

Various Factors Affect Product Properties in Apple Cider Production

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Abstract

Different parameters in cider processing were evaluated using different cultivars of Norwegian-grown table apples measuring the quality of cider. Seven different apple cultivars were mixed into four different apple juice mixtures. In this experiment, we evaluated the maturation of the apples along with commercial cider yeast and spontaneous alcoholic fermentation. Other parameters were fermentation temperature and filtration along with content of polyphenols, organic acids and volatile compounds that was analysed as an effect of the fermentation process. Succinic acid was the major organic acid in apples and ciders. The different apple juice mixtures did not reveal pyruvic and acetic acids but they appeared in relatively high amount in the ciders. The level of citric acid increased from apple to cider. Chlorogenic acid was the major polyphenolic compound found from 13-109 mg L⁻¹ in the apple juice mixtures and between 27-200 mg L⁻¹ in the ciders. The higher alcohol 3-methyl-1-butanol appeared in relatively large amounts in all the ciders (91-166 mg L⁻¹). The average content of acetaldehyde increased during the fermentation process, from apple juice mixtures 2.75 mg L⁻¹ and 14.65 mg L⁻¹ in the ciders. The content also increased for ethyl acetate with levels at 0.1 mg L⁻¹ in the apple juice mixture and 20 mg L⁻¹ in the cider. In the sensory evaluation experiment, the ciders produced from the apple cultivars Aroma, Gravenstein and Summerred got higher scores in fruitiness and complexity compared to the other apple juice mixtures.

Keywords: Cider; Apples; Alcoholic Fermentation; Polyphenols; Aromatic Compounds

1 Introduction

Apple (*Malus* sp.) originates from Central Asia and has been cultivated thousands of years mostly in cooler climates in Asia and Europe. In Scandinavia (nearly up to the Arctic Circle), apple has been known for more than 1000 years. The Nordic climate with long days and cool nights during summer gives a fresh acidic-sweet distinct aroma to the apples. In addition, the Nordic climate results in slow maturation of fruits in this region compared to apples grown

in the South of Europe (Redalen, 1991).

Most of the apples in Norway are grown for fresh consumption, but there are long traditions for using apples in various dishes in addition to juice and cider making. Since the end of 1700 in the west coast area of Norway, Hardanger, cider was commercially produced. The cultivars used in this cider production are usually high in acids and low in polyphenols. Phenolic compounds in apples are found to be the most important contributors to cider qualities, such as complex taste and body, astringency and keepability (Alonso-

Salces et al., 2001; Leforestier et al., 2015; Sanoner, Guyot, Marnet, Molle & Drilleau, 1999; Verdu et al., 2013). Previously, rowanberries were used as an ingredient in producing apple wine and cider due to lack of tannins in apples (Erken, 1932). Today cider producers are experimenting with mixing different dessert apples, Crab apples and cider apples along with hops to give cider more tannins and taste that is more complex. Importation of cider apple trees for cultivation in Norway is of great interest for apple growers, because cider apples have a higher content of polyphenols than dessert apples (Bamforth, 2004).

According to Tsao, Yang, Xie, Sockovie and Khanizadeh (2005), antioxidant activity is positively correlated with the total phenolic concentration in apples, whereas from in vitro studies flavan-3-ols/procyanidins were found to be the most important contributors to antioxidant activity in apples. In a study by Sanoner et al. (1999), they found the most important individual polyphenols in cider apples to be the procyanidin B2 and (-)-epicatechin, though the proportion of the polyphenol classes varied greatly among apple cultivars.

The selection of proper yeast is important for developing good sensory properties in the final product. The use of wild yeast fermentation (spontaneous fermentation) is less predictable but might give a product of more distinguished aromatic profile. However, there will always be a risk of microbiological spoilage and off-flavours. Traditional cider makers often add sulphite to prevent contamination when using wild yeast. Inoculating with selected yeast strains isolated from a cider of good quality might be a good alternative to wild yeast fermentation. Yeast strains of *Saccharomyces cerevisiae*, *S. bayanus* or *S. bayanus* var. *uvarum* will also produce ciders of good quality. The taste of a cider is a consequence of many different biochemical interactions that occur as the result of the selection of apples as a raw material and the multiple steps in the fermentation process. Cider styles vary between countries and regions. Ciders can be hazy or clear, still or carbonated, colourless to brownish, pasteurized or unpasteurized (Bamforth, 2004).

Today, there is an increased interest for renew-

ing the old traditions of using apples as raw material for juice, cider and apple spirit. Of particular focus in Norway, is the use of apple cultivars with a higher content of polyphenolic compounds or the use of old cultivars or other ingredients for increasing the content of polyphenols in the product. New regulations in Norway (since 2015), allow farmers to produce and distribute wine and cider with alcohol content up to 22 % ABV directly from the farm. Four different juice mixtures from seven apple cultivars produced different ciders in this experiment. In order to evaluate the influence of various processing variables on cider quality fermentation methods, temperature and time, yeast strain, filtration and addition of hops were evaluated.

2 Materials and Methods

2.1 Apples

Seven apple cultivars were included in this experiment of product development using Norwegian grown apples for cider production. The apple cultivars used were Sunrise, Discovery, Aroma, Gravenstein, Summerred, Jonagold and Torstein. The apples were harvested in the fruit orchard at the Norwegian University of Life Sciences (NMBU) at Aas (10° 77' E, 59° 67' N) at harvest maturity stage in September and October 2015. All the apple cultivars were stored at +3 °C and 85 % RH in normal atmosphere before juicing. The cultivars were stored between 19 and 65 days before juicing, depending on the harvest date of each cultivar. The different ciders were processed and analysed at the Norwegian University of Life Sciences (NMBU), Ås, Norway, 2015. For details of the parameters, see Table 1.

