Comparison of physicochemical properties of commercial UHT-treated plant-based beverages and cow's milk

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Milk or plant-based beverages? (Photo: Gabriela Brändle, Agroscope)

Abstract

A comparison of consumer-relevant physicochemical and technofunctional properties was performed between plant-based beverages (PBBs) and cow's milk treated at ultra-high temperatures. The PBBs' viscosities and pH values were similar to or higher than those in cow's milk. The PBBs were less white, and their mean particle sizes were usually considerably larger than those of cow's milk. Foam heights were quite different, from 41.5 mm to 173 mm at room temperature (milk foam height: 134.8 mm) and 50.9 mm to 203.6 mm at 60 °C (milk foam height: 179.3 mm), with a median bubble size radius (root mean square) of 14.0–149.5 µm (milk bubble size: 18 µm) and 31.0–175.5 µm (milk bubble size: 82.8 µm). Our correlation revealed that phytic acid (PA) might affect foam height at 60 °C, the temperature of interest for the consumption of hot beverages. This may be of interest, as PA might be reduced in these beverages for nutritional reasons.

Key words: milk, plant-based beverages, technofunctional properties, foam height, foamability, phytic acid, proteins.

Introduction

The number of plant-based beverages (PBBs) on supermarket shelves is increasing. To ensure a long shelf life, they are treated at ultra-high temperatures (UHT) and placed beside UHT-treated cow's milk (milk) in the shelves. From a processing point of view, grinding the raw material can be the first major processing step in producing a PBB, usually after pretreatment, such as soaking. Grinding in water is a key step because it generally determines the particle size distribution (PSD) in a PBB. The separation of undissolved materials from watery slurry using different types of separation methods, before or after homogenisation, provides a soluble extract (Sethi *et al.*, 2016). In cases in which such a separation is not allowed, which is mostly the case for protein sources that fall under new food regulations, all material remains in the product. A subsequent heating step, usually a form of UHT treatment that follows the addition of further ingredients, leads to a product with a long shelf life at room temperature (RT).

Homogenisation of PBBs prevents creaming and/or sedimentation during storage. In milk, major proteins (caseins) are colloidal soluble, with a diameter of around 50–500 nm (Fox & Brodkorb, 2008). The PSD of homogenised milk is typically dominated by fat globules < 1.7 µm (Di Marzo *et al.*, 2016). Sethi *et al.* (2016) found that for PBBs, the PSDs after homogenisation were between 5 and 20 µm, which is considerably higher than that in milk. In the case of PBBs, PSDs are often the result of overlaying the PSDs of fat globules and those of nonfat particles. Heat treatment is necessary to ensure microbial stability at ambient temperatures for storage periods of up to 6 months. In general, the final UHT processing of 130–150 °C for a few seconds (e.g. 140 °C for 2.3 s) takes place after homogenisation. PBBs are equally treated with UHT to ensure long shelf life (Sethi *et al.*, 2016).

Different processing designs and compositions (e.g. the addition of further ingredients, such as starch or stabilisers) affect PBBs' product functionalities, such as their foam properties. In recent years, drinks with frothed milk have become increasingly popular, making the foam properties of PBBs of great interest if they are to be used as substitutes for milk. In general, there are two major mechanisms for generating milk foam, one via the injection of gas and the other through agitation. The foaming process directly impacts bubble size distribution (Ho *et al.*, 2019). In coffee specialities, such as cappuccinos or latte macchiatos, air or steam is injected through a nozzle. As soon as gas bubbles are introduced into the system, surface-active molecules start to populate the air–fluid interface. In milk foam, milk proteins typically stabilise the interface (Huppertz, 2010). Insufficient stabilisation leads to coalescence of gas bubbles, thus increasing the bubble sizes and decreasing the bubble count number. Polar lipids can also stabilise foam through the Gibbs-Marangoni mechanism (Bos *et al.*, 1997). However, foam containing both proteins and fatty acids has no additive effect (Wilde *et al.*, 2004). In contrast, small amounts of free fatty acids can destabilise foams, especially at RT.

Processing and formulation can influence the PSD, which plays an important role in a PBB's functional properties, such as its viscosity, separation stability and foamability (McClements *et al.*, 2019; Morales *et al.*, 2015). Fibre content and fibre profile affect viscosity (Dikeman & Fahey, 2006); fat content affects foamability (Ho *et al.*, 2020; Kamath *et al.*, 2008; Walstra, 2003); and protein content affects foaming capacity (Huppertz, 2010). In general, additives can alter rheological properties (McClements *et al.*, 2019) and foamability (Balerin *et al.*, 2007). Starch is supposed to contribute to emulsion stability (Zhao *et al.*, 2019); calcium ions can bind to proteins (Taherian *et al.*, 2008; J. V. Silva *et al.*, 2018) and alter their properties; and salt can affect protein solubility and consequently foamability (Bera & Mukherjee, 1989). Phytic acid (PA) is supposed to bind to proteins and decrease their solubility, enzymatic activity and digestibility (Kamath, Huppertz, *et al.*, 2008; Kumar *et al.*, 2010). PA is supposed to alter a PBB's foaming properties; however, little information is available in the literature (Kumar *et al.*, 2010). In the future, it might be possible to reduce the PA content in the next generation of beverages. Therefore, it would be interesting to investigate in the future whether PA or other components in the formulation of PBBs might influence their physicochemical properties.

The main objective of this study was to analyse the selected physicochemical properties of PBBs and milk in a comparative study with a special emphasis on foam properties. In total, 27 UHT-treated PBBs purchased from main Swiss retailers (80 % market volume) were analysed and compared with two UHT-treated whole-milk samples. Based on packaging data and the performed analysis of the PA content, a second key objective was to reveal the fundamental factors affecting the foam properties of all 27 PBBs, regardless of their raw material sources, to identify the impact of PA and subsequent research topics in the field.

Materials and Methods

Samples

A total of 29 samples were collected from the main supermarkets in Switzerland. Samples included were almond (4), cashew (2), coconut (3), hemp (1), oat (4), rice (5), soy (7) and spelt (1) UHT-treated PBBs. They were compared to UHT cow's milk with 3.5 % fat (two different brands). Before analysis, they were kept at RT (21 ± 1 °C) and freshly opened for analysis. Table 1 displays an overview of the products and their labelled ingredients.

pH

The pH of the liquid samples (50 ml) was measured in duplicate at RT using a SevenMulti pH meter (Mettler-Toledo, Switzerland), which was calibrated before the experiments with buffer solutions of pH 4 and 7.

Viscosity

Measurements of viscosity were taken in duplicate at 20 °C using an MCR 301 rheometer equipped with a DG 26.7 double gap measuring system (both by Anton Paar, Switzerland). A sample of 3.8 ml was transferred into the device, and measurements started after temperature equilibration. Viscosity was measured over shear rates between 0.1 and 500 s^{-1}, with a logarithmic distribution of 56 data points. The values at shear rates of 10 s^{-1} and 100 s^{-1} were used to compare the samples.

Colour

Colour was measured using a CM-700d Konica Minolta (Sensing Europe, B.V.) handheld spectrophotometer. The device was calibrated twice, first with a zero-calibration box and then with a white calibration cap. The sample was poured into a rectangular cell (50 × 38 mm) with an optical path of 10 mm placed in a horizontal position. The measurements were performed in duplicate at RT. The whiteness index was calculated according to the instructions of the International Commission on Illumination's Colorimetry committee (Robertson, 1990).

