



# Impact of conventional and innovative processing conditions on organoleptic and nutritional properties of applesauce from organic and conventional production systems

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## ABSTRACT

The impact of cultivar and production system was studied over two years on organoleptic and nutritional characteristics of apple fruits and then, on purees processed with both conventional convection cooking under vacuum and fast innovative microwave cooking. The main factors affecting the content and composition of sugars, organic acids, volatiles, polyphenols, and fibre were in the decreasing order, cultivar, year, and production system. Regarding processing, the fast innovative microwave cooking led to puree with a higher viscosity but with a lower polyphenol content compared to the convection cooking. Microwave cooking seemed to better preserve fibre but limit the diffusion of minor phenolic components, coming from skin and pips to puree and increase polyphenols degradation due to excessively high temperature spot. These results indicate that apple puree processed under vacuum did not present lower organoleptic and nutritional quality compared with fresh apple, as far as sugars, acids, volatiles, polyphenols and fibres were concerned.

## 1. Introduction

The growing interest in the potential of apple and derived products in terms of their biological value and improvement on human health is the result of their wide consumption and availability at any time of the year. Due to their popularity, apple is one of the most important fruit sources of health-promoting phenolic compounds in the diet of many societies (Asma et al., 2023). Apple health effects are mainly linked to its phenolic and fibre contents (Aprikian et al., 2003). Epidemiological studies have revealed an inverse correlation between both apple consumption and flavonoid content and coronary mortality (Hertog et al., 1993; Hyson, 2011; Knekt et al., 1996; Koutsos et al., 2015). Nowadays a frequent way to consume products which are rich in phenolic compounds and fibres is to eat applesauce or puree. These products are often presented as a healthy and ready-to-eat, convenient alternative to chocolate bars. They can be used to promote fruit consumption and they contribute to the five portions per day or 400 g/day of fresh, frozen or

canned produces recommended by the World Health Organization (WHO).

The physicochemical composition of fruit and fruit derived products, i.e. phenolic compounds, sugars, acids and fibres, are significantly influenced by several abiotic and biotic factors such as cultivar, agricultural practices, i.e. conventional vs organic production system, ripening stages, pedoclimatic conditions, i.e. climate and soil, and processing conditions (Guyot et al., 2002; Le Bourvellec et al., 2011; Le Bourvellec et al., 2015; Romero et al., 2016; Ruan et al., 2007; Valavanidis et al., 2009; Wojdyło et al., 2008). Trees grown under organic system are assumed to be more exposed to stresses than trees grown under conventional system. As a result, a higher polyphenol content is expected as shown by a metanalysis (Barański et al., 2014). However, results are controversial. Depending on the study, polyphenol content is higher, lower, or with limited or no variation in organic apple than in conventional one (Barański et al., 2014; Le Bourvellec et al., 2015; Mikulic-Petkovsek et al., 2010; Średnicka-Tober et al., 2020;

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Valavanidis et al., 2009; Yuri et al., 2012). The observed discrepancies may be due to the great variability of practices, with comparison of neighbouring farms presenting different pedoclimatic conditions, orchard designs, cultivars, sampling methods and analyses. Moreover, the aforementioned studies are mainly related to raw fruits, while far fewer studies are devoted to fruit-derived products processed from these raw fruits.

Fruit heat processing improves palatability, extends shelf life and destroys micro-organisms that cause spoilage. However, high temperatures also impact the content of health-promoting compounds such as polyphenols, aroma compounds and rheological properties leading to different levels of puree viscosity (Renard & Maingonnat, 2012; Marszałek et al., 2016; Le Bourvellec et al., 2018; Buegy, Rolland-Sabaté, Leca, Falourd, et al., 2021; Lan et al., 2022). These changes are related to the health, sensory with textural, odour and taste characteristics, and then impact the quality and acceptance of fruit-derived products by consumers. Nowadays, consumers demand minimally processed fruit-derived products with fresh flavour, natural appearance, and free from chemical additives and preservatives.

Microwave technology has been used in food industry because it enables shorter cooking times and saves energy. Its main applications are drying, heating and sterilization which are applied on a large variability of fruit and vegetables but also on meat and fish (Guo et al., 2017). In general, microwave technology is used for microbial decontamination (Cañumir et al., 2002; Picouet et al., 2009) or as pre-treatment step before cooking (Oszmiański et al., 2008). Microwave heating better preserves food qualities than conventional one (Marszałek et al., 2016; Zhou et al., 2022). However, to the best of our knowledge, it has never been used as cooking treatment or only has been used to cook small amount of apple or even one apple at a time (Lan et al., 2022; Picouet et al., 2009).

In this study, the use of microwave heating has been proposed as an alternative to conventional heating to preserve organoleptic characteristics and thermolabile compounds of apple, due to the shorter cooking time required for solids, resulting in faster enzyme inactivation. Microwave heating also has the advantage to use entire fruit without chopping, avoiding the use of chemical preservatives. For conventional cooking, fruits are trimmed, cored, chopped, cooked at temperature between 90 and 98 °C for about 5 min where preservative can be added, and finally pasteurized for 10–15 min in boiling water. Thus, in this context, it is important to evaluate the capacity of microwave treatment to maintain apple organoleptic and nutritional qualities, with no added preservatives, in comparison to conventional processing.

This study was designed to evaluate the impact of the mentioned heating conditions on apple nutritional and organoleptic qualities. The comparison covered two apple cultivars, 'Pinova' and 'Szampion', in two production systems, organic and conventional orchards, from two geographical origins located 11 km away. Two different processing conditions were then applied on the different batches of apples. The first one was a conventional process using convective heating followed by refining. The convective heating corresponded to hot break condition (HB), referring to a hot grinding at 95 °C, performed under vacuum to inactivate enzymes and prevent oxidation. The second process was innovative insofar as apples were directly microwaved, without grinding, in presence of vapour and then refined to obtain puree. All raw and processed products were then characterized by mid-infrared spectroscopy (MIR) as a rapid tool to evaluate physicochemical changes, and by classical methods to evaluate color, texture (on raw fruits), rheology (on purees), and content of sugars, organic acids, polyphenols and volatile compounds, and content and composition of cell walls. The following research questions were addressed in this paper:

1. Did the organoleptic and nutritional qualities of raw apples differ according to year, variety and production system?

2. Did the organoleptic and nutritional qualities of apple puree differ from those of raw apples according to year, variety and production system?
3. Were the differences observed in raw apple due to year, variety and production system, maintained after processing?
4. Did the organoleptic and nutritional qualities of applesauce differ according to processing conditions, and in particular do the softer conditions, i.e., rapid and innovative microwave cooking, result in applesauce of the same quality as raw apples?

## 2. Material and methods

### 2.1. Standards and chemicals

Ethanol and acetone were from Fisher Scientific (Strasbourg, France). Acetonitrile of HPLC grade and acetic acid were from Fischer Scientific (Pittsburgh, PA, USA). 5'-caffeoylquinic acid, (+)-catechin, (–)-epicatechin, sucrose, glucose, fructose, sodium carbonate, sodium hydroxide, NaBH<sub>4</sub>, N-methylimidazole, acetic anhydride, lignin alkali, toluene- $\alpha$ -thiol, inositol, galacturonic acid and citric acid were from Sigma-Aldrich (Darmstadt, Germany). Phloretin, para-coumaric acid, quercetin and cyanidin-3-O-galactoside were obtained from Extrasynthese (Lyon, France). Sugar standards (arabinose, fucose, galactose, xylose, mannose and rhamnose) and phloridzin were obtained from Fluka (Buchs, Switzerland). Malic acid was obtained from R-Biopharm (Darmstadt, Germany). Methanol-d<sub>3</sub> was from Acros Organics (Geel, Belgium).

### 2.2. Plant material and sample preparation

Two apple cultivars were used, 'Pinova' and 'Szampion' grown in both organic and conventional orchards. The two orchards are scientifically managed at the Institute of Horticulture in Skierniewice, central Poland. The organic orchard is run in accordance with the principles of organic farming as set out in Regulation (EU) 2018/848 of the European Parliament and of the Council of 30 May, 2018. The orchard is annually inspected and certified by one of the accredited organic farming certification bodies in Poland (Agro Bio Test PL EKO 07). The conventional orchard is managed in accordance with the generally applicable guidelines for conventional orchard production in the European Union. The practices used for fertilisation and plant protection during apple production in the two orchards and the two years, 2019 and 2020, are recap in Table 1. The agricultural practices for organic and conventional systems were the same for the two years. The amount of nitrogen was applied similarly in organic and conventional orchards.

Moreover, as it was further found that the soil of the conventional orchard contained sufficient amounts of phosphorus and potassium, they were only added in the organic orchard, especially potassium. The soil in the conventional experimental orchard at Dąbrowice (51° 55' 17" N, 20° 05' 46" E) is podzolic, sandy loam soil with a loamy subsoil with an average organic matter content of about 1.3 %. The soil in the organic experimental orchard in Nowy Dwór-Parcela (51° 51' 56" N, 20° 15' 44" E) is similar to that in Dąbrowice and is a podzolic, sandy loam soil with an average organic matter content of about 1.8 %.

In terms of plant protection strategy, for the organic orchard, mineral oil, copper, sulphur and mechanical weed control were used whereas for conventional orchard 34 sprays were applied with 22 different synthetic pesticides. Fungicides were used most frequently, followed by herbicides and insecticides with equal frequency. The described agrotechnical systems were typical of modern arboriculture, in both conventional and organic systems. As far as rootstocks are concerned, in conventional orchard the apple trees grew on P14 rootstock and in organic orchard on M26 rootstock. Both rootstocks are of medium-low growth strength, so-called semi-dwarf. The tree spacing was similar in both orchards. Fruit load was lower in organic system with on average 20 t ha<sup>-1</sup> than in conventional system with on average 30 t ha<sup>-1</sup>.

**Table 1**

Agrotechnology of apple trees (fertilisation and plant protection) in 2019 and 2020.