2.2 Analyses measuring apple fruit quality

For evaluation of maturity stage and general quality on the various apple cultivars, ground colour, firmness, starch, titratable acidity and soluble solids were measured on five apples from each cultivar.

Analyses were evaluated in triplicate.

Colour

For evaluation of ground colour, a colour chart based on the cultivar Golden Delicious was used, ranging from 1–9, where 1 is green and 9 is yellow. The numbers indicate the degradation of chlorophyll in the apple skin, which is degrading during maturity, revealing the presence of carotenoids (yellow) when the apples are mature.

Firmness

A penetrometer (Fruit pressure tester FT 327, Italy) was used for measuring fruit firmness. Apples were peeled with a fruit peeler at three different places around the apple equator to remove the apple skin before measuring, resulting in 3 x 5 measurements per apple cultivar. Results are presented as kg/cm².

Starch

The starch content in the apples was tested, the apples were cut in half and the surfaces of one-half were soaked in potassium iodine for approximately 10 seconds. The apples were compared to a colour chart with a range from 1-9, where 1 indicates 100 % presence of starch degraded to 9 (no starch).

Acidity

Titrateable acidity was measured using 10 mL of apple juice diluted with distilled water, using an automatic titrator (Titrator 716 DMS Titranox, Metrohm, Switzerland) using 0.1M NaOH and phenolphthalein as indicator. Results are expressed as % TA. The calculation was based on the malic acid equivalent 67, and is regarded as synonymous to the % of malic acid in the sample.

Total Soluble solids

For total soluble solids (TSS) measurements, a digital refractometer (ATAGO, USA) was used for measurements. Results are expressed as % TSS in the juice and referred to as Brix degree (°B).

2.3 Production of apple cider

Various processing parameters were included in the production of apple ciders.

Pressing of apple juice

All the apples were washed in cold water then crushed in a Speidel fruit mill (Speidel, Germany) and pressed in a 20 L Speidel hydro press (Speidel, Germany). Each press lasted for approximately 15 minutes. All the equipment was cleaned between crushing and pressing of each cultivar. The different apple juices were mixed according to information in Table 1.

Apple juice mixture and selection of apple cultivars

Cultivars Sunrise and Discovery are apples that mature early. They were pressed after 19 days of storage (apple juice blend A) and 62 days of storage (apple juice blend B). Cultivars Aroma, Gravenstein and Summerred are apples that mature later. They were pressed after 19 days of storage (apple juice blend C). The apple juice blend D contained the cultivars Aroma, Gravenstein, Summerred, Jonagold and Torstein and were pressed after 35 days of storage. The selection was done in order to include early and late cultivars in addition to storage time before pressing.

Fermentation and yeast addition

Either the apple juices were inoculated with cider yeast (M02 from Mangrove Jack's – *Saccharomyces bayanus*) or not (spontaneous fermentation). Fermentation temperatures were 10 °C. For some batches, fermentation was started at 20 °C for 2 days and then continued at 10 °C. Fermenting vats were either 5 L or 30 L and experiments were done in triplicate. The ciders were fermented until dryness. Fermentation rate slowed down with time, and when no further reduction in % TSS was observed the end point was between 4 and 5 °Brix.

Sugar addition

Due to the relatively low levels of TSS in the apples, we chose to increase the sugar level by adding white table sugar to the apple juice mixtures, except cider style 6, to start the fermentation. From an average of 11.7 °Brix the level increased by adding approximately 18 g sugar L⁻¹ to reach 13.5 °Brix (Table 1).

Addition of hops

Dessert apples are often low in tannin and astringency. In an attempt to add more aroma and body to the cider, hops were added. The hops, Amarillo and Cascade, were added in two different batches of cider in a quantity of 1 g L⁻¹, stored at 20 °C for 3 days before bottling and maturation.

Filtering

Some of the cider batches were filtered using a Colombo[®] 6-INOX system (Rover Pompe, Italy). Filters used were Rover 4: 10 µm and Rover 12: 1,5 µm. Filtering was applied as a process parameter to evaluate the effect on clarity and taste profile.

Carbonation and bottling

At the end of fermentation, the ciders were chilled down to 1-3 °C, extracted from the lees, bottled with addition of external CO₂, using beer gun equipment before capping and bottle pasteurization (66 °C for 30 minutes).

2.4 Chemical analyses

Polyphenols

Polyphenols were analysed according to Guyot et al., although we omitted the thiolysis step. Only the native polyphenols were analysed (Guyot, Marnet, Sanoner & Drilleau, 2001). Phenolic compounds were identified by HPLC on the basis of their retention time and their characteristic fragmentation pattern in comparison with available standards. The polyphenol standard solutions were (+)-catechin, (-)-epicatechin, procyanidin B1, procyanidin B2, phloretin, phloridzin,

chlorogenic acid, caffeic acid, rutin and quercetin.

Total phenolic compounds (TP)

Total phenols were analysed according to the Folin Ciocalteu method modified as described by (Volden et al., 2008). Quantifications were obtained by reporting the absorbance at 765 nm to a calibration curve of gallic acid and are expressed as mg equiv. GAE 100 mL⁻¹ of sample.

Nitrogen

Total nitrogen was analysed by the Kjeldahl method, according to IDF 2001, and expressed as mg N L⁻¹.

Free amino acids

Free amino acids were analysed by HPLC, based on a method by (Bütikofer & Ardö, 1999). The following standards were used for identification of the amino acids: L-aspartic acid, L-glutamic acid, L-asparagine, L-serine, L-glutamine, L-histidine, Glycin, L-threonin, L-citrulline, L-arginine, L-alanine, GABA, L-tyrosine, L-valin, L-metionin, L-norvalin, L-isoleucin, L-phenylalanin, L-tryptophane, L-leucin, L-ornitin and L-lysin. Only a limited number of amino acids are presented and expressed as mg L⁻¹.

