

In this project aspects of ice cider production were evaluated. Relevant literature was reviewed and production aspects tested and evaluated practically. The practical parts of this project were performed at Centrum för Innovativa Drycker (Innovative Beverages Centre) pilot hall at SLU Balsgård, Kristianstad.

Theoretical background

Ice cider definition

Ice cider is a fermented, alcoholic beverage made from apple juice concentrated by cold. The resulting product is similar to a dessert wine with intensely concentrated apple aromas, sweetness and acidity. In Quebec, the origin of ice cider production, official regulation of what is to be defined as an ice cider has been established. The regulations includes a pre-fermentation sugar content of not less than 30 °Brix of the apple juice, a finished fermented product with a residual sugar content of not less than 130 g/l, containing 7-13 % alcohol. Furthermore, the concentrated juice should be achieved by natural cold only and the producer should also be the grower of the main part of the apples (Quebec government, 2015). The full regulations are given in Table 1. Ice cider production has also spread across the border from Canada to the US, mainly Vermont. The Vermont definition of an ice cider is adopted from the Quebec definition (www, Vermont Ice Cider, 2015).

Table 1. *Quebec ice cider regulations.*

The Quebec official regulations for Ice cider:
"Ice cider": cider obtained by the fermentation of juice of apples that has a pre-fermentation sugar content of not less than 30° Brix achieved solely by natural cold, producing a finished product with a residual sugar content of not less than 130 g per litre and an actual alcoholic strength of more than 7% by volume but not more than 13% by volume;
Flavoured cider, ice cider, strong cider, light cider, liquoreux cider and cocktail cider may be artificially injected with carbon dioxide provided that the volume of dissolved carbon dioxide per volume of finished product is 1.5 to 2.5 or 3.5 to 5.5.
Ice cider produced by a holder must be made from apples grown by the holder and the pressing of the apples and subsequent stages of the production process must take place at the holder's establishment.
Despite the foregoing, a holder of a cider maker permit may subcontract the pressing of his or her apples in Québec for the purposes of producing an ice cider, provided that no juice concentration or freezing is performed during such pressing and that a traceability system be implemented and maintained, in respect of the apples used by the permit holder to make all of the holder's ice cider, between the raw material and the finished product certified by an accredited certification body, approved by the Minister of Agriculture, Fisheries and Food.
In addition, the permit holder may make ice cider by using no more than 50% of Québec apples not grown by the permit holder.
The use of artificial cold in the production of ice cider is permitted only for purposes of malic precipitation, provided the temperature is not lower than -4 °C
Source: Quebec government, 2015

Ice cider products

The concept of ice cider is evolving and spreading, and apart from Canada and the USA examples can be found in Spain, Sweden and Denmark. A list of examples of ice cider producers and their products are given in Table 2. The first producer in Sweden, Brännland Cider, has been active since 2012 and is located in Umeå with apples also sourced from Skåne and Finland (www, Brännland, 2015).

Producer	Region	Apple cultivars	Alcohol	Residual sugars	Total Acids
Antolino Brongo antolinobrongo.com	Quebec, Canada	*Cryomalus 2009:* MacIntosh 48 %, Spartan 16 %, Lobo 16 %, Empire 11 %, Cortland 9 %	9 %	150 g/l	9 g/l
Brännland Iscider, www.brannland.se	Sweden	*Iscider 2014*: Rödluvan, Silva, Slava Petersburg, Säfstaholm, Belle de Boskoop, Cortland, Mutsu, Rubinola	11 %	155 g/l	14,5 g/l
		Iscider 2013: Antonovka, Mutsu, Spartan, Säfstaholm	10 %	190 g/l	19 g/l
Clos Saragnat www.saragnat.com	Quebec, Canada	*L'original 2011* – various heirloom and wild varieties, including cider apple varieties	11 %	No data	No data
Coteau Rougemont coteaurougemont.com	Quebec, Canada	Cidre de glace Reserve: McIntosh, Cortland, Empire, Spartan	10 %	No data	No data
Domaine Pinnacle	Quebec, Canada	Ice Cider - (no data)	12 %	180 g/l	No data
Eden Ice Cider www.edenicecider.com	Vermont, USA	*Heirloom blend 2012*: MacIntosh, Empire, Russet, Calville Blanc, Esopus Spitzenburg, Ashmeads Kernel	10 %	150 g/l	16 g/l
		Honeycrisp 2012: Honeycrisp 100 %	10 %	150 g/l	17 g/l
		Northern Spy 2011: Northerns Spy 100 %	10 %	150 g/l	12 g/l
La Face Cachée de la Pomme www.lafacecachee.com	Quebec, Canada	*Neige Premiere 2012*: MacIntosh 55%, Cortland, Spartan	12 %	160 g/l	8.4 g/l
		Neige Reserve 2012 (oak barrel fermented): Honey Crisp, Russet, Northern Spry	12.5 %	170 g/l	8.9 g/l
		Neige Methode Traditionnelle (sparkling): MacIntosh 55 %, Corland 17 %, Honey Crisp, Lobo, Empire, Honey Gold, Spartan, Gala	11 %	160 g/l	9.5 g/l
Malus Mama, www.malusmama.com	Baskien	(cider apple varieties)	11 %	130 g/l	(no data)

Table 2. *Examples of Ice cider producers and products.*

Apple quality

As is the case for other wines, ice cider quality will be heavily dependent on the starting material. The apple cultivars, their ripeness, the growth place, cultivation practice and post-harvest handling will all have an effect on the finished product. The weather conditions also have its effect, making each years harvest differ from the next. Differences in the acids and phenolic compounds of the apples will highly influence the qualitative traits of the finished product. These aspects are further discussed in the following sections.

Apple organic acids

Malic acid constitutes around 90% of the acid content of apples, the rest being citric acid, succinic acid, and traces of several other acids. The main acids in apples have been observed to decrease during ripening and storage (Ackerman et al., 1992). Acids are of major relevance for flavour perception. The dominant flavour of organic acids is

sourness, but also contribute to bitterness and astringency in different ways depending on the type of acid. There are also other flavour characteristics of individual acids - citric acid has a fresh acidity in a different way compared to malic acid, while succinic acid has a salty, bitter component, and acetic acid has another characteristic flavour (Whiting, 1976). The astringency and sourness of lactic, acetic and citric acid has been demonstrated to decrease with increasing pH-level, and acids were found to be differentially sour at equal pH (Lawless et al., 1996). In addition to their importance for flavour, some acids are also important in binding SO2 and reducing the amount of free SO2 in the medium. Acids are also metabolised by the yeast in different ways; succinic acid is not metabolised easily while malic acid and citric acid are readily metabolised by the yeast, resulting in flavour changes (Whiting, 1976).
In a review Whiting (1976) describes the fate of organic acids during fermentation: malic acid can either be formed or broken down during fermentation, depending on yeast strain and fermentation conditions. Succinic acid is formed throughout the fermentation, but particularly during the early stages. Citric acid can be formed early during fermentation, and then taken in to the cells and catabolized later during fermentation process. Volatile acids are formed during fermentation; the amounts are variable depending on yeast strain, medium and physical factors during the fermentation. The main volatile acid, acetic acid, is formed most rapidly in the beginning of fermentation and its formation depends on yeast strain, medium and fermentation conditions. Reduced levels of biotin, pantothenic acid or thiamine in the medium increases acetate formation (Whiting, 1976). Zhang et al. (2008) studied the evolution of organic acid content in apple cider during cider fermentation, and demonstrated that pyruvic, lactic, succinic, and acetic acid are produced during fermentation (Zhang et al., 2008). The yeast cells strive to maintain a fairly constant intracellular pH-level; excretion of acids in to the medium surrounding them is an effect of the yeasts control system, and is depending on concentrations of various enzymes and co-factors. Selection of yeast strain and controlling the pH-level, nitrogen level, thiamine level and SO2 addition are tools that can be used control the acid metabolism and to achieve the desired product (Whiting, 1976).

Apple phenolics

Phenolic compounds are of high importance for quality, flavour, colour in wine and cider. The mouthfeel of a fruit beverage is largely due to polyphenols (in combination with acids and fibres), and particularly to procyanidins, and procyanidins are also associated to bitterness and asringency (Lea, 1992). The colour of apple juice is due to phenolic oxidation, of procyanidins and phenolic acids, and haze in apple juice is often due to procyanidin polymerization (Lea, 1992). Apple phenolics include chlorogenic acid, p-coumaroyl acid, dihydrochalcones, epicatechin and procyanidin (Burda et. al, 1990, Vamos-Vigyazo et al., 1976, Lea & Timberlake, 1974). The levels of phenolics vary greatly in different apple cultivars cultivars (Francini & Sebastiani, 2013, Matthes & Schmitz-Eiberger, 2012) and are located in different parts of the apple (Francini & Sebastiani, 2013). For dessert apples the total polyphenolic content have in one study been observed to range from 154 to 178 mg/L, while cider apple cultivars ranged from 261 to 970 mg/L (Kahle et al., 2005). Another study indicated polyphenolic content of Asturian ciders to range between 446-1180 mg/gallic acid equivalents/L (Madrera et al., 2006). The concentration of phenolic compounds has been observed to decrease sharply during early development of the fruit and to then remain relatively constant during the maturation (Burda et al., 1990). Polyphenols have been observed to remain fairly stable during cold storage and storage in controlled atmosphere, while storage in room temperature leads to lower concentration of polyphenols (Burda et al., 1990, Matthes & Schmitz-Eiberger, 2012) The level of phenolics extracted into the apple juice during pressing is also depending on processing conditions (Lea & Timberlake, 1978).

Apples in ice cider

Most Canadian commercial ice cider manufacturers mainly use apple cultivars grown for direct consumption, cooking or juice, such as MacIntosh, Spartan, Lobo, Honeycrisp, Empire and others (see Table 2). Most commonly several different apple varieties are blended to achieve a more complex flavour, however some single-variety ice ciders also occur. On the contrary from regular cider, bitter cider apple varieties with higher content of polyphenols are not used to large extent for mainstream ice cider. However, some producers, including the ice cider inventor Christian Barthomeuf, advocate bitter cider variants, wild apples or heirloom varieties as producing a more interesting ice cider (www, The Globe and Mail, 2012). The larger scale way for ice cider production is to after harvest keep the apples in cold storage until the outdoor temperature is sufficiently low, and the apples are then pressed and the juice processed. Some producers employ a cryoextraction method similar to ice wine production, leaving the apples to hang on the branch until frozen. This allows the apples to ripe longer and exposes the apples for the sun and wind and several freeze-thaw cycles (www, The Globe and Mail, 2012). This method is more labour intensive and only very few varieties of apples stay on the branch long enough. This mode is similar to that of ice wine production - ice wine grapes freeze naturally on the wine and go through several freeze-thaw cycles before harvesting that affects the chemical profile of the ice wine juice, due to e.g. breakdown of tissues, oxidation processes, microbial processes (Inglis et al., 2006:2).

Apple terroir

Cultivation practices and growth place of the apples are affecting the fruit and juice quality. Just as for grapes in wine production, the terroir of the apples has an effect on the juice produced. Apple juice from the same apple cultivar has been demonstrated to have different content of sugar, acid, phenolic compounds and minerals depending on the growth place of the apple (Rumpunen et al., 2015). Furthermore, sensory evaluation shows that these differences give a perceivable difference in flavour of the juice. A higher level of sugars and phenolic compounds in the juice resulted in a higher appreciation score from consumers. Synthesis of phenolic compounds is higher when cider apple trees are grown under reduced nutrient supply, resulting in a final product with more bitterness and astringency (Lea, 1978). The concentration of polyphenols is also observed to be higher with a decreased crop load for the tree (Stopar et al., 2002).

Cryoconcentration techniques

Cryoconcentration is a technique utilizing the natural phenomenon that a more concentrated phase is separated from the initial solution during thawing. Just as water in the form of steam is removed from a solution through heating during evaporation, water in frozen form is removed from the solution during the cryoextraction. Cryoconcentration, or cryoseparation, techniques are commonly used in the food and nutraceutical industries. Nutritional and sensory qualities of freeze-concentrated fruit juice are higher than juice concentrated of evaporation due to the low processing temperatures, which avoid undesirable chemical and biochemical changes and minimize the loss of sensory properties in the juice (Sanchez et al., 2009).

There are a number of techniques available, such as suspension crystallization, progressive cryoconcentration, eutectic cryoconcentration, partial block- and complete block cryoconcentration (Aider & De Halleux, 2009). While some techniques are complicated and require advanced equipment, the cryoconcentration technique used for ice cider production is a simple type of complete block cryoconcentration; a container (of variable size) of juice is frozen solid and the concentrate extracted by gravity during the thawing. This technique does not require advanced equipment and

can be done with natural, outside temperatures, thus making it an environmentally friendly technique.

Aider & Halleux (2008a) found in a study of maple sap complete block cryoconcentration that the freezing temperature does not influence the process efficiency or product quality. Furthermore, they observed that three subsequent steps of cryoconcentration were beneficial for the process efficiency while not reducing the product qualities (Aider & Halleux, 2008a). Similar results have been observed for cherry and apricot juice (Aider & Halleux, 2008b). Bayindirli et al. (1993) found in a study of apple juice concentration that lowering the freezing temperature during the freezing stage shorten the time to achieve a certain concentration level, but does not alter the concentration level in itself. They also found that a multi-stage cryoconcentration process can achieve higher concentration compared to single-stage process (Bayindirli et al., 1993).
The freezing point of apple juice at different concentrations was determined by Auleda et al. (2011), presented in Table 3. A juice with 40 °Brix will freeze at -6.7 °C, thus at least this temperature would need to be reached in order to concentrate it to this extent.

Table 3. *Freezing points of concentrated apple juice (adapted from Auleda et al., 2011)*

Concentration (°Brix)	**Apple juice freezing point (°C)**
10	-1.07
15	-1.74
20	-2.50
25	-3.20
30	-4.10
35	-5.20
40	-6.70

Cryoconcentration has been observed to be very efficient in maintaining aroma qualities in orange juice (Braddock & Marcy, 1985), pineapple juice (Braddock & Marcy, 1985) and black currant juice (Dette & Jansen, 2009). Braddock & Marcy (1985) studied cryoconcentration of pineapple juice and found no significant differences between fresh juice and cryoconcentrated samples for % acid, vitamin C, browning index or colour. The aroma numbers and ascorbic acid content of the cherry and apricot juices has been observed to be very high, compared to conventional concentration methods such as evaporation. (Aider & Halleux, 2008b).
Cryoextraction method is based on the same principle as cryoconcentration, with the difference that the whole fruit is frozen, and also pressed in a frozen state. This process shares similarities with that for producing ice wine, where frozen grapes are pressed to achieve the concentrated grape juice. This requires powerful equipment that can press the whole frozen fruit. There is very limited data on the biochemical properties of cryoconcentrated apple juice. To our knowledge there are no studies investigating the level of concentration effect on acids or the on phenolic compounds

in apple juice, nor the subsequent fermentation of juice cryoconcentrated to different extent.

A small scale cryoextraction method for ice cider production

For a small-scale ice cider producer, a simple complete block cryoextraction process can be applied as described by Leger (2010). In summary, the process is described in the following section.

The production starts when outside temperatures are projected to be sufficiently low. The apples that have been stored since harvest are washed, sorted and pressed. The juice is filled in containers. Depending on the production scale different containers can be used; typically intermediate bulk container (IBC) can be useful. Optionally, preservatives, enzymatic treatment and filtering can be done at this stage. The containers are placed outside in freezing temperatures until frozen solid. The time for this is variable depending on temperature and container volume. For a large IBC of 1000 L in -7 C it will take around 4 weeks. The containers are then moved inside for defrosting. The melting juice is drained off from the bottom of the tank and collected, and the °Brix-level is continuously measured. The °Brix-level of the collected juice should be above 32°, which will be 20-25 % of the original juice volume. The juice is then transferred to fermentation tanks for subsequent fermentation process.

Fermenting high sugar juice

The scientific documentation on ice cider fermentation is very limited. To our knowledge there is only one scientific study published on ice cider fermentation. A study from Korea by Choi et al. (2012) investigates the use of a sugar-tolerant *Saccharomyces cerevisiae* strain versus an industrial wine *S. cerevisiae* strain. The study shows that the more sugar-tolerant strain performed better and resulted in a more rapid increase of alcohol content. The organic acid distribution after fermentation was similar between the strains and the results from sensory evaluation was also similar. However, the distribution of alcohol species was somewhat different and the sugar-tolerant yeast strain scored higher colour intensities (Choi et al., 2012). Apart from this study, no scientific documentation on ice cider fermentation has been found. In lack of this, learning from other closely related field of ice wine fermentation can be made. This field is also not extensively documented, but more so than for ice cider.

