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Research paper

The sensory and physicochemical properties of an α-lactalbumin enriched whey protein and the contribution of minerals to the sensory profile

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ABSTRACT

Due to the lower proportions of α-lactalbumin existing in whey protein, compared to β-lactoglobulin, less research has focused on its potential application and utilisation. The sensory profile of α-lactalbumin was determined by comparing a commercial whey protein with α-lactalbumin-enriched and α-lactalbumin-deficient samples. When assessed by a trained sensory panel, the α-lactalbumin-enriched sample was associated with a significantly higher slipperiness perception, which correlated with a reduction in instrumental friction and an altered tribology profile. However, this sample was perceived to have significantly higher bitter taste and peppery mouthfeel which, through further investigation, was attributed to be a result of an increased free mineral content as an artifact of additional processing. This study highlights the contribution of α-lactalbumin and minerals to the sensory profile of whey and the need for research to facilitate optimal application and utilisation.

1. Introduction

1.1. Separation of whey protein components

Whey protein exists as a mixture of proteins including: β-lactoglobulin (48-58%); α-lactalbumin (13-19%); glycomacropeptides (12-20%); lactoferrin (2%); lactoperoxidase (0.5%); bovine serum albumin (6%); and immunoglobulins (8-12%) (Etzel, 2004; Madureira et al., 2007). The differing proportions of these proteins have led researchers to speculate that their contribution to the sensory and physicochemical properties of whey protein may vary. Existing research in this area has focused on β-lactoglobulin as the major constituent of whey. However, the other proteins also need characterisation, as it is possible that the different physiochemical and sensory attributes of the other whey proteins may suit different applications. Supporting this is the observation that increasing the α-lactalbumin: β-lactoglobulin ratio is associated with an increase in heat stability (Crowley et al., 2016), better calcium binding (Barone et al., 2020), and decreased bulk density (Barone et al., 2019). The physicochemical differences occurring as a result of increase α-lactalbumin give weight to the suggestion that altering the composition of whey protein could expose different potential applications. Previous literature has detailed a range of methods to separate these components, including membrane fractionation, ion exchange chromatography, and electro-separation (El-Sayed & Chase, 2011; Giles et al., 2026; Nath et al., 2022). It has been noted that these different methods to increase α-lactalbumin are all likely to result in a final product with differing properties (Barone et al., 2019). The authors have previously demonstrated membrane fractionation as a methodology to separate proteins in WPI based upon their molecular weights to isolate two protein streams; an α-lactalbumin enriched (AL-E) filtrate and an α-lactalbumin deficient (AL-D) retentate (Giles et al. 2026). These fractions were yet to be characterised, presenting a novel opportunity to understand the sensory contribution of α-lactalbumin in a commercially relevant whey product.

1.2. Commercial and research applications of α-lactalbumin

One potential use for α-lactalbumin is infant formula: whilst it

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typically accounts for 13-19% of total protein content in whey, its levels in human breastmilk are much higher at around 75% (Kamau et al., 2010). In addition, the amino acid sequences of human and bovine α-lactalbumin are very similar, leading to a strong commercial interest in the separation of α-lactalbumin from whey protein for the production of infant formula aiming to closely emulate the protein profile of breastmilk (Heine et al., 1991; Jakopovic et al., 2016; Ponchon et al., 2024). In addition, α-lactalbumin has a high proportion of the branched chain amino acids leucine and isoleucine (Etzel, 2004): these promote muscle protein synthesis in older adults (Katsanos et al., 2006), meaning there may be additional health benefits from isolation of this protein as highlighted by recent studies into exercise science (Qin et al., 2017, 2019). The high tryptophan content of α-lactalbumin, and its effect on neurological processes (Layman et al., 2018), has also been highlighted as having the potential to support neurological function and sleep in adults. Jakopovic et al., 2016 detailed a range of other potential health benefits of α-lactalbumin including anti-cancer activity, antimicrobial activity, immunoactivity properties and antiviral activity. However, the sensory and physicochemical profiles of α-lactalbumin are not fully understood. This is important with regards to commercial applications: whey protein has some negative sensory attributes that limit consumer acceptance and uptake by different demographics (Bull et al., 2017; Norton et al., 2020). A key driver for disliking of whey protein beverages is mouthdrying (Zhang et al., 2020), which is thought to be a result of increased oral friction resulting from mucoadhesion (Giles et al., 2024). To better understand the contribution of α-lactalbumin to the sensory and physicochemical profile of whey products, this paper will profile and compare the α-lactalbumin enriched and deficient products (AL-E and AL-D) generated in Giles et al. (2026) to each other, and to the source whey protein.