Organic acids

Organic acids were analysed using HPLC as described by (Moe, Porcellato & Skeie, 2013). Organic acids for standard solutions were citric, pyruvic, succinic, lactic and acetic acids (all from Sigma). Malic acid was analysed in the apples by titration method but was not analysed in the ciders.

Volatile compounds

Volatile compounds were analysed using a head-space gas chromatography system (HSGC) according to (Gronnevik, Falstad & Narvhus, 2011). Peaks were externally identified and quantified using standard solutions of the following compounds: acetaldehyde, 2-butanone, ethyl

acetate, 2-methyl-1propanol, 2-methyl-butanal, 3-methyl-butanal, 3-methyl-1-butanol, 2-methyl-1-butanol, 2-methyl-1propanal, diacetyl, 1-butanol, 2-butanol, acetoin, iso-butylacetate, dimethylsulfide, acetone, 2,3pentadion. 2-hexanol, hexanal, isoamyl acetate, ethyl hexanoate, 3-carene, R-(+)-limonene, ethyl heptanoate, ethyl octanoate, b-citronellol, ethyl nonanoate, ethyl decanoate, phenylethyl alcohol, ethanol, 1-propanol.

Sensory evaluation

A semi-trained sensory panel (20 judges) evaluated the drinking quality of the ciders. The panel evaluated haze, aroma, sweetness, acidity, bitterness, fruitiness, complexity and aftertaste using a scale from 1-10, where 1 indicated low level and 10 indicated high level of the individual property. For calibration of the judging panel, the commercially produced “Somersby dry apple cider” was used as a reference. The cider samples were served chilled (6 °C) in a quantity of 40 mL per sample.

Statistical analyses

Statistical analyses were performed with Minitab statistical software version 17 (Minitab Ltd., UK). One-way analyses of variance (ANOVA) were performed on the experimental data. For Principal Component Analyses (PCA), the statistical programme R was performed on the effect on cider quality.

3 Results and Discussions

The maturity of apples at the time of harvest might influence yield as well as chemical composition of the apple juice. If the apples need to be stored before pressing, time of storage and various storage conditions such as temperature, atmosphere, humidity and light will also affect the fruits (Børve & Vangdal, 2009). Apples harvested for this experiment were stored for 19-65 days, depending on cultivar and harvesting time, at +3 °C at 85 % RH and normal atmosphere until pressing. This was due to the differences in maturity time of the different cultivars in addition to evaluate the effect of apple storage on

cider quality.

Apple firmness declined during storage while no significant effect appeared on the other physiological properties. The apple cultivars Torstein and Jonagold were firmest at the time of pressing with 9.3 and 8.5 kg/cm², respectively. For the less firm qualities, with firmness of 4-5 kg/cm², there were problems during pressing, with apple pulp packed up in the press. This problem was especially severe for the cultivars Sunrise and Summerred that had been stored for 59 and 42 days after harvest, respectively, before pressing. The content of soluble solids varied between cultivars. We found the highest level in the cultivar Jonagold (13.9 °B) and the lowest in Summerred (10.9/10.5 °B) (Table 2). A high portion of the soluble solid content in apples is sugar, and cultivars grown in northern countries, usually have lower content of soluble solids and higher content of titratable acidity than cultivars grown further south. (Jolicoeur, 2013) indicated ideal acidity and specific gravity levels in apple juice for cider production to be 5-6.5 g L⁻¹ (malic acid) and 1060-1075 (SG) (14.9-18.2 °B). Titratable acidity in our apples was between 0.57 and 0.84 %. A higher acidity level is crucial for the taste and freshness of the product and can be important to balance the sugar level. We found no effect of storage on the level of fermentable sugars and acidity in the apple cultivars in this experiment. Due to the relatively low levels of soluble solids in the apples, the sugar level was increased in most of the apple juice mixtures before the start of the fermentation process by adding white table sugar. The ciders fermented until dryness. Fermenting at 20 °C using M02 cider yeast finished within 10 days. When fermentation started at 20 °C for a couple of days and continued at 10 °C, the fermentation time increased to 13 days. Fermentation at lower temperatures resulted in longer fermentation times, 24-29 days when using M02 yeast and 39 and 56 days for the spontaneously fermented ciders (Table 1).

In apples, the content of nitrogen is affected by cultivar, soil, fertilization, climatic conditions and the age of the apple trees (Milosevic, Milosevic & Mladenovic, 2019; Planchon, Lateur, Dupont & Lognay, 2004). To prevent fermentation from stopping, a sufficiently high amount of nitrogen is necessary (Lea, 2015). Lea recom-

mended a nitrogen level of approximately 100 mg N L⁻¹. If the nitrogen content is too high, fermentation might continue even after most of the fermentable sugars are used (Jolicoeur, 2013). Jolicoeur defined typical values for cider; 50 mg N L⁻¹ is regarded as low and fermentation might be incomplete, 80-120 mg N L⁻¹ is the range for most cider apple juices, 120-150 mg N L⁻¹ is regarded as rich and 300 mg N L⁻¹ is regarded to be high and unsuitable for cider production. Unstable yeast growth might lead to the development of undesirable aromatic components giving the cider an unpleasant taste. In order to get a smooth fermentation process, mixing of apple juices from different cultivars is possible to obtain a good mixture. By blending our apple juice, we obtained nitrogen content between 138-166 mg N L⁻¹ (Table 3). Using Jolicoeur's definition, our apple juice mixtures were all rich in nitrogen. The most important amino acids, asparagine, glutamine and aspartic acid, accounted for 85-95 % of the total amino acids. L-asparagine accounted for about half of the amino acids. Content of amino acids decreased during fermentation (Table 3), indicating their importance as nutrients for the yeast. Alberti et al. (2016) also reported a significant decrease in the content of most amino acids during cider fermentation.