Ice wine is made from juice of grapes that have frozen on the wine, harvested below 8 °C, and pressed while frozen. The resulting juice should hold a minimum of 35 °Brix. In a study of the composition of 348 ice wines, of different origins, years, grape varieties and style, it was established that ice wine juice typically has a pre-ferment total soluble solids of 39 °Brix, resulting in a wine of typically 10.6 % ethanol, a residual sugar level of 207g/L, and volatile acid level of 1.18 g/L (Soleas & Pickering, 2007). The high sugar level and the developing ethanol level poses significant stress on the yeast, and typically fermentation ends while there is still a high level of residual sugar left (Nurgel et al., 2004). Ice wine fermentations are often slow and are prone to becoming stuck (Inglis et al., 2006a). Inglis et al. (2006a) found that increasing the ice wine-juice concentration decreases yeast growth, sugar consumption rate, total amount of sugar consumed and the amount of ethanol produced. Furthermore, it increased the proportion of sugar metabolism diverted to production of glycerol and acetic acid, in order to cope with the osmotic stress placed by the high-sugar solution (Inglis et al., 2006a). The high sugar concentration induces an osmotic stress on the yeast cells, that up-regulates genes involved in the synthesis of acetic acid and glycerol (Erasmus et al., 2004). Aspects of ice wine fermentations were studied by Kontkanen at al. (2004). It was found that ice wine fermentations benefit from using a higher yeast inoculum level than that of standard wines (0.2 g/L) - inoculating 0.5 g/L was observed to

increase the yeast biomass accumulation, sugar consumption and the amount of ethanol produced. Also, the high sugar fermentation was observed to benefit from using a stepwise acclimatizing procedure when inoculating the yeast rather than a direct inoculation. Allowing the yeast cells to acclimatize to a high osmotic pressure in this way increased the sugar consumption and the amount of ethanol produced. Furthermore, it was observed that adding a yeast nutrient during yeast rehydration increased the biomass accumulation, increased the viable cell concentration, reduced the fermentation time, and reduced the rate of acetic acid production, however it also reduced the amount of ethanol produced as a function of sugar concentration (Kontkanen et al., 2004)

Selecting yeast strains

Selecting the appropriate yeast strain has effect both on the fermentation efficiency and the qualities of the finished product. In a study by Erasmus et al. (2004), the type of yeast strain used for Riesling ice wine fermentations was demonstrated to have effect on the fermentation rate, acetic acid production and glycerol formation. Subsequent sensory analysis showed significant differences in overall quality, sulphur-like aroma and colour with different yeast strains. Furthermore, the formation of acetic acid and glycerol due to osmotic stress by highly concentrated juice was observed to be yeast-strain dependent (Erasmus et al., 2004)

Non-saccharomyces strains

Early stages of spontaneous, alcoholic fermentation are dominated by growth of non-*Saccharomyces* yeasts. As higher concentrations of ethanol are produced, the fermentation is taken over by the most ethanol-tolerant yeast species *Saccharomyces* (Bely et al., 2008). Even though the non-*Saccharomyces* species are mainly active during the initial stages of fermentation, they still have an influence the wine quality. One of the non-*Saccharomyces* yeasts that have been studied is *Torulaspora delbrueckii*, which can have a positive effect on the taste and aroma of alcoholic beverages (Ciani & Maccarelli, 1998) while displaying a high fermentation purity (defined as the ratio between volatile acidity and ethanol produced) (Ciani & Maccarelli, 1998, Comitini et al., 2011). Alcoholic fermentation with *T. delbrueckii* is specifically characterized by production of succinic acid (Ciani & Maccarelli, 1998). The capability of *T. delbrueckii* to handle high osmotic pressure makes it of special interest to high sugar fermentations. Bely et al. (2008) studied the impact of using *T. delbrueckii* culture alone or in combination with a strain of *Saccharomyces cerevisiae* on high-sugar grape must fermentation. It was demonstrated that a pure *T. delbrueckii* culture did not respond to high osmotic pressure by high acetic acid production, but fermentations were sluggish and produced low levels of ethanol and low biomass yields. A mixed culture of both *T. delbrueckii* and *S. cerevisiae* was observed to reduce the amount volatile acidity remarkably compared to pure *S. cerevisiae* culture, while still reaching the required alcohol level (14 % in this case). In this study it was further observed that a mixed culture from the start of fermentation had a better effect on reducing the volatile acidity compared to using a step-wise inoculation, adding the S. cerevisiae culture 5 days after the *T. delbrueckii*. This is in contrast to the recommendations form the manufacturer, where *T. delbrueckii* is recommended to use first, followed by inoculation of *S. cerevisiae* (Lallemand, 2015)

Cider flavour development

The flavour compounds in ciders was reviewed by Williams (1974). The main bearers of flavour in apple cider are sugars, acids and phenolics. The various sugar species basically contribute to sweetness, the different acids mainly to sourness and the phenolics to bitterness and astringency. Furthermore, various volatile compounds (such as alcohols, esters, acids, carbonyls and acetyls) interact with the olfactory

system and are also highly important for the flavour sensation The flavour-bearing compounds in ciders are derived from the fruit as well as being formed during fermentation. During the fermentation the yeast's biosynthetic pathways produce many flavour compounds, as end products or intermediates, when the yeast strive to produce energy for its own growth. The fermentation conditions, such as temperature, aeration, adding yeast nutrients, has an effect on the aroma compounds produced, but the single most important factor is the yeast strain used for fermentation (Williams, 1974).

Ice wine flavours

Sugar and acids suppress each other in flavour perception (Noordeloos & Nagel, 1972, Martin et al., 2002), and it is the balance between the two that is important for consumer appeal. For ice wine the high residual sugar level needs to be balanced by the acidity level, and the residual sugar:acid ratio is an important indicator for taste sensation. The sugar:acid ratios of some examples of ice ciders are give in Table 4. Physical viscosity is also an important aspect of the overall perception, and the physical viscosity of ice wine can typically be twice that of regular table wine (Nurgel, 2004). The residual sugar content influences the perception of viscosity the most. Ethanol has a moderate effect and glycerol content only a nominal effect (Nurgel & Pickering, 2005). Acetic acid is the main volatile acid in wine and can give a vinegar-like aroma even in relatively low concentrations (Nurgel et al., 2004), and is thus an important factor influencing wine quality. Acetic acid is especially important to ice wines as larger amounts can be produced during fermentation, as previously discussed. Elevated levels of acetic acid are considered a wine fault and the content limited by law in many countries. In Canada the maximum allowed level of volatile acidity, expressed as acetic acid, in ice wine is 2.1 g/L (www, Foreign Affairs, Trade and Development of Canada, 2015). Actual levels of acetic acid in Canadian ice wines have been observed to range from 0.49 to 2.29 g/L (Nurgel et al., 2004).

Table 4. *Sugar:TA ratio of example ice cider products.*

Product	Residual sugar g/L	Total Acidity g/L	Sugar:TA ratio
Antolino Brongo Cryomalus 2009	150	9	**16.7**
Brannland Iscider 2014	155	14.5	**10.7**
Brannland Iscider 2013	190	19	**10.0**
Eden Heirloom Blend 2012	150	16	**9.4**
Eden Honeycrisp 2012	150	17	**8.8**
Eden Northern Spy 2011	150	12	**12.5**
La Face Cachee Neige Premiere 2012	160	8.4	**19.0**
La Face Cachee Neige Reserve 2012	170	8.9	**19.1**
La Face Cachee Neige Methode Traditionelle 2012	160	9.5	**16.8**
		Average	**13.7**

Sensory analysis

There are a number of sensory analysis methods available. The methods can generally be grouped as analytical methods (such as difference tests and descriptive tests) and affective methods (consumer tests) (Gustafsson, 2014). As described by Gustafsson et al. (2014), ranking test is a type of analytical test and uses a method where more than two samples are ranked according to a specific attribute, such as sweet, sweeter, sweetest. By using this method, information of the internal ranking of the samples for this particular attribute is gained. In a ranking test different attributes describing the

appearance, smell, flavour or texture of the sample can be tested, but only one attribute at a time is tested. The participants should have a common understanding of the meaning of the attribute, and samples should be served in different orders to each participant (Gustafsson et al., 2014).

Aim

This project aims to investigate and document some of the aspects of ice cider production process for Swedish conditions. The ambition is to start building experience and knowledge useful for ice cider production in Sweden. There are many aspects important to the production ice cider - apple varieties, juice extraction method, fermentation conditions, yeast strains used for the fermentation, storage - to name a few. In this short-term project, focus lies on understanding some of the biochemical aspects of the cryoconcentration process and subsequent fermentation, and the effect of yeast strain on fermentation progress and flavour development.

Specifically, the aim of this project is:

1. Investigate the effect of cryoconcentration on the biochemical composition of the juice, and establish:
    a. Cryoconcentration effect on total phenolic content in the juice
    b. Cryoconcentration effect on total acids content in the juice

2. Evaluate different levels of juice concentration effect on fermentation
    a. Concentration effect on fermentation rate
    b. Concentration effect on level of ethanol produced
    c. Concentration effect on total phenolic content in the juice after fermentation
    d. Concentration effect on total acidity, organic acids and acetic acid in the juice during fermentation

3. Evaluate different yeast strains for fermentation, and establish:
    a. Yeast strain effect on fermentation rate
    b. Yeast strain effect on level of ethanol produced
    c. Yeast strain effect on total phenolic content in the juice after fermentation
    d. Yeast strain effect on total acidity, organic acids and acetic acid in the juice during fermentation
    e. Yeast strain effect of the flavour of the ice cider

Materials and methods

Plant material

Four cultivars of dessert apple were obtained February 2015 from a local grower (Gedenryds Frukt, Österslöv, Kristianstad), where the fruit had been kept in cold storage since harvest 2014. The cultivars were 'Aroma', 'Ingrid Marie', 'Cox Orange' and 'Mutsu' (Table 5).

Table 5. *Apple cultivars used for juice production.*

Apple variety	**Approx. amount (kg)**
Mutsu	30
Cox Orange	30
Ingrid Marie	30
Aroma	60

Juice extraction and concentration

In order to produce the concentrated juice a combination of cryoextraction and cryoconcentration method was used in a stepwise manner. A flow-chart of the procedure is outlined in *Figure 1*. The apples were frozen solid in -20 °C freezer, and then placed in 6 °C until the inner temperature of the apples reached around -4 °C. The semi-defrosted apples were pressed in a hydraulic bucket water press. Total soluble solids (TSS) measured as °Brix was continuously monitored using a portable digital refractometer (Atago). After the initial cryoextraction process the resulting juice had a concentration of 25 °Brix. Thus, a second step of cryoconcentration was done by freezing the juice in -20 °C in plastic bags (bag-in-box bags), and extracting concentrated juice. For the concentration study the juice was collected in fractions of 39, 34, 29 and 24 °Brix, and for the yeast study until the juice had 32 °Brix. The pectolytic enzyme Panzym Univers (Begerow) was added at 0.04 mL/L and Panzym Pro SP-L (Begerow) at 0.06 mL/L. The yeast nutrient VitamonCombi (Erbslöh) was added to the juice at 0.5 g/L.

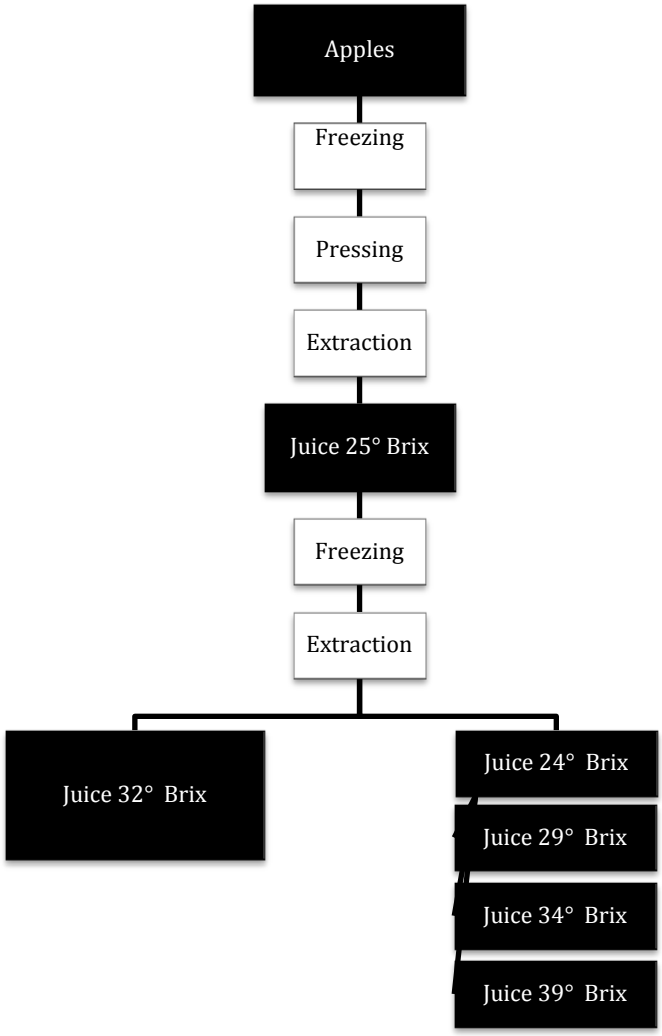


Figure 1. Flow chart of cryoextraction/cryoconcentration process.

Yeast strains

Seven *Saccharomyces cerevisiae* yeast strains and one *Torulaspora delbrueckii* were used for the ice cider fermentations (Table 6). The commercially available yeast strains were selected for being tolerant to high °Brix juice, tolerate higher concentration of ethanol and colder fermentation temperatures (based on statement from manufacturer). The commercial yeast nutrient Go-Ferm Protect Evolution (Lallemand) was used for all yeast strains.

Table 6. *Selected yeast strains used for study.*

Commercial name	Manufacturer	Strain
Biodiva TD291	Lallemand	*Torulaspora delbrueckii*
SIHA A7	Begerow	*Saccharomyces cerevisiae cerevisiae*
Lalvin DV10	Lallemand	*Saccharomyces cerevisiae bayanus*
Lalvin ICV Okay	Lallemand	*Saccharomyces cerevisiae cerevisiae*
Lalvin Rhone 2226	Lallemand	*Saccharomyces cerevisiae cerevisiae*
Lalvin R2	Lallemand	*Saccharomyces cerevisiae bayanus*
Lalvin QA23	Lallemand	*Saccharomyces cerevisiae bayanus*
Uvaferm Exence	Lallemand	*Saccharomyces cerevisiae cerevisiae*

Study design – yeast strains

A total of 7 different fermentations were studied. 3 *Saccharomyces cerevisiae* strains were used alone, and 4 *Saccharomyces cerevisiae* strains were used in combinations with a *Torulaspora delbrueckii* strain. The combinations are outlined in Table 7.

Table 7. *Yeast strain combinations used for study.*

Trial no.	Yeast strain 1	Yeast strain 2
1	Biodiva TD291	Lalvin DV10
2	Biodiva TD291	Lalvin ICV Okay
3	Biodiva TD291	Lalvin QA23
4	Biodiva TD291	Uvaferm Exence
5	Lalvin R2	-
6	Lalvin Rhone 2226	-
7	SIHA A7	-

Study design – juice concentration

For all four fractions of variable concentration the commercial *S. cerevisiae* yeast-strain Lalvin R2 (Lallemand) was used for fermentation.

Table 8. *Juice concentration fraction study design.*

Trial no.	Fraction	TSS	Yeast strain
1	F1	39 °Brix	Lalvin R2
2	F2	34 °Brix	Lalvin R2
3	F3	29 °Brix	Lalvin R2
4	F4	24 °Brix	Lalvin R2

Fermentation

The inoculation procedure (including rehydration and acclimatization) is outlined in detail below. The yeasts were rehydrated in the presence of the yeast nutrient Go-Ferm Protect Evolution (Lallemand). After rehydration, the yeasts were acclimatized to the high °Brix-juice by a stepwise procedure to reduce the hyperosmotic stress response of the yeasts. The yeasts were inoculated at 0.35 g/L. All the yeasts and juice concentrations were fermented in duplicate. The fermentations were kept at 20 °C for the initial days, and then kept in 7 °C for the length of the fermentation. The yeast study fermentations were carried out in 750 mL glass bottles fitted with airlocks, while the juice concentration study fermentations used 300 mL bottles fitted with airlocks. The bottle weight was measured at irregular intervals during the fermentation to give an indication of the speed of the fermentation. The fermentation continued until the speed slowed down, for a total of 45 days. The fermentations were transferred to new bottles and samples for analysis were taken. The bottles were then placed again in 7 °C for continued fermentation.

Yeast inoculation

The yeast rehydration procedure was according to the instruction from the manufacturer (Lallemand) and acclimatized to the high °Brix-juice similar to the method described by Kontkanen et.al, 2004:

S. cerevisiae yeasts*:*

1. 0.6 g Go-Ferm Protect Evolution was dissolved in 5 g clean water (43 °C, kept in water bath), and when the solution was cooled to 40 °C (in water bath) 0.5 g yeast was rehydrated in the solution. The solution was placed in 37 °C heat chamber.
2. When the yeast was clearly active and foaming (30-45 min), equal amount of ice cider juice diluted to 50 % and adjusted to 37 °C was added and gently stirred.
3. After 1 hour, equal amount of the 50 % ice cider juice was added.
4. After 1 hour, equal amount of the 50 % ice cider juice was added and the solution kept at 20 °C.
5. After 1 hour, equal amount of ice cider juice (20 °C) was added.
6. After 1 hour, the solution was divided in two equal parts and added to the two 0.75 L fermentation bottles and ice cider juice added to the fill the bottles.