Cultivation system	Localization	Fertilizer	Dose of fertilisers and time of application	Plant protection system	N (kg ha <sup>-1</sup> )	K (kg ha <sup>-1</sup> )	P (kg ha <sup>-1</sup> )
Organic system	Nowy Dwór Parcela 51°87'03"N, 20°24'74"E	Azocor 105 -organic nitrogen	300 kg ha <sup>-1</sup> in tree rows (March)	Pests: spider mites, weevils paraffin oil 10 L ha <sup>-1*</sup> <i>Cydia pomonella</i> – <i>CpGv</i> ( <i>Cydia Pomonella Garnulosis virus</i> ), 100 mL ha <sup>-1*</sup> , <i>Tortricidae</i> - <i>Bacillus thuringiensis</i> subsp. <i>Kurstaki</i> 1 kg ha <sup>-1*</sup> , <i>Dysaphis plantaginea</i> and <i>Aphis pomi</i> - <i>potassium horticultural soap</i> 20 L ha <sup>-1*</sup>	54.3	16	100
		Cattle manure	30 t ha <sup>-1</sup> in tree rows (April)				
		EmFarma - microbiological fertilizer	20 L ha <sup>-1</sup> in tree rows (April)				
		Rokohumin - multi-ingredient organic fertilizer	10 L ha <sup>-1</sup> in tree rows (May)				
		Ecovigor - fertilizer based on marine algae	5 L ha <sup>-1</sup> (June)				
Conventional system	Dąbrowice 51°91'77"N, 20°10'59"E	Ammonium nitrate	100 kg ha <sup>-1</sup> (April)	Weeds: mechanical regulation	49.5	0	0
		KristaLeaf (fruit controller)	1 kg ha <sup>-1</sup> (April)				
		Urea	0.5 % (April & October)	In totally 34 treatments including: 16 with fungicides, 9 with herbicides 9 with insecticides (22 different pesticides were used)			
		Bolero -boron fertilizer	0.5 L ha <sup>-1</sup> (April)				
		Mg sulphate	1 % (May)				
		Wapnovit Turbo -calcium fertilizer plus Mg, B, Cu, Mo, Zn- foliar application	30 L ha <sup>-1</sup> (June to September)				
		Radkovit 0.5 carbonate lim with Mg - carbonate-magnesium fertilizer lime	1.5 t ha <sup>-1</sup> lime (October)				

\*: ready-made product.

Plant protection in conventional system – a list of pesticides: Agrostar 360 SL (glyphosate), Basta 150 SL (glufosinate ammonium), Captan 80 WG (captan), Chwastox 750 SL (MCPA), Chwastox Extra 300 SL (MCPA), Coragen 200 SC (chlorantraniliprole), Delan 700 WG (dithianon), Delan Pro (dithianon and dipotassiumphosphonate), Difo 250 EC (difenoconazole), Fontelis 200 SC (penthiopyrad), Kaptan zawiesinowy 50 WP (captan), Karate Zeon 050 CS (lambda-cyhalothrin), Kerb 400 SC (propryzamide), Merpan 80 WG (captan), Miedzian Extra 350 SC copper oxychloride, Ortus 05 SC (fenpyroximate), Pirimor 500 WG (pirimicarb), Runner 240 SC (methoxyfenozide), Select Super 120 EC (clethodim), Superam 10 AL (adjuvant: benzenesulfonic acids, alkyl derivatives, sodium salts and alcohols, branched, ethoxylated) Teppeki 50 WG (flonicamid), Thiram Granuflo 80 WG (tiuram).

The fruits were randomly harvested in the two orchards. For each batch of apples (orchard, cultivar and year), about 55 kg of fruits at a commercial maturity stage were transported to Avignon (France) and stored at 1 °C until processing. Each batch of apples was divided in three sub-batches: 1) raw apple characterisation (3 × 10 fruits), 2) conventional processing in purees (3 batches × 2.5 kg) and 3) innovative microwave processing in purees (3 batches × 2.5 kg). Purees were prepared and characterized for their biochemical composition and texture properties at INRAE Avignon as well as raw apples.

### 2.3. Raw apple fruits

Measurements of color and texture were firstly performed on each apple of each sub-batch (10 apples × 3 replicates). Then, for biochemical analysis, for each replicate, fruits were divided and cut into pieces as described in Le Bourvellec et al. (2011) and immediately frozen in liquid nitrogen. Apple pieces were gathered and then divided in three bags, A, B and C. Bags A were freeze-dried, stored at –20 °C, and used for characterisation of phenolic compounds and cell walls. Bags B were stored at –20 °C and used for mid-infrared spectroscopy, determination of sugars and organic acids. Finally bags C were stored at –80 °C and used for volatile compounds analysis.

### 2.4. Apple processing conditions

Two types of processing conditions were used. The first one was a convection cooking using hot-break (HB). The second one was an innovative microwave cooking applied directly on raw apples. For each cultivar and production system, three replicates were done for each processing condition.

### 2.5. Convection cooking

Approximately 2.5 kg of apples were cut into 12 pieces and processed under vacuum using a cooker-cutter robot (RoboQbo Qb8–3, RoboQbo, Bentivoglio, Italy), equipped with two micro-serrated cutting blades and a mixing blade. Fruits were cooked with a standard hot-break recipe at 95 °C for 10 min at a 3000-rpm grinding speed, then cooled down to 65 °C while maintaining the grinding speed. The total heat treatment time was 24 min. The cook value of the heating process was estimated at 7.05 (Holdsworth, 2007). Puree was then refined using an automatic sieve (Robot Coupe C80, Robot Coupe SNC, Vincennes, France) of 0.5 mm, to remove skin pieces and particles larger than the sieve opening. Finally, purees were conditioned in three hermetically sealed plastic bags: the first one was cooled at room temperature (22.5 °C) until the next-day measurement of rheology, mid-infrared spectra acquisition and biochemical measurements, the second one was freeze-dried (5 days, at –20 °C, Cryonext, Montpellier, France) and stored at –20 °C for cell wall and polyphenol characterisation and the third one was stored at –80 °C and used for volatile compounds characterisation.

### 2.6. Fast innovative microwave processing

Apples (batches of approximately 600 g, 3 to 5 apples) were half-cut and placed in a microwave oven (Samsung CM1529, Samsung Electronics Industry, Seoul, South Korea). A dish containing 10 mL of water was also placed in the microwave oven cavity. Apples were cooked for 6 min at 1.5 kW power and then refined as described above. The cook value of this microwave heating was estimated at 6.90, considering the average temperature of the heated fruit to reach 102 °C upon measurement. This confirmed the equivalence between both processes

(Rinaldi et al., 2021). After cooking, the 5 batches were combined and then refined until 2.5 kg of puree were obtained for each apple set. Purees were conditioned as described above in three bags.

## 2.7. Quality traits characterisation

Physicochemical characterisation was performed on raw apples before processing and on purees prepared with two types of processing conditions for each cultivar and production system (for both raw apples and purees three replicates for each sample set).

## 2.8. Mid-infrared spectroscopy on fruits and purees

As previously described (Bureau et al., 2013), mid-infrared spectra were collected at 23 °C with a Bruker Tensor 27 FTIR spectrometer (Wissembourg, France) equipped with a horizontal attenuated total reflectance (ATR) sampling accessory and deuterated triglycine sulphate (DTGS) detector. The homogenized raw apples and processed purees were placed at the surface of the zinc selenide crystal providing six internal reflections into the samples. The sample consistency, a thick liquid, allowed a good contact between sample and crystal and did not require pressing. The samples were scanned at wavenumbers ranging from 4000  $\text{cm}^{-1}$  to 650  $\text{cm}^{-1}$ , and corrected against the background spectrum of air. The spectrum of each sample was obtained by taking the average of 32 scans. The crystal was cleaned between measurements with deionized water and dried with lint-free tissue. Instrument control and spectral collection were performed using OPUS software (version 4.0 Bruker, France) supplied by the equipment manufacturer.

## 2.9. Color characterisation on fruits and purees

The apple color was measured by surface reflectance with a Konica-Minolta CM-400 chromameter (Minolta Co. Ltd., Osaka, Japan) with two measurements per fruit on the opposite sides (un-blushed and blushed sides). The color of purees was measured with the same equipment using a glass cell made of optical glass with a path of 10 mm. Nine measurements were made for each puree. The color was expressed with coordinates CIE 1976  $L^*a^*b^*$  based on D65 illuminant and 0° view angle, illumination area diameter 8 mm. In the  $L^*a^*b^*$  space,  $a^*$  represents the green-red color (scale from -60 to +60) and  $b^*$  the blue-yellow color (scale from -60 to +60).  $L^*$  coordinate represents lightness, where  $L^* = 0$  is completely black, and  $L^* = 100$  is completely white.

## 2.10. Texture of fruits and purees

The raw apple texture was determined by a puncture test using a multi-purpose texturometer TaPlus (Lloyd, Ametek, Elancourt, France). The chosen probe was a flat cylinder tip (2.0 mm diameter) which penetrates up to a depth of 17 mm into each peeled section of apple, at constant penetration rate (1  $\text{mm s}^{-1}$ ). Fruit texture was expressed as its firmness, i.e., the load (in N) at the plateau of the distance-load curve.

The texture of apple purees was expressed as their rheological properties. Analysis was conducted at 22.5 °C as previously reported (Buerge, Rolland-Sabaté, Leca, Falourd, et al., 2021) using a stress-controlled rheometer (Physica MCR301) equipped with a Peltier cell (CPTD-200) and a vane measuring cylinder system (CC27/SFL100/6 W) from Anton Paar (Graz, Austria). Flow curves were performed by varying the shear rate from 10 to 250  $\text{s}^{-1}$ . Since plant cell dispersions such as apple puree are pseudoplastic fluids, the flow curves were modelled with the Ostwald-de Waele equation. The equation parameters are the consistency index  $K$  describing the consistency at rest (the higher the  $K$  value, the more viscous the product) and the flow index  $n$  describing the ability to flow as a function of the shear rate (i.e., the slope of the flow curve: the farther the  $n$  value is from 1, the stronger the fluid reacts to either flowing or “packing”) (Lan et al., 2020). Also, the mouth-feel viscosity was defined as the apparent viscosity at 50  $\text{s}^{-1}$  shear rate,

the latter being assessed as the shear rate of food oral processing. Since the products did not show discrepancies depending of the year (not shown), texture data were combined to provide more robust datasets for statistical analyses.

## 2.11. Chemical analysis methods

### 2.11.1. Determination of dry matter content (DMC) in fruits and purees

The DMC was estimated from the weight of freeze-dried samples upon reaching a constant weight (freeze-drying, 5 days, Cryonext, Montpellier, France) and expressed in % Fresh Weight (FW) (Le Bourvellec et al., 2015).

### 2.11.2. Determination of sugars and organic acids in raw fruits and purees

Sugars (glucose, fructose and sucrose) and organic acids (malic acid and citric acid) were quantified using an enzymatic method with kits for food analysis (Boehringer Mannheim Co., Mannheim, Germany) and expressed in  $\text{g kg}^{-1}$  FW (Le Bourvellec et al., 2015). These measurements were performed with a SAFAS flx-Xenius XM spectrofluorimeter (SAFAS, Monaco).