3.1 Organic acids

Citric and succinic acids were found in all apple juice mixtures while pyruvic, lactic and acetic acids were not detected (Table 3). The sum of citric and succinic acids was 1033 mg L⁻¹ for apple juice blend A, 1178 mg L⁻¹ for blend B, 1462 mg L⁻¹ for blend C and 1616 mg L⁻¹ for blend D. In the ciders, styles 1, 2 and 3, from blend A and B, together with style 4 from blend C, appeared more bitter and acidic and lower in sweetness than the other ciders (Figure 2).

After the fermentation process, pyruvic and acetic acids were present in all the cider samples. Content of succinic acid increased during fermentation for all the ciders except styles 2 and 6 which were spontaneously fermented. Lactic acid did not appear in the apple juice but was present in many of the ciders. In the malo-lactic fermentation, the yeast is able to degrade malic acid to

ethanol, amyl alcohol, succinic acid, lactic acid and isobutanol with help from CO₂. The malo-lactic step is often desired when using dessert apples in cider production, due to their high acidity (Jolicoeur, 2013). The sensory evaluation of acidic taste did not correspond to measured acidity, indicating the importance of the malo-lactic transformation in giving a less acidic feeling.

3.2 Polyphenolic compounds

Content of phenolic compounds varied between the different apple juice mixtures (Table 4). The levels of the various polyphenols were lower than reported by other researchers (Kahle, Kraus & Richling, 2005; Wojdylo, Oszmianski & Laskowski, 2008). We also found a variation in the single components in the process from apple juice to cider. This is comparable to what was reported by (Laaksonen, Kuldjarv, Paalme, Virkki & Yang, 2017) but slightly different to results from (Ye, Yue & Yuan, 2014) who found decreases in most of the polyphenols in the apple juice to cider. Chlorogenic acid was the most abundant polyphenol in the apple juice mixtures, followed by procyanidin B2 and procyanidin B1, (+)-catechin and (-)-epicatechin. We found increases in caffeic acid during fermentation while for chlorogenic acid the results were more variable. This is in contrast to observations from Alberti et al. (Alberti et al., 2016) who observed decreases in both these components in ciders compared to apple juice. Quercetin and phloretin did not appear in the apple juices and rutin only in one sample. Phloretin appeared in most of the ciders. The highest level was observed in cider style 6. Quercetin and rutin were detected in all the ciders and in much higher quantity than the apple juice mixture they were made from, indicating a metabolization of rutin and quercetin during the fermentation process. We found a decrease in TP (Folin) from raw material to cider. For the single polyphenols no such change was observed (Table 4). On average, the TP content in apples and ciders from apple juice blend A and B was higher than in apples and ciders from blend C and D. In the sensory evaluation, cider styles 1, 2 and 3 from blend A and B were also evaluated to be more bitter than the

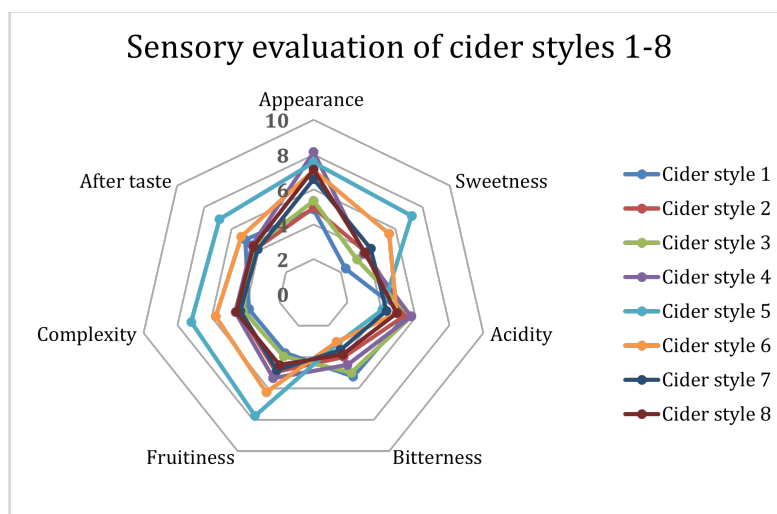


Figure 1: Sensory evaluation of cider styles 1-8. Sensory attributes were appearance, sweetness, acidity, bitterness, fruitiness, complexity and aftertaste