T. delbrueckii:

1. 2.6 g Go-Ferm Protect Evolution was dissolved in 5 g clean water (43 °C, kept in water bath) and when the solution cooled to 40 °C (in water bath), 2.1 g yeast was rehydrated in the solution. The solution was placed in 30 °C heat chamber.
2. When the yeast was clearly active and foaming (30-45 min), the equal amount of ice cider juice diluted to 50 % and adjusted to 30 °C was added and gently stirred.
3. After 1 hour, equal amount of the 50 % ice cider juice was added.
4. After 1 hour, equal amount of the 50 % ice cider juice was added and the solution kept at 20 °C.
5. After 1 hour, equal amount of ice cider juice (20 °C) was added.
6. After 1 hour, the solution was added to a larger vessel and ice cider juice was added to a total of 6 L.
7. The day after the inoculation of *T. delbrueckii*, the secondary cultures of *S. cerevisiae* were rehydrated and acclimatized according to the procedure described previously.
8. The *S. cerevisiae* cultures were added to the fermentation 0.75 L bottles, and the *T. delbrueckii*-inoculated juice was added to fill the bottles.

Concentration study

For the concentration study the yeast Lalvin R2 was used. The inoculation procedure was the same as for *S. cerevisiae* described previously, except after rehydration the yeast solution was divided into 4 equal parts and the ice cider juice of variable concentration was added in the stepwise manner described previously. An additional step of acclimatization to the highest concentrated fraction was added. The solution was distributed in two bottles per fraction in a final volume of 0.2 L.

Analytical assays

Ethanol, residual sugar, acetic acid, organic acids

For analysis of ethanol and sugar, 30 µL of each sample was directly injected into the HPLC apparatus (LC-10AD Shimadzu), which was equipped with an autosampler, SCL 10A System Controller, and a RID-10A Refractive Index Detector. Separation was performed at 40 ° C on a Column Rezex-Organic Acid H+ 300x7.80 mm (Phenomenex) The mobile phase was water, at a flow rate of 0.8 mL/min, and run time 20 min Quantification was performed in comparison with calibration curves and the area ethanol and sugar peaks were recorded and integrated using the data system Class VP. For the analysis of organic acids, the procedure was the same except the Detector SPD-10AV UV-Vis detector and the Column Allure Organic Acids 5 um 250 x 4.6 mm particle size 5 um (Restek) were used, the injection volume was 5 µL, the mobile phase was buffer pH2.8.

Total acid automatic titration

Total acidity was analyzed by automatic titration (Radiometer Copenhagen). 5 mL of the pre-fermentation juice and post-fermentation ciders samples were diluted in 15 mL distilled water. The samples were analyzed in duplicates. Titration was made with 0.1 M NaOH until pH 8.4, and the acidity calculated as concentration of malic acid.

Total phenols

Spectrophotometric analysis using the Folin-Ciocalteau's method was used to determine total phenolic content of the pre-fermentation juice and the post-fermentation ciders. The samples were analyzed in duplicates. Phenolic extraction was made of 1 mL sample and 9 mL 90 % methanol, placed on a vibrating plate in 7 °C for 15 hours. The samples were centrifuged at 4500 rpm for 4 minutes. 5 µL of the extract, 95 µL of 5 % ethanol, 200 µL Folin-Ciocalteau's reagent, 2 mL 15 % Na_2CO_3 and 1 mL distilled water was added to cuvettes. The mixture was kept for 1 hour in room temperature, and the absorbance at 765 nm measured in a UV-VIS scanning spectrophotometer (UV-1650 PC Shimadzu). The absorbance was compared to a standard curve of gallic acid, and the content in the sample was calculated as mg gallic acid equivalents (GAE)/L.

Statistics

The average of sample duplicates in the study and correlations were calculated in Excel. The sample standard deviation was also calculated in Excel and added as error bars in charts.

Sensory evaluation

The sensory analysis was performed Wednesday 27th of May 2015, at Kristianstad Hogskola sensory laboratory, with 23 participants. One participant's answers were excluded due to a technical error in the equipment. Each sample was assigned a three-letter code. The code was randomly computer generated. The participants evaluated the 7 samples in respect to the attributes sweetness, acidity, bitterness, flower flavours, alcohol flavours, overall smell intensity and overall liking. For the overall smell intensity and the overall liking they were also encouraged to leave comments for each of the samples.

Statistics

The significance of the differences among the samples in the sensory analysis was calculated using Friedmans formula. Level of significance (Friedman) was 5 %.

Results

Cryoconcentration process

The analysis of the juice processed to fractions with different concentrations shows that acids and phenolic compounds, expressed as mg Gallic Acid Equivalents (GAE)/mL, were concentrated to the same extent as the sugar in the cryoconcentration process (

Figure 2. and

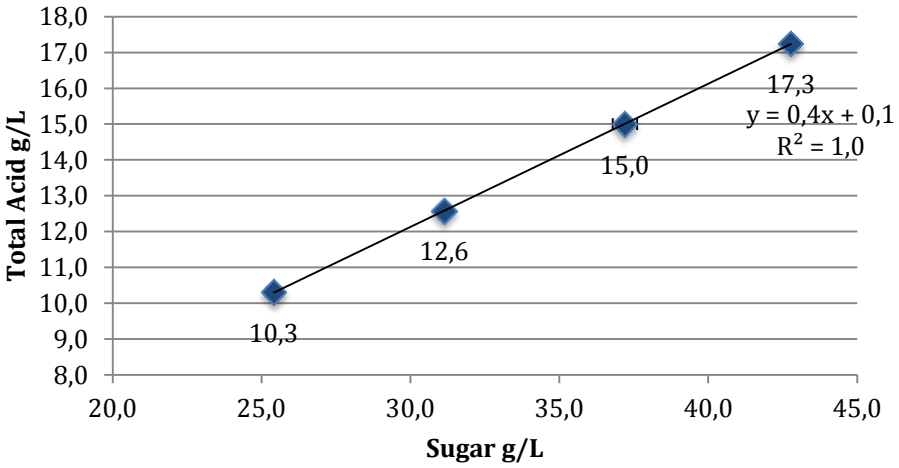


Figure 4.). HPLC-analysis of sugar content (glucose, fructose and saccharos combined) in the four fractions indicated a slightly higher level of sugar than that of the initial °Brix-measurement across all four fractions. There was a direct linear relationship found between the sugar concentration and the acidity of the samples (

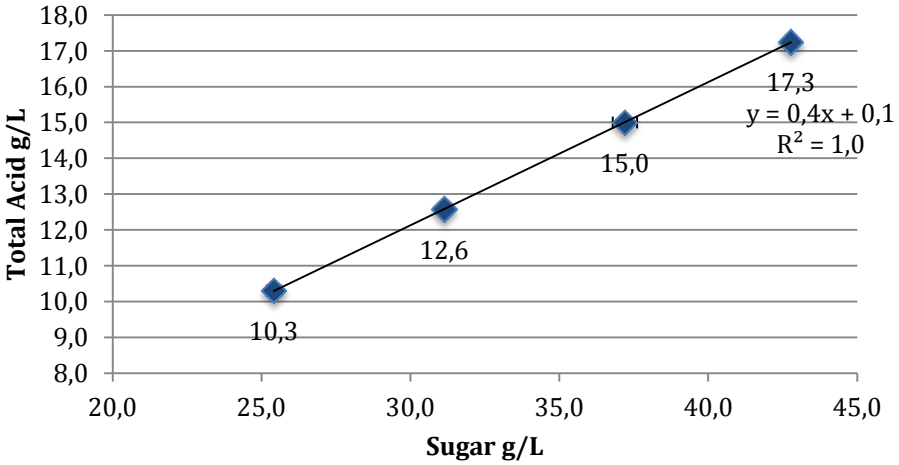


Figure 4). There was also a direct linear relationship between the sugar content (analyzed by HPLC) and total phenolic content (

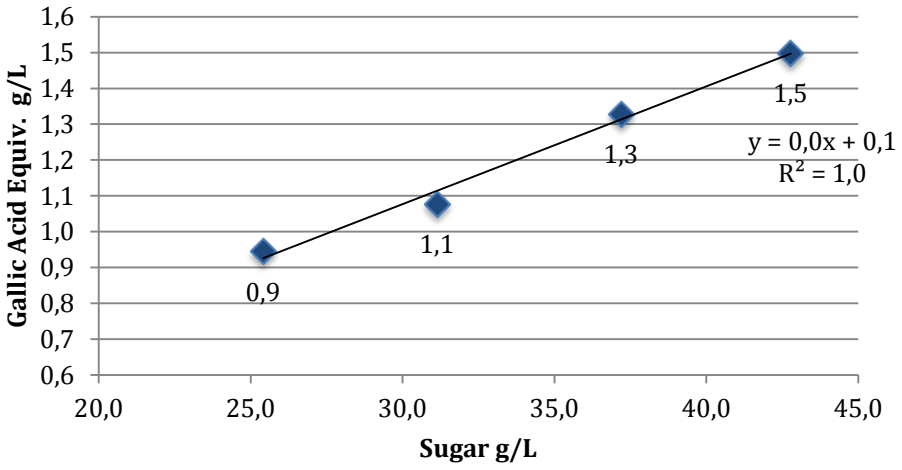
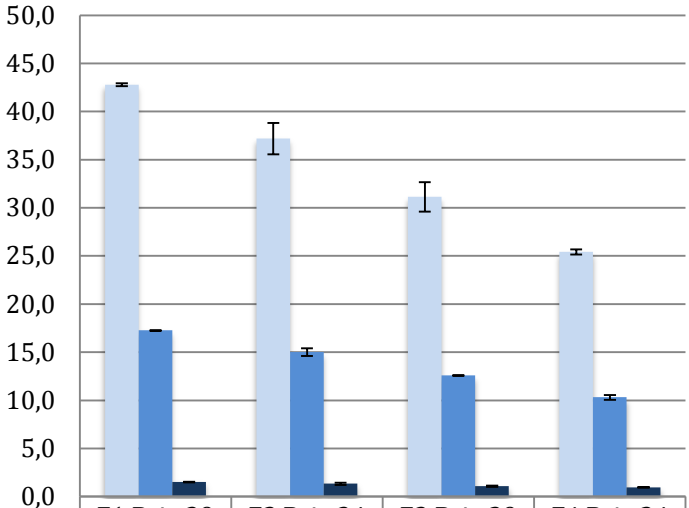
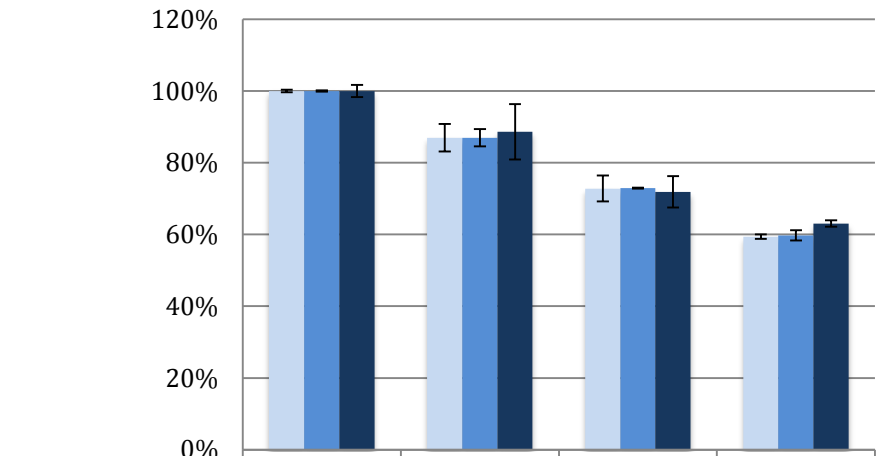


Figure 5).



	F1 Brix 39	F2 Brix 34	F3 Brix 29	F4 Brix 24
Sugar (g/L)	42,8	37,2	31,1	25,4
TA (g/L)	17,3	15,0	12,6	10,3
Gallic Acid Equiv. (mg/mL)	1,5	1,3	1,1	0,9

Figure 2. Pre-fermentation Sugar, Total Acid and Phenolic content (Gallic acid equivalents) of the four juice fractions of different concentrations after cryoconcentration, absolute values.



	F1 Brix 39	F2 Brix 34	F3 Brix 29	F4 Brix 24
Sugar	100%	87%	73%	59%
TA	100%	87%	73%	60%
Gallic Acid Equiv.	100%	89%	72%	63%

Figure 3. Pre-fermentation Sugar, Total Acid and Phenolic content (Gallic acid equivalents) of the four juice fractions of different concentrations after cryoconcentration, expressed as percentage of F1.

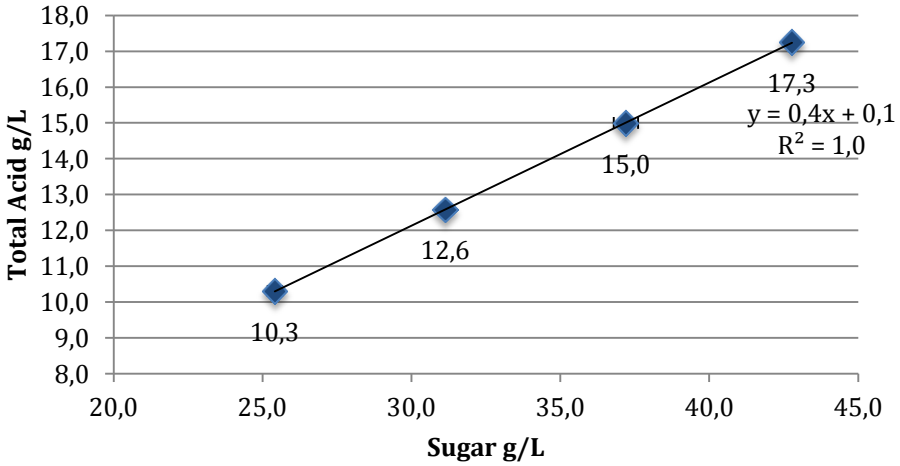


Figure 4. Total acidity plotted against the sugar concentration of the juice fractions, pre-fermentation.

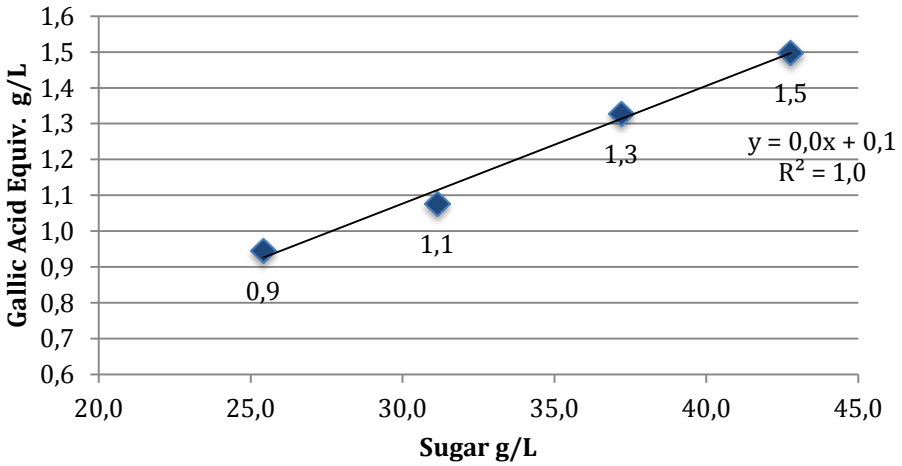


Figure 5. Phenolic content, expressed as gallic acid equivalents, plotted against sugar concentration of the juice fractions, pre-fermentation.

Ice cider juice

The concentrated apple juice used for all the subsequent yeast fermentations was analysed biochemically, the properties presented in Table 9. The HPLC analysis of sugar content (glucose, fructose and saccharos combined) was 36.8 %, and the total acid measured to 14.8 g/l.

Table 9. *Pre-fermentation juice properties*.

Measurement	Value	Stddev
Sugar %	36.8 %	0.1
TA g/L	14.8 g/L	0.2
Malic acid mg/mL	*17.7 g/L*	0.0
Citric acid mg/mL	*0.3 g/L*	0.0
GAE g/L	1.2 g/L	0.0

Fermentation rate

The rate of fermentation was monitored by measuring the loss of weight from the fermentation bottles at irregular intervals. The loss of weight for the yeast trial is presented in Figure 6, and for the concentration trial in Figure7.

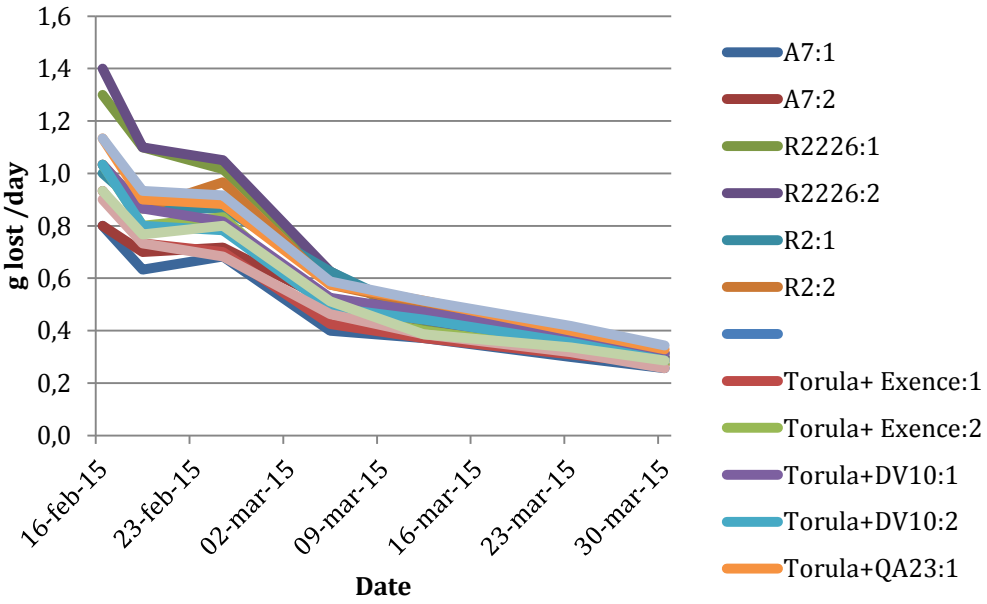


Figure 6, Decrease of weight of bottles during fermentation, yeast strain trial.