1.3. Potential contribution of minerals to sensory profile of whey protein

In addition to proteins, whey contains a range of minerals, with calcium and potassium typically present at the highest levels (González-Weller et al., 2023). The contribution of these minerals to the sensory profile of whey protein is unknown, with no study existing to the authors knowledge that compares the sensory effect of different mineral levels in a whey medium. However, he contribution of minerals to the taste of water has been thoroughly documented. When dissolved in water, multiple minerals can contribute to bitter, metallic and astringent sensations (Delompré et al., 2019). In water it has been shown that potassium and calcium are more bitter than corresponding levels of sodium (Vanderklaauw & Smith, 1995). In addition, calcium salts have been associated with bitterness, sourness, astringency and metallic perception in water (Ben Abu et al., 2018). This highlights the potential for minerals to influence the sensory profile of whey; thus, demineralisation may present a novel method to improve whey protein's associated taste and mouthfeel.

1.4. Aims and objectives

To the author's knowledge, no comprehensive profiling of α-lactalbumin has been completed meaning the sensory and physicochemical properties of this protein are not known. The primary objective of the study was to compare an α-lactalbumin-enriched (AL-E) protein, an α-lactalbumin deficient (AL-D) protein, and a commercial source whey protein (WP) to understand the contribution of α-lactalbumin to the sensory and physicochemical profile of whey. A secondary objective of the study was to understand the contribution of minerals to the sensory profile of whey protein. The hypothesis is that a reduction in mineral content would reduce bitter and metallic perception.

2. Methodology

2.1. Sample production

A commercial sample of pH neutral whey protein was provided by Volac Whey Nutrition Ltd (Hertfordshire, UK) in a liquid form for membrane fractionation. Fractionation was completed on liquid whey protein using an industrially relevant polyethersulfone membrane on a large pilot-scale membrane fraction unit. This was manufactured by Axium (Swansea, UK) and utilises a 3838 housing with a maximum pressure of 3.5 bar. The rig was operated at 3 bar applied pressure using two Sanitary Ultrafiltration Spiral-Wound Element membrane cassettes. The subsequent filtrate and retentate were passed through a reverse osmosis unit (Axium, Swansea, UK) to increase the concentration of dissolved solids, and spray dried (FT80 tall form spray dryer, Armfield Ltd, Hampshire, UK). This generated AL-E and AL-D samples: this methodology has been described further in Giles et al. (2026). Whey protein from the same liquid batch was provided in a powdered form after commercial spray drying, generating the whey protein (WP) sample. The production process for these three samples has been summarised in Fig. 1.

Analysis was completed on the products in a powdered state and as suspensions. To prepare 10% w/v suspensions, 25 g (±0.5 g) powder was made up to 250 mL using bottled water (Harrogate Spring Water, Harrogate, North Yorkshire, UK) and stirred using a magnetic stirrer for 60 min at room temperature (19 ± 2 °C). This water has average levels of 8 mg/L sodium, 57 mg/L calcium, 19 mg/L magnesium, and 37 mg/L chloride. Samples were refrigerated overnight (16-20 h) prior to use and analysed within 24 h of production.

2.2. Static and temporal sensory profiling

A screened and trained sensory panel (n = 11; females 10, males 1) based at the MMR Sensory Science Centre in the UK (MMR Research Worldwide Ltd, Wokingham, UK) participated in this study. The panel training for this study and data collection sessions were run by the University researchers. The panel were given further training on mouthfeel attributes used for whey protein profiling (minimum 3 h). Sensory evaluation was carried out in a temperature-controlled room (23 ± 2 °C) in isolated booths. A consensus vocabulary was developed by the panel during training and using reference standards (26 attributes; 2 appearance, 4 aroma, 8 taste and flavour, 5 mouthfeel, 7 after-effects). Samples were evaluated in duplicate according to a balanced design using unstructured line scales (scaled 0-100) with appropriate anchors. Panellists were able to see previous scores within the same tasting session. After-effects were scored after a 30 s delay. Samples were presented monadically in black cups (30 mL). Low salt crackers (Carr's water crackers, McVities, UK) and warm filtered tap water were provided as palate cleansers between samples during an enforced break (2 min). Evaluation was carried out under artificial daylight. Sensory questions were presented, and data collected, on Compusense (cloud version, Ontario, Canada).