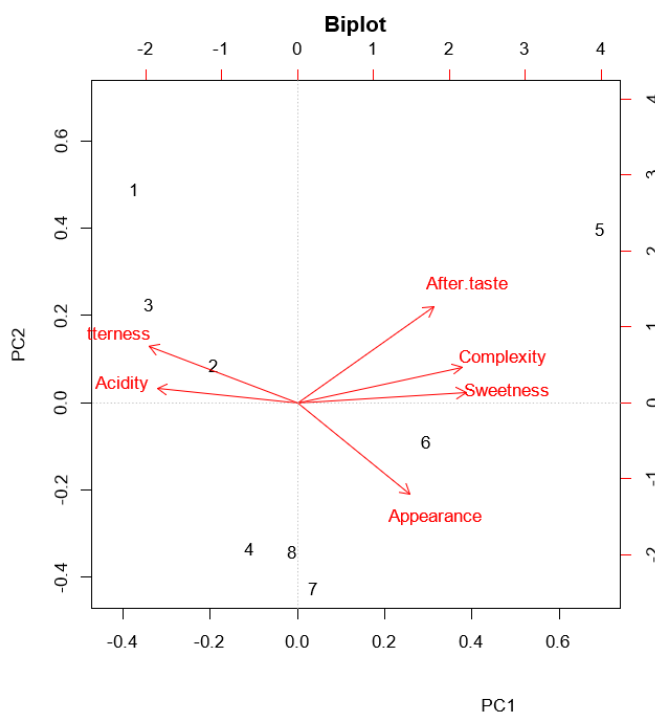


Figure 2: Biplot of cider styles 1-8 and sensory properties

Table 1: Parameters in cider processing

Cider style	Apple juice blend	Batch size (L)	Yeast type	Hop Hop	Soluble solids (°B)	Temp (°C)	Fermentation (days)	Filter Filter
1	A	5	M02		13.5*	20	10	-
2	A	30	Sp. ferm.		13.5*	10	56	4
3	B	5	M02		13.5*	10	24	4
4	C	5	M02		13.5*	20-10	13	-
5	C	30	M02	Am	13.5*	20-10	13	4
6	C	5	Sp. ferm.		10.6	10	39	4
7	C	5	M02		13.5*	10	24	12
8	D	30	M02	Cas	13.5*	10	29	4

Apple juice mixtures:

A: 'Sunrise' 50 % and 'Discovery' 50 %, stored 19 days before pressing

B: 'Sunrise' 50 % and 'Discovery' 50 %, stored 65 days before pressing

C: 'Aroma' 40 %. 'Gravenstein' 20 %. 'Summerred' 40 %, stored 19 days before pressing

D: 'Torstein' 15 %. 'Jonagold' 15 %. 'Aroma' 20 %. 'Summerred' 20 %. 'Gravenstein' 30 %, stored 35 days before pressing

*: adjustment of % TSS by sugar addition

For fermentation, various yeasts were added

M02 – Cider yeast (Mangrove Jack's. UK)

Spontaneous fermentation – no yeast addition

Hops: Am: Amarillo. Cas: Cascade

Table 2: Fruit quality at two maturity stages after storage of the apple cultivars Sunrise, Discovery, Aroma, Gravenstein, Summerred, Jonagold and Torstein at the time of juicing.

Apple cultivar	Firmness	Ground colour	Starch content	Soluble solids	Titrateable acidity	Nitrogen	TP
	kg/cm ² ± STD	1-9	1-10	°B	% TA ± STD	mg N L ⁻¹ ± STD	GAE 100mL ⁻¹ ± STD
Sunrise ^a	7.26 ± 0.42	6.7	7.9	12.1	0.58	220 ± 6.1	35.3 ± 5.63
Sunrise ^c	5.37 ± 0.18	7.9	10.0	12.0	0.57 ± 0.02		27.1 ± 2.07
Discovery ^a	8.16 ± 0.33	8.0	10.0	11.8	0.82	65 ± 1.02	81.5 ± 0.13
Discovery ^c	6.07 ± 0.94	8.0	10.0	11.7	0.61 ± 0.05		101.7 ± 0.60
Summerred ^a	5.37 ± 0.33	7.3	9.7	10.9	0.82 ± 0.01	329 ± 18.2	15.0 ± 0.31
Summerred ^b	4.54 ± 0.11	7.4	10.0	10.5	0.77 ± 0.05		12.1 ± 0.15
Aroma ^a	6.19 ± 0.02	7.1	9.4	11.9	0.77 ± 0.01	125 ± 14.4	51.9 ± 0.94
Aroma ^b	5.68 ± 0.33	6.9	10.0	11.2	0.80 ± 0.01		11.2 ± 0.91
Gravenstein ^a	6.40 ± 0.03	6.2	9.9	11.5	0.68 ± 0.02	98 ± 12.8	49.2 ± 2.15
Gravenstein ^b	5.12 ± 0.26	7.2	9.9	11.5	0.66 ± 0.04		44.8 ± 1.56
Jonagold ^b	9.31 ± 0.19	7.3	9.7	13.9	0.77 ± 0.04	91 ± 13.1	19.4 ± 0.47
Torstein ^b	8.54 ± 0.11	6.5	6.9	12.4	0.84 ± 0.02	212 ± 15.2	113.3 ± 1.25

^{abc} Days of storage of apples before pressing a: 19 days, b: 35 days, c: 62 days

TP: total phenols (Folin Ciocalteu)

Table 3: Content of organic acids, amino acids and total nitrogen from raw material to cider.

Apple juice blend	Cider style	Citric acid mg L ⁻¹	Pyruvic acid mg L ⁻¹	Succinic acid mg L ⁻¹	Lactic acid mg L ⁻¹	Acetic acid mg L ⁻¹	Tot N mg N L ⁻¹	L-asp acid mg L ⁻¹	L-glut mg L ⁻¹	L-asp mg L ⁻¹
A		28	nd	1005	nd	nd	138±3.3	87.70	57.50	176.5
	1	37	75	1792	nd	145		nd	nd	nd
	2	120	70	1321	55	182		nd	nd	nd
B		47	nd	1131	nd	nd	149±2.7			
	3	60	60	1563	22	98				
C		56	nd	1370	nd	nd	151±15.1	141.00	49.90	370.7
	4	100	46	1831	nd	112		nd	0.44	nd
	5	88	75	1818	nd	137		0.27	0.73	0.16
	6	76	26	1233	24	89		nd	nd	nd
	7	77	38	1488	nd	208				
D		71	nd	1545	nd	nd	166±9.7	147.60	62.20	424.6
	8	108	49	2112	41	110		nd	nd	nd