### 2.11.3. Cell wall preparation and characterisation

Cell wall yield was evaluated by extracting and weighing alcohol insoluble solids, using the protocol established by Le Bourvellec et al. (2011). Cell wall composition was evaluated by analysing neutral sugars, galacturonic acids and methanol as described by Renard and Ginies (2009).

### 2.11.4. Polyphenols

Polyphenols were measured by high-performance liquid chromatography (HPLC)-diode array detection (DAD) after thioacidolysis using a method described by Le Bourvellec et al. (2011).

### 2.11.5. Volatile compounds

The analysis of volatiles was done by a dynamic headspace trapping on 10 g of homogeneous powder prepared by grinding raw apple with liquid nitrogen and on 10 g of processed apple purees, all samples stored at -80 °C. Each sample was put in a flask and added with anhydrous  $\text{CaCl}_2$  salt in order to improve the compounds volatilization and inhibit enzymatic oxidation. Flasks were then thermostated at 50 °C in a water bath. After 5 min of stabilization, a flow of nitrogen, an inert gas at 50  $\text{mL min}^{-1}$ , swept into the headspace and carried away volatile compounds. The latter were trapped during 5 min and concentrated on Tenax TA tubes (60/80 mesh, PerkinElmer, USA). The adsorbed volatiles were then desorbed using an automatic thermal desorber (ATD 650 Perkin Elmer, USA). A split of 40 % of the Tenax trap content was injected at 250 °C with a flow at 1  $\text{mL min}^{-1}$  in a gas chromatograph (Trace 1300, ThermoFisher Scientific, USA) equipped with a TG-WAXMS (30  $\text{m} \times 0.25$  mm i.d., 0.5  $\mu\text{m}$  film thickness) and coupled to a mass spectrophotometer (ISQ LT, ThermoFisher Scientific, USA). During thermal desorption, the trap was heated at 250 °C, allowing volatiles to be desorbed from the trap to a cold secondary trap (4 °C) before injection. The oven temperature ranged from 40 °C to 240 °C with an increase of 5 °C per minute. The mass detection was at 70 eV in a mass range from 33 to 250  $m/z$  at 0.2 s intervals. Volatiles were identified by comparing their spectra with the NIST (National Institute of Standards and Technology) database and using the software Chromeleon 7 (ThermoFisher Scientific, USA). Results were done as peak areas.

### 2.11.6. Quality Guidelines

The used methods in this work comply with the frame of Quality reference of INRAE (Quality Guidelines for the research and experimental units, Version 2, March 2013), which goes beyond reliability and traceability to also include a process driven approach from the beginning of the research question to quality of methods, of biological materials and safety of data. A data management plan was established as it



is mandatory for European project.

## 2.12. Statistical analysis

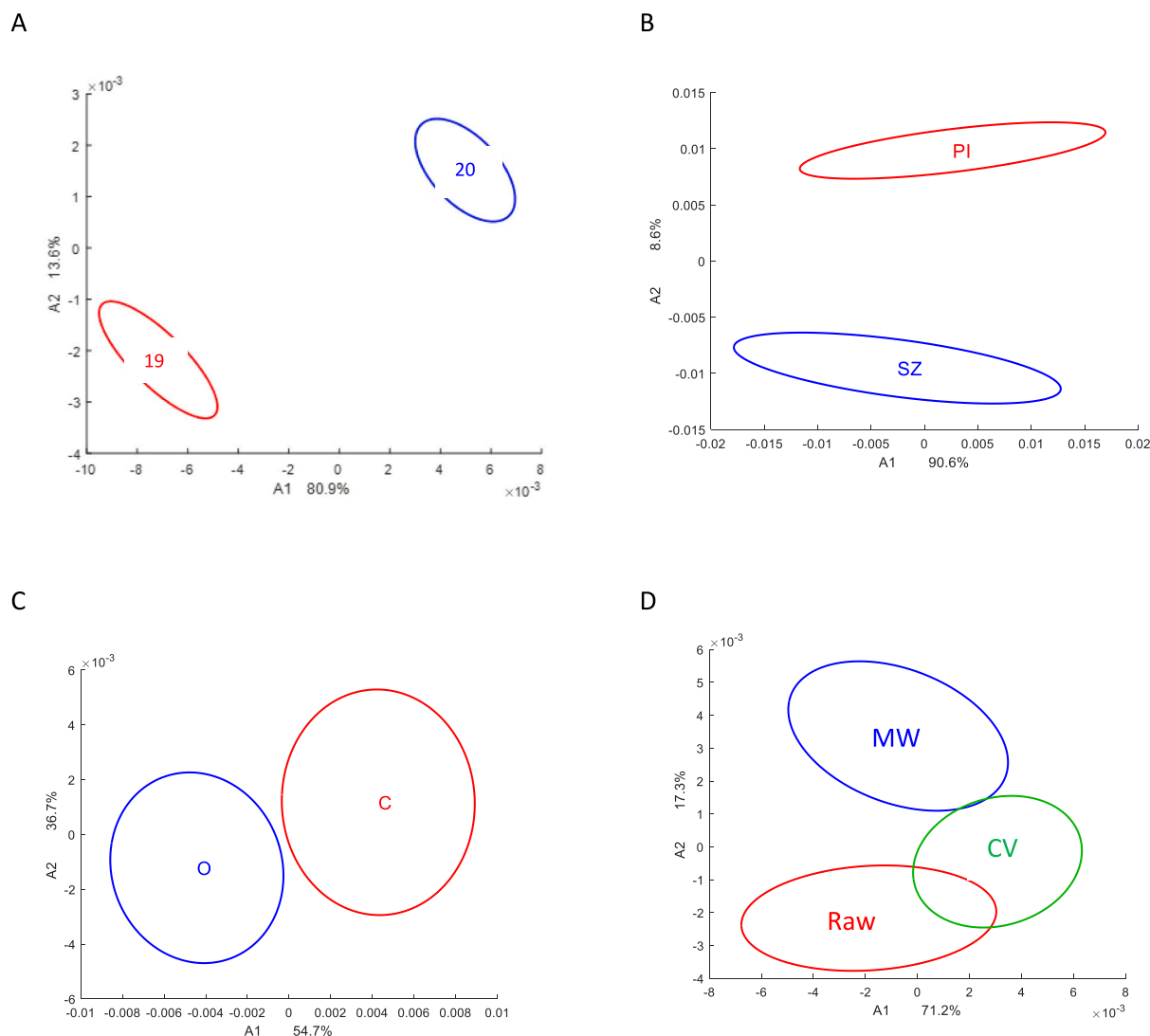
Results were presented as mean values, and their reproducibility was expressed as pooled standard deviation. Pooled standard deviations were calculated for each series of replicates using the sum of individual variances weighted by the individual degrees of freedom (Box et al., 1978). The differences between conditions were expressed as averaged values and standard deviations. Analysis of variance (ANOVA) using Fisher's test (F) was used to compare the mean values depending on cultivar, management system, year and processing. It was performed using the XLSTAT package of Microsoft Excel and using R language and environment (R Core Team, 2022) on texture data. Differences were considered to be significant at  $p < 0.05$ . Spectral data management including pre-processing, in particular standard normal variate (SNV) to correct multiplicative interferences and variations in baseline shift, ANOVA and Principal Component Analysis (PCA) were performed using MATLAB 7.5 (Mathworks Inc. Natick, MA) software with the SAISIR package (Cordella & Bertrand, 2014).

## 3. Results and discussion

### 3.1. Discrimination of raw and processed products based on mid-infrared spectra

Given the mid-infrared spectral region from 2000 to 900  $\text{cm}^{-1}$  gathers bands of different molecular vibrations such as stretching or bending of C=O, C—C, C—O, C—H, O—H or C—O—H, the obtained spectra are representative of apple composition (Bureau et al., 2019). In particular spectra bring information about the apple major compositional characteristics including dry matter, soluble solids, titratable acidity, pH, malic acid, and also some data on alcohol insoluble solids and total phenolic compounds after sample preparation and purification (Bureau et al., 2012; Lan et al., 2021). Therefore, spectra represent a good way to have an overview on both apple and puree composition and an evaluation of factors affecting their composition.

According to ANOVA performed on the spectral data, all studied factors significantly affected apple and puree compositions, and are classified by their decreasing effect using the Fisher values (F) in the following order: year (2019 and 2020,  $F = 600$ ) > cultivar (Pinova and



**Fig. 1.** Plots of PCA (Principal Component Analysis) representing the variability of raw and processed apples based on their mid-infrared spectra. A: spectral area in the range 1600–1500  $\text{cm}^{-1}$  for year (19: 2019 and 20: 2020), B: spectral area in the range 1100–900  $\text{cm}^{-1}$  for cultivar (PI: 'Pinova' and SZ: 'Szampion'), C: spectral area in the range 1800–1500  $\text{cm}^{-1}$  for production system (O: organic system in the Nowy Dwór Parcela farm and C: conventional system in the Dąbrowice farm), and D: spectral area in the range 1600–1400  $\text{cm}^{-1}$  for processing condition (Raw: raw apples, CV: convection cooking and MW: innovative microwave cooking). Ellipses represent 95 % of the variability of each apple set.

Szampion,  $F = 250$ ) > processing (raw, convection and microwave cooking,  $F = 90$ ) > production system (organic in the Nowy Dwór Parcela farm and conventional in the Dąbrowice farm,  $F = 18$ ). It means that the effect of year was significantly stronger than that of cultivar, itself higher than that of processing, the production system having the least impact. ANOVA also allowed to identify spectral areas which specifically varied with the studied factors. The specific spectral areas were 1600–1500  $\text{cm}^{-1}$  for year, 1100–900  $\text{cm}^{-1}$  for cultivar, 1600–1400  $\text{cm}^{-1}$  for processing and 1800–1500  $\text{cm}^{-1}$  for production system. These specific spectral areas were then chosen to illustrate the variability using Principal Component Analyses (PCA) (Fig. 1). Both, studied years and varieties were clearly discriminated (Fig. 1 A and B). For the other two factors, production system and processing, the groups of apple products were partly overlapped (Fig. 1 C and D). The two production systems, combining both two geographical origins located 11 km away and agricultural practices, were distinct on the axis 1 (Fig. 1 C). The raw apples (Raw) and processed apples using convection (CV) were rather separated on the axis 1 representing 71 % of the total variability whereas the raw apples and the innovative microwaved processed apples (MW) were separated on the axis 2 representing only 17 % of the total variability (Fig. 1D).