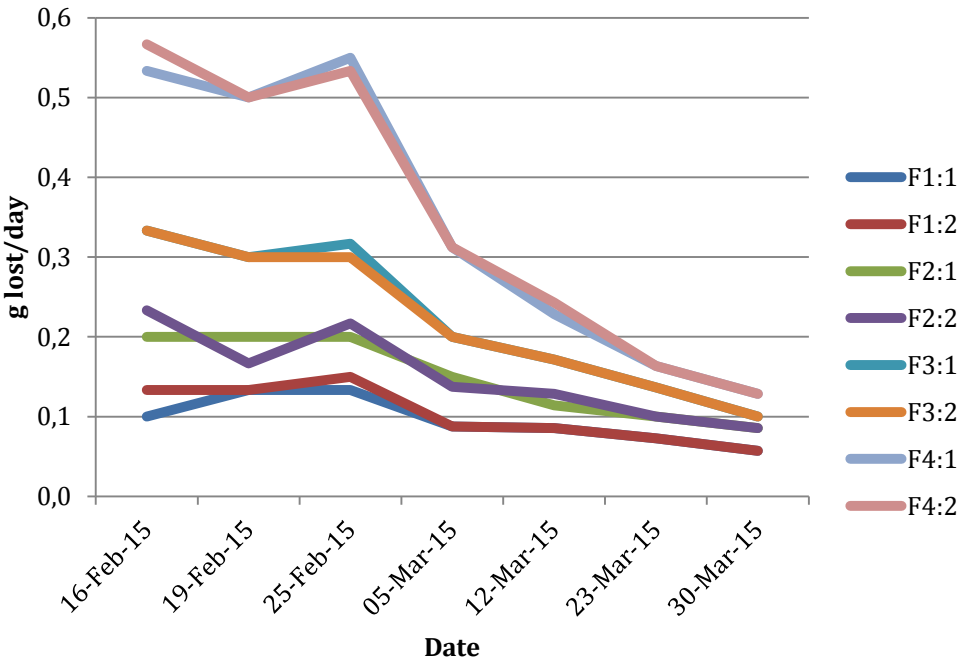
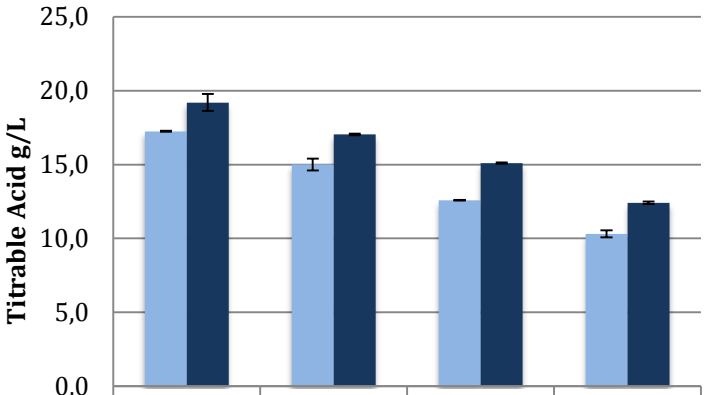


Figure7. Decrease of weight of bottles during fermentation, concentration trial.

Concentration fractions study

Titrable Acid

The titrable acid levels of the post-fermentation fraction samples, analyzed using the automated titration method, were higher than the pre-fermentation fraction samples for all the sugar concentration levels (*Figure 8*). For the most concentrated fraction F1 the increase was 11 % after fermentation, for second most concentrated fraction F2 14 %, followed by F3 20 % and F4 also 20 %.

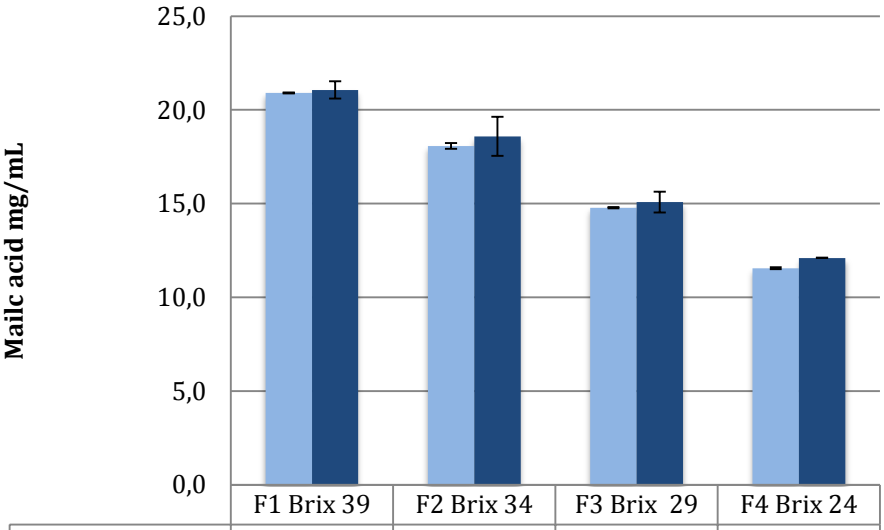


	F1 Brix 39	F2 Brix 34	F3 Brix 29	F4 Brix 24
Pre-fermentation TA	17,3	15,0	12,6	10,3
Post-fermentation TA	19,2	17,0	15,1	12,4

Figure 8. Titrable Acid content of the ice cider juice fractions, pre- and post-fermentation.

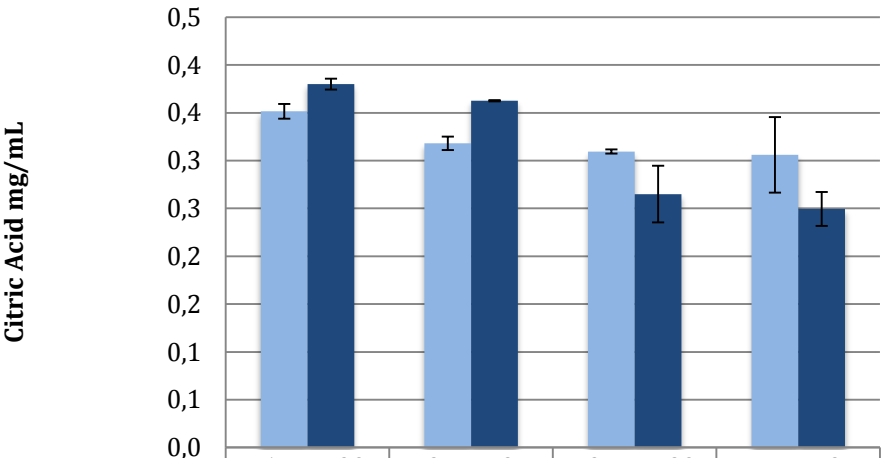
Organic acids

HPLC analysis of malic acid showed similar levels in samples pre- and post-fermentation for all the four concentration fractions (*Figure 9* and *Figure 10*). HPLC analysis of citric acid showed a higher level post-fermentation for the two most concentrated fractions F1 (11 % increase) and F (8 % increase) 2, and lower levels for fractions F3 (4 % decrease) and F4 (8 % decrease), compared to pre-fermentation.



	F1 Brix 39	F2 Brix 34	F3 Brix 29	F4 Brix 24
Malic pre-ferm.	20,9	18,1	14,8	11,6
Malic post-ferm.	21,1	18,6	15,1	12,1

Figure 9. Malic acid content of the ice cider juice fractions, pre- and post-fermentation.



	F1 Brix 39	F2 Brix 34	F3 Brix 29	F4 Brix 24
Citric pre-ferm	0,4	0,3	0,3	0,3
Citric post-ferm	0,4	0,4	0,3	0,2

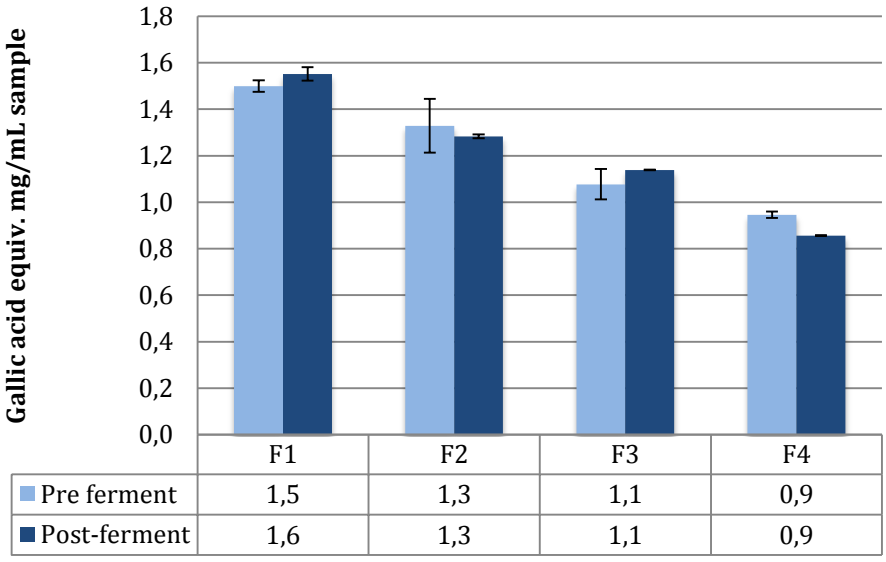
Figure 10. Citric acid content of the ice cider juice fractions, pre- and post-fermentation.

Acetic Acid

HPLC analysis of acetic acid in the four different concentration fraction samples did not detect any levels, either pre- or post-fermentation.

Phenolic compounds

The amounts of phenolic compounds were on similar levels in the pre- and post-fermentation juice fractions (*Figure 10*).



	F1	F2	F3	F4
Pre ferment	1,5	1,3	1,1	0,9
Post-ferment	1,6	1,3	1,1	0,9

Figure 10. Phenolic compounds (GAE) in the ice cider juice fractions, pre- and post-fermentation.

Ethanol

The amount of ethanol produced after 45 days of fermentation was for fraction F1 2.6 %, for F2 3.8 %, for F3 7.4 % and for F4 9.9 %. The ethanol level produced by fermentation was detected to be negatively correlated to the pre-fermentation sugar level in the fractions (

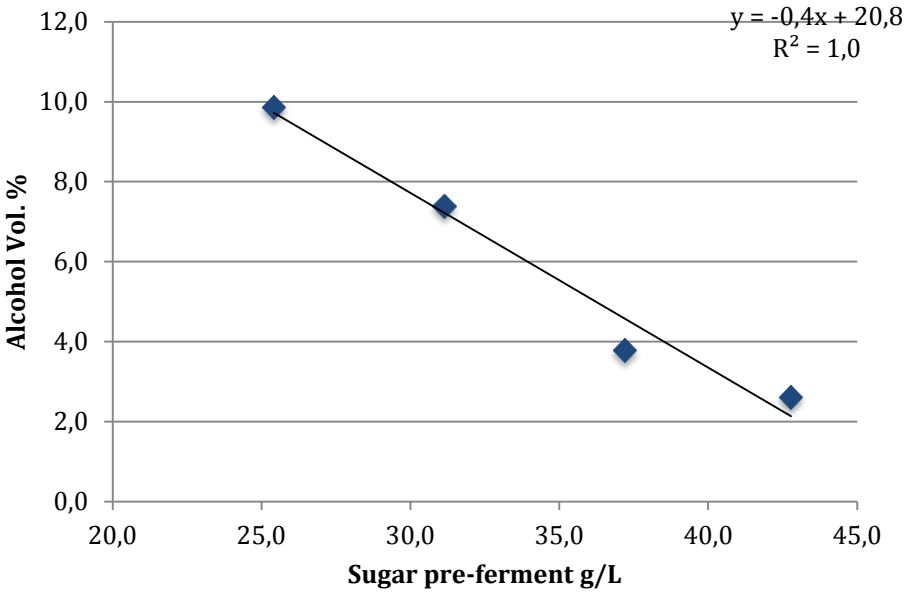


Figure 11). This was also manifested in the comparisons of the sugar level of the fractions pre- and post-fermentation (*Figure 12*); the post-fermentation sugar level was only 9.5 g/L in the least concentrated fraction (15.9 g/L sugar has been consumed), and 39.0 g/Lin the most concentrated fraction (3.8 g/L sugar has been consumed).

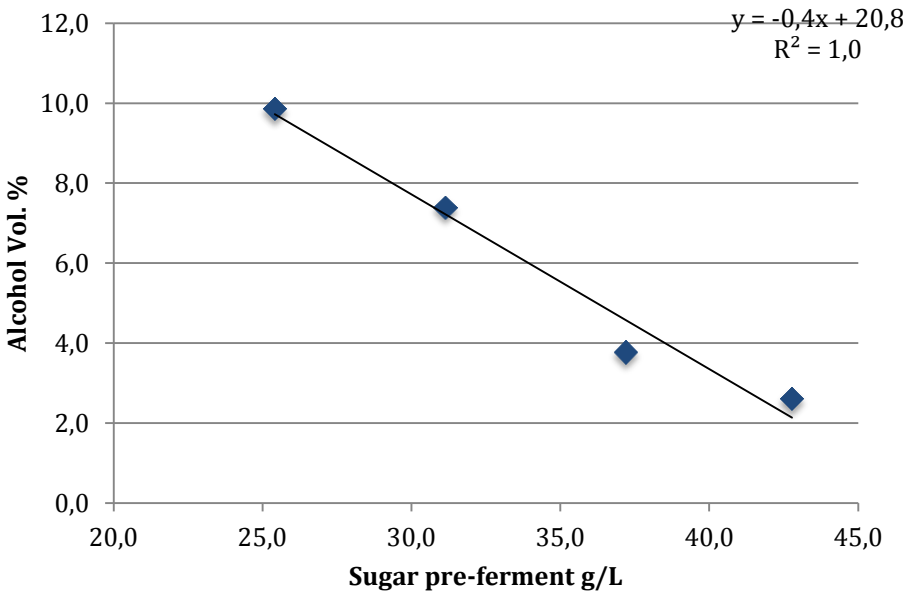
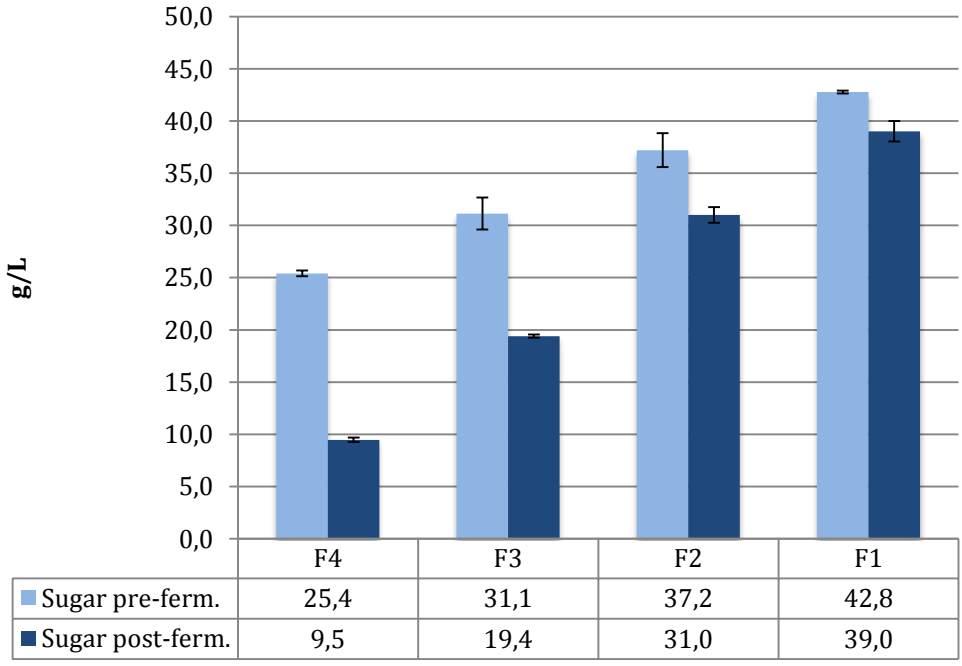


Figure 11. Ethanol (%) plotted against sugar concentration of the juice fractions, post-fermentation.



	F4	F3	F2	F1
Sugar pre-ferm.	25,4	31,1	37,2	42,8
Sugar post-ferm.	9,5	19,4	31,0	39,0

Figure 12. Sugar level in the juice fractions pre- and post- fermentation.