Table 4: Content of polyphenols from raw material to cider

AB	C	CT	EC	B1	B2	CA	CAF	PLZ	XPL	QUE	RU	TOT	TP		
		mg L ⁻¹	mg L ⁻¹	mg L ⁻¹	mg L ⁻¹	mg L ⁻¹	mg L ⁻¹	mg L ⁻¹	mg L ⁻¹	mg L ⁻¹	mg L ⁻¹	mg L ⁻¹	mg L ⁻¹	GAE mL ⁻¹	100
A		2.8	4.4	8.1	18	109	1	0.1	nd	nd	0.3	143		60	
	1	1.6	5.9	6.6	15	121	1.2	0.6	nd	nd	1.6	153		41	
	2	1.5	4	5	12	87	1.1	0.3	0.1	0.03	0.9	112		23	
B		4.1	1	9.3	15	89	1.7	0.2	nd	nd	nd	121		68	
	3	4.7	1.4	nd	18	200	2.9	0.1	0.2	nd	6.9	234		38	
C		1.4	2	8.1	4.3	28	0.1	0.2	nd	0.04	nd	44		26	
	4	12	4.8	9	5	27	0.4	3.6	0.1	0.1	1.4	63		14	
	5	18	5.3	6.5	4	28	0.5	5.8	0.1	nd	2.9	71		16	
	6	8.3	5.7	4.1	4.8	28	0.6	4.1	0.6	0.01	2	58		9	
D		3.3	4.4	2.7	4.9	13	0.1	0.1	nd	nd	nd	29		17	
	8	2.1	2.0	6.0	7.2	19	0.3	1.0	0	nd	0.5	38		11	

AB: apple juice blend; C: cider style; CT: (+)-catechin, EC: (-)-epicatechin, B1: Procyanidin B1, B2: Procyanidin B2, CA: Chlorogenic acid, CAF: Caffeic acid PLZ: Phloridzin, XPL: Phloretin, QUE: Quercetin, RU: Rutin, TOT: Sum Polyphenols, TP: Total Phenols – Folin Ciocalteu

other cider styles (Figure 2).

3.3 Volatile compounds

Esters are the dominant volatile compounds in ciders (Fan, Xu & Han, 2011) and are associated with the desirable taste of the product. Ethyl acetate is important for the sensory character in both wine and cider giving the product a fruity taste. Ethyl acetate originates from the apples and during the fermentation process. Ethyl acetate was the dominant ester in this experiment for ciders, though only registered in minor level in the apples. All the ciders contained substantial amounts of ethyl acetate and iso-amyl acetate that together with phenyl ethyl acetate, isobutyl acetate, ethyl hexanoate and hexyl octanoate are regarded to be the most important esters for

fruitiness and for general cider quality (Xu, Fan & Qian, 2007). Butyl butyrate (fruity flavour) appeared in the apple juices and not in the ciders. We found no difference in formation of volatile compounds between ciders inoculated with M02 yeast and the spontaneously fermented ciders. During the fermentation process of cider, considerable amounts of the higher alcohols: phenyl ethyl alcohol, 2-methyl-1-propanol, 1-propanol, 3-methyl-1-butanol and 2-methyl-1-butanol developed. These components are important for the fruity and characteristic cider taste of the product. 3-methyl-1-butanol and ethyl hexanoate (taste of ripe fruit) were not present in the apple juices, but found in relatively high amounts in the ciders (Table 5).

Acetaldehyde and diacetyl are important for the aromatic profile of fermented products (Berry &

Table 5: Content of volatile compounds from raw material to cider

Apple juice blend	Cider style	Acetald ehyde mg L ⁻¹	Acet one mg L ⁻¹	Ipropanol mg L ⁻¹	Diacetyl mg L ⁻¹	Ethyl acetate mg L ⁻¹	2meth.1-prop mg L ⁻¹	Butyl butyrat e mg L ⁻¹	3-methyl-1-butanol mg L ⁻¹	2-methyl-1-butanol mg L ⁻¹	Butyl acetate mg L ⁻¹	Ethyl hexanoate mg L ⁻¹	Phenyl ethyl alcohol mg/L
A	1	1.64	0.14	2.52	0.01	0.04	0.17	0.08	0	1.17	2.99	0	0.10
	2	11.62	0.32	7.89	0	14.59	14.86	nd	116.53	25.13	1.12	0.65	25.37
B	3	11.68	0.27	5.81	0.01	12.15	21.88	nd	125.37	14.19	1.16	0.19	20.94
	4	1.07	0.04	1.92	0	0.02	0.17	0.08	0	0.58	3.27	0	0
C	5	26.31	0.30	7.37	0.02	17.57	12.07	nd	90.41	21.29	1.17	0.33	19.20
	6	4.58	0.12	0.64	0.09	0.09	0.19	0.37	0	0.55	1.76	0	0.20
D	7	6.53	0.30	11.93	0	21.96	30.98	nd	165.82	31.63	0.66	0.65	34.26
	8	26.65	0.57	13.33	0.08	22.90	26.41	nd	148.70	27.77	0.62	0.48	34.53
	9	11.76	0.32	8.87	0.00	38.86	17.93	nd	96.75	17.24	0.34	0.30	19.23
	10	8.90	0.22	5.83	0.02	21.44	22.01	nd	130.45	8.26	0.75	0.46	24.70
	11	4.11	0.12	0.61	0.01	0.09	0.23	0.35	0	0.63	1.24	0	0.09
	12	19.98	0.32	10.20	0	19.82	10.01	nd	97.48	21.40	0.79	0.61	21.81

Slaughter, 2003). Low levels of acetaldehyde can give a fruit nice taste of green fruit, but high levels will give an unpleasant taste. Higher levels were present in cider styles 3, 5 and 8. Sensory evaluation of style 3 showed high scores for acidity and bitterness and low scores for fruitiness and sweetness. On the opposite side, style 5 (Amarillo hop added) got high scores for fruitiness, complexity, aftertaste and sweetness and style 8 (Cascade hop added) got relatively low scores on most attributes except appearance. We did not find any correlation between yeasts and the level of acetaldehyde in the ciders. Cider style 3 was slightly lower in higher alcohols than cider styles 1 and 2. These ciders were made from the same apple cultivars, but at different stages of maturity at the time of pressing. Although we did not find significant differences in chemical composition between the raw materials at different maturity, the levels most likely influenced the fermentation process and the formation of volatile compounds.