The variability observed between years, cultivars, production systems and processing conditions was large enough to be visualised in mid-infrared, mainly in terms of biochemical properties such as dry matter, sugars and organic acids.

### 3.2. Fruits and puree color

Fruit color was determined on side exposed to sunlight, i.e., the blushed side, and on the un-blushed side. Both  $L^*$  and  $a^*$  coordinates of the fruit un-blushed side were significantly different depending on the cultivar ( $p < 0.001$ ) with higher values in 'Pinova' cultivar than in 'Szampion' in accordance with their respective genetic origin (Table 2). The production system affected fruit  $L^*$ ,  $a^*$  and  $b^*$  coordinates ( $p < 0.001$ ,  $p < 0.001$  and  $p < 0.01$  respectively). In particular, both  $L^*$  and  $b^*$  coordinates were higher in fruits from conventional production system, meaning they were brighter and yellower, than fruits from organic

**Table 2**

Fruit color determination using  $L^*$ ,  $a^*$ ,  $b^*$  coordinates for un-blushed and blushed sides depending on apple cultivar, production system and year.

	Unblushed side			Blushed side		
	$L^*$	$a^*$	$b^*$	$L^*$	$a^*$	$b^*$
<b>2019</b>						
<b>Pinova</b>						
conventional	74.8	4.7	51.9	43.8	45.7	24.0
organic	68.5	14.1	44.8	47.8	38.9	27.0
<b>Szampion</b>						
conventional	70.8	0.7	48.5	48.3	34.5	28.7
organic	68.0	1.4	47.7	49.8	30.1	30.5
<b>2020</b>						
<b>Pinova</b>						
conventional	77.8	−6.8	47.9	56.0	23.1	32.0
organic	72.0	−0.2	48.1	53.6	21.4	32.0
<b>Szampion</b>						
conventional	71.1	−10.9	46.7	53.9	18.1	32.7
organic	66.5	−4.0	44.5	46.9	24.4	27.4
<b>SD</b>	1.0	1.6	1.0	1.2	2.2	1.1
<b>Statistics: F-value and significance</b>						
Cultivar	35	30	4	0	14	2
	***	***	ns	ns	**	ns
Production system	48	28	13	1	1	0
	***	***	**	ns	ns	ns
Year	4	91	4	38	104	21
	ns	***	*	***	***	***

SD: pooled standard deviation, ddl = 64, ns: non-significant, \*: significant at  $p \leq 0.05$ , \*\*: significant at  $p \leq 0.01$ , \*\*\*: significant at  $p \leq 0.001$ . Values are means of 30 replicates.

production system. However,  $a^*$  coordinate was higher in fruits from organic production system, meaning that fruits were less green with more diffuse red over-pigmentation (un-blushed and blushed sides less differentiated and contrasted), than fruit from conventional production. Year had an effect on  $a^*$  and  $b^*$  coordinates with higher values in 2019 than in 2020 which would be linked with a difference of ripening stage giving greener and less ripen fruits (Table 2).

Concerning fruit blushed side,  $a^*$  coordinate was the only parameter which was significantly different between cultivars ( $p < 0.001$ ), with higher values in 'Pinova' than in 'Szampion' meaning that these fruits were redder, and so associated with higher anthocyanin content (Table 5). The production system did not modify the fruit blushed  $L^*$ ,  $a^*$  and  $b^*$  coordinates. For year,  $L^*$  and  $b^*$  coordinates were higher in 2020 than in 2019 and  $a^*$  coordinate was lower in 2020 than in 2019, which could be explained by a difference of ripening stages between the two years, as already mentioned for the un-blushed side.

The process had an impact on puree color in comparison with that of raw apples (Tables 2 and 3).  $L^*$ ,  $a^*$  and  $b^*$  coordinates were lower after processing whatever the processing conditions (Table 3) than the raw fruit coordinates.  $L^*$  coordinate was shift to darker values,  $b^*$  coordinate decreased towards low values and  $a^*$  coordinate to negative values indicating a loss of red and yellow colors (Ibarz et al., 2000).

Puree  $L^*$ ,  $a^*$  and  $b^*$  coordinates were significantly modified by all studied factors, in the following order: year > > cultivar > > processing conditions > production system for  $L^*$  coordinate, cultivar > > processing conditions > year > production system for  $a^*$  coordinate, and cultivar > > year > processing conditions > production system for  $b^*$  coordinate. Puree discoloration could be due to both non-enzymatic browning and degradation due to thermal treatments as conditions were well adapted to prevent enzymatic oxidation (hot break and

**Table 3**

Puree color determination using  $L^*$ ,  $a^*$ ,  $b^*$  coordinates depending on apple cultivar, production system, processing condition and year.

	process	$L^*$	$a^*$	$b^*$
<b>2019</b>				
<b>Pinova</b>				
conventional	CV	48.7	−0.5	16.1
organic	CV	49.2	−0.8	17.8
conventional	MW	44.5	1.8	19.9
organic	MW	47.0	0.1	18.7
<b>Szampion</b>				
conventional	CV	48.4	−3.2	15.6
organic	CV	48.3	−2.5	15.9
conventional	MW	50.7	−3.0	14.7
organic	MW	51.0	−3.1	17.0
<b>2020</b>				
<b>Pinova</b>				
conventional	CV	46.0	−1.1	13.5
organic	CV	45.6	0.7	15.5
conventional	MW	50.1	−4.5	19.0
organic	MW	48.6	−4.5	18.9
<b>Szampion</b>				
conventional	CV	46.5	−2.5	11.9
organic	CV	47.6	−0.9	13.1
conventional	MW	46.0	−3.1	12.4
organic	MW	46.4	−0.9	13.6
<b>SD</b>		0.3	0.2	0.3
<b>Statistics: F-value and significance</b>				
Cultivar		16	105	468
		**	***	***
Production system		5	17	139
		*	***	**
Processing condition		10	40	163
		**	***	***
Year		71	29	234
		***	***	***

CV: convection cooking, MW: innovative microwave cooking, SD: pooled standard deviation, ddl = 96, ns: non-significant, \*: significant at  $p \leq 0.05$ , \*\*: significant at  $p \leq 0.01$ , \*\*\*: significant at  $p \leq 0.001$ . Values are means of 9 replicates.

vacuum). Several reactions may have occurred such as destruction of pigments, caramelisation and Maillard condensation (Ibarz et al., 2000).

To summarize, all studied factors, year, cultivar, production system and processing conditions had an effect on the color of fruit and puree after processing and concerned all  $L^*$ ,  $a^*$  and  $b^*$  coordinates. However, in this work, no sensory analysis was carried out to determine whether the measured differences were perceptible by experts or even by consumers. As statistical differences were high between conditions, the color of the processed purees can influence consumer acceptability.

### 3.3. Texture: fruit firmness and puree viscosity

The overall values of raw fruit texture (firmness) varied significantly between the two cultivars (mean firmness was 2.4 N for 'Pinova' and 1.3 N for 'Šampion'). In detail, the harvest year had a significant impact on fruit firmness for 'Šampion' ( $p < 0.001$ ) but not for Pinova ( $p > 0.1$ ). For 'Pinova' ( $p < 0.05$ ) in 2019 slightly significant differences were observed between the two production systems, whereas there was no significant difference for 'Šampion' ( $p > 0.1$ ). In 2020 however, both cultivars had significantly different fruit firmness values depending on the production system ( $p < 0.01$ ) (Fig. 2). Apart from 'Pinova' in 2020, the range of fruit firmness for conventional apples was significantly larger than that of organic fruits (standard deviation ratio of Conventional over Organic fruits ranging from 1.1 to 2.8). This meant, despite the overall discrepancies between conventional and organic fruits (Fig. 1), that the firmness was more homogeneous among fruit grown under organic system than under conventional. However, this was only observed for two harvest years and other agronomic and pedoclimatic factors not considered in this study could have an impact on the fruit firmness. Also, it must be kept in mind that the differences of fruit firmness due to the cultivar were considerably higher (F-value = 305) than the differences due to the production system (F-value = 6).

The viscosity parameter of processed purees highlighted significant

differences between the two cultivars ( $p < 0.05$ ) but not between the two harvest years ( $p > 0.1$ , data not shown) and the production systems ( $p > 0.1$ , data not shown). The processing conditions had an overall significant impact on the puree viscosity ( $p < 0.05$ ). In general, microwaved purees were more viscous than the convection-cooked ones except for 'Pinova' organic apples for which the viscosity was the same for both the convection (CV) and the innovative microwave (MW) cooking (Figs. 3 A and B).

Mouth-feel viscosity also highlighted slightly significant differences ( $p < 0.05$ , F-value = 5) due to both, cultivar and production system (Fig. 4). However, the processing condition had the strongest impact on the viscosity ( $p < 0.05$ , F-value = 30), confirming the importance of processing conditions on final characteristics of processed purees, in particular for viscosity. Systematically, the mouth-feel viscosity of purees produced by microwave cooking (MW) was higher than the one of purees produced by convective cooking (CV). The difference in viscosity observed between the purees produced by the two processing conditions could be due to differences in dry matter content, as the innovative microwave treatment resulted in a puree with a higher dry matter content than the puree obtained using convection cooking (Table 4). Puree viscosity is a key organoleptic property for consumers. The main levers to modulate it are the processing conditions (thermo-mechanical conditions) and in a lesser extent the cultivar. These factors should be taken into account by fruit processors.

### 3.4. Sugar and organic acid contents

Sugars and organic acids define the taste balance and, with volatile compounds, they also define the apple flavour. Among sugars, sucrose and fructose were the major ones in apple (Table 4), while glucose was the least abundant. Malic acid was the main organic acid, followed by citric acid as already observed in apple (Aprea et al., 2017; Le Bourvellec et al., 2015; Mikulic-Petkovsek et al., 2007; Oszmiański et al., 2018).