Yeast strain study

Titratable acidity

The analysis of total acidity level (

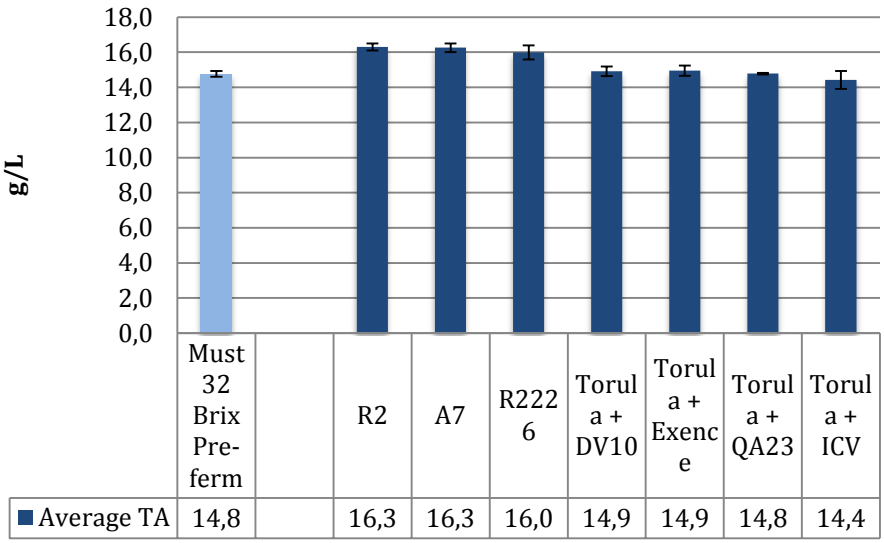


Figure 13) in the *T. delbrueckii* + *S. cerevisiae* samples post-fermentation did not show any significant differences from the pre-fermentation juice. The samples fermented with *S. cerevisiae* alone had however higher content of titrable acidity than that of the pre-fermentation juice, on average 16.2 g/l compared to 14.8 g/l, an increase by 9 % (*Figure 13*).

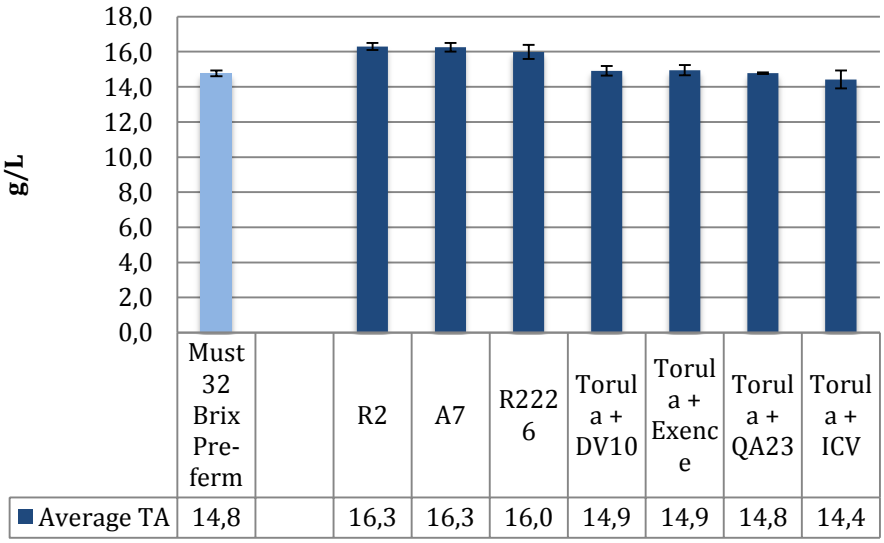


Figure 13. Total Acid for the different yeast cultures post-fermentation, and the original juice pre-fermentation.

Organic acids

HPLC analysis of the yeast strain fermentation samples showed a decreased content of malic acid, in the range of 4-9 % after fermentation (*Figure 14*). HPLC analysis showed a 3-20 % increase of citric acid in the different yeast strain fermentation samples (*Figure 15*).

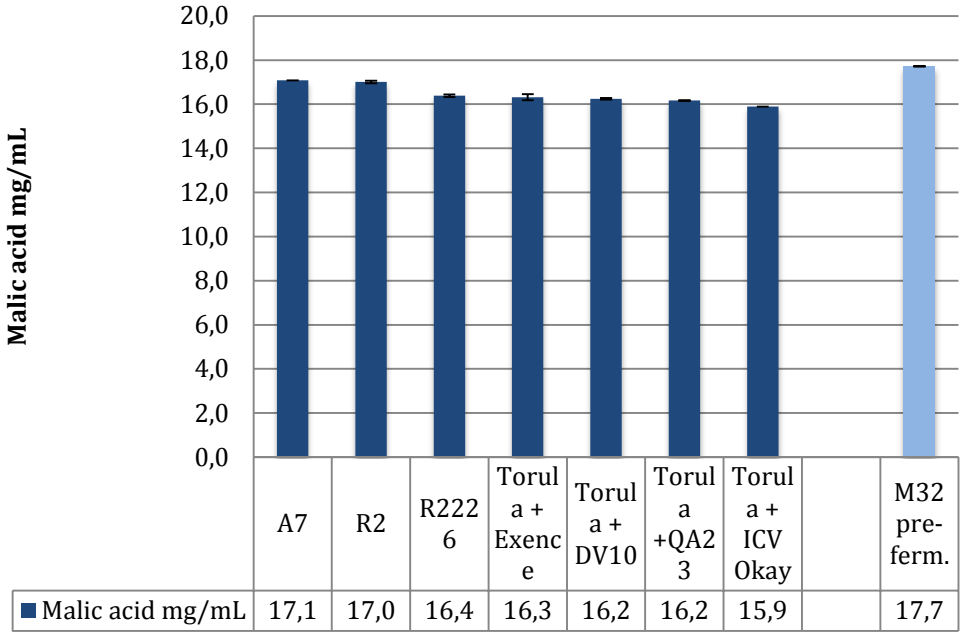


Figure 14. Malic acid in yeast strain fermentations and the pre-fermentation juice.

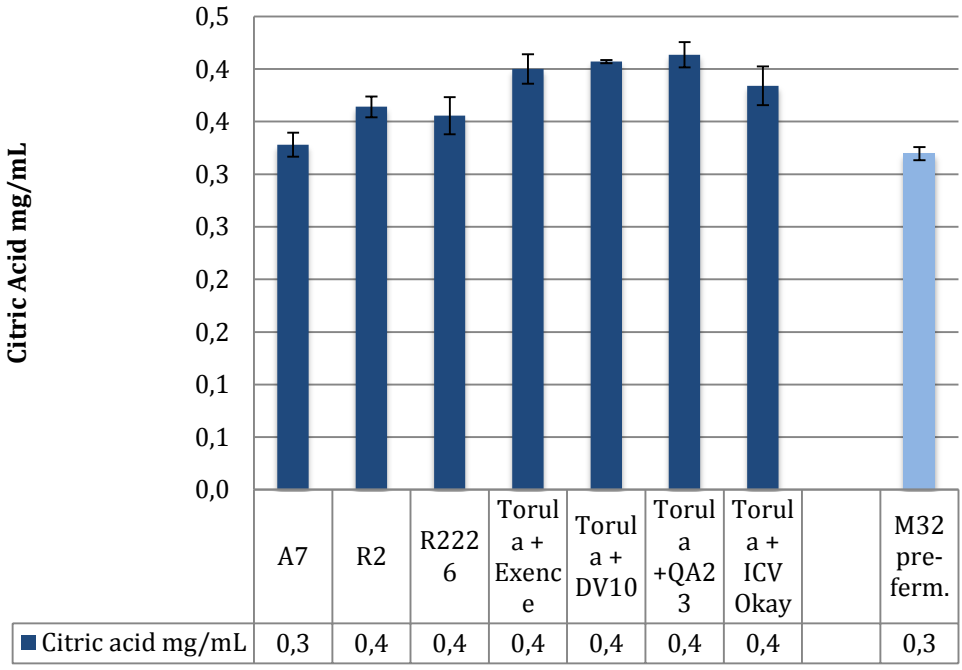


Figure 15. Citric acid in yeast strain fermentations and the pre-fermentation juice.

Acetic Acid

HPLC analysis of acetic acid did not detect any levels in the seven different yeast fermentation samples or in the pre-fermentation juice.

Phenolic compunds

The analysis of amount of phenolic compounds, GAE, (*Figure 16*) showed similar levels in the pre-fermentation juice and the post-fermentation yeast strain samples.

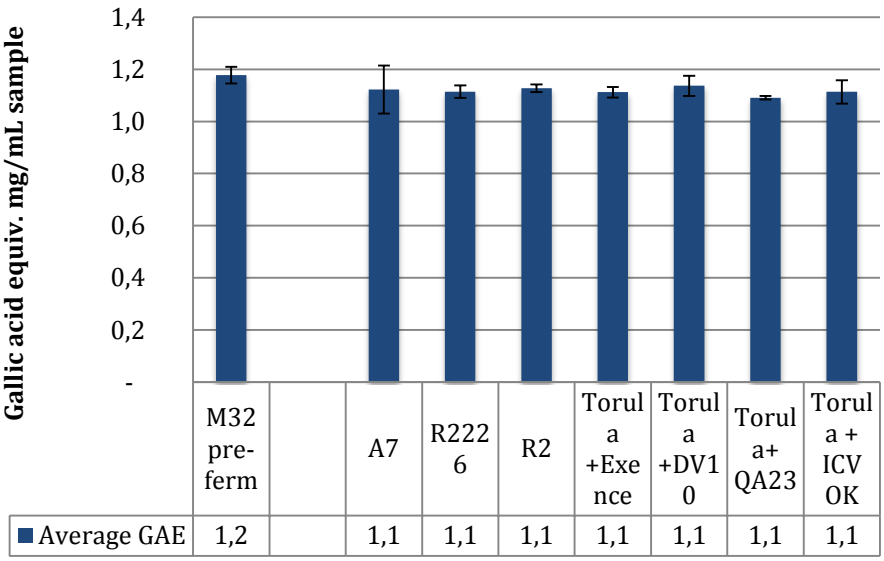


Figure 16. Phenolic compounds, expressed as Gallic acid equivalents, in the ice cider juice fractions, pre- and post-fermentation.

Ethanol

After 45 days of fermentation, the level of ethanol produced was variable between the different yeast strains, the highest level 6.7 % produced by the *S. cerevisiae*-strain R2226 and the lowest level 5.0 % by the *S. cerevisiae*-strain A7. On average the ethanol level produced in the two groups is similar, however more variable in the *S. cerevisiae-group*; the average level of ethanol produced by the 3 *S. cerevisiae* alone-group is 6.0(+/-0.8) %, and by the *T. delbrueckii* + *S. cerevisiae*-group the average ethanol level produced is 6.3 (+/- 0.4) %.

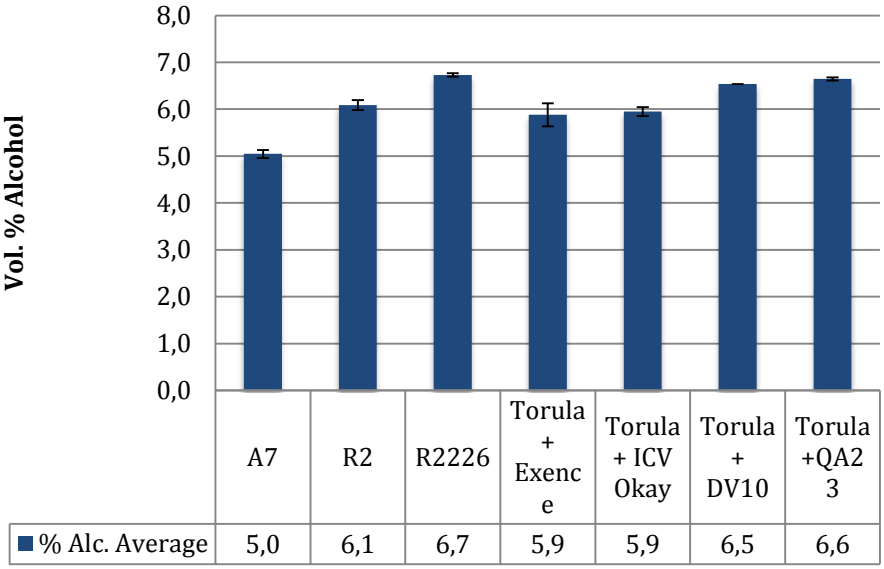


Figure 17. Ethanol (%) in the different yeast cultures.

Residual Sugar

The amount of residual sugar (glucose, fructose and saccharos combined) analyzed by HPLC was at 22-26 % for the different yeast strain fermentation samples (*Figure 18*).

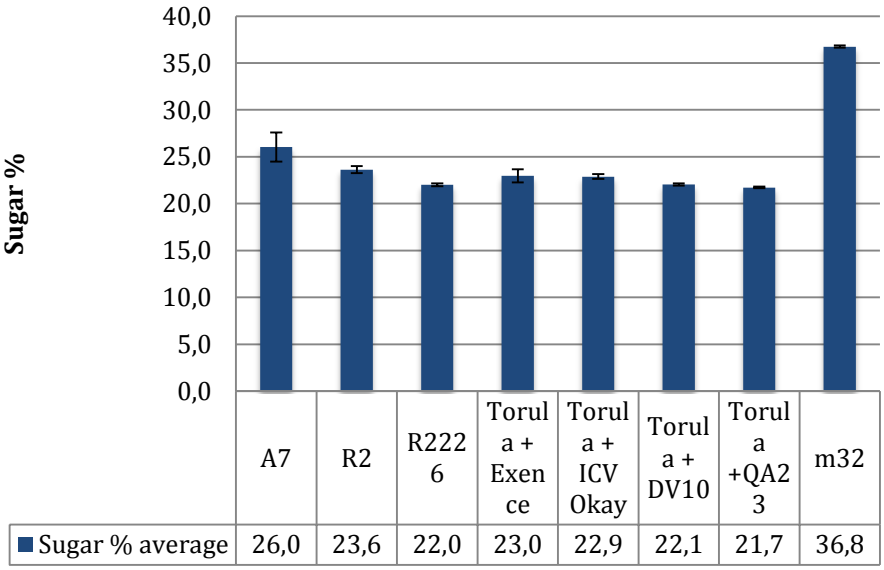


Figure 18. Residual sugar level for the yeast strain fermentations.

Sugar: acid ratio

The sugar to acid ratios are ranging between 14 to 16 for the different yeast strains, and are given in Table 10.

Table 10. Sugar:acid ratio, yeast strain trial.

Yeast strain	Sugar level g/l	Titrable acid g/l	Sugar:acid ratio
A7	260	16.3	**16**
R2	236	16.3	**14**
R2226	220	16.0	**14**
Torula + Exence	230	14.9	**15**
Torula + ICV Okay	229	14.4	**16**
Torula + DV10	221	14.9	**15**
Torula +QA23	217	14.8	**15**
		Average	***15***

Sensory Analysis

The sensory analysis showed overall small differences between the samples. In a few attributes there were significant differences identified. The full results from the sensory analysis are given in Appendix 1. In summary, the results for each attribute were:

- Sweetness: For the sweetness attribute, the sample perceived as most sweet (Yeast A7) and the sample perceived as least sweet (Yeast R2226) were significantly different from most of the other samples. For the samples in between, the differences were not significant.

- Acidity: The acidity attribute gave less clear results compared to the sweetness attribute. The sample perceived as least acid (yeast Torula + QA23) was significantly different from 3 of the other samples, that were ranked as having the highest acidity (Yeast R2, R2226, followed by Torula + DV10). Except for that, the differences rated were not significant.

- Bitterness: For the attribute bitterness a few significant differences were found. The sample perceived as most bitter (Yeast R2226) were significantly different from the two least bitter samples (yeast A7 and yeast Torula + ICV Okay), and the sample perceived as the second most bitter (Yeast R2) significantly differed from the least bitter (yeast A7). Apart from that the differences among samples were not significant.

- Flowery flavours: For the attribute of flower flavour, no significant differences were perceived.

- Alcohol flavour: For the attribute alcohol flavour, the difference was only large enough to be significant between the least alcohol flavour (yeast A7) and the most alcohol flavour (yeast Torula + DV10).

- Smell intensity: The panellists were also asked to rate the overall smell intensity. In this no significant differences were detected.

- Overall appreciation: The panellists were also asked to rank which sample the appreciated the most overall. For most samples the rating was similar and there were no significant differences, except for the least appreciated sample (yeast R2) that was significantly different to all except the second least appreciated (yeast A7) and the third least appreciated (yeast Torula + QA23).

Discussion

Small-scale ice cider production

Aspects of the ice cider production in this project is discussed in the following section:

Apples

The apples selected for the ice cider will highly influence the qualitative traits of finished icecider. The apple cultivars (Aroma, Mutsu, Ingrid Marie and Cox Orange) in this project were utilized because they were available at that time from the producer. They may not be the most suitable for ice cider production, but since optimizing the apple cultivar was not the objective of this project this was acceptable. The ripeness of the apples was also not considered in this project.

Juice extraction

During the course of this project, different methods to extract the juice were tested. Although not quantified, there were clear differences observed between juice produced by cryoextraction and cryoconcentration methods. The personal observation was that juice pressed from frozen apples with the cryoextraction method was more fresh and bright in colour, but with less intensity of the colour, compared to the cryoconcentrated juice. The flavour was similarly fresh and bright, lacking any 'oxidized' flavours. Based on the observations it can be speculated that the oxidation of the flavour and colour compounds (phenolics) is occurring to less extent in the cryoextracted juice. For the production of the juice used for the subsequent fermentation trials, a combination of cryoextraction and cryoconcentration was used. The initial cryoextraction process was beneficial since it served as a primary concentration step and the subsequent handling of juice became easier due to the smaller volumes. Also, the small-scale equipment used for this project has the practical advantage of being easy enough to handle for one person. It would certainly be beneficial to do only a single cryoextraction procedure to achieve a juice above 32 °Brix. If the process conditions, temperature and equipment are optimized, this could likely be achieved. With the equipment available it was found that the initial juice extracted was at low °Brix, followed by juice stable around 25 °Brix, until the apples defrosted more and °Brix -level decrease. Colder apples at the start of pressing resulted in longer processing time, but not higher °Brix -levels. However, with more powerful equipment and colder apples most likely the juice extracted would be at higher °Brix -levels. Using 10-liter plastic bags for freezing and extraction is an efficient way for small scale trials since the freezing and thawing is fast, but quickly becomes too laborsome for larger volumes. Larger vessels such as buckets, tanks, IBCs with tap in the bottom would be the natural choice for processing larger volumes. The amount of high °Brix-juice extracted depends on the sugar content of the initial juice of the specific apple cultivar, but was typically around 20 % (from unconcentrated juice). The exchange could however likely be improved with process optimization (e.g. by re-freezing cycles of the discarded juice). In this study ice cider was paralleled to ice wine fermentation.