Particle Size Distribution (PSD)

The particle size was analysed in duplicate using an LS 13 320 Beckman Coulter (Switzerland) laser diffraction particle size analyser. The samples were arbitrarily diluted with demineralised water and injected into a liquid handling unit filled with demineralised water to reach an obscuration value between 8 % and 12 % before the measurements started. The volumetric-weighted PSD q3 and associated distribution parameters were calculated from the intensity profile of the scattered light by the instrument's software using the Fraunhofer optical model.

Phytic Acid (PA)

For PA analysis, 50 ml of each sample was frozen to –20 °C and then freeze-dried with the LyoCube 4–8 LSC Freeze Dryer (Christ, Germany). PA analysis was performed in duplicate using a Megazyme K-PHYT kit (Romer Labs, Butzbach, Germany). The analysis consisted of an extraction of 1 ± 0.01 g of the samples at RT, followed by an enzymatic dephosphorylation reaction and a colorimetric determination of released phosphorus according to the manufacturer's instructions. The samples were measured at 655 nm in a UV5Nano spectrophotometer (Mettler-Toledo, Switzerland).

Foam Analysis

Measurements for the foam analysis were performed using a DFA100 Dynamic Foam Analyzer (KRÜSS, Germany). The camera height was set to 100 mm, and infrared light (λ = 850 nm) was used for height illumination. A 50-ml sample to be foamed was placed in a glass vessel with a diameter of 40 mm and equipped with a 16–40 µm, Ø 30 mm filter (FL4533-G3, KRÜSS, Germany). Air was added to the liquid via the filter, 0.3 L/min for 30 s. The initial foam height (IFH) was measured 20 s after the end of foaming. The continuous foam height measurement was stopped after 90 s. The temperatures used were RT and 60 ± 2 °C to mimic a consumption situation. For the measurement at 60 °C, a 50-ml sample was heated separately until the target temperature was reached (modified according to a previous study; Oetjen *et al.*, 2014). DFA100 software (KRÜSS, Germany) calculated the size distribution of the bubbles and characteristically deviated numbers. For the quantification of the median bubble size (mBS) and foam height instabilities – namely, bubble size instability (BSIS) and foam height instability (FHIS) – mBS and foam height (FH) were taken at 0 s and 90 s after foaming and were calculated according to Equations (1) and (2), respectively:

$$FHIS\ (\%) = \frac{FH\ (90s) - FH\ (0s)}{FH\ (0s)} \times 100, \qquad 1$$

where FH (0 s) and FH (90 s) are the foam heights at 0 s and 90 s, respectively, after foaming, and

$$BSIS\ (\%) = \frac{BS\ (90s) - BS\ (0s)}{BS\ (0s)} \times 100, \qquad 2$$

where BS (0 s) and BS (90 s) are the mBS determined as the radius (root mean square [rms]) at 0 s and 90 s, respectively, after foaming.

Statistical Analysis

Statistical analysis was conducted on all samples from two replicate trials ($n = 2$) using R software (R Core Team, 2013; http://www.R-project.org/).

A correlation test (using the library ggcorrplot) among all products' parameters (inclusive of the contents of fat, protein, vitamins, minerals, salt and PA) and the physical parameters was performed using Spearman's correlation due to the absence of normal distribution. Significance was accepted at $p < 0.05$.

A linear model (lm(foam_height60~phytic_acid)) was applied to test the significant impact of PA on FH at 60 °C. R software was also used for principal component analysis (PCA, library ggplot) and a heatmap (library metabolomics) that included the physical parameters (pH, colour measurement [L*, a*, b*, Whiteness], viscosity [viscosity sheer rate 10, 100 (s^{-1})], particle size [D50, D10, D90], foam parameters [foam height at room temperature and 60 °C (FH-RT; FH-60) and foam height instability at room temperature and 60 °C (FHIS_RT and FHIS_60)] and the PA content).

Results and Discussion

Composition

Table 1 shows the ingredients of the different PBBs and milk as listed on the packaging of each sample, and Table 2 gives an overview of the nutritional composition of the PBBs as indicated on the packaging. The PA concentration for each sample was analysed in our lab.

Table 1 | **Products and Their Ingredients as Listed on the Packaging.**

Sample	Ingredients
Milk 1	bovine milk, UHT, 3.5 % fat
Milk 2	bovine milk, UHT, 3.5 % fat
Almond 1	water, almonds 7 %, sea salt
Almond 2	water, sugar, almonds (2.3 %), tricalcium phosphate, sea salt, stabilisers (carob gum, gellan), emulsifier (sunflower lecithin), vitamins (riboflavin/B$_2$ [0.21 mg/100 ml], B$_{12}$ [0.38 µg/100 ml], E [1.8 mg/ml], D$_2$ [0.75 µg/100ml])
Almond 3	water, almond paste 8 %, raw cane sugar, cooking salt
Almond 4	water, almond (2 %), mineral salt: calcium phosphate, sodium chloride, stabilisers: gellan (soya) and locust bean gum, emulsifier: sunflower lecithin, acidity regulator: sodium bicarbonate, natural flavourings, sweeteners: steviol glycosides, vitamins: D (0.75 µg/100ml) and B$_1$ (0.21 mg/100ml)
Cashew 1	water, cashew paste 6 %, rice flour 3 %, sea salt; all agricultural ingredients come from organic production
Cashew 2	water, cashew kernels 6.5 %, sea salt
Coconut 1	water, coconut extract 8 %, sea salt
Coconut 2	water, coconut extract 6.6 %, raw cane sugar, natural flavouring, sea salt, thickening agent: E 407, stabiliser: E 418; total content: 98.2 % without water, of which sugar types compensate for the majority
Coconut 3	water, coconut milk (5.3 %; coconut cream, water), rice (3.3 %), tricalcium phosphate, stabilisers (carrageenan, guar gum, xanthan), sea salt, vitamins (B$_{12}$ [0.38 µg/100ml], D$_2$ [0.75 µg/100ml]), flavourings
Hemp 1	water, hemp flour 5 %, sunflower oil, corn starch, sea salt
Oat 1	water, whole oats 11 %, sunflower oil, sea-red algae 0.4 %, sea salt
Oat 2	water, whole oats 11 %, sunflower oil, sea salt
Oat 3	water, oats 11 %, sunflower oil, sea salt; all agricultural ingredients come from organic production.
Oat 4	water, oats (10 %), inulin, sunflower oil, tricalcium orthophosphate, maltodextrin, sea salt, stabiliser (gellan gum), vitamins (riboflavin/B$_2$ [0.21 mg/100ml], B$_{12}$ [0.38 µg/100ml], D$_2$ [0.75 µg/100ml])
Rice 1	water, rice powder 8.5 % (rice syrup, rice starch, rice flour), sunflower oil, rice starch, cane sugar raw, rice maltodextrin, sunflower lecithin, thickener: E 407
Rice 2	water, rice 14 %, sunflower oil, calcium-containing red algae (Lithothamnium calcareum) 0.4 %, sea salt
Rice 3	water, rice 14 %, sunflower oil, marine red algae Lithothamnium calcareum) 0.4 %, sea salt
Rice 4	water, rice flour 14 % (Italy), sunflower oil, calcium-containing red algae powder (Lithothamnium calcareum), sea salt
Rice 5	water, rice flour 14 %, sunflower oil, sea salt; all agricultural ingredients come from organic production; contains natural sugars
Soy 1	water, soybeans 8.5 %
Soy 2	water, soybeans 7.2 %, sugar, calcium phosphate, table salt, vitamin B$_2$ (0.1 mg/100ml), vitamin D (0.4 µg/100ml), vitamin B$_{12}$ (0.2 µg/100ml)
Soy 3	water, soybeans 8 %, cane sugar, seaweed Lithothamnium 0.4 %, sea salt
Soy 4	water, shelled soybeans (5.9 %), sugar, tricalcium phosphate, acidity regulator (monopotassium phosphate), sea salt, flavour, stabiliser (gellan), vitamins (riboflavin/B$_2$ [0.21 mg/100ml], B$_{12}$ [0.38 µg/100ml], D$_2$ [0.75 µg/100ml])
Soy 5	water, organic soybeans 9 %
Soy 6	soy drink 99 % (water, soybeans 7 %), calcium phosphate, stabiliser: gellan
Soy 7	soy drink 97 % (water, soybeans 7 %), sugar, calcium phosphate, stabiliser: gellan, natural flavour, salt, vitamin D (0.75 µg/100ml)
Spelt 1	water, spelt flour (11 %), sunflower oil, sea salt