Generally, cider styles 4-7, all made from C apple juice mixture, tend to be higher in ethyl acetate, 2-methyl-1-propanol, 3-methyl-1-butanol and phenylethyl acetate than ciders from the other apple juice mixtures. These components are important for the fruity taste of the cider. These ciders also scored higher in the sensory evaluation of attributes like sweetness, fruitiness, complexity and aftertaste (Figure 1) and showing the same pattern in the PCA plot (Figure 2). Style 6 got higher scores for most of the attributes except bitterness compared to style 2. This indicates that apple juice mixture C was preferable for making cider in this experiment. Consequently, selection of apple cultivars is important for making a cider of good sensory properties.

3.4 Filtering and clarity

Filtering of the cider before bottling will also affect the appearance. Cider style 7 using filter number 12, became clearer, but lost some colour and taste attributes. Sensory evaluation showed that this cider got relatively low scores on most attributes except appearance. We found more haze in style 1 (apples stored a shorter time be-

fore pressing) and thus too many pectin substances in the juice. Bamforth (2004) recommended a maximum 2 % starch in the apples at the time of pressing, meaning that all of the pectin substances would be sufficiently degraded. On the other hand, overripe apples will provide low acidity, and give the cider a taste of “boiled apples”. This corresponds with our findings that style 3 was clearer but contained some soluble solids that remained after filtration.

This cider was characterised to be less fresh and with a hint of boiled taste.

3.5 Sensory evaluation

In the sensory evaluation, ciders made from the apple juice mixtures A and B, styles 1-3, got lower scores in most attributes compared to ciders from the mixtures C or D. Styles 4-7 scored higher in attributes like fruity/flowery taste, complexity and aftertaste, criteria that usually are regarded as positive attributes for cider. We found a high correlation ($r=0.853$, $p<0.01$) between fruitiness and content of catechin in the ciders. Evaluation of cider made from the cultivars Sunrise and Discovery had low sweetness, being acidic with a strong sour aftertaste (Figure 1 and 2). Average TP content in ciders from A and B juice mixtures was 34 GAE 100 mL^{-1} , while for ciders from C and D juice mixtures the average was 12.6 GAE 100 mL^{-1} . TP content was positively correlated ($r=0.847$, $p<0.01$) to bitterness in ciders in the sensory evaluation. Cider style 5 was evaluated to be sweeter, less acidic and less bitter than the other ciders, while cider style 8 got much lower scores on these attributes (Figure 1 & 2). Selection of hops for the cider during processing is important for development of a good aromatic profile.

4 Conclusion

The most important factor for the impact on cider quality was the various mixtures of apple juice with different selection of apple cultivars and apple maturity. Fermentation temperature, hop addition and filtering also affected the product properties. The addition of hops was successful for one of the styles. Choosing a

proper hop variety that goes well with the taste profile of the cider is essential.

The ciders made from apple juice mixture C (apple cultivars Aroma, Gravenstein and Summerred) got superior sensory characteristics compared to cider mixtures A and B (cultivars Sunrise and Discovery) and D (cultivars Aroma, Gravenstein, Summerred, Jonagold and Torstein).

Ciders made from Sunrise and Discovery were higher in phenolic compounds as well as total phenols though they were ranked lower in sensory evaluation.

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References

- Alberti, A., Machado dos Santos, T. P., Ferreira Zielinski, A. A., Eleuterio dos Santos, C. M., Braga, C. M., Demiate, I. M. & Nogueira, A. (2016). Impact on chemical profile in apple juice and cider made from unripe, ripe and senescent dessert varieties. *LWT-food Science and Technology*, *65*, 436–443. doi:[10.1016/j.lwt.2015.08.045](https://doi.org/10.1016/j.lwt.2015.08.045)
- Alonso-Salces, R. M., Korta, E., Barranco, A., Berrueta, L. A., Gallo, B. & Vicente, F. (2001). Determination of polyphenolic profiles of basque cider apple varieties using accelerated solvent extraction. *Journal of Agricultural and Food Chemistry*, *49*(8), 3761–3767. doi:[10.1021/jf010021s](https://doi.org/10.1021/jf010021s)
- Bamforth, C. (2004). *Fermented beverage production*, 2nd edn. Edited by AGH Lea and JR Piggott. Kluwer Academic/Plenum Publishers, New York, 2003. 423 pp ISBN 0-306-47275-9. *Journal of the Science of Food and Agriculture*, *84*(11), 1442–1442. doi:[10.1002/jsfa.1707](https://doi.org/10.1002/jsfa.1707)
- Berry, D. R. & Slaughter, J. C. (2003). Alcoholic beverage fermentations. In *Fermented beverage production* (pp. 25–39). Springer.
- Børve, J. & Vangdal, E. (2009). Physiological storage decay on apples at low temperature and low o_2 . In *X international controlled and modified atmosphere research conference 876* (pp. 399–400).
- Bütikofer, U. & Ardö, Y. (1999). Quantitative determination of free amino acids in cheese. *International Dairy Federation*.
- Erken, H. S. (1932). *Stor kokebok for større og mindre husholdninger : Kokning, bakning, syltning, råsyltning, vinlegning, slaktning, hermetikk, sykekost og dietkost, menyer, anretning*. Oslo: Aschehoug.
- Fan, W., Xu, Y. & Han, Y. (2011). Quantification of volatile compounds in chinese ciders by stir bar sorptive extraction (sbse) and gas chromatography-mass spectrometry (gc-ms). *Journal of the Institute of Brewing*, *117*(1), 61–66. doi:[10.1002/j.2050-0416.2011.tb00444.x](https://doi.org/10.1002/j.2050-0416.2011.tb00444.x)
- Gronnevik, H., Falstad, M. & Narvhus, J. A. (2011). Microbiological and chemical properties of Norwegian kefir during storage. *International Dairy Journal*, *21*(9), 601–606. doi:[10.1016/j.idairyj.2011.01.001](https://doi.org/10.1016/j.idairyj.2011.01.001)
- Guyot, S., Marnet, N., Sanoner, P. & Drilleau, J.-F. (2001). Direct thiolysis on crude apple materials for high-performance liquid chromatography characterization and quantification of polyphenols in cider apple tissues and juices. In *Methods in enzymology* (Vol. 335, pp. 57–70). Elsevier.
- Jolicoeur, C. (2013). *The new cider maker's handbook: A comprehensive guide for craft producers*. Chelsea Green Publishing.
- Kahle, K., Kraus, M. & Richling, E. (2005). Polyphenol profiles of apple juices. *Molecular Nutrition & Food Research*, *49*(8), 797–806. doi:[10.1002/mnfr.200500064](https://doi.org/10.1002/mnfr.200500064)
- Laaksonen, O., Kuldjärvi, R., Paalme, T., Virkki, M. & Yang, B. (2017). Impact of apple cultivar, ripening stage, fermentation type and yeast strain on phenolic composition of apple ciders. *Food Chemistry*, *233*, 29–37. doi:[10.1016/j.foodchem.2017.04.067](https://doi.org/10.1016/j.foodchem.2017.04.067)
- Lea, A. (2015). *Craft cider making*. Crowood.