Ice wine comparison

As previously discussed, ice wine grapes freeze naturally on the wine and go through several freeze-thaw cycles before harvesting that affects the chemical profile of the ice wine juice (due to e.g. breakdown of tissues, oxidation processes, microbial processes) (Inglis et al., 2006b). Thus, this is not necessarily comparable to icecider juice produced by cryoconcentration techniques. Even though there likely are differences in

the chemical composition, the osmotic stress on the yeast cells due to the high sugar content nevertheless likely share similarities.

Fermentation conditions

The fermentation temperature is an important aspect that was not investigated in this project. The fermentation temperature was kept low during most of the fermentation, in order to reduce the amount of detrimental by-products and off-flavours that may be produced during the fermentation from stressed yeast strains. This also means that the speed of fermentation was very low. As volatile acids were not detected in any of the samples that were analyzed, even the most concentrated fractions, the formation of by-products does not seem to be a problem at this fermentation rate and temperature. The choice of proper fermentation temperature is a balance between flavour development, by-product formation and speed of fermentation. There could possibly be room to increase the fermentation temperature used in this project in order to achieve a more efficient fermentation, without production of high volatile acids and off-flavours.

Fermentation rate

Since the temperature was kept low, the fermentation took longer than expected in this project. It was hard to predict in the beginning exactly how long it would take to achieve the required alcohol content. At the time of analysis only 5-6.7 % ethanol had been produced by the different yeast strains, and 2.6-9.9 % for the concentration fractions. In retrospect, it would have been beneficial to either wait longer to do the sampling for analysis or use a higher fermentation temperature to achieve a faster process, to achieve higher ethanol content at the time of analysis.

Yeast acclimatization

In this project yeast rehydration was performed in a step-wise manner in order to acclimatize the yeast to the high-sugar conditions, as described by Kontkanen et al. (2004) to be successful during ice wine production. In the initial phase of this project, a first fermentation was performed without these procedures, and this fermentation did not start properly. This juice was also SO2 treated. Without confirmation that this was the reason, this could indicate that the yeast could not handle the osmotic pressure and/or the SO2 level used. Thus, procedures to support the yeast are recommended but would need to be further evaluated an optimized. The acclimatization of the yeast is also performed to reduce the amount of volatile acids produced. In these fermentations no acetic acid was observed. Juice treatment procedure was not the focus of this study and thus not extensively discussed or evaluated, but this likely too has an effect on the finished product.

Cryoconcentration

Cryoconcentration effect on biochemical composition of the juice

The effect of cryoconcentration on apple juice composition has not previously been reported. The analysis of the cryoconcentration shows that concentrations of several important compounds are directly and linearly correlated to the sugar level. The amount of total soluble solids measured as °Brix indicated a slightly lower level than the sugar concentration indicated by HPLC-analysis. The phenomenon is equally observed across all four concentration fractions. This would usually be the opposite case. This could potentially be explained by evaporation of the samples during handling before the HPLC-analysis.

Cryoconcentration effect on total phenolic content in the juice

This study showed that the phenolics are concentrated to the same extent as sugars during the cryoextraction/cryoconcentration process. A direct linear relationship was

observed between the phenolics content and the sugar content of the fractions samples. The amount of phenolics is likely however depending on the mode of preparation of the apple juice, since phenolics are not evenly distributed in the fruit, and processing conditions affect the extraction of these. It is thus likely that a cryoextraction process will give different amounts compared to a cryoconcentration process.

Cryoconcentration effect on total acids content in the juice

The amount of total acid was also concentrated in the same extent as sugar during the cryoextraction/cryoconcentration process. Also here a direct linear relationship was observed. When comparing results form the automated titration method and the HPLC analysis of organic acids, the HPLC-analysis indicate slightly higher levels of acids than the automated titration. The explanation for this would need further investigation.

Effect of different levels of juice concentration on fermentation

Concentration effect on fermentation rate and ethanol produced

There was a clear difference in fermentation rate observed between the different concentration fractions. Fermentation rate of the ice cider was negatively correlated to the initial concentration of the juice – a higher concentration of sugar resulted in a lower level of ethanol. This is in line with studies of ice wine fermentation, as observed by Inglis et al. (2006a). This is likely due to the hyperosmotic stress on the yeast cells, as previously described for ice wine fermentation. Furthermore, Inglis et al. (2006a) found that fermenting grape juice above 42 °Brix is very difficult. The findings in this study give support to that ice cider fermentation is similar in this respect.

Concentration effect on total phenolic content in the juice after fermentation

The results from this study do not indicate that there is a change in phenolic content during fermentation.

Concentration effect on titratable acidity, organic acids and acetic acid in the juice during fermentation

The amount and distribution of acids are indicated in this study to be altered during fermentation. The amount of total acids increase by 11-20 % after fermentation, the amount of malic acid remained on similar levels, while the amount of citric acid both increased (for the most concentrated fractions) and decreased (for the least concentrated fractions). According to previous studies the amount of acids typically increase during fermentation (Whiting, 1976), but the process is complex with influence of medium, yeast strain, pH, and more, and changes during the course of fermentation. Previous studies have demonstrated that pyruvic, lactic, succinic, and acetic acid are produced during cider fermentation (Zhang et al., 2008). The different rate of fermentation in the different concentration factors could be one of the factors responsible for the different changes of acids. Acetic acid was not detected even for the most highly concentrated samples. Other studies indicate that this could be a problem during high-sugar fermentation (Inglis, 2006a), and to see some development of acetic acid was expected. The production of acetic acid mainly occurs during the initial phase of fermentation (Whiting, 1976) so the limited progress of fermentation for the most concentrated fractions is likely not the explanation. The steps taken to limit the production (reducing the osmotic stress on yeast cells and low fermentation temperature) could be functional. However, to verify the detected levels and find possible explanations would require further investigations.

Yeast strain effect

Yeast strain effect on fermentation rate and ethanol produced

It was demonstrated in this project that the amount of ethanol produced is influenced by choice of yeast strain; yeast R2226 producing the highest level of almost 7 % and yeast A7 the lowest, 5 %, after 45 days of fermentation. The fermentation would need to continue further to achieve a finished product. The level of ethanol produced was slightly higher when *T. delbrueckii* was used, but the difference was small. An observation is that the variation was smaller in *T. delbrueckii* treatments, indicating that fermentation may be more predictable using *T. delbrueckii*. Evaluating the effect of *T. delbrueckii* alone was not the main goal of this study, and such study would need to be repeated and enlarged to make conclusions of the *T. delbrueckii*- effect on fermentation rate.

Yeast strain effect on total phenolic content in the juice after fermentation

The results form this study does not indicate that there was a change in phenolic content during fermentation.

Yeast strain effect on total acidity, organic acids and acetic acid in the juice during fermentation

The level of total acidity was observed to be influenced by yeast strain in this project. The *T. delbrueckii*-containing cultures did not display an increased acidity, while the pure *S. cerevisiae* cultures displayed an increase in total acidity by 9 %. This is in line with previous findings that yeast culture influence the acid progression during fermentation (Whiting, 1976). As discussed previously, the evolution of acids during fermentation is a complex process influenced by many factors.

Yeast strain effect of the flavour of the ice cider

Even though the differences are small and ranking the samples was deemed difficult by the participants in the sensory evaluation, some significant differences were identified. The yeast A7-sample was perceived as being the most sweet, and this is also the sample with the highest residual sugar level and lowest ethanol level as analyzed by HPLC. This was also the sample perceived as having the least alcoholic flavour. The sample perceived as least sweet, yeast R2226, has among the lowest residual sugar level and the highest ethanol level. Thus, these biochemical traits were perceivable by the panelists. For acidity and bitterness there was no pattern observed in connection to the biochemical analysis. The flower flavours and the overall smell intensity was probably not different enough to be perceived by the panelists or interpreted in different ways. There was no clear winner in the overall liking rating of the samples, but there was however a loser. The yeast R2 was significanlty less appreciated compared to the other yeasts by the participants.

The sugar:acid ratio is of high importance for flavour perception, as discussed previously. There is a high residual sugar level in the ice ciders for all the yeast strains in this project, due to the fact that the fermentation has not proceeded long enough at time of analysis. The acidity is however also at a high level, making the sugar:acid ratio for all the yeast strains to fall within the range of commercial ice ciders.

Conclusions and further analysis

Ice cider production process

Producing ice cider is highly feasible for a small-scale operation, but it requires knowledge about a number of factors concerning the raw material and the fermentation process. Based on learning's from ice wine fermentations, it is concluded that using a higher yeast inoculation rate, acclimatizing the yeast to high sugar conditions, and use of yeast micronutrients is beneficial to achieve an efficient fermentation without excessive production of volatile acids. Based on observations made in this project, which is in line with the research on ice wine production, a juice with too high concentration will be very difficult to ferment. The choice of apple cultivars and their ratio will have a large impact on the finished product. Apples have different flavour profiles, different levels of acidity, and different phenolic content, thus the blend of apple cultivars will be of high importance. This was however not studied in this project. A subsequent step would be to further study the effect of apple cultivar, and this would require acquisition of apples at different times during the harvest since different cultivars ripen at different times. It could also be desirable to use apples with higher amount of polyphenols (cider apple varieties), to possibly achieve an ice cider with more character. The ripeness level of the apples would be of further interest to evaluate and the mode of storage of the apples before pressing. The exposure of climate conditions is a requirement for ice wine production, and this has an effect of the biochemical composition of the grape juice and the flavour of the finished ice wine. Thus, to investigate the comparable effect on apples, and ice cider, would be of high interest. The effect of pectolytic enzymes, SO2 treatment, and other juice treatments would also be interesting to evaluate. The most efficient mode of inoculation of yeast, and the use of yeast nutrients during rehydration and fermentation could also be of interest to evaluate further. The optimal temperature during fermentation will be yeast strain dependent, and would need to be further evaluated. The length of fermentation, resulting in different ethanol and residual sugar level, is also of high importance for the style of the finished product. This would also be highly interesting to evaluate. Finally, the post-fermentation handling has not been a subject in this project due to time constraint. However, the qualitative properties highly depend on storage of the product and the flavour characteristics perceived will surely be different after a certain time of storage. The possibilities of develop the ice cider by storage time and conditions (such as oak barrels) would be highly interesting to elaborate further.

Cryoconcentration

It was demonstrated in this project that concentrating the apple juice by cold has an equally concentrating effect on sugars, total acids and total phenolics. This has to our knowledge not been previously demonstrated. However, the analysis is limited in number of samples and replicates, and does not analyse individual components. Efficiency of the cryoconcentration technique can be further optimized to achieve a higher output. A step-wise concentration process could be an option. The differences between cryoconcentration and cryoextraction in biochemical composition of the apple juice would be highly interesting to further investigate, as this can result in very different product qualities. The amounts of phenolic compounds are not evenly distributed throughout the apple and are highly depending on the mode of preparation of apple juice. Thus, it would be interesting to further evaluate and optimize the procedure for juice concentrate extraction. Also, since a clear difference was observed in the flavour and colour between cryoconcentrated juice and cryoextracted juice, it would be very interesting to further evaluate the mode of production effect on biochemical composition.

Juice concentration effect

It was demonstrated in this project that the level of juice concentration achieved will highly influence the fermentation kinetics and output. It will be the basis of the style of product produced. The juice needs to be sufficiently concentrated to produce an ice cider with the alcohol content and residual sugar level that defines the product. On the other hand, the juice should not be excessively concentrated due to the hyperosmotic stress placed upon the yeast cells in a high-sugar juice that result in slow and potentially stuck fermentations. The juice needs to hold a minimum of 32 °Brix, while a juice of above 42°Brix will be very difficult to ferment.

Yeast strain selection

It was demonstrated in this project that the selection of yeast strain has an effect on level of ethanol produced, and acid development during fermentation of ice cider. The sensory evaluation also indicated some yeast dependent differences among the samples, mainly in connection to residual sugar level, ethanol and acidity. The yeast R2 was perceived as less liked compared to the other yeast strains.

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Popular scientific summary

Ice cider product development

Ice cider is a dessert wine made from apple juice that has been concentrated by freezing. In this project aspects of producing ice cider were investigated. The freeze concentration process, the impact of different levels of concentration of the apple juice, and the impact of yeast strain used for fermentation was investigated. Aspects of ice cider production have not been previously scientifically documented.

Ice cider originates from Canada, where the product since it's dawn in the 1990'ies has become popular and successful. Ice cider is a natural product made from only concentrated apple juice. In addition to high quality apples, the original production method requires cold temperatures in winter. Ice cider is a high value product, that could be suitable production in Sweden to a larger extent than today. This project aims to investigate and document some of the aspects of ice cider production process for Swedish conditions. The ambition is to start building experience and knowledge useful for ice cider production in Sweden.

In freeze-concentration techniques water is removed from the solution in frozen form. The freeze-concentration can be done by freeze-thawing apple juice (cryoconcentration), or by pressing whole, frozen fruit (cryoextraction). The level of juice concentration achieved in the freeze-concentration step will highly influence the finished product. Concentrated juice contains a high amount of sugars. It has previously been demonstrated in ice wine production that the high sugar level places stress on the fermenting yeast. That makes ice wine fermentation difficult and too much bi-products, such as acetic acid, can be formed. The yeast strain selected for fermentation is known to influence fermentation rate and output, including flavour, for cider and ice wine. The sugar level in the juice is often measured in °Brix.

Results:

- ***Freeze-concentration:*** It was demonstrated in this project that concentrating the apple juice by cold has an equally concentrating effect on sugars, the acidity and phenolic compounds (sometimes called tannins).

- ***Concentration effect:*** It was demonstrated that the concentration highly influenced speed and rate of fermentation: the higher the sugar level, the slower the fermentation. Production of acetic acid was not detected, even for the highest concentrations tested. In order to produce a good ice cider, the juice needs to be sufficiently concentrated to produce the alcohol content and residual sugar level that defines the product, while not be too concentrated and place excessive stress upon the yeast cells, resulting in slow and potentially stuck fermentations. The juice needs to hold a minimum of 32 °Brix, while a juice of above 42°Brix will be very difficult to ferment.

- ***Yeast strain effect:*** It was demonstrated in this project that the selection of yeast strain has an effect on level of ethanol produced, and acid development during fermentation of ice cider. The level of phenolics was found to remain fairly stable across different yeast strains during fermentation. The yeast strain also had an effect on the flavours and overall liking of the ice cider by consumers, demonstrated in sensory analysis. Thus, the yeast strain selected is important for the product both in terms of production efficiency and for the flavour development.

Appendix 1. Sensory Analysis results

Description
Ranking is a method used when the test objective is to compare several samples according to a single attribute (e.g. sweetness, bitterness, preference or crunchiness). During a ranking test each panellist receives at least 3 samples. The panellist must rank the samples according to the attribute of interest. This method is particularly useful when samples are to be screened for later analysis.