Table 2 | Nutritional Composition of the Plant-Based Beverages Examined in This Study

Sample	Fat	Protein	Carbo-hydrates	Sugar	Fibres	Salt	Phytic acid (PA)
	(g/100 ml)	(g/100 ml)	(g/100 ml)	(g/100 ml)	(g/100 ml)	(g/100 ml)	(mg/100ml)
Milk 1	3.7	3.5	5.0	5.0	0.0	0.10	0.00
Milk 2	3.5	3.2	4.9	4.9	0.0	0.10	0.00
Almond 1	3.3	1.1	<0.5	<0.5	0.7	0.14	33.6 ± 0.9
Almond 2	1.1	0.4	2.4	2.4	0.4	0.14	26.4 ± 1.7
Almond 3	4.5	1.5	3.0	2.5	0.5	0.06	78.9 ± 41.0
Almond 4	1.1	0.0	0.5	0.2	0.0	0.19	34.9 ± 15.9
Cashew 1	2.5	1.2	4.5	2.4	1.6	0.1	85.4 ± 4.6
Cashew 2	2.8	1.0	0.9	<0.5	0.5	0.09	68.8 ± 13.2
Coconut 1	<0.5	1.7	1.3	1.3	<0.5	0.06	20.1 ± 4.7
Coconut 2	2.5	<0.5	3.5	3.5	<0.5	0.10	11.0 ± 1.9
Coconut 3	0.9	0.1	2.7	1.9	0.1	0.13	20.8 ± 2.5
Hemp	2.5	0.0	2.0	0.0	0.0	0.10	74.9 ± 25.2
Oat 1	1.4	0.6	6.0	5.2	<0.5	0.13	12.5 ± 4.6
Oat 2	1.4	0.6	6.0	5.2	<0.5	0.13	18.9 ± 1.3
Oat 3	1.5	0.5	7.0	4.0	1.0	0.09	8.8 ± 11.2
Oat 4	1.5	0.3	6.6	3.2	1.4	0.09	27.3 ± 1.7
Rice 1	2.5	<0.5	11.0	2.0	<0.5	0.00	9.0 ± 4.5
Rice 2	1.5	<0.5	11.0	4.2	<0.5	0.07	3.3 ± 1.1
Rice 3	1.1	<0.5	9.9	7.1	<0.5	0.07	3.6 ± 1.7
Rice 4	1.0	0.5	9.0	7.0	0.5	0.01	7.5 ± 2.1
Rice 5	1.0	0.5	9.0	7.0	0.5	0.01	2.9 ± 1.8
Soy 1	2.0	3.5	1.5	0.0	1.5	0.10	155.7 ± 2.8
Soy 2	2.0	3.5	5.0	4.0	<0.5	0.10	116.4 ± 11.1
Soy 3	1.9	3.1	1.7	0.7	<0.5	0.11	98.1 ± 10.0
Soy 4	1.8	3.0	2.5	2.5	0.5	0.09	271.5 ± 9.2
Soy 5	2.5	4.0	1.5	0.5	0.5	0.02	176.0 ± 16.1
Soy 6	2.0	3.6	<0.5	<0.5	0.5	0.07	143.2 ± 6.8
Soy 7	2.0	3.6	2.3	2.3	0.5	0.09	118.8 ± 4.8
Spelt 1	1.5	0.8	6.2	5.7	<0.5	0.13	24.8 ± 5.3

Note: Data are taken from packaging information.

According to Tables 1 and 2, PBBs show a heterogeneous composition among those with different raw materials and, to a lesser extent, among those with the same raw material source (e.g. almond, coconut, rice). In terms of their nutrient composition, when comparing, for example, nut-based products with cereal-based or legume-based products, the differences in the botanical origin and composition of the raw materials become clear. However, the industrial production processes are not known in detail. Although the processing might be similar for many of the PBBs, the processing parameters usually have a strong impact, for example, on protein and fat release or the PSD of raw material constituents (Table 2). In combination with filtering and high-pressure homogenisation treatments, this could lead to observed differences in the nutritional composition of products based on the same raw material source. Furthermore, it is very likely that the PA content of the beverages will be reduced through processing in the future to increase protein bioavailability. A comparison of the protein content of milk with that of the PBBs shows that most samples have considerably lower protein concentrations, except for products containing soya.

Several PBBs contain additives, such as stabilisers (e.g. gellan [*n* = 6], carob gum [*n* = 1], carrageenan [*n* = 1], guar gum [*n* = 1], xanthan [*n* = 1]), emulsifiers (e.g. lecithin [*n* = 3]), starches (*n* = 2), sugars (*n* = 10) and additional oil (sunflower oil [*n* = 11]). Such additives are usually used to reduce destabilisation effects, such as sedimentation or creaming, or to adjust creaminess or mouthfeel in general. In addition, other properties, such as foaming, can be altered. To improve nutritional value or taste, ingredients like sugars, salts, vitamins and calcium are often added as well. PA, in contrast, is an antinutritional factor often present in seeds that can bind to proteins, resulting in reduced protein bioavailability (Yu *et al.*, 2012). Soya beverages exhibited the highest concentrations, with 154 ± 4.0 mg/100 ml. This is consistent with the results in the literature on soya beans, which shows that they contain higher amounts of PA than other legumes or cereals (Egli *et al.*, 2002). Almond, cashew and hemp samples showed lower concentrations than soya but higher than coconuts, oat, spelt and rice. Rice drinks had the lowest PA concentrations, with 5 ± 1.0 mg/100 ml, and no PA was found in milk, as assumed. An explanation for the low concentration of PA in rice drinks could be either a very low concentration in the raw material or elimination through processing. According to the literature, untreated rice has PA concentrations similar to those of untreated oats, but processing can reduce PA in rice better than in the other sources (Gilani *et al.*, 2012). PA is usually located in protein-rich sites, such as the aleuronic layer in monocotyledonous seeds, and it is well distributed among the kernels in dicotyledonous seeds (Schlemmer *et al.*, 2009).