- Leforestier, D., Ravon, E., Muranty, H., Cornille, A., Lemaire, C., Giraud, T., ... Branca, A. (2015). Genomic basis of the differences between cider and dessert apple varieties. *Evolutionary Applications*, 8(7), 650–661. doi:[10.1111/eva.12270](https://doi.org/10.1111/eva.12270)
- Milosevic, T., Milosevic, N. & Mladenovic, J. (2019). Tree vigor, yield, fruit quality, and antioxidant capacity of apple (*malus x domestica* borkh.) influenced by different fertilization regimes: Preliminary results. *Turkish Journal of Agriculture and Forestry*, 43(1), 48–57. doi:[10.3906/tar-1803-109](https://doi.org/10.3906/tar-1803-109)
- Moe, K. M., Porcellato, D. & Skeie, S. (2013). Metabolism of milk fat globule membrane components by nonstarter lactic acid bacteria isolated from cheese. *Journal of Dairy Science*, 96(2), 727–739. doi:[10.3168/jds.2012-5497](https://doi.org/10.3168/jds.2012-5497)
- Planchon, V., Lateur, M., Dupont, P. & Lognay, G. (2004). Ascorbic acid level of belgian apple genetic resources. *Scientia Horticulturae*, 100(1-4), 51–61. doi:[10.1016/j.scienta.2003.08.003](https://doi.org/10.1016/j.scienta.2003.08.003)
- Redalen, G. (1991). Lær å dyrke frukt : Dyrkingsmåter som du lykkes med : Omtale av 103 fruktsorter. Oslo: Det norske hageselskap ; I kommisjon hos Grøndahl.
- Sanoner, P., Guyot, S., Marnet, N., Molle, D. & Drilleau, J. F. (1999). Polyphenol profiles of french cider apple varieties (*malus domestica* sp.) *Journal of Agricultural and Food Chemistry*, 47(12), 4847–4853. doi:[10.1021/jf990563y](https://doi.org/10.1021/jf990563y)
- Tsao, R., Yang, R., Xie, S., Sockovie, E. & Khanizadeh, S. (2005). Which polyphenolic compounds contribute to the total antioxidant activities of apple? *Journal of Agricultural and Food Chemistry*, 53(12), 4989–4995. doi:[10.1021/jf048289h](https://doi.org/10.1021/jf048289h)
- Verdu, C. F., Gatto, J., Freuze, I., Richomme, P., Laurens, F. & Guilet, D. (2013). Comparison of Two Methods, UHPLC-UV and UHPLC-MS/MS, for the Quantification of Polyphenols in Cider Apple Juices. *Molecules*, 18(9), 10213–10227. doi:[10.3390/molecules180910213](https://doi.org/10.3390/molecules180910213)
- Volden, J., Borge, G. I. A., Bengtsson, G. B., Hansen, M., Thygesen, I. E. & Wicklund, T. (2008). Effect of thermal treatment on glucosinolates and antioxidant-related parameters in red cabbage (*brassica oleracea* l. ssp capitata f. rubra). *Food Chemistry*, 109(3), 595–605. doi:[10.1016/j.foodchem.2008.01.010](https://doi.org/10.1016/j.foodchem.2008.01.010)
- Wojdylo, A., Oszmianski, J. & Laskowski, P. (2008). Polyphenolic compounds and antioxidant activity of new and old apple varieties. *Journal of Agricultural and Food Chemistry*, 56(15), 6520–6530. doi:[10.1021/jf800510j](https://doi.org/10.1021/jf800510j)
- Xu, Y., Fan, W. & Qian, M. C. (2007). Characterization of aroma compounds in apple cider using solvent-assisted flavor evaporation and headspace solid-phase microextraction. *Journal of Agricultural and Food Chemistry*, 55(8), 3051–3057. doi:[10.1021/jf0631732](https://doi.org/10.1021/jf0631732)
- Ye, M., Yue, T. & Yuan, Y. (2014). Evolution of polyphenols and organic acids during the fermentation of apple cider. *Journal of the Science of Food and Agriculture*, 94(14), 2951–2957. doi:[10.1002/jsfa.6639](https://doi.org/10.1002/jsfa.6639)