Study
Number of panelists: 22
Number of products: 8
Number of records: 22

Product information (if applicable)

Product information

	codes	image
1	210	A7
2	682	Torula DV10
3	158	Torula Exence 1
4	948	Torula ICV Okay
5	547	R2
6	842	R2226
7	795	Torula QA23

Results
Kön(Q1)
Summary

	Results
Kvinna(2)	54.55%
Man(1)	45.45%
Top 2 Box (%)	100%
Bottom 2 Box (%)	100%

Ålder(Q2)
Summary

	Results
61 – 75 år(4)	45.45%
46 – 60 år(3)	18.18%
31 – 45 år(2)	4.55%
19 – 30 år(1)	31.82%
Top 2 Box (%)	63.64%
Bottom 2 Box (%)	36.36%

ranking SÖTMA(Q3)
Ranks

	1	2	3	4	5	6	7
anonymous_024mfddlra	3	6	1	2	4	5	7
anonymous_0lvc4rdmyi	7	5	4	2	3	1	6
anonymous_5vzpi45kpu	7	5	3	4	2	1	6
anonymous_8e4w1gknrq	7	2	6	4	5	1	3
anonymous_8xjz8ggc93	7	5	2	6	4	1	3
anonymous_944hefqece	5	3	7	2	6	4	1
anonymous_96ujwykndd	4	1	6	3	5	2	7
anonymous_9z65lftsip	6	4	5	7	3	1	2
anonymous_cagypnt5xb	5	7	3	6	2	1	4
anonymous_f1dk1i8o5m	4	7	1	5	2	3	6
anonymous_gzltvg7v71	7	5	6	3	2	1	4
anonymous_j3v0ihkodz	7	6	3	2	5	4	1
anonymous_l4rzlchg7j	6	2	3	4	5	7	1

anonymous_l5aw4q06m2	4	1	6	7	5	3	2
anonymous_mdekrgyxt8	3	5	7	2	4	1	6
anonymous_mhcbcahd59	7	4	6	5	3	1	2
anonymous_n9jxpjx3ul	6	7	4	2	1	3	5
anonymous_se1zk6nnp3	6	5	4	2	1	3	7
anonymous_t2x2ycc98c	1	6	2	4	5	3	7
anonymous_tmnbvk7n6g	6	3	5	1	7	2	4
anonymous_zgswxslmck	6	3	1	7	5	2	4
anonymous_zo9oy82idd	6	2	4	7	3	5	1

Sizes by ranks

	1	**2**	**3**	**4**	**5**	**6**	**7**
1	1	2	3	1	2	9	4
2	0	3	2	7	4	3	3
3	2	3	4	2	4	5	2
4	3	2	4	4	3	2	4
5	2	6	2	2	7	2	1
6	7	3	5	2	1	0	4
7	7	3	2	4	1	1	4
Total	22	22	22	22	22	22	22

Friedman test

	Friedman
chi-squared	21.31
df	6
p.value	0.002

Level of significance (Friedman): 5%

Rank.sum

	1(A)	**2(B)**	**3(C)**	**4(D)**	**5(E)**	**6(F)**	**7(G)**
1	4.55%	9.09%	13.64%	4.55%	9.09%	40.91%	18.18%
2	0%	13.64%	9.09%	31.82%	18.18%	13.64%	13.64%
3	9.09%	13.64%	18.18%	9.09%	18.18%	22.73%	9.09%
4	13.64%	9.09%	18.18%	18.18%	13.64%	9.09%	18.18%
5	9.09%	27.27%	9.09%	9.09%	31.82%	9.09%	4.55%
6	31.82%	13.64%	22.73%	9.09%	4.55%	0%	18.18%
7	31.82%	13.64%	9.09%	18.18%	4.55%	4.55%	18.18%
Rank sum	120	94	89	87	82	55	89
Mean rank	5.45	4.27a	4.05A	3.95A	3.73A	2.5ABCDeG	4.05A
StDev.	1.65	1.91	1.94	2.01	1.64	1.68	2.21

Level of significance for the Mean Rank (LSD): A<5%; a<10%

LSD.rank.theory

	x
1	28.09

Signif.

	1	**2**	**3**	**4**	**5**	**6**	**7**
1		0.07	**0.031***	**0.021***	**0.008***	**0***	**0.031***
2	0.07		0.727	0.625	0.402	**0.006***	0.727
3	**0.031***	0.727		0.889	0.625	**0.018***	1
4	**0.021***	0.625	0.889		0.727	**0.026***	0.889
5	**0.008***	0.402	0.625	0.727		0.06	0.625
6	**0***	**0.006***	**0.018***	**0.026***	0.06		**0.018***
7	**0.031***	0.727	1	0.889	0.625	**0.018***	

OUTPUT TYPE NOT DEFINED: 15

Kommentarer(Q4)

Panelist	**Remark**
A0002	De klara dryckerna var rätt lika i sötma. De matta något beskare

A0003	Ganska sött över lag
A0005	Svårt att särskilja. De minst söta smakade konstgjort. Ingen smakade riktigt naturlig sötma.
A0006	Kännas sötma vid drickande och efter i munnen. Den med mindre sötma haft fint syrlig smak från början
A0008	De blev sötare efter hand jag smaka den ordning dom stod i :-)
A0011	842 trevlig med små bubblor, förhöjer smaken.
A0012	De allra flesta var i mitt tycke väldigt söta, nästan lite för söta
A0013	Tyckte att alla var väldigt söta, dock var 795 mjukast när man svalde,
A0014	Svårt att känna eftersom syrligheteh var hög i alla prover
A0015	Strävheten påverkar sötman för mig. Upplever alla som söta och svårt att rangordna.
A0016	Dem ligger relativt nära varandra men 210 mest söt och aningen tunn i smak
A0017	682 ohyggligt söt,
A0018	Svårt och osäkert. Alla är ju söta.
A0019	Det var svårt, men så här får det bli!
A0021	För mycket sötma i 210, 948, 795, 547.
A0022	De tre sötaste låg nära varandra. 795 var på gräsen till sur.

ranking Syra(Q5)

Ranks

	1	2	3	4	5	6	7
anonymous_024mfddlra	3	7	1	2	5	4	6
anonymous_0lvc4rdmyi	7	2	3	5	6	4	1
anonymous_5vzpi45kpu	4	1	5	3	6	7	2
anonymous_8e4w1gknrq	4	7	3	2	5	1	6
anonymous_8xjz8ggc93	3	4	5	2	7	6	1
anonymous_944hefqece	3	7	5	1	6	2	4
anonymous_96ujwykndd	3	7	4	6	2	5	1
anonymous_9z65lftsip	5	7	4	6	3	1	2
anonymous_cagypnt5xb	5	4	2	3	6	7	1
anonymous_f1dk1i8o5m	4	2	3	5	7	6	1
anonymous_gzltvg7v71	5	2	1	4	7	6	3
anonymous_j3v0ihkodz	6	1	3	7	5	4	2
anonymous_l4rzlchg7j	5	6	1	7	2	4	3
anonymous_l5aw4q06m2	6	7	2	3	5	1	4
anonymous_mdekrgyxt8	6	3	2	1	5	7	4
anonymous_mhcbcahd59	1	3	4	2	6	7	5
anonymous_n9jxpjx3ul	2	1	5	6	7	4	3
anonymous_se1zk6nnp3	1	2	6	3	4	7	5
anonymous_t2x2ycc98c	2	4	5	3	6	7	1
anonymous_tmnbvk7n6g	4	6	7	5	2	3	1
anonymous_zgswxslmck	1	6	7	2	4	5	3
anonymous_zo9oy82idd	5	6	3	4	2	1	7

Sizes by ranks

	1	2	3	4	5	6	7
1	3	3	3	2	0	4	7
2	2	4	3	5	4	1	3
3	4	2	5	5	1	1	4
4	4	3	3	2	2	5	3
5	5	0	5	3	5	2	2
6	3	4	1	3	6	3	2
7	1	6	2	2	4	6	1
Total	22	22	22	22	22	22	22

Friedman test

	Friedman
chi-squared	11.18
df	6
p.value	0.083

	codes	image
1	210	A7
2	682	Torula DV10
3	158	Torula Exence 1
4	948	Torula ICV Okay
5	547	R2

		6	842	R2226
		7	795	Torula QA23

Level of significance (Friedman): 5%

Rank.sum

	1(A)	**2(B)**	**3(C)**	**4(D)**	**5(E)**	**6(F)**	**7(G)**
1	13.64%	13.64%	13.64%	9.09%	0%	18.18%	31.82%
2	9.09%	18.18%	13.64%	22.73%	18.18%	4.55%	13.64%
3	18.18%	9.09%	22.73%	22.73%	4.55%	4.55%	18.18%
4	18.18%	13.64%	13.64%	9.09%	9.09%	22.73%	13.64%
5	22.73%	0%	22.73%	13.64%	22.73%	9.09%	9.09%
6	13.64%	18.18%	4.55%	13.64%	27.27%	13.64%	9.09%
7	4.55%	27.27%	9.09%	9.09%	18.18%	27.27%	4.55%
Rank sum	85	95	81	82	108	99	66
Mean rank	3.86	4.32	3.68e	3.73e	4.91	4.5	3BEF
StDev.	1.75	2.32	1.81	1.88	1.74	2.22	1.9

Level of significance for the Mean Rank (LSD): A<5%; a<10%

LSD.rank.theory

	x
1	28.09

Signif.

	1	**2**	**3**	**4**	**5**	**6**	**7**
1		0.485	0.78	0.834	0.108	0.329	0.185
2	0.485		0.329	0.364	0.364	0.78	0.043*
3	0.78	0.329		0.944	0.06	0.209	0.295
4	0.834	0.364	0.944		0.07	0.235	0.264
5	0.108	0.364	0.06	0.07		0.53	0.003*
6	0.329	0.78	0.209	0.235	0.53		0.021*
7	0.185	0.043*	0.295	0.264	0.003*	0.021*	

OUTPUT TYPE NOT DEFINED: 15

Kommentarer(Q6)

Panelist	**Remark**
A0002	Svårt. Alla känns så söta
A0005	Mycket svårt att särа på proven. Får återigen en konstgjord eftersmak på flera prov.
A0006	Jag tyckte om syrligheten i dem dricker som jag kände mest syrlighet. Väldighet frisk smak i munnen och även eftersmak.
A0008	Alla var väldigt syrliga mycket svårt att välja
A0011	Ger en angenäm strävhet på tungan.
A0013	Tyckte att det kunde vara högre syra på alla
A0014	682 smakade tom lite alkohol
A0015	Vissa av proverna hade en fräsch syra medansvar andra var åt garvsyrahållet
A0016	842 stack ut mest av alla på syra
A0017	Syran för mycket för att vara gott på de 3sista
A0018	Svårt igen. Osäker. Många är lika i syra.
A0019	Ok
A0021	Syran finns, dock inte lika tydlig som sötman.
A0022	Fruktsmake gör det svårt att välja.

Rangordning BESKA(Q7)

Ranks

	1	**2**	**3**	**4**	**5**	**6**	**7**
anonymous_024mfddlra	3	7	1	2	4	5	6
anonymous_0lvc4rdmyi	2	3	6	4	7	5	1
anonymous_5vzpi45kpu	1	6	3	7	5	4	2

anonymous_8e4w1gknrq	1	3	4	2	5	7	6
anonymous_8xjz8ggc93	6	3	4	5	7	1	2
anonymous_944hefqece	4	6	1	3	5	2	7
anonymous_96ujwykndd	2	5	3	7	4	6	1
anonymous_9z65lftsip	2	1	7	3	6	5	4
anonymous_cagypnt5xb	5	6	2	3	4	7	1
anonymous_f1dk1i8o5m	3	2	5	4	6	7	1
anonymous_gzltvg7v71	1	4	3	6	7	2	5
anonymous_j3v0ihkodz	5	3	1	4	6	2	7
anonymous_l4rzlchg7j	2	1	7	5	4	6	3
anonymous_l5aw4q06m2	2	1	6	5	4	7	3
anonymous_mdekrgyxt8	5	2	4	1	6	7	3
anonymous_mhcbcahd59	3	4	5	1	2	6	7
anonymous_n9jxpjx3ul	7	6	4	3	2	1	5
anonymous_se1zk6nnp3	1	2	6	5	4	7	3
anonymous_t2x2ycc98c	6	5	2	1	3	7	4
anonymous_tmnbvk7n6g	1	3	6	4	2	7	5
anonymous_zgswxslmck	1	6	5	3	7	4	2
anonymous_zo9oy82idd	6	7	1	3	2	5	4

Sizes by ranks

	1	**2**	**3**	**4**	**5**	**6**	**7**
1	6	3	4	3	0	2	4
2	5	3	2	2	4	3	3
3	3	5	3	6	1	0	4
4	1	2	4	4	6	2	3
5	3	2	3	4	3	4	3
6	3	5	4	1	4	3	2
7	1	2	2	2	4	8	3
Total	22	22	22	22	22	22	22

Friedman test

	Friedman
chi-squared	11.05
df	6
p.value	0.087

	codes	**image**
1	210	A7
2	682	Torula DV10
3	158	Torula Exence 1
4	948	Torula ICV Okay
5	547	R2
6	842	R2226
7	795	Torula QA23

Level of significance (Friedman): 5%

Rank.sum

	1(A)	**2(B)**	**3(C)**	**4(D)**	**5(E)**	**6(F)**	**7(G)**
1	27.27%	13.64%	18.18%	13.64%	0%	9.09%	18.18%
2	22.73%	13.64%	9.09%	9.09%	18.18%	13.64%	13.64%
3	13.64%	22.73%	13.64%	27.27%	4.55%	0%	18.18%
4	4.55%	9.09%	18.18%	18.18%	27.27%	9.09%	13.64%
5	13.64%	9.09%	13.64%	18.18%	13.64%	18.18%	13.64%
6	13.64%	22.73%	18.18%	4.55%	18.18%	13.64%	9.09%
7	4.55%	9.09%	9.09%	9.09%	18.18%	36.36%	13.64%
Rank sum	69	86	86	81	102	110	82
Mean rank	3.14EF	3.91f	3.91f	3.68F	4.64	5	3.73f
StDev.	2.01	2	2	1.76	1.73	2.14	2.05

Level of significance for the Mean Rank (LSD): A<5%; a<10%

LSD.rank.theory

	x
1	28.09

Signif.

	1	2	3	4	5	6	7
1		0.235	0.235	0.402	0.021*	0.004*	0.364
2	0.235		1	0.727	0.264	0.094	0.78
3	0.235	1		0.727	0.264	0.094	0.78
4	0.402	0.727	0.727		0.143	0.043*	0.944
5	0.021*	0.264	0.264	0.143		0.577	0.163
6	0.004*	0.094	0.094	0.043*	0.577		0.051
7	0.364	0.78	0.78	0.944	0.163	0.051	

OUTPUT TYPE NOT DEFINED: 15

Kommentarer(Q8)

Panelist	Remark
A0005	Väldigt svårt att skilja på proven. Jag kände ingen uttalad besk smak på något prov.
A0006	Blandning av sötma syrlighet med det beska smak ger helheten av detta dricka. Nummer 795 var best.
A0008	Mycket svårt att särskilja
A0013	Upplevde ingen som direkt besk.
A0014	948 158 och 842 kraftigaste beskan
A0015	547 har en mjuk honungs smak som för mig uppfattas blommig. 682 har en stickigare blommton
A0017	De 3 första har beska,därefter kan jag egentligen inte rangordna ärligt,de har jag bara lagt i raden
A0018	Lättare men ändå svårt. Snarlika i två grupper tror jag. 4 vänstra och 3 högra.
A0019	Ok
A0021	Inte så tydlig beska på den som hade mest beska men jag kunde känna en ton av den.
A0022	De faller in i 2grupper inom varje liknade smak

Rank blommighet(Q9)

Ranks

	1	2	3	4	5	6	7
anonymous_024mfddlra	4	2	1	6	3	7	5
anonymous_0lvc4rdmyi	7	4	3	2	5	1	6
anonymous_5vzpi45kpu	1	2	5	6	3	4	7
anonymous_8e4w1gknrq	1	7	6	3	5	2	4
anonymous_8xjz8ggc93	4	5	6	3	2	7	1
anonymous_944hefqece	1	7	6	2	4	3	5
anonymous_96ujwykndd	5	1	7	3	4	6	2
anonymous_9z65lftsip	5	7	4	3	2	1	6
anonymous_cagypnt5xb	6	2	5	7	1	4	3
anonymous_f1dk1i8o5m	1	7	4	2	3	5	6
anonymous_gzltvg7v71	6	3	2	4	7	1	5
anonymous_j3v0ihkodz	4	1	6	7	5	2	3
anonymous_l4rzlchg7j	1	5	4	7	6	2	3
anonymous_l5aw4q06m2	6	1	5	7	2	4	3
anonymous_mdekrgyxt8	5	7	4	3	6	1	2
anonymous_mhcbcahd59	1	6	4	2	3	5	7
anonymous_n9jxpjx3ul	7	6	4	3	1	2	5
anonymous_se1zk6nnp3	6	7	4	1	3	2	5
anonymous_t2x2ycc98c	5	1	6	3	7	4	2
anonymous_tmnbvk7n6g	1	3	6	2	5	4	7
anonymous_zgswxslmck	7	6	2	3	1	4	5
anonymous_zo9oy82idd	1	7	5	6	4	3	2

Sizes by ranks

	1	2	3	4	5	6	7
1	8	4	1	1	3	4	1
2	0	3	2	5	3	5	4
3	0	2	1	8	5	2	4

4	3	1	7	1	3	6	1
5	4	2	4	0	4	2	6
6	4	3	6	3	2	1	3
7	3	7	1	4	2	2	3
Total	22	22	22	22	22	22	22

Friedman test

	Friedman
chi-squared	4.75
df	6
p.value	0.576

	codes	image
1	210	A7
2	682	Torula DV10
3	158	Torula Exence 1
4	948	Torula ICV Okay
5	547	R2
6	842	R2226
7	795	Torula QA23

Level of significance (Friedman): 5%

Rank.sum

	1(A)	2(B)	3(C)	4(D)	5(E)	6(F)	7(G)
1	36.36%	18.18%	4.55%	4.55%	13.64%	18.18%	4.55%
2	0%	13.64%	9.09%	22.73%	13.64%	22.73%	18.18%
3	0%	9.09%	4.55%	36.36%	22.73%	9.09%	18.18%
4	13.64%	4.55%	31.82%	4.55%	13.64%	27.27%	4.55%
5	18.18%	9.09%	18.18%	0%	18.18%	9.09%	27.27%
6	18.18%	13.64%	27.27%	13.64%	9.09%	4.55%	13.64%
7	13.64%	31.82%	4.55%	18.18%	9.09%	9.09%	13.64%
Rank sum	85	97	99	85	82	74	94
Mean rank	3.86	4.41	4.5	3.86	3.73	3.36c	4.27
StDev.	2.38	2.42	1.54	2.01	1.86	1.87	1.86

Level of significance for the Mean Rank (LSD): A<5%; a<10%

LSD.rank.theory

	x
1	28.09

Signif.