Physicochemical Characteristics

The common pH value of whole milk at 25 °C is between 6.5 and 6.7 (McCarthy & Singh, 2009). Table 3 shows a comparable mean pH value for the analysed milk samples. In contrast, most PBBs had higher pH values compared to milk. Rice-based beverages had a pH value close

Table 3 | Product Properties of Plant-Based Beverages

Sample	pH (–)	Colour space			Whiteness index (–)	Particle size			Viscosity
		L* (–)	a* (–)	b* (–)		D10 (µm)	D50 (µm)	D90 (µm)	η0 (Pa.s)
Milk 1	6.6	82.2	−1.2	6.4	81.0	0.2 ± 0.0	0.6 ± 0.0	0.7 ± 0.0	2.1 ± 0.0
Milk 2	6.6	83.5	−1.2	6.9	82.1	0.2 ± 0.0	0.6 ± 0.0	0.8 ± 0.0	2.1 ± 0.0
Almond 1	6.7	80.5	−0.1	3.3	80.2	2.1 ± 0.0	5.4 ± 0.1	11.4 ± 0.1	2.5 ± 0.0
Almond 2 (a,b,c)	7.2	72.7	−0.1	9.2	71.2	0.8 ± 0.0	4.7 ± 0.1	21.1 ± 0.8	5.6 ± 0.0
Almond 3	6.7	81.3	0.9	6.6	80.1	2.5 ± 0.4	8.0 ± 0.3	24.6 ± 4.1	2.0 ± 0.0
Almond 4 (a,b,c)	7.5	70.5	−0.5	2.4	70.4	0.6 ± 0.0	2.7 ± 0.2	8.1 ± 0.2	3.2 ± 0.0
Cashew 1	7.1	73.6	0.2	4.4	73.2	0.4 ± 0.0	5.2 ± 0.2	20.2 ± 5.6	2.3 ± 0.0
Cashew 2	6.9	77.3	0.2	3.8	77.0	0.7 ± 0.0	3.3 ± 0.3	10.2 ± 0.9	2.2 ± 0.0
Coconut 1	6.4	52.8	−1.6	−2.3	52.7	0.5 ± 0.0	3.9 ± 0.0	8.8 ± 0.1	1.3 ± 0.0
Coconut 2 (a,b)	7.7	67.5	−0.8	−0.1	67.5	1.3 ± 0.0	4.7 ± 0.2	17.7 ± 1.7	6.7 ± 0.1
Coconut 3 (a,c)	7.2	70.1	−0.9	−0.8	70.1	0.6 ± 0.0	1.6 ± 0.0	4.4 ± 0.0	17.4 ± 0.0
Hemp (d)	6.8	67.2	0.4	13.6	64.5	2.9 ± 0.1	8.6 ± 0.5	17.8 ± 1.6	14.3 ± 0.1
Oat 1 (c)	7.4	72.1	−1.2	6.5	71.3	0.3 ± 0.0	0.6 ± 0.0	0.8 ± 0.0	1.7 ± 0.0
Oat 2	6.8	70.4	−1.0	5.6	69.8	0.4 ± 0.0	0.8 ± 0.0	2.8 ± 0.2	1.7 ± 0.0
Oat 3	6.9	71.3	−1.5	5.6	70.7	0.2 ± 0.0	0.4 ± 0.0	0.7 ± 0.0	1.7 ± 0.0
Oat 4 (a,c,d)	6.8	71.8	−1.6	10.6	69.8	0.8 ± 0.0	2.8 ± 0.0	18.8 ± 0.9	6.5 ± 0.0
Rice 1 (b,d)	7.0	71.9	−0.0	7.6	70.9	0.9 ± 0.0	1.9 ± 0.1	5.0 ± 0.7	13.5 ± 9.5
Rice 2 (c)	8.0	69.9	−1.2	2.8	69.7	0.3 ± 0.0	0.7 ± 0.0	2.1 ± 0.6	1.6 ± 0.0
Rice 3 (c)	7.7	70.2	−1.2	2.4	70.1	0.4 ± 0.0	1.5 ± 0.0	7.6 ± 0.1	1.6 ± 0.0
Rice 4 (c)	7.7	68.5	−0.7	3.9	68.1	0.4 ± 0.0	2.4 ± 0.3	8.7 ± 1.2	1.6 ± 0.0
Rice 5	7.2	71.4	−0.9	2.2	71.3	0.3 ± 0.0	2.9 ± 0.4	9.0 ± 1.0	1.6 ± 0.0
Soy 1	6.6	78.0	−0.4	9.5	76.1	0.2 ± 0.0	0.5 ± 0.0	1.2 ± 0.0	2.4 ± 0.0
Soy 2 (c)	6.6	79.2	−0.7	12.1	75.9	0.2 ± 0.0	0.7 ± 0.1	4.0 ± 0.3	2.2 ± 0.0
Soy 3 (c)	8.0	70.9	−0.3	11.5	68.7	0.2 ± 0.0	0.8 ± 0.0	4.6 ± 0.2	2.5 ± 0.0
Soy 4 (a,c)	7.1	76.6	−0.9	15.4	72.0	0.4 ± 0.0	2.0 ± 0.0	6.7 ± 0.1	5.8 ± 0.1
Soy 5	6.9	79.1	−0.0	11.5	76.1	0.4 ± 0.0	0.5 ± 0.0	2.1 ± 0.0	4.5 ± 0.2
Soy 6 (a,c)	7.3	75.2	−0.8	8.6	73.7	0.2 ± 0.0	0.5 ± 0.0	1.7 ± 0.0	2.2 ± 0.0
Soy 7 (a,c)	7.1	75.4	0.11	9.1	73.8	0.3 ± 0.0	1.9 ± 0.0	12.7 ± 0.1	5.6 ± 0.0
Spelt	6.6	66.5	−1.1	5.8	66.0	1.4 ± 0.1	5.4 ± 0.2	12.4 ± 0.5	2.1 ± 0.2

Note: D_{10}, D_{50}, D_{90}: volume-weighted particle diameter representing the diameter that separates the smallest particles accounting for 10 %, 50 % and 90 %, respectively, of the overall volume.
The standard deviation of pH values and of the values for the colour space and whiteness index are below 2 % with respect to their base values and are not listed in the table for better readability.
a = with stabilisers, b = with emulsifiers, c = with calcium, d = with vitamins.

to 8, which was higher than all the other products we analysed, which is also consistent with the literature (Mäkinen *et al.*, 2015). The pH strongly impacts solubility and thus foaming properties as well (Molina Ortiz & Wagner, 2002). From a manufacturer's point of view, an increased pH promises higher yields. Increased pH values might be a consequence of composition, ingredients used (McClements *et al.*, 2019), processing at a basic pH (Rustom *et al.*, 1991) or differences in the amino acid spectrum found in the raw material sources (McCarthy & Singh, 2009).