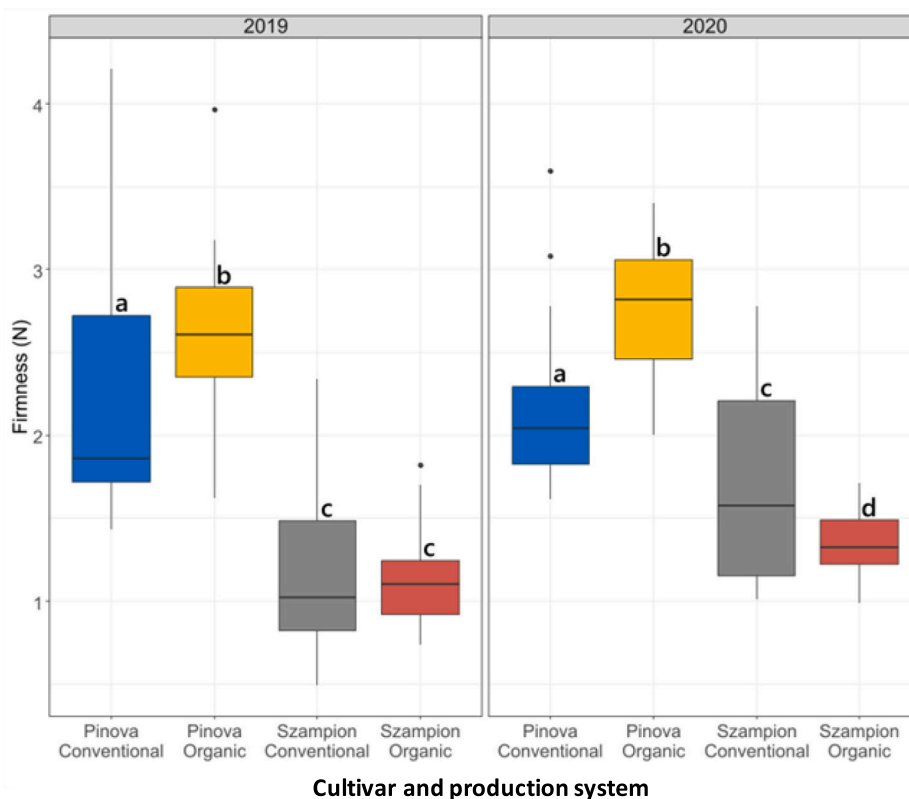
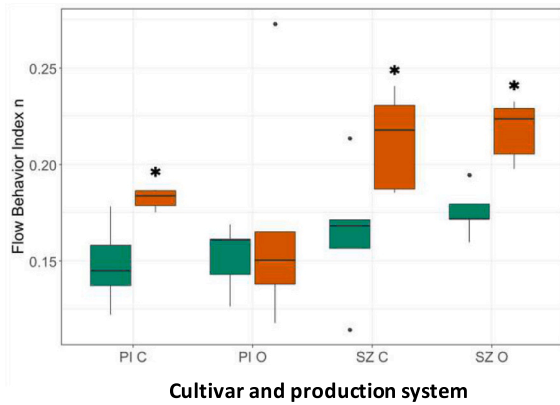
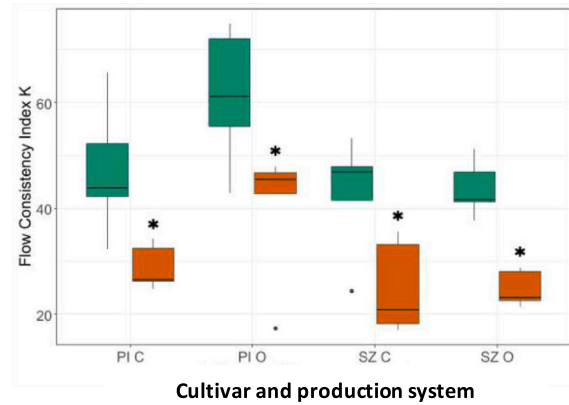


Fig. 2. Fruit firmness of the four different sets of apples ('Pinova' and 'Šampion', organic and conventional systems) in 2019 (left) and 2020 (right). Boxplots show the median, first and fourth quartiles, and outliers (black dots). Letters a, b, c, d stand for statistical significance according to Tukey post-hoc test ( $\alpha = 95\%$ ).

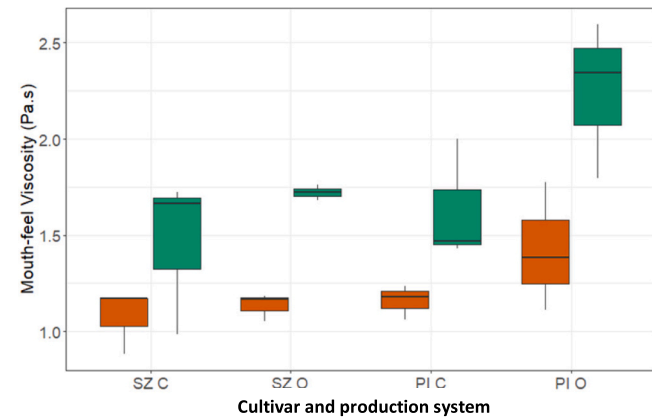
A



B



**Fig. 3.** Flow behaviour (A) and consistency (B) indices depending on the matrix properties (PI: ‘Pinova’, SZ: ‘Šampion’, O: Organic, C: Conventional) for each processing condition (■: convection cooking, ■: innovative microwave cooking). Star symbol (\*) indicates statistical significance ( $p < 0.05$ ) between processes.



**Fig. 4.** Mouth-feel viscosity (apparent viscosity at a shear rate of  $50 \text{ s}^{-1}$ ) depending on the sample type (PI: ‘Pinova’, SZ: ‘Šampion’, O: Organic, C: Conventional) and the processing condition (■: convection cooking, ■: MW: innovative microwave cooking). Star symbol (\*) indicates statistical significance ( $p < 0.05$ ) between processes.

The amount of sucrose, fructose and glucose ranged between  $56.0 \text{ g kg}^{-1}$  FM (‘Šampion’ organic 2020) and  $119.7 \text{ g kg}^{-1}$  FM (‘Pinova’ organic 2019),  $66.7 \text{ g kg}^{-1}$  FM (‘Pinova’ organic 2020) and  $101.9 \text{ g kg}^{-1}$  FM (‘Šampion’ conventional 2019) and  $14.2 \text{ g kg}^{-1}$  FM (‘Pinova’ conventional 2020) and  $19 \text{ g kg}^{-1}$  FM (‘Šampion’ conventional 2019) respectively, which is in agreement with previous reports (Aprea et al., 2017; Mikulic-Petkovsek et al., 2007). Malic acid content ranged from  $4.2 \text{ g kg}^{-1}$  FM (‘Pinova’ conventional 2019) to  $7.0 \text{ g kg}^{-1}$  FM (‘Pinova’ organic 2019).

Fructose and sucrose contents were significantly different depending on the cultivar as expected, contrary to glucose ones. At the same time, fructose and sucrose contents were not modified by the production system (conventional vs organic) whereas glucose content was significantly higher in fruit from organic management system. The content of malic acid was higher too in these organic fruits. Roussos and Gasparatos (2009) compared apple grown under both organic and conventional production systems and did not find any difference neither in the total soluble solid contents nor in the titratable acidity. Roth et al. (2007) also found that sugar and organic acid contents of apples coming from different regions and different production systems did not differ significantly. In another comparative study, ‘Golden delicious’ apples were found to have a lower sugar content in fruits grown under organic system than in fruits grown under integrated system (Jakopic et al., 2012).

**Table 4**

Dry matter (%), sugars ( $\text{g kg}^{-1}$  fresh weight), and organic acids ( $\text{g kg}^{-1}$  fresh weight) contents depending on apple cultivar, production system, processing condition and year.

	Process	DM	SUC	GLC	FRU	MA	CA
<b>2019</b>							
<b>Pinova</b>							
conventional	Raw	18.1	88.8	16.4	69.2	4.2	1.7
organic	Raw	20.2	119.4	17.2	82.7	5.7	1.5
conventional	CV	17.8	83.8	15.1	92.1	4.6	0.8
organic	CV	20.3	77.8	15.8	93.3	4.4	0.9
conventional	MW	18.2	74.1	14.0	75.3	3.6	0.5
organic	MW	20.5	65.5	16.1	81.0	5.5	0.9
<b>Šampion</b>							
conventional	Raw	18.7	105.0	19.0	101.9	5.2	1.6
organic	Raw	14.7	88.9	18.3	79.5	5.5	1.6
conventional	CV	19.9	80.6	20.4	120.0	4.8	0.8
organic	CV	16.3	68.6	19.7	100.5	6.0	0.9
conventional	MW	20.2	51.4	16.4	93.3	2.5	0.9
organic	MW	18.4	61.5	19.7	82.9	6.0	0.9
<b>2020</b>							
<b>Pinova</b>							
conventional	Raw	16.8	61.4	14.2	67.5	7.0	1.5
organic	Raw	18.4	76.2	14.7	66.7	6.3	0.1
conventional	CV	15.6	49.5	16.5	62.0	7.3	0.8
organic	CV	17.2	31.5	23.5	56.4	7.3	0.1
conventional	MW	16.8	54.4	15.0	70.5	5.1	0.1
organic	MW	19.0	33.8	19.8	76.8	5.2	0.3
<b>Šampion</b>							
conventional	Raw	15.5	58.7	17.0	67.4	5.0	0.3
organic	Raw	17.3	56.0	17.5	74.5	6.1	0.3
conventional	CV	15.0	50.7	19.1	79.3	6.5	0.7
organic	CV	17.1	40.4	15.3	72.6	5.4	0.7
conventional	MW	16.6	38.1	11.8	59.9	3.6	0.4
organic	MW	18.1	34.0	19.9	64.0	4.5	0.2
SD		2.08	7.24	2.08	4.91	0.50	0.36
<b>Statistics: F-value and significance</b>							
Cultivar		7.7	4.6	2.0	15.4	3.9	0.0
		**	*	ns	**	ns	ns
Production system		4.6	1.3	4.3	1.1	9.6	0.7
		*	ns	*	ns	**	ns
Processing condition		4.3	33.6	1.1	7.0	12.5	4.4
		*	***	ns	**	***	*
Year		26.4	98.6	0.1	95.6	18.2	14.7
		***	***	ns	***	**	**

Raw: Raw fruit, CV: convection cooking, MW: innovative microwave cooking, DM: Dry Matter, SUC: sucrose, GLC: glucose, FRU: Fructose, MA: Malic Acid, CA: Citric Acid, SD: pooled standard deviation, ddl = 48, nd: not detected, ns: non significant, \*: significant at  $p \leq 0.05$ , \*\*: significant at  $p \leq 0.01$ , \*\*\*: significant at  $p \leq 0.001$ . Values are means of 3 replicates.