Sequential profiling was completed using the same trained sensory panel: this followed the methodology described in (Giles et al., 2025a). Suspensions were presented in 8 black cups, with 5 mL of suspension being added to each cup. Panellists consumed the first cup and immediately scored the attributes. The attributes were scored for after-effect after 30 s and again after a further 30 s. The next cup was consumed 90 s after the first. This was repeated cyclically until a total of 8 cups had been consumed. This was completed in an open room with the panel leader instructing panellists when to consume and score the samples. Attributes for assessment were selected based on their temporal relevance for this beverage type, as detailed in (Giles et al., 2025a): this led to limited mouthfeel (n = 4) and taste (n = 2) attributes being scored. Samples were evaluated in duplicate according to a balanced design. Carr's water crackers and warm filtered tap water were provided as

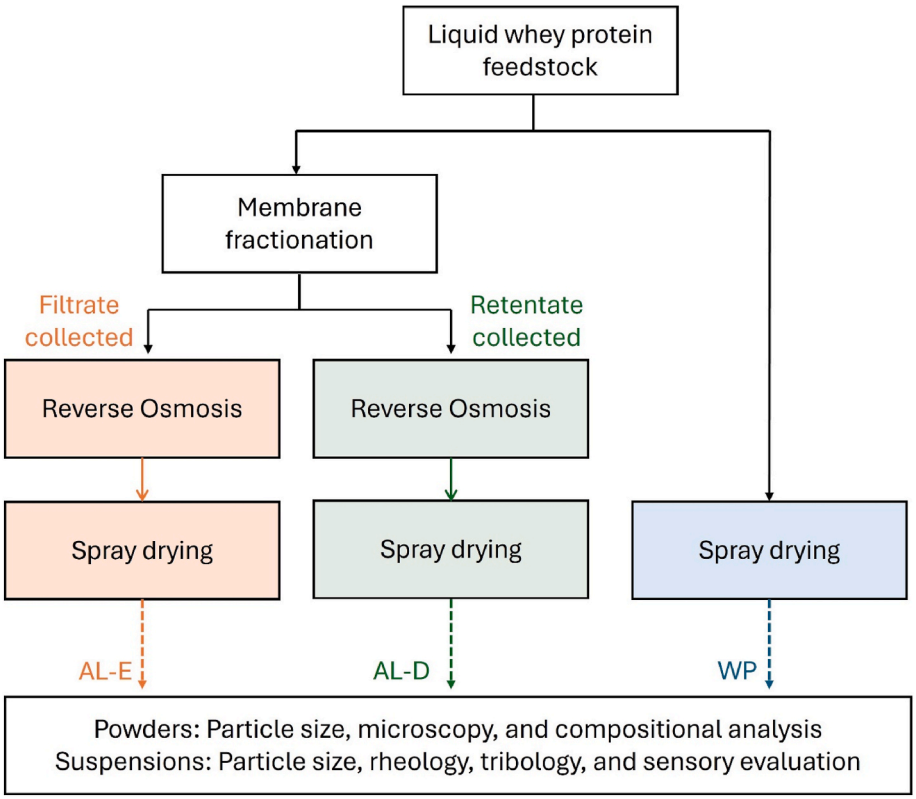

Fig. 1. Schematic to show sample production stages for the whey protein (WP), α-lactalbumin-enriched (AL-E), an α-lactalbumin deficient (AL-D) protein streams. These were analysed in powdered form and as 10% w/v suspensions.

palate cleansers between samples during an enforced break (2 min). Evaluation was carried out under artificial daylight. Sensory questions were presented, and data was collected on Compusense (cloud version, Ontario, Canada).

2.3. Rheology

Rheological properties of whey protein samples were analysed using an oscillatory rheometer (MCR 302, Anton Paar, St. Albans, UK) fitted with a 40 mm diameter smooth rotating plate adjusted to 25 °C. Work was completed in a temperature-controlled room (19 ± 1 °C) with samples acclimatised to room temperature for 1 h prior to recording. After loading the sample onto the lower plate surface, a rest time of 5 min prior to measurement was established for sample relaxation and temperature equilibration. Amplitude sweeps of the samples were obtained by applying an oscillation at a frequency of 1 Hz for strain values ranging from 0.01% to 10% in 12 steps. A strain of 10% was then chosen in the linear viscoelastic region for frequency sweeps, where frequency was varied from 100 to 0.01 Hz. Viscosity was recorded through measurements from shear rates of 0.001-1000 s^{-1} in 42 logarithmic steps.

2.4. Tribology

Tribological measurements were performed with the oscillatory rheometer (MCR 302, Anton Paar, St. Albans, UK) equipped with a tribology cell attachment (T-PTD200, BC12.7, Anton Paar, St Albans, UK). A ball-on-three-pin tribo-pair with a glass probe and three polydimethylsiloxane (PDMS) pins (6 mm pin height), inclined at 45° to the base, was used. Analysis was completed in a temperature-controlled room (19 ± 1 °C) with samples acclimatised to room temperature for 1 h prior to recording. Temperature was controlled at 25 °C and a normal force of 1 N was applied. To measure the friction coefficient, samples were added to the tribology cup to the level of the top of the pins and the friction coefficient was measured as a function of sliding speed. Sliding speed between 1E-05 m/s and 1 m/s was used as the measurement window. One measurement consisted of three runs using the same pins, after which the pins were replaced. The data from the second run was selected for further analysis.

2.5. Microscopy

Scanning electron microscopy (SEM) was performed at 5-20 kV using the four quadrant back scatter electron detector on a SEM Stereoscan 360 (Cambridge Instruments, UK). Samples were mounted onto small SEM aluminium stubs via sticky conducting carbon tabs and the sample excess removed through tapping. Samples were then sputter coated with a thin layer of gold, using an Edwards S150B Sputter Coater, prior to examination in the SEM. Images were taken in four areas of the sample stub at a range of magnifications: images chosen were representative of the larger pool of images selected.

2.6. Particle size

Particle size of dry powders was measured using a Malvern Mastersizer 3000 (Malvern Instruments, UK) with the Mastersizer software (version 3.81) and Aero S attachment, connected to a DustControl DC 1800 Eco vacuum. Particle size of suspensions was mirrored using the Hydro EV attachment.

2.7. Mineral composition

Mineral composition was determined by ALS (Cambridgeshire, UK) following a UKAS-accredited standardised protocol of digestion in 50% nitric acid to decompose the organic matter, followed by the addition of an internal standard and subsequent dilution. Levels of calcium, potassium, magnesium, phosphorus, iron and zinc were then determined by

multi-point calibrations on a Thermo iCAP 7600 Duo Inductively Couple Plasma Optical Emission Spectrometer.

2.8. Colorimetry

Colorimetry of the powders was determined by measuring the Commission Internationale d'Eclairage CIE LAB coordinates (L*, a* and b*) using a CR-400 Chroma Meter (Konica Minolta, Tokyo, Japan) with a Granular Materials Attachment (CR-A50) from the same supplier. A white calibration tile was used to calibrate the instrument before colour measurements.

2.9. Statistical analysis

All instrumental data was analysed using Excel (version 2312). For rheology of suspensions, three analytical repeats were taken at each recording session and samples were prepared on two separate days, leading to six values being obtained for each suspension. For tribology, particle size and colorimetry, each sample was prepared in triplicate and the average of the three datasets used for analysis. Sensory assessment was completed in duplicate according to a balanced design, as is standard for single sip sensory profiling. Sensory questions were analysed using SenPaq (Qi Statistics, Kent, UK). A mixed model two-way ANOVA was used where the sample was the fixed effect and the panellists the random effects, with both effects tested against the sample by panellist interaction. Tukey HSD tests were used for multiple pairwise comparisons to assess significance between samples, at a significance value of $p < 0.05$.

3. Results & discussion

The protein profile of the powders was discussed in Giles et al. (2026): here it was shown that the AL-E sample contained 53% α-lactalbumin and 9.6% β-lactoglobulin, compared with AL-D which contained 18.9% and 66.6% of these proteins, respectively (Table 1). Whilst these are not pure samples, the high proportion of α-lactalbumin present in the AL-E sample, over double the amount in commercial source whey protein (WP), enables it to give strong indications of the sensory and physicochemical properties of this protein. The authors recognise that these changes could also be a result of decreasing the β-lactoglobulin content, and thus will discuss the results in the context of increasing the α-lactalbumin:β-lactoglobulin ratio (Table 2). It was reported previously that the protein level between the three samples was comparable (Giles et al. 2026), however, future studies could also investigate the levels of other macronutrients such as