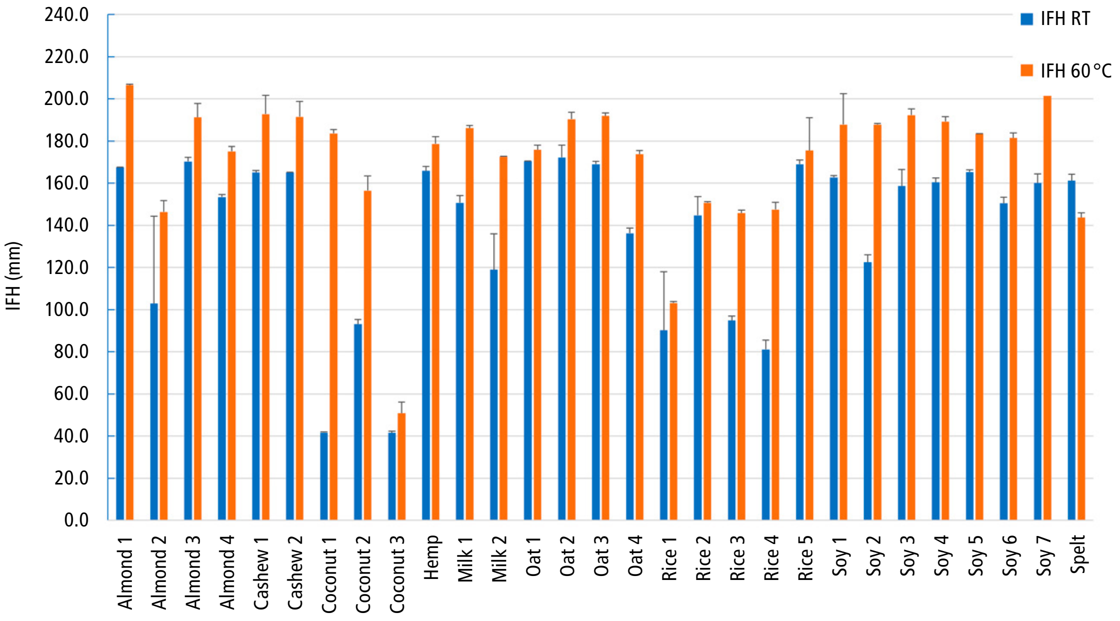
Both milk samples showed higher lightness values (L*: 82.18 and 83.46) than all other samples, which subsequently affected the index of whiteness. The positive values in b* determine the yellow characteristic in milk, and the negative values in a* give it a pale green tone (McClements *et al.*, 2019). As for the PBBs, greenish-yellow tones were present, except for the coconut-based samples, which had a more blueish tone. Milk exhibited the highest whiteness index values for all samples. Except for the two almond-based samples, the index of whiteness for all PBBs was found to be lower compared to milk (52.69–76.97). The literature confirms the values obtained in the present analysis, attributing the difference in the index of whiteness to the ingredients and the process itself (Jeske *et al.*, 2017).

Both milk samples exhibited a median particle size (mPS) of 0.6 µm, which coincides with other studies (Durand *et al.*, 2003; Ho *et al.*, 2019; Jeske *et al.*, 2017; Mäkinen *et al.*, 2015) and can be expected after homogenisation. For PBBs, mPS was determined to be between 0.4 and 8.6 µm. For PBBs, an mPS of up to 20 µm can usually be found (Sethi *et al.*, 2016), which impacts phase separation due to the sedimentation and/or creaming of the PBBs (Taherian *et al.*, 2008). Large particles, if nonfat, can be perceived as sandy, mealy or gritty, which is usually considered undesirable. In PBBs, the overall PSD usually consists of groups of smaller particles, such as solved proteins, and larger particles, which are mainly plant material fragments or fat globules (Ho *et al.*, 2019). The viscosity levels of many samples were comparable to those of milk (2 mPa·s). Only a few samples exhibited considerably higher viscosities; however, this could be attributed to the addition of starch, hydrocolloids or fermentation. Most of the samples containing stabilisers and/or emulsifiers (except soy 6) showed higher viscosity than milk.

Foaming Properties

Initial Foam Height and Foam Height Instability

The effect of foaming temperature on IFH is shown in Figure 1. For foaming at high temperatures, the literature reports that milk proteins start to denature, leading to altered foam properties (Ho *et al.*, 2019; Oetjen *et al.*, 2014; Silva *et al.*, 2008, 2018). In the present study, FH-60



Note: Initial foam height (IFH) at RT (blue) and at 60 °C (red). All data points represent the mean of a duplicate measurement; black arrows indicate the standard deviation.

Figure 1 | Mean Values and Standard Deviation of the Initial Foam Height for Foaming at Room Temperature.

Table 4 | Foam Height Instability and Bubble Size Instability for Foams Produced at Room Temperature and 60 °C.

Sample	Foaming at RT				Foaming at 60 °C			
	FHIS %	StDev %	BSIS %	StDev %	FHIS %	StDev %	BSIS %	StDev %
Milk 1	−19.2	1.4	66.7	3.5	−2.7	0.4	22.2	0.7
Milk 2	−30.6	16.8	n.d.	n.d.	−2.1	0.1	20.0	0.6
Almond 1	−0.2	0.3	60.9	11.3	−4.4	0.4	62.4	3.0
Almond 2 (a,b,c)	−56.7	17.4	n.d.	n.d.	−13.4	18.7	n.d.	n.d.
Almond 3	0.4	0.3	42.1	4.4	−2.3	0.7	29.5	0.6
Almond 4 (a,b,c)	−28.5	6.3	n.d.	n.d.	−8.5	6.0	72.8	4.1
Cashew 1	−0.9	0.7	46.5	7.7	−3.3	0.6	28.0	7.1
Cashew 2	−1.2	1.2	56.3	6.0	−3.0	0.1	34.6	19.2
Coconut 1	−0.1	0.2	n.d.	n.d.	−3.3	0.2	12.7	12.1
Coconut 2 (a,b)	−4.1	4.9	n.d.	n.d.	−0.4	0.1	n.d.	n.d.
Coconut 3 (a,c)	4.3	10.1	n.d.	n.d.	−0.1	0.2	n.d.	n.d.
Hemp	−0.2	0.5	73.8	12.7	−3.1	0.4	52.5	10.5
Oat 1 (c)	−0.1	0.3	22.0	0.5	−2.7	3.3	21.3	4.0
Oat 2	−0.2	0.3	15.4	1.5	−2.8	0.5	10.4	0.3
Oat 3	−0.3	0.4	22.5	3.8	−3.4	0.0	17.9	4.2
Oat 4 (a,c,d)	−0.1	0.2	−66.2	230.4	−1.9	0.1	41.1	4.2
Rice 1 (b,d)	−17.7	13.3	n.d.	n.d.	−1.8	1.8	n.d.	n.d.
Rice 2 (c)	−0.1	0.4	−481.3	761.8	−1.6	0.2	10.7	2.0
Rice 3 (c)	−5.6	1.8	n.d.	n.d.	−1.7	1.8	−84.5	83.0
Rice 4 (c)	−18.9	26.7	n.d.	n.d.	−3.1	1.7	10.3	0.4
Rice 5	−0.4	0.1	20.8	5.7	3.6	9.3	7.1	2.6
Soy 1	−0.1	0.1	29.4	11.2	−2.0	1.2	48.0	2.8
Soy 2 (c)	−54.9	8.1	n.d.	n.d.	−2.9	0.3	58.5	1.5
Soy 3 (c)	−2.1	0.2	36.4	13.8	−2.8	1.3	36.0	4.5
Soy 4 (a,c)	0.4	0.4	66.9	1.6	−2.5	0.0	36.3	29.9
Soy 5	0.2	0.2	69.3	1.5	−1.6	0.4	56.0	3.0
Soy 6 (a,c)	−0.7	0.9	38.1	1.3	−3.1	0.6	46.5	1.2
Soy 7 (a,c)	−0.4	0.1	56.4	0.3	−3.3	1.0	27.5	3.5
Spelt	0.0	0.0	63.2	43.2	−4.1	1.5	22.7	3.1

Note: RT: room temperature. FHIS: change in the foam height at 90 s after foaming, expressed relative to the initial foam height after foaming. BSIS: change of the median bubble size at 90 s after foaming, expressed relative to the initial median bubble size after foaming. Foam height/median bubble size: negative values represent a reduction in foam height / median bubble size; positive values represent an increase. All measurements were performed in duplicate; n.d. = not detected; a = with stabilisers, b = with emulsifiers, c = with calcium, d = with vitamins.

was usually found to be higher than IFH at RT, which accords with the results from the literature (Kamath, Huppertz, *et al.*, 2008). Increased temperatures lead to a decrease in viscosity, which could lead to improved incorporation of gas into a fluid phase (Martínez-Padilla *et al.*, 2015).