In 'Cortland' and 'McIntosh' apples higher soluble solids contents were measured in fruits grown under organic system than in fruits grown under conventional system while there was no significant difference in titratable acidity (DeEll & Prange, 1992). However, in Gala apples, a lower titratable acidity was found in fruits grown under both conventional and integrated systems compared to fruits grown under organic system (Peck et al., 2006).

Sucrose and fructose contents were higher in 2019 than in 2020 whereas malic acid content was higher in 2020 than in 2019. This observed year effect could be due to a difference in ripening stage between fruits, as already mentioned for color (Table 2), as fruit ripening is known to impact balance between sugars (sweetness) and organic acids (acidity) contents (Hulme & Rhodes, 1970).

Sucrose, fructose, malic and citric acid contents were significantly lower in purees than in fruits. They were also lower in innovative microwaved puree than in convention-cooked puree (Table 4). In kiwifruit puree, thermal treatments using a constant temperature of 90 °C maintained to reach the target pasteurization value of  $P_{8.3^{\circ}\text{C}}^{85^{\circ}\text{C}} = 5$  min, do not lead to significant changes in sugars, i.e., glucose, fructose and sucrose, and organic acids (Yi et al., 2016). Moreover, the content of sucrose and reducing sugars do not change in peach puree concentrate (Aktag & Gökmen, 2021). However, it was previously shown that during apple or apricot puree processing, sucrose and organic acids tend to decrease with heat treatment without a clear trend for glucose and fructose as these sugars also come from sucrose hydrolysis (Ibarz et al., 2000; Le Bourvellec et al., 2018).

### 3.5. Volatile compounds

Among all volatile compounds in apple fruit and purees, a total of 20 key aroma compounds were identified and analysed in this study. These apple volatile compounds can be mainly grouped in several chemical classes such as ester (isobutyl acetate, butyl acetate, 2-methyl-1-butylacetate, propyl acetate), aldehyde ((E)-2 hexenal, 2 octenal, benzaldehyde, butanal, hexanal, nonanal, pentanal), alcohol (ethanol, propanol, 1-butanol, 2-methyl-1-butanol, pentanol, hexanol), ketone (6-methyl-5-hepten-2-one, acetone) and sesquiterpene ( $\alpha$ -farnesene), as already described in apple (Apréa et al., 2012; Liberatore et al., 2021; Tanaka et al., 2015; Yi et al., 2017). To better visualize and understand the composition of volatile compounds depending on cultivar, year, production system and processing conditions, data were investigated using a Principal Component Analysis (PCA) (Fig. 5).

The first two axes (PC1 and PC2) accounted for 91 % of the total variance, with PC1 and PC2 explaining 79.5 % and 11.6 % of the total variance, respectively. The first PC-score (PC1) was explained by high and positive contribution of isobutyl acetate, butyl acetate, 2-methyl-1-butylacetate, propyl acetate, (E)-2 hexenal, butanal, propanol, 1-butanol, 2-methyl-1-butanol, hexanol, pentanal, acetone,  $\alpha$ -farnesene, hexanal, pentanol, 2 octenal, and 6-methyl-5-hepten-2-one. The second PC-score had a negative contribution of ethanol, nonanal and 2 octenal (Fig. 5 A).

For the factors, year and cultivar, the groups of apple products were partly overlapped (Figs. 5 B and C) indicating a possible similarity of volatile compounds between apples grown in different years and from different cultivars. Both, production systems and processing conditions were quite discriminated (Fig. 5 D and E).

The two production systems, combining two geographical origins located 11 km away and agricultural practices, were overlapped on axis 1 representing 79.5 % of the total variability and separated on axis 2 representing 11.6 % of the total variability (Fig. 5 D). Hence, apple volatile compound composition was slightly influenced by the production system, and apple grown under organic and conventional systems showed similar volatile compound profile, although some differences occurred. Propyl acetate, butyl acetate, butanal,  $\alpha$ -farnesene, propanol and butanol were higher in conventional fruit than in organic fruit while

ethanol, 2 octenal and nonanal were higher in organic fruit than in conventional fruit (data not shown). Tanaka et al. (2015) have shown that apple from conventional system contained higher levels of most volatile components particularly esters and alcohols than apple from organic system. Roth et al. (2007) have shown that production system had no clear effect on apple volatile compounds. Moreover, sensory panellists could not differentiate apple from organic and conventional systems in terms of fruit off-flavour at harvest (DeEll & Prange, 1992).

The raw apples (Raw) and processed purees, whatever the applied processing conditions, i.e., convection (CV) and microwave (MW) cooking, were clearly separated on axis 1 representing 79.5 % of the total variability, indicating a huge change in the volatile compounds between raw and processed apples. Except for ethanol, 2 octenal, benzaldehyde, nonanal and pentanal, other volatiles were detected in higher amount in raw fruit than in processed purees (data not shown). Regardless the processing conditions, the loss of volatile compounds may be due to temperature-induced degradation, as demonstrated for strawberry puree (Teribia et al., 2021). The convection cooked (CV) and the innovative microwaved purees (MW) were separated only on axis 2 representing only 11.6 % of the total variability (Fig. 5 E), showing another effect on volatile compounds depending on the processing conditions. Except for esters and acetone, 2 hexenal, 2-methyl-1-butanol,  $\alpha$ -farnesene, hexanal, volatile compounds were detected in higher amount in convection puree than in microwaved puree. Excessive high temperature spot in microwave processing may have degraded volatile compounds. Moreover, the higher viscous texture of microwaved puree may also have reduced the release of volatile compounds during headspace analysis. Indeed, it has been shown that an increased thickness in pectin gel was inversely related to aroma release (Boland et al., 2004; Bolland et al., 2006), hence the higher viscosity of microwaved puree compared to convection one may involve a greater interaction between matrix and volatile compounds and then limit their release (Fig. 4).

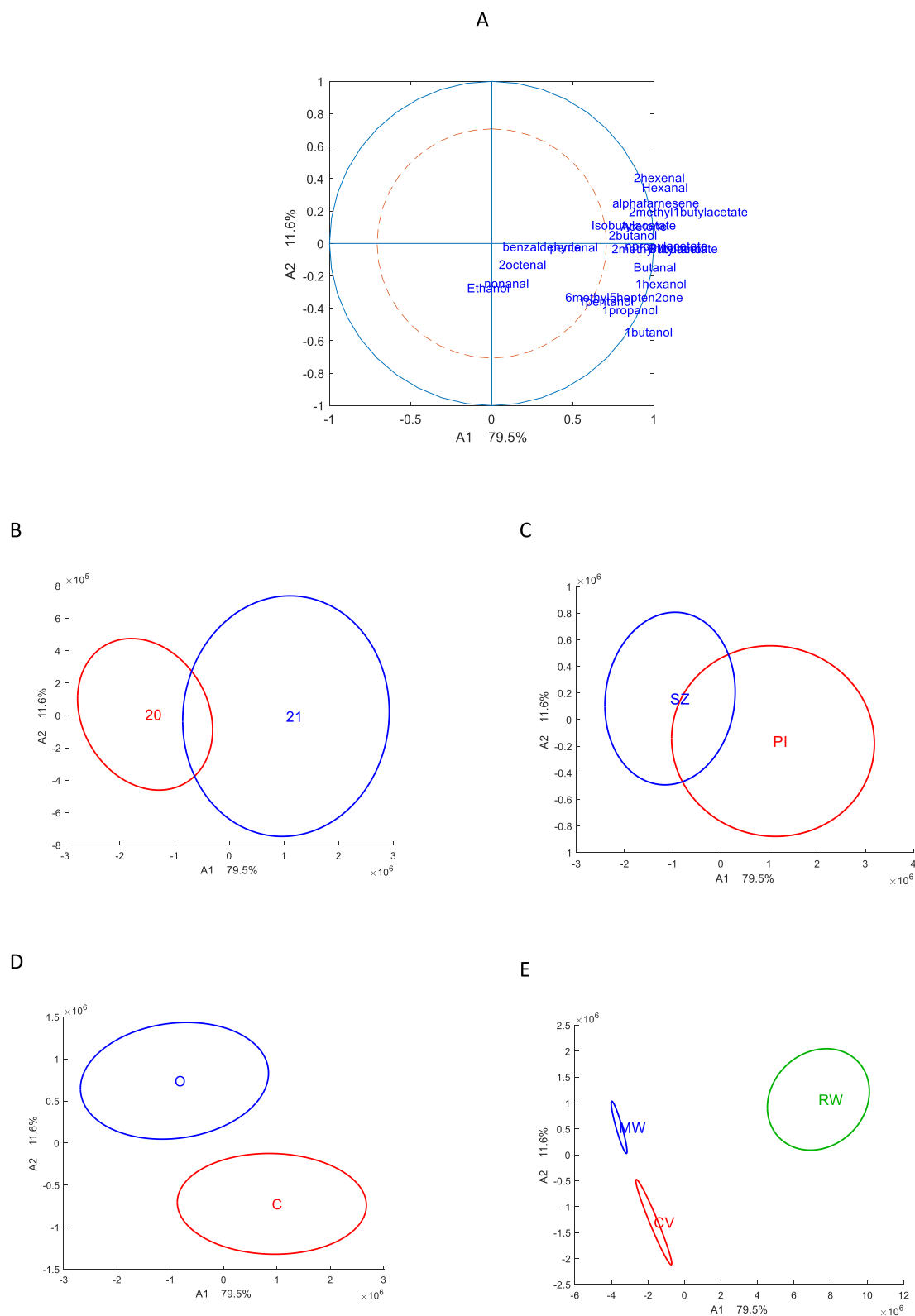
As for puree viscosity the main levers to modulate volatile composition were processing conditions that should be taken into account by fruit processors. In a lesser extent the production system could also affect them even if its effect was not clearly understood.

### 3.6. Polyphenol contents

Five major phenolic groups, corresponding to hydroxycinnamic acids (caffeoylquinic acid and para-coumaroylquinic acid), flavan-3-ols ((+)-catechin, (–)-epicatechin and proanthocyanidins), dihydrochalcones (phloridzin and phloretin xyloglucoside), flavonol (hyperosid, isoquercitrin, reynoutrie, guajaverin, avicularin and quercitrin) and anthocyanins (cyanidin-3-O-glucoside), with a total of fourteen individual compounds were identified and quantified in the raw apples of 'Şampion' and 'Pinova' cultivars (Table 5). The sum of phenolic compounds determined by thioacidolysis and HPLC-DAD analysis ranged from 0.78 mg g<sup>-1</sup> ('Şampion' conventional 2020) to 1.85 mg g<sup>-1</sup> FW ('Pinova' organic 2020) in the raw fruits, in agreement with the usual range in dessert apple (Guyot et al., 2002; Le Bourvellec et al., 2011; Le Bourvellec et al., 2015; Wojdyło et al., 2008).