	1	2	3	4	5	6	7
1		0.402	0.329	1	0.834	0.443	0.53
2	0.402		0.889	0.402	0.295	0.108	0.834
3	0.329	0.889		0.329	0.235	0.081	0.727
4	1	0.402	0.329		0.834	0.443	0.53
5	0.834	0.295	0.235	0.834		0.577	0.402
6	0.443	0.108	0.081	0.443	0.577		0.163
7	0.53	0.834	0.727	0.53	0.402	0.163	

OUTPUT TYPE NOT DEFINED: 15

Kommentarer(Q10)

Panelist	Remark
A0005	Inga kommentarer att komma med, förutom att det var näst intill omöjligt att riktigt rangordna dem.
A0006	Eftersmak i munnen lämnar fint känsla. Lukten som sprider sig vid drickande
A0008	Mycket lika
A0011	547, härlig får en tänka på skolavslutning, när Syrén blommar!
A0016	795 känns mest blommig mest karaktär
A0017	Mest blommig är det som är tydligast,annars kan jag inte urskilja så mycket blomning

A0018	Så snarlika
A0022	Den jag valde som mest blommig var för riktigt blomsmak

Q11(Q11) Alkoholsmak

Ranks

	1	2	3	4	5	6	7
anonymous_024mfddlra	2	6	1	5	3	4	7
anonymous_0lvc4rdmyi	3	1	5	4	7	6	2
anonymous_5vzpi45kpu	1	6	5	2	7	3	4
anonymous_8e4w1gknrq	4	5	7	6	2	3	1
anonymous_8xjz8ggc93	4	6	7	3	2	5	1
anonymous_944hefqece	2	6	1	5	3	4	7
anonymous_96ujwykndd	4	6	3	5	1	2	7
anonymous_9z65lftsip	4	6	3	5	7	1	2
anonymous_cagypnt5xb	5	6	2	3	7	4	1
anonymous_f1dk1i8o5m	2	6	4	1	3	5	7
anonymous_gzltvg7v71	1	4	2	6	7	3	5
anonymous_j3v0ihkodz	5	1	2	3	4	6	7
anonymous_l4rzlchg7j	1	4	6	3	7	5	2
anonymous_l5aw4q06m2	5	1	6	4	7	3	2
anonymous_mdekrgyxt8	7	4	6	5	3	1	2
anonymous_mhcbcahd59	2	6	4	1	3	5	7
anonymous_n9jxpjx3ul	6	4	5	3	1	2	7
anonymous_se1zk6nnp3	3	7	2	4	1	5	6
anonymous_t2x2ycc98c	4	2	6	3	7	5	1
anonymous_tmnbvk7n6g	4	5	3	2	7	6	1
anonymous_zgswxslmck	2	7	1	5	3	4	6
anonymous_zo9oy82idd	1	2	3	6	4	7	5

Sizes by ranks

	1	2	3	4	5	6	7
1	4	3	3	2	3	2	5
2	5	2	4	2	2	2	5
3	2	0	4	6	6	4	0
4	6	4	2	3	2	4	1
5	3	2	3	6	0	6	2
6	1	9	4	3	0	3	2
7	1	2	2	0	9	1	7
Total	22	22	22	22	22	22	22

Friedman test

	Friedman
chi-squared	5.12
df	6
p.value	0.528

	codes	image
1	210	A7
2	682	Torula DV10
3	158	Torula Exence 1
4	948	Torula ICV Okay
5	547	R2
6	842	R2226
7	795	Torula QA23

Level of significance (Friedman): 5%

Rank.sum

	1(A)	2(B)	3(C)	4(D)	5(E)	6(F)	7(G)
1	18.18%	13.64%	13.64%	9.09%	13.64%	9.09%	22.73%
2	22.73%	9.09%	18.18%	9.09%	9.09%	9.09%	22.73%
3	9.09%	0%	18.18%	27.27%	27.27%	18.18%	0%
4	27.27%	18.18%	9.09%	13.64%	9.09%	18.18%	4.55%
5	13.64%	9.09%	13.64%	27.27%	0%	27.27%	9.09%
6	4.55%	40.91%	18.18%	13.64%	0%	13.64%	9.09%
7	4.55%	9.09%	9.09%	0%	40.91%	4.55%	31.82%
Rank sum	72	101	84	84	96	89	90

Mean rank	3.27Be	4.59	3.82	3.82	4.36	4.05	4.09
StDev.	1.72	1.99	1.99	1.53	2.38	1.65	2.56

Level of significance for the Mean Rank (LSD): A<5%; a<10%

LSD.rank.theory

	x
1	28.09

Signif.

	1	2	3	4	5	6	7
1		0.043*	0.402	0.402	0.094	0.235	0.209
2	0.043*		0.235	0.235	0.727	0.402	0.443
3	0.402	0.235		1	0.402	0.727	0.675
4	0.402	0.235	1		0.402	0.727	0.675
5	0.094	0.727	0.402	0.402		0.625	0.675
6	0.235	0.402	0.727	0.727	0.625		0.944
7	0.209	0.443	0.675	0.675	0.675	0.944	

OUTPUT TYPE NOT DEFINED: 15

Kommentarer(Q12)

Panelist	Remark
A0005	Ytterligare ett svårt test. Ingen med uttalad alkoholsmak hittades.
A0006	Kännes vid drickande och luktande
A0007	682 & 795 för mkt alkoholsmak
A0008	Verkar vara ganska lika
A0012	Något stickningar smaker i vissa smakprov. Påminner mer om en lite söt och klibbig hostmedicin än en alkoholhaltig dryck. Gäller både konsistens och smak hos vissa
A0013	Pytte lite alkoholskak i alla,
A0015	Vissa smakar nästan lite jäst
A0017	Den med mest smak av alkohol tydlig,de övriga mindre/lika alkosmak
A0018	Små skillnader. Svårt igen. Bränner inte till i gommen.
A0022	De tre sista hade tydlig alkoholsmak

Q13(Q13) Overall smell intensity

Ranks

	1	2	3	4	5	6	7
anonymous_024mfddlra	3	5	2	1	4	7	6
anonymous_0lvc4rdmyi	3	5	7	4	1	2	6
anonymous_5vzpi45kpu	6	5	2	1	3	4	7
anonymous_8e4w1gknrq	6	1	5	3	7	4	2
anonymous_8xjz8ggc93	3	1	4	5	2	7	6
anonymous_944hefqece	4	5	2	6	1	3	7
anonymous_96ujwykndd	5	7	3	1	4	2	6
anonymous_9z65lftsip	2	5	1	6	7	3	4
anonymous_cagypnt5xb	5	6	4	1	7	3	2
anonymous_f1dk1i8o5m	4	1	6	3	7	5	2
anonymous_gzltvg7v71	3	4	1	2	7	5	6
anonymous_j3v0ihkodz	7	6	5	3	2	1	4
anonymous_l4rzlchg7j	3	6	2	7	5	1	4
anonymous_l5aw4q06m2	5	6	4	3	2	1	7
anonymous_mdekrgyxt8	7	4	2	3	6	5	1
anonymous_mhcbcahd59	2	6	5	1	3	7	4
anonymous_n9jxpjx3ul	2	1	5	4	7	6	3
anonymous_se1zk6nnp3	7	6	4	2	1	3	5
anonymous_t2x2ycc98c	5	6	3	2	7	4	1
anonymous_tmnbvk7n6g	3	2	5	7	4	1	6
anonymous_zgswxslmck	4	6	1	5	3	2	7
anonymous_zo9oy82idd	5	3	1	2	7	6	4

Sizes by ranks

	1	2	3	4	5	6	7
1	0	4	4	5	3	4	2
2	3	1	5	4	3	3	3
3	6	1	2	5	3	4	1
4	3	2	4	2	3	3	5
5	5	5	5	2	1	3	1
6	2	8	1	2	1	2	6
7	3	1	1	2	8	3	4
Total	22	22	22	22	22	22	22

Friedman test

	Friedman
chi-squared	8.08
df	6
p.value	0.232

Level of significance (Friedman): 5%

Rank.sum

	1(A)	2(B)	3(C)	4(D)	5(E)	6(F)	7(G)
1	0%	18.18%	18.18%	22.73%	13.64%	18.18%	9.09%
2	13.64%	4.55%	22.73%	18.18%	13.64%	13.64%	13.64%
3	27.27%	4.55%	9.09%	22.73%	13.64%	18.18%	4.55%
4	13.64%	9.09%	18.18%	9.09%	13.64%	13.64%	22.73%
5	22.73%	22.73%	22.73%	9.09%	4.55%	13.64%	4.55%
6	9.09%	36.36%	4.55%	9.09%	4.55%	9.09%	27.27%
7	13.64%	4.55%	4.55%	9.09%	36.36%	13.64%	18.18%
Rank sum	94	97	74	72	97	82	100
Mean rank	4.27	4.41	3.36g	3.27beg	4.41	3.73	4.55
StDev.	1.64	1.99	1.79	1.98	2.34	2.05	2.02

Level of significance for the Mean Rank (LSD): A<5%; a<10%

LSD.rank.theory

	x
1	28.09

Signif.

	1	2	3	4	5	6	7
1		0.834	0.163	0.125	0.834	0.402	0.675
2	0.834		0.108	0.081	1	0.295	0.834
3	0.163	0.108		0.889	0.108	0.577	0.07
4	0.125	0.081	0.889		0.081	0.485	0.051
5	0.834	1	0.108	0.081		0.295	0.834
6	0.402	0.295	0.577	0.485	0.295		0.209
7	0.675	0.834	0.07	0.051	0.834	0.209	

OUTPUT TYPE NOT DEFINED: 15

Komentarer(Q14)

1

Panelist	Remark
A0001	Stark
A0008	Klar dryck ,
A0012	Vitt vin
A0013	Söt Honungs doft .
A0014	Fläder
A0015	Stark lite söt doft
A0022	Jäst fruktdoft

2

Panelist	Remark

A0001	Sötare
A0008	Klar lite gulare färg, äppeldoft
A0012	Luktar vitt vin, svag doft
A0013	Ej lika söt i smak men ger lite i doft
A0014	Parfym
A0015	Lakritstoner
A0017	Luktat sött
A0022	Behaglig doft

3

Panelist	Remark
A0001	Stark
A0008	Grumlig, lätt jästdoft
A0012	Mild doft, luktar inte så mycket
A0014	Krydda
A0015	Svagt äpple
A0022	Minst doft

4

Panelist	Remark
A0001	Kval,mig
A0008	Halvklar, lätt jäsdoft, lätt söt smak
A0012	LuktAr som cider brukar lukta
A0014	Äppel
A0015	Äpple svagt
A0022	Inget upphetsande

5

Panelist	Remark
A0001	Mer alkoholdoft
A0007	Snarlik 842 & 158
A0008	Klart genomskinlig, lätt doft , sötsur
A0012	Svag doft av vitt vin
A0013	Svag men ändå bäst utan att sticka ut
A0014	Träd
A0015	Strakt mogen
A0017	Luktar mycket vinägerlikt
A0022	Tydlig fruktdoft

6

Panelist	Remark
A0001	Lätt doft
A0008	Aning grumsig, lätt doft halvt surast
A0012	Mild lukt
A0015	Sött äpple
A0016	Alla ligger väldigt nära men 842 känns mest doft i
A0022	Tydlig bra doft

7

Panelist	Remark
A0001	Lätt doft
A0008	Klar dryck , äppeldoft , syrlig smak
A0012	Luktar medicin, väldigt från lukt
A0013	Tunn men ändå behaglig
A0014	Parfym
A0015	Svagt friskt
A0018	Färgen tydliggör annan doft avvikande
A0022	Alkoholdoft

Q15(Q15) Overall liking

Ranks

	1	2	3	4	5	6	7
anonymous_024mfddlra	2	5	1	4	3	6	7
anonymous_0lvc4rdmyi	2	1	7	4	6	5	3

anonymous_5vzpi45kpu	5	7	1	6	3	4	2
anonymous_8e4w1gknrq	7	3	4	6	2	5	1
anonymous_8xjz8ggc93	3	6	5	2	1	7	4
anonymous_944hefqece	5	6	2	7	3	4	1
anonymous_96ujwykndd	5	2	6	3	4	7	1
anonymous_9z65lftsip	5	7	4	6	3	1	2
anonymous_cagypnt5xb	7	1	6	5	3	2	4
anonymous_f1dk1i8o5m	4	6	2	3	1	5	7
anonymous_gzltvg7v71	7	3	6	5	1	4	2
anonymous_j3v0ihkodz	2	3	4	5	7	6	1
anonymous_l4rzlchg7j	1	6	5	2	3	4	7
anonymous_l5aw4q06m2	5	6	4	3	2	1	7
anonymous_mdekrgyxt8	6	4	7	3	2	1	5
anonymous_mhcbcahd59	1	2	5	3	4	6	7
anonymous_n9jxpjx3ul	7	3	6	4	1	5	2
anonymous_se1zk6nnp3	3	6	4	2	1	5	7
anonymous_t2x2ycc98c	4	2	3	7	1	6	5
anonymous_tmnbvk7n6g	3	7	5	6	4	1	2
anonymous_zgswxslmck	1	5	7	2	6	3	4
anonymous_zo9oy82idd	5	4	2	6	1	3	7

Sizes by ranks

	1	**2**	**3**	**4**	**5**	**6**	**7**
1	3	2	2	0	7	4	4
2	3	3	3	4	3	1	5
3	3	4	1	5	6	2	1
4	2	2	5	3	3	4	3
5	6	2	4	3	0	5	2
6	1	6	4	5	2	4	0
7	4	3	3	2	1	2	7
Total	22	22	22	22	22	22	22

Friedman test

	Friedman
chi-squared	8.16
df	6
p.value	0.226

	codes	**image**
1	210	A7
2	682	Torula DV10
3	158	Torula Exence 1
4	948	Torula ICV Okay
5	547	R2
6	842	R2226
7	795	Torula QA23

Level of significance (Friedman): 5%

Rank.sum

	1(A)	**2(B)**	**3(C)**	**4(D)**	**5(E)**	**6(F)**	**7(G)**
1	13.64%	9.09%	9.09%	0%	31.82%	18.18%	18.18%
2	13.64%	13.64%	13.64%	18.18%	13.64%	4.55%	22.73%
3	13.64%	18.18%	4.55%	22.73%	27.27%	9.09%	4.55%
4	9.09%	9.09%	22.73%	13.64%	13.64%	18.18%	13.64%
5	27.27%	9.09%	18.18%	13.64%	0%	22.73%	9.09%
6	4.55%	27.27%	18.18%	22.73%	9.09%	18.18%	0%
7	18.18%	13.64%	13.64%	9.09%	4.55%	9.09%	31.82%
Rank sum	90	95	96	94	62	91	88
Mean rank	4.09	4.32	4.36	4.27	2.82aBCDFg	4.14	4
StDev.	2.04	2.01	1.89	1.7	1.79	1.96	2.41

Level of significance for the Mean Rank (LSD): A<5%; a<10%

LSD.rank.theory

	x

1	28.09

Signif.

	1	2	3	4	5	6	7
1		0.727	0.675	0.78	0.051	0.944	0.889
2	0.727		0.944	0.944	0.021*	0.78	0.625
3	0.675	0.944		0.889	0.018*	0.727	0.577
4	0.78	0.944	0.889		0.026*	0.834	0.675
5	0.051	0.021*	0.018*	0.026*		0.043*	0.07
6	0.944	0.78	0.727	0.834	0.043*		0.834
7	0.889	0.625	0.577	0.675	0.07	0.834	

OUTPUT TYPE NOT DEFINED: 15

Q16(Q16)

1

Panelist	Remark
A0002	Alltför söt
A0008	Bra
A0009	Den enda som föll mig i smaken.
A0011	Frisk sommar dag
A0012	Alldeles för söt
A0013	För söt
A0015	Lite bubblig men för sträv smak
A0022	Bra smak

2

Panelist	Remark
A0002	Smakar äpple, men lite väl besk
A0007	För mkt alkoholsmak
A0008	Bra
A0011	Mer intensiv både på gott och ont.
A0012	God
A0015	Söt men halsbränna varning
A0018	Klart är bra
A0022	Medelklass

3

Panelist	Remark
A0008	Bättre
A0011	Trevlig aromatisksmak
A0012	Också för besk och söt på samma gång
A0015	Mild och snäll
A0022	Nej

4

Panelist	Remark
A0002	God smak av äpple
A0008	Fin färg
A0011	Dessertenvin över det, passar nog bra till en ostbricka
A0012	Uppskattar den milda smaken
A0015	Mest stark inte så mycket mer smak
A0022	Bra smak

5

Panelist	Remark
A0002	Balanserad smak av sötma och beska
A0008	God
A0011	En friskhet i det söta.
A0012	Uppskattar inte alls. Brännande och söt på samma gång
A0015	Jäst
A0017	Sur
A0018	Grumsigt är inte bra

A0022	Inget upphetsande

6

Panelist	Remark
A0002	Sötbesk
A0008	God
A0011	En kryddighet.
A0012	Något för söt för min smak
A0015	Ju värmer ju starkare smak och intensivare
A0022	Neutral

7

Panelist	Remark
A0007	För mkt alkoholsmak
A0008	Bra
A0011	Vanilj och honung.
A0012	Uppskattar inte alls. Besk och smakar som hostmedicin pga den sötbeska smaken
A0013	Bäst balanserad mellan syrlighet sötma och alkohol samt en klar och fin färg.
A0015	Söt men mild
A0016	Bäst smak
A0017	Mest drickbar
A0018	Ju klarare vätska desto bättre
A0022	Lagom sötma bra smak