Several PBBs showed higher IFH compared to milk at RT. The highest IFH measured at RT originated from an oat drink, although compared to other samples, the protein concentration was rather low. Coconut- and rice-based samples showed deficits in foamability, as indicated by lower IFHs. Table 4 provides an overview of FHIS and BSIS. The FHIS was higher at RT for most of the beverages. Most of the PBBs showed a lower FHIS_RT compared to milk and an equivalent FHIS_60.

Initial Bubble Size and Bubble Size Instability

Figure 2 shows the initial bubble size (IBS) after foaming at RT and 60 °C. PBBs of rice and oat showed the largest mBS, and in contrast, the PBBs of soy had the smallest by trend.

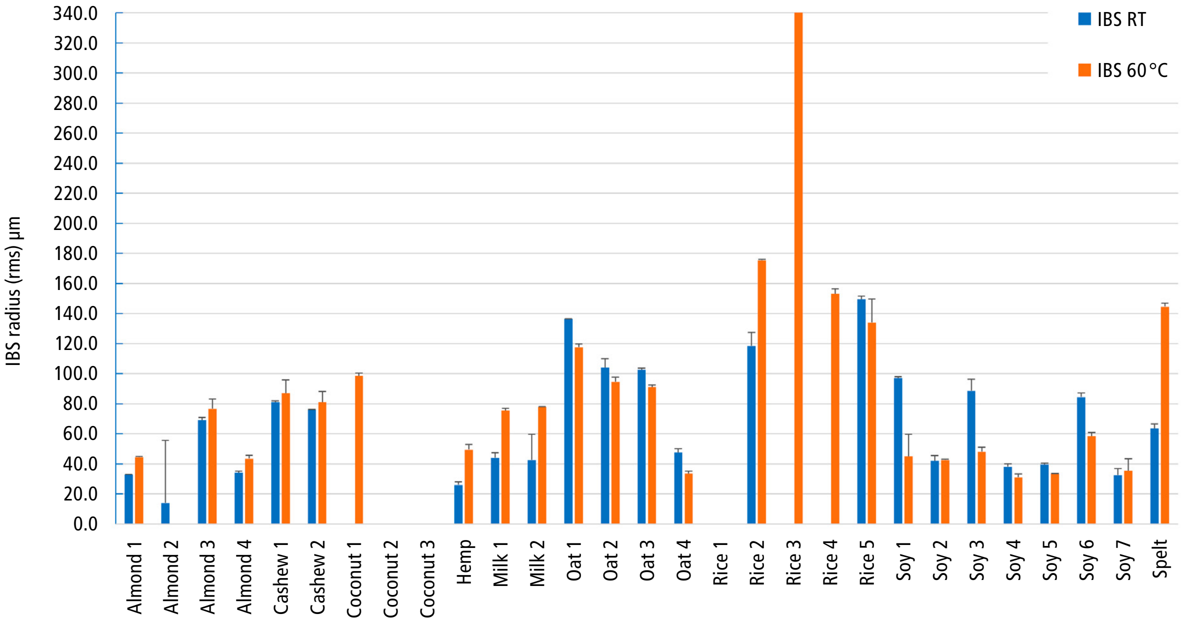
For BSIS, the picture is more complex, as quantitatively larger instabilities can be observed for RT foams compared to 60 °C foams (Table 4). Instabilities vary widely not only across PBBs but also between PBBs of the same raw material type. While the former is likely due to the different compositions of the raw materials, the manufacturing process is likely responsible for the latter. Table 4 shows that in general, the mean bubble sizes increased, usually due to coalescence. Compared to milk, almond and soya PBBs showed increased coalescence over time. BSIS might be a result of the fat content at RT, an observation that has been previously reported in foaming experiments with milk (Kamath, Huppertz, *et al.*, 2008; Kamath, Wulandewi *et al.*, 2008). In some PBBs in the rice and spelt groups, however, the mean bubble size was observed to be reduced. Since spontaneous bubble formation can be ruled out, this effect is probably due to the collapse of larger bubbles.

Correlation Analysis

The correlation analysis in Figure 3 shows that within the plant-based samples, higher fat and protein contents were correlated with lower pH. Furthermore, fat and protein content are important factors in determining the final colour impressions of the samples. With increasing concentrations of fat and protein and increasing particle sizes, the L* and b* values will also increase (McClements *et al.*, 2019). In addition, PSDs affect light propagation, causing a difference in colour (Martínez-Padilla *et al.*, 2015; Stocker *et al.*, 2017). Riboflavin (B_2) is correlated specifically with the b* value, as this micronutrient is well known to add a yellow characteristic (Kearsley & Rodriguez, 1981). PA also showed a significant positive correlation with the a* and b* values of the products, although the origin of this correlation is unclear.

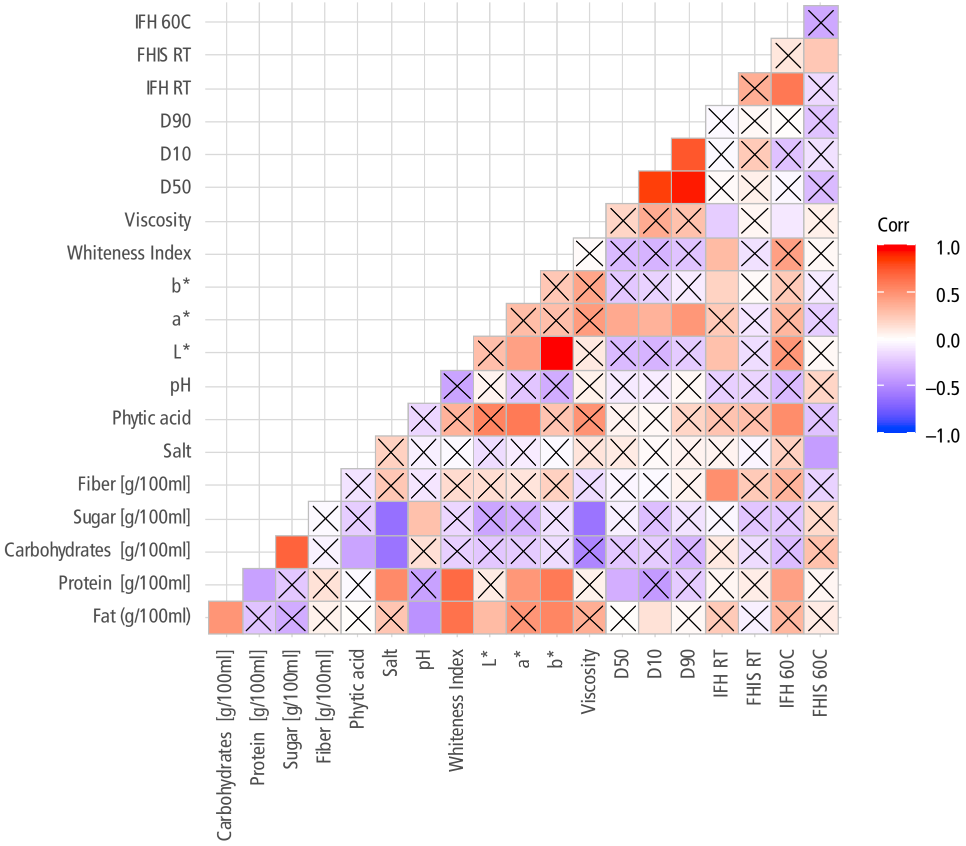
A rather strong positive correlation between PA and protein content suggests a general association with the seeds. A negative correlation with carbohydrates and sugar, which is often an additive, was also observed. The interaction between protein and PA depends on the number of positively charged groups in the proteins available to interact with negatively charged ends in PA. The interactions become stronger at a lower pH (Cheryan & Rackis, 1980). In addition, the concentration of PA depends on the origin of the plant material itself, such as the variety and cultivation conditions.

The protein concentration showed a significant positive correlation with IFH at 60 °C. Fat did not correlate with IFH in either cold or hot foaming. Fat/lipids in general can have a detrimental effect on IFH, but free fatty acids have also been shown to improve foamability (Huppertz, 2010). Olive oil, for example, has been shown to be beneficial for the foaming process (Kamath & Deeth, 2011) at RT rather than for hot foaming (Hatakeyama *et al.*, 2019). In the case of the analysed samples, sunflower oil was often found in rice, coconut and oat samples. Our main finding was a positive correlation for foaming at 60 °C for the PA content and IFH. This is very interesting for application. The mechanism behind this could be that the binding of PA to protein becomes weaker, and more protein is unbound and improves foamability. Darby *et al.* (2017) underlined this in their study, showing that the interaction of PA and protein is temperature dependent. Therefore, it is very likely that a reduction



Note: IBS = initial bubble size radius in µm at room temperature (RT; blue) and at 60 °C (red). All data points represent the mean of a duplicate measurement; black arrows indicate the standard deviation. Samples with very low formability (Coconut 2, Coconut 3, Rice 1) did not reach the camera position for IBS analysis; therefore, they could not be detected.

Figure 2 | Mean Values and Standard Deviation of the Initial Bubble Size for Foaming.



Note: Colours indicate the strength of the correlation. A cross denotes that the significance level was not reached. The significance level was set at $p < 0.05$.

Figure 3 | Correlation Matrix of All Properties.

in PA content might also be beneficial for foamability. Fibre content has a significant positive correlation with IFH for foaming at RT. The mechanism so far remains unclear; however, it seems possible that fibres in natural matrices are associated with proteins, which could award them surface affinity. Additionally, the fibre content was correlated with the viscosity at RT, which is in line with other studies (Dikeman & Fahey, 2006). In addition, viscosity was correlated with FHs-RT and FHs60.
IFH at RT and 60 °C foaming showed a significant positive correlation, indicating that the foaming principles were very similar. Concerning FHIS at RT, a significant positive correlation was found with FH-RT.
We did not include the IBS and BSIS data in our correlation analysis because some beverages did not reach the camera height due to bad foamability; therefore, there were missing values. However, we applied a linear model to see the effect of PA content on IBS and found that it significantly ($p < 0.04$) reduced IBS and BSIS. The reason for this remains unclear. Previous studies have suggested that the addition of chelating agents can alter foam properties (Ward *et al.*, 1997). A previous study found that the addition of calcium did not affect foamability (Deeth & Lewis, 2015). More interestingly, the addition of a calcium chelator like ethylenediaminetetraacetic acid (EDTA) was found to improve foaming properties, as it liberates more protein that can stabilise the air–water interface (Ward *et al.*, 1997). Considering PA as a calcium chelator, it can be speculated whether PA also alters the solubility of proteins, leading to different surface activities. Regarding foaming temperature, foaming at higher temperatures compared to RT results tendentially in smaller IBS (Figure 2). For milk, this has been observed in the literature (Borcherding *et al.*, 2008).
We further performed a PCA (Figure 4) and created a heatmap (Figure 5) with the physical parameters and the PA content to see how the beverages were clustering.
The PCA (Figure 4) showed a medium contribution of PC1 (25 %) and PC2 (23 %) to the overall variability of the plotted data. In the biplot, the distance between milk samples and protein drinks indicates a relationship between them in terms of PA and physical properties, in which a shorter distance indicates similarity and longer distances indicate dissimilarities. The PCA revealed similarities in PA content, FH-RT, FHIS_RT, FH-60, index of whiteness and b* mainly with soy drinks that contain

the highest amounts of PA and good foamability. Further similarities were found in terms of a* and particle sizes between almond drinks 1–3 and the hemp drink, which contained the largest medium particles (D50). In addition, there were also similarities in pH among the three rice beverages (2–4) that showed slightly higher pH values than the other drinks. The two milk samples showed the closest similarities with three oat drinks and six soy samples (total $n=9$) out of the 27 beverages.

Additionally, a heatmap was created to evaluate which PBBs clustered most with the milk samples (Figure 5). A dark red colour indicates the highest values, whereas a bright yellow colour indicates the lowest values for the single categories (the distinct physical properties and PA). The vertical axis groups the physical parameters and PA, and the horizontal axis represents the beverages. The milk samples were surrounded by three oat and three rice drinks, and six soy drinks were in the same cluster; the other beverages (hemp, spelt, almond, coconut and cashew) were not in the same cluster.

One limitation of the study was that we had no insight into the exact processing of the analysed products; therefore, the influence of the processing on the final product characteristics was not determined. It should be considered that many of the analysed PBBs contained stabilisers, which could potentially result in foams with improved properties (Krempel *et al.*, 2019). Foaming was probably further influenced by different processing strategies, processing conditions and prehydrolysis of proteins (Zayas, 1997). These effects were not included in the analysis.

Conclusions

This study investigated selected physicochemical and technofunctional properties, with a focus on the foaming and foam properties of PBBs as compared with milk. In particular, the influence of PA content was analysed. Highly diverse properties were found, even in the same groups of products. However, some products showed similarities with the milk samples according to hierarchical clustering and PCA analysis.

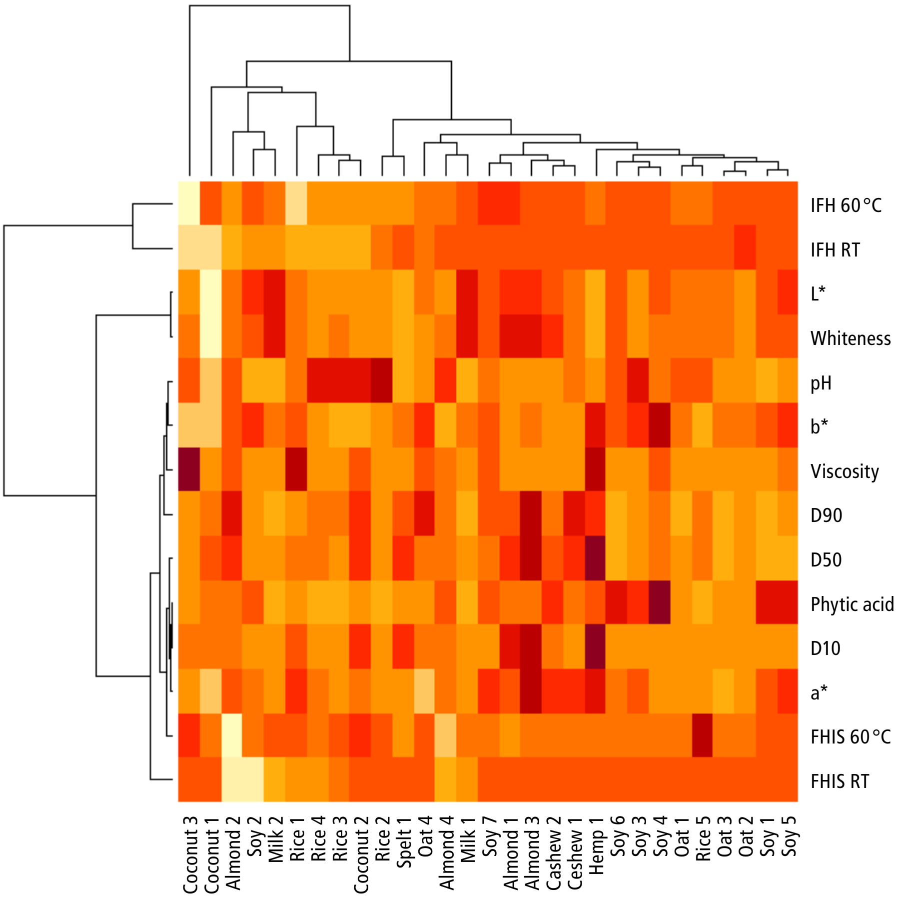
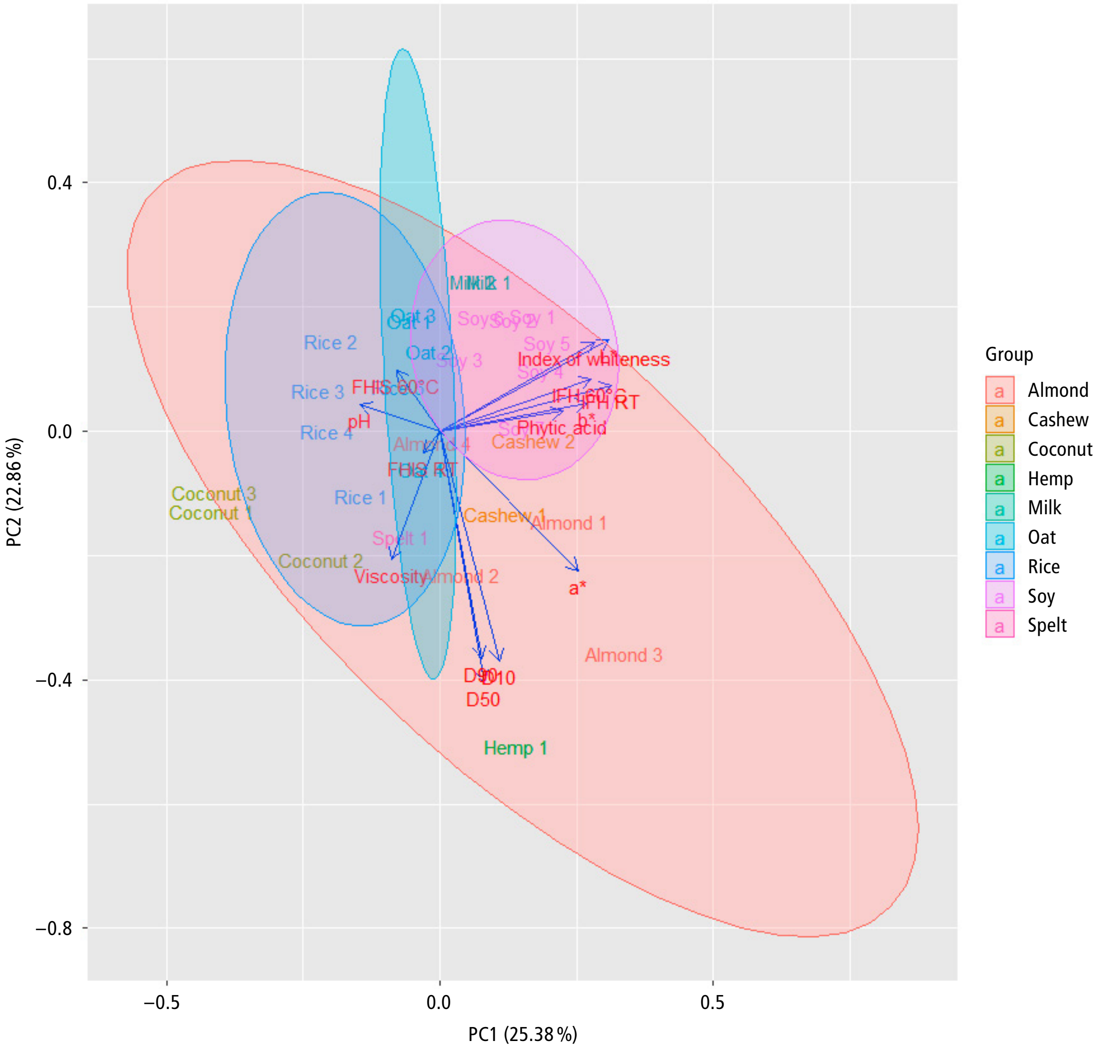


Figure 4 | Principal Component Analysis of the Selected Properties of the Analysed Beverages for the Physical Parameters and Phytic Acid Content.



Note: IFH = initial foam height, FHIS = foam height instability.

Figure 5 | Selected Properties of the Analysed Beverages for the Physical Parameters and Phytic Acid Content.

This was not surprising, as, in contrast to milk, the protein composition and functionality are different and the raw materials are naturally present in a dry state and need to be dispersed in water. Although processing principles might be similar for all the materials (mixing the raw material with water, eventual addition of further additives or enzymes, grinding, removing nondissolved particles, heat treatment), the concrete design of the processes could widely impact, for example, the composition, PSD and extraction of antinutritive substances and, thus, a high number of product features. As a result, product properties can be very different, even when the origin of the raw material is the same. Interestingly, some PBBs showed good foamability, comparable to or even better than that of milk.

Despite these uncertainties (extraction process, additional ingredients), it was suggested that PA potentially affects foaming properties, showing a significant positive correlation with FH-60. This might be due to the weaker binding to proteins at higher temperatures. As the concentrations of proteins and PA in the products are positively correlated, it is impossible to differentiate their individual impacts. Seen from a technological point of view, there is evidence that PA can alter foam properties, particularly hot foams. Thus, PA content can impact foamability, and the interactions of protein and PA might be temperature dependent. The impact of viscosity showed tendentially lower IFH but also less foam degradation with increasing viscosity. Grinding and homogenisation parameters further strongly determine PSD; therefore, mPS strongly depends on processing conditions. In contrast, fibres seem to be beneficial for IFH and low BSIS for cold foams. The mechanism is not clear so far, although fibres and proteins are known to often be associated with surface activity. However, the results suggest that for foam design reasons, it could be worth looking deeper into the role of natural, nonpurified fibres, which are usually found in huge quantities in the raw material sources for PBBs. Here again, we assume the potential for such technofunctional components to be released during the extraction process and suggest using them as natural ingredients to tailor foam and foaming properties in the future.

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Conflict of Interest

The authors declare that they have no conflicts of interest.

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