Except for the degree of polymerisation of procyanidins and catechin content, cultivar had a significant impact on all polyphenols quantified in raw fruits as already observed in previous studies (Le Bourvellec et al., 2011; Le Bourvellec et al., 2015; Wojdyło et al., 2008).

The effect of production system was also significant for all phenolic compounds except for anthocyanins. For 'Pinova' cultivar, phenolic contents were higher in fruit from organic system than in fruit from conventional system (1.54 mg g<sup>-1</sup> FW vs 1.63 mg g<sup>-1</sup> FW for conventional and organic fruit respectively in 2019 and 1.41 mg g<sup>-1</sup> FW vs 1.85 mg g<sup>-1</sup> FW for conventional and organic fruit respectively in 2020). However, for 'Şampion' cultivar, phenolic contents were higher in fruit from conventional system in 2019 than in fruit from organic system (1.25 mg g<sup>-1</sup> FW vs 1.05 mg g<sup>-1</sup> FW for conventional and organic fruit



**Fig. 5.** Plots of PCA (Principal Component Analysis) representing the variability of raw and processed apples based on their volatile compounds. A: Correlation circle of volatile compounds loadings on PC1 and PC2, B: Sample map of scores on PC1 and PC2 depending on the year, C: Sample map of scores on PC1 and PC2 depending on the cultivar, D: Sample map of scores on PC1 and PC2 depending on the production system, and E: Sample map of scores on PC1 and PC2 depending on the processing condition. The code corresponds to the year (19 for 2019 or 20 for 2020), the cultivar (PI: 'Pinova', SZ: 'Szampion'), the production system (C: conventional, O: organic), and the processing condition (Raw: raw apples; CV: convection cooking, MW: innovative microwave cooking). Ellipses represent 95 % of the variability of each apple set.

Table 5

Phenolic compounds (mg g<sup>-1</sup> fresh weight) contents depending on apple cultivar, production system, processing condition and year.

	Process	CAT	EPI	PCA	mDP	XPL	PL	5CQA	pCoQA	TotFl.	TotA.	Tot
<b>2019</b>												
<b>Pinova</b>												
conventional	Raw	0.04	0.10	0.97	6.9	0.03	0.07	0.18	0.0004	0.011	0.04	1.54
organic	Raw	0.03	0.10	1.10	6.0	0.03	0.06	0.21	0.0009	0.07	0.03	1.63
conventional	CV	0.04	0.11	0.89	5.9	0.03	0.05	0.21	nd	0.11	0.02	1.46
organic	CV	0.04	0.12	1.13	5.5	0.04	0.08	0.31	0.0005	0.09	0.02	1.82
conventional	MW	0.03	0.07	0.63	7.3	0.02	0.03	0.17	0.0006	0.04	0.004	1.00
organic	MW	0.04	0.11	0.72	6.5	0.03	0.05	0.26	0.0013	0.03	0.004	1.23
<b>Szampion</b>												
conventional	Raw	0.04	0.10	0.88	6.4	0.02	0.03	0.08	nd	0.09	0.01	1.25
organic	Raw	0.03	0.09	0.78	5.2	0.01	0.03	0.07	0.0002	0.04	0.01	1.05
conventional	CV	0.04	0.12	0.91	5.6	0.02	0.03	0.09	nd	0.09	0.005	1.30
organic	CV	0.04	0.13	0.68	5.3	0.02	0.04	0.08	nd	0.06	0.004	1.05
conventional	MW	0.03	0.10	0.88	7.4	0.02	0.02	0.11	nd	0.06	0.003	1.22
organic	MW	0.04	0.11	0.66	5.8	0.02	0.02	0.08	nd	0.04	0.01	0.98
<b>2020</b>												
<b>Pinova</b>												
conventional	Raw	0.02	0.09	0.97	6.0	0.02	0.04	0.18	0.0014	0.07	0.01	1.41
organic	Raw	0.03	0.13	1.23	5.8	0.04	0.08	0.25	0.0018	0.07	0.02	1.85
conventional	CV	0.03	0.14	0.97	6.1	0.02	0.03	0.22	0.0023	0.11	0.01	1.55
organic	CV	0.03	0.16	1.11	5.2	0.03	0.06	0.25	0.0040	0.11	0.01	1.77
conventional	MW	0.03	0.10	0.93	5.4	0.02	0.03	0.23	0.0023	0.05	0.01	1.40
organic	MW	0.03	0.13	1.01	4.9	0.03	0.04	0.33	0.0034	0.06	0.01	1.63
<b>Szampion</b>												
conventional	Raw	0.02	0.07	0.52	5.5	0.01	0.03	0.08	0.0049	0.04	0.01	0.78
organic	Raw	0.02	0.12	0.85	5.3	0.02	0.03	0.08	0.0133	0.04	0.01	1.19
conventional	CV	0.04	0.15	0.82	5.5	0.01	0.02	0.10	0.0116	0.08	0.01	1.24
organic	CV	0.05	0.20	1.19	5.7	0.02	0.04	0.11	0.0226	0.07	0.01	1.71
conventional	MW	0.03	0.15	0.65	6.4	0.01	0.03	0.09	0.0115	0.03	0.01	1.01
organic	MW	0.05	0.19	1.07	6.2	0.02	0.04	0.12	0.0221	0.06	0.01	1.59
<b>SD</b>		<b>0.003</b>	<b>0.009</b>	<b>0.056</b>	<b>0.27</b>	<b>0.002</b>	<b>0.004</b>	<b>0.011</b>	<b>0.0004</b>	<b>0.011</b>	<b>0.003</b>	<b>0.069</b>
<b>Statistics: F-value and significance</b>												
Cultivar		3.7	15.0	35.7	0.7	228.6	125.2	893.2	805.0	13.8	24.5	113.8
		ns	**	***	ns	***	***	***	***	**	***	***
Production system		4.2	36.1	25.5	23.7	57.4	42.0	53.2	218.2	6.3	0.1	41.0
		*	***	***	***	***	***	***	***	*	ns	***
Processing condition		13.9	36.4	11.1	9.5	16.4	17.1	21.5	62.7	22.5	25.0	18.7
		***	***	**	**	***	***	***	***	***	***	***
Year		17.2	59.3	13.7	15.2	4.5	1.9	14.7	1676.1	0.5	5.5	18.7
		**	***	**	**	*	ns	**	***	ns	*	***

Raw: Raw fruit, CV: convection cooking, MW: innovative microwave cooking, CAT: (+)-catechin, EPI: (–)-epicatechin, PCA: procyanidins, mDP: average degree of polymerisation of procyanidins, XPL: phloretin-2-O-xyloglucoside, PL: phloridzin, 5CQA: 5-O-caffeoylquinic acid, pCoQA: para-coumaroylquinic acid, TotalFl: total flavonols, TotalA: total anthocyanins, Tot: total phenolic, SD: pooled standard deviation, ddl = 48, nd: not detected, ns: non significant, \*: significant at  $p \leq 0.05$ , \*\*: significant at  $p \leq 0.01$ , \*\*\*: significant at  $p \leq 0.001$ . Values are means of 3 replicates.

respectively) whereas it was the opposite in 2020 (0.78 mg g<sup>-1</sup> FW vs 1.05 mg g<sup>-1</sup> FW for conventional and organic fruit respectively).

Some authors found that organic production system led to higher fruit phenolic compound contents compared to conventional production system (Barański et al., 2014; Mikulic-Petkovsek et al., 2010; Średnicka-Tober et al., 2020) or to a limited or non-existent impact (Le Bourvellec et al., 2015; Valavanidis et al., 2009; Yuri et al., 2012). Nutrient supply to plants can affect their secondary metabolism, but there is clearly no simple mechanism explaining the dilemma between secondary metabolite synthesis and growth, even if the growth differentiation equilibrium hypothesis postulates that investment in secondary compounds can lead to growth limitation (Herms & Mattson, 1992). Hence, an increase in nitrogen supply reduces flavonoid accumulation in apple tree leaves (Rühmann et al., 2002; Leser & Treutter, 2005) and fruits (Awad & de Jager, 2002). However, the total nitrogen supply was very close in the two studied orchards but the nature of their supply, i.e., mineral vs organic - and therefore its availability for trees - may be different. Moreover, procyanidin degree of polymerisation was lower in organic fruit than in conventional fruit as well as flavonol contents. Surprisingly, the production system had no impact on anthocyanin contents. It is generally assumed that organic orchards are more exposed to biotic (plant-parasitic nematodes, fungi and bacteria) and abiotic (salt, drought, and UV) stresses than orchards with synthetic inputs. In response to stresses, plants would increase biosynthesis of their

secondary metabolites such as phenolic compounds and especially flavonoids that are involved in different biological activities in plants. Flavonols and anthocyanins are mainly located in fruit skin and may be expected to be present in higher concentration in organic fruit due to their involvement in stress protection (Landi et al., 2015; Winkel-Shirley, 2002; Zhuang et al., 2023). However, this was not confirmed here. Moreover, it is known that procyanidins interact with proteins leading to their inhibition and this mechanism could be involved to control biotic infestation. This inhibition mechanism is mainly driven by structural conformation and composition of procyanidins and especially their degree of polymerisation, i.e., the number of their constitutive units (Le Bourvellec & Renard, 2012). Hence, a higher procyanidins degree of polymerisation may be expected in fruit grown under more stressed conditions like organic orchards. However, it was not confirmed here.

The processing had a significant impact on content of all phenolic compounds (Table 5). The microwave cooking gave purees with less polyphenols which are present in apple peel and pips, such as quercetin derivatives, dihydrochalcones and anthocyanins compared to purees obtained by the convection cooking. This may be due to a lower diffusion of polyphenols from the peel and pips to the flesh as the innovative microwave process is much faster, i.e., 8 min, than the convection cooking one, i.e., 24 min. However, a degradation by thermal treatment during processing could also be involved as procyanidins and flavonol contents were lower in microwaved puree than in convection purees.

The procyanidin degree of polymerisation was higher in convection puree than in microwaved puree probably in relationship with oligomer degradation during heat treatment (Le Bourvellec et al., 2013). Anthocyanin content was lower in purees than in raw fruit, and lower in microwaved purees than in convection ones. This could probably be due to their degradation as they are known to be sensitive to high temperatures, and especially during microwave cooking as excessive high temperature spot can be reached. Moreover, 5-O-caffeoylquinic and para-coumaroylquinic acid contents were lower in fruit than in purees which might be due to better extraction efficiency of these compounds from puree than from fruit for analysis (Le Bourvellec et al., 2018). Even if polyphenols were degraded during processing their content and composition were close to that of raw fruit.

### 3.7. Cell wall content and composition

The alcohol insoluble solids (AIS) contents of apple flesh varied from 2.4 % ('Şampion' organic 2019) to 3.7 % ('Pinova' organic 2019) fresh weight (Table 6), in accordance with previously published works (1.5–2.7 % fresh weight) (Buergy, Rolland-Sabaté, Leca, Falourd, et al., 2021; Le Bourvellec et al., 2011; Massiot & Renard, 1997; Renard, 2005a) except for 'Pinova' organic 2019 which could be due to a less good sample preparation and probably a higher peel ratio known to

contain more AIS material than flesh (Massiot & Renard, 1997). But, these results were consistent with the observed differences of fruit firmness between cultivars, since AIS yield in 'Pinova' was significantly higher than 'Şampion', which would explain a denser microstructure, and thus firmer fruit. The cell wall contents were very stable in our study as they were affected neither by the cultivar nor by the production system nor by the year. However, cell wall contents were significantly lower after processing whatever the cooking conditions (convection vs microwave). During processing, the high temperature (95 °C) may have contributed to the depolymerisation of pectins by acid hydrolysis and  $\beta$ -elimination leading to their solubilization and elimination during AIS preparation (Buergy, Rolland-Sabaté, Leca, Falourd, et al., 2021; Liu et al., 2021; Renard, 2005b).

The neutral sugar composition of AIS was very close in the two apple cultivars (Table 6). It reflected the macromolecular composition of apple cell wall: cellulose composed of glucose, highly methylated pectins relatively rich in xylogalacturonan, fucogalactoxyloglucan and mannan. Their contents are within the usual ranges (Le Bourvellec et al., 2011; Massiot & Renard, 1997; Renard, 2005b). Among neutral sugars (rhamnose, fucose, arabinose, xylose, mannose, galactose, non-cellulosic and cellulosic glucose) and galacturonic acid that are constitutive of apple cell wall, only arabinose and galactose were significantly lower in 'Şampion' cultivar whereas non-cellulosic glucose was

**Table 6**

Cell wall content and composition including AIS yields (% fresh weight), neutral sugars and galacturonic acid contents (mg g<sup>-1</sup> dry matter of AIS) determined in apples and purees AIS depending on apple cultivar, production system, processing and year.

	process	Yield	Rha	Fuc	Ara	Xyl	Man	Gal	NcGl	CGl	Gal A	MeOH	DM
<b>2019</b>													
<b>Pinova</b>													
conventional	Raw	3.1	8	6	88	49	13	53	13	202	291	33	76
organic	Raw	3.7	10	8	117	63	16	83	14	267	267	33	81
conventional	CV	2.3	8	8	89	48	15	60	21	229	253	31	72
organic	Cv	2.5	8	8	98	46	13	82	20	215	232	31	78
conventional	MW	2.6	9	8	83	46	14	57	15	238	241	32	83
organic	MW	2.5	9	8	96	51	15	79	19	245	271	31	56
<b>Şampion</b>													
conventional	Raw	3.00	9	7	79	47	13	42	18	215	244	24	67
organic	Raw	2.4	11	8	74	49	16	51	15	228	291	21	48
conventional	CV	2.7	8	7	80	44	14	47	25	222	299	32	71
organic	CV	2.3	9	8	75	46	15	51	24	231	328	33	68
conventional	MW	3.0	8	8	73	45	13	43	23	221	255	31	81
organic	MW	2.9	9	9	66	47	15	52	23	250	285	31	72
<b>2020</b>													
<b>Pinova</b>													
conventional	Raw	3.0	10	7	89	43	16	58	11	201	314	34	73
organic	Raw	3.2	8	7	103	46	18	66	12	208	322	34	69
conventional	CV	2.9	11	10	104	56	19	74	21	265	299	40	88
organic	CV	2.7	9	9	116	51	19	82	21	223	314	39	83
conventional	MW	2.3	10	10	107	57	19	76	15	275	285	25	59
organic	MW	2.2	10	10	112	54	19	76	16	253	296	25	57
<b>Şampion</b>													
conventional	Raw	2.5	10	7	79	47	17	44	13	234	353	37	70
organic	Raw	2.9	8	7	90	50	15	59	25	206	306	33	72
conventional	CV	2.7	9	8	82	49	16	52	22	240	366	40	72
organic	CV	2.7	10	8	99	46	17	75	34	236	325	36	74
conventional	MW	2.3	10	10	106	60	19	61	25	279	311	24	51
organic	MW	2.3	11	6	112	54	12	83	46	257	271	36	88
<b>SD</b>		0.11	0.41	0.45	3.74	2.12	0.85	2.63	1.90	8.52	9.35	1.34	3.74
<b>Statistics: F-value and significance</b>													
Cultivar		3.7	0.04	3.4	24.3	1.6	3.6	47.7	29.6	0.01	5.4	0.7	1.5
	ns	ns	ns	***	ns	ns	ns	***	***	ns	*	ns	ns
Management system		0.01	0.02	0.4	8.7	0.2	0.3	43.3	7.9	0.08	0.1	0.01	0.06
	ns	ns	ns	**	ns	ns	ns	***	**	ns	ns	ns	ns
Processing condition		11.0	0.78	4.7	1.4	1.8	0.4	8.3	12.3	7.4	4.6	12.8	3.0
	***	ns	*	ns	ns	ns	ns	**	***	**	*	***	ns
Year		0.9	4.7	2.6	19.5	1.1	14.4	13.6	2.4	1.4	29.9	10.1	0.03
	ns	*	ns	***	ns	ns	**	**	ns	ns	***	**	ns

AIS: Alcohol Insoluble Solids, Raw: Raw fruit, CV: convection cooking, MW: innovative microwave cooking, Rha: rhamnose, Fuc: fucose, Ara: arabinose, Xyl: xylose, Man: mannose, Gal: galactose, Gal A: galacturonic acid, NcGl: non cellulosic glucose, CGl: glucose from cellulose, MeOH: methanol, DM: pectins' degree of methylation, SD: pooled standard deviation, ddl = 48, ns: non-significant, \*: significant at  $p \leq 0.05$ , \*\*: significant at  $p \leq 0.01$ , \*\*\*: significant at  $p \leq 0.001$ . Values are means of 3 replicates.

significantly lower in 'Pinova' cultivar. This could be related to differences between 'Pinova' and 'Šampion' cultivars in pectin side chains, i. e., arabinan and galactan, and hemicellulose structures (Le Bourvellec et al., 2011; Liu et al., 2021).

The effect of production system on AIS composition was very weak and the difference was only significant for galactose, arabinose and non-cellulosic glucose, with lower contents in conventional fruit than in organic fruit. However, for both cultivar and production system an additional effect could be due to a difference in fruit ripening stage, known to affect cell wall composition (Brummell, 2006), and already mentioned in this paper for fruit color, sugars and organic acids. In general, cell wall modifications during fruit maturation involve hydrolysis of neutral sugars from pectin side chains, depolymerization and solubilization of both pectins and hemicelluloses (Brummell, 2006). Here, the pectin degree of methylation was the same between cultivars and was not modified either by production system or by year.

Cell wall composition was modified after processing with different trends depending on the processing conditions. Both convection and microwave processing led to an increase in fucose, galactose and non-cellulosic glucose. This tendency may be related to the pectin degradation by acid hydrolysis or/and  $\beta$ -elimination reactions, giving an apparent increase of these neutral sugars constitutive of hemicelluloses which were not affected by thermal treatment (Buerge, Rolland-Sabaté, Leca, Falourd, et al., 2021; Liu et al., 2021; Renard, 2005b). The microwave processing led to an increase of rhamnose, xylose and cellulosic glucose contents and to a decrease of pectin degree of methylation, and galacturonic acid and methanol contents. Even if the microwave process is very fast, i.e., 8 min, compared to the conventional one, i.e., 24 min, excessive high temperature spot may have been reached as temperature distribution during microwave heating can be non-uniform (Zhang et al., 2023) leading to an extensive pectin depolymerisation and degradation by acid hydrolysis or/and  $\beta$ -elimination mechanisms.

#### 4. Conclusions

This study had a look at several factors that may impact raw apple and the corresponding processed purees, in terms of organoleptic (texture, color and taste) and nutritional properties such as fibres and polyphenols.

The characteristics of raw apple were clearly affected by year (2019 vs 2020) and cultivar ('Pinova' vs 'Šampion') while production system had less influence (organic vs conventional systems). These data confirmed therefore our previous study performed in another environment in France (Le Bourvellec et al., 2015) which was done using well-documented system experiment in order to eliminate bias induced by uncontrolled environmental external factors. Hence, orchard production system significantly affected polyphenol contents, but this effect depended on year and cultivar.

The other aspect studied in this paper was the fruit processing into puree comparing convection and microwave cooking. An original way to process apple was tested here including only two steps which were first, cooking entire apples in microwave oven with low quantity of water, and second, refining apple to obtain puree, free of large particles of skins and pips. The advantages were the speed of cooking, a few minutes, and the obtained purees with interesting organoleptic characteristics such as a brighter color and a more viscous texture, but less polyphenols.

However, a multifactorial analysis approach such as LCA (Life Cycling Analysis) is needed to take into account not only the consumer acceptability but also the economic, environmental and technical evaluation of this innovative microwave process in order to study its scale up transfer to industry.

#### CRediT authorship contribution statement

**Sylvie Bureau:** Writing – original draft, Supervision, Project administration, Funding acquisition, Formal analysis,

Conceptualization. **Alexandre Leca:** Writing – review & editing, Validation, Supervision, Methodology, Investigation. **Barbara Gouble:** Writing – review & editing, Supervision, Methodology, Data curation. **Caroline Garcia:** Supervision, Methodology, Data curation. **Witold Danelski:** Data curation. **Ewelina Hallmann:** Data curation. **Renata Kazimierzczak:** Data curation. **Dominika Średnicka-Tober:** Writing – review & editing, Data curation. **Ewa Rembiałkowska:** Writing – review & editing, Funding acquisition, Conceptualization. **Carine Le Bourvellec:** Writing – review & editing, Supervision, Project administration, Investigation, Funding acquisition, Formal analysis, Conceptualization.

#### Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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#### Data availability

Data will be made available on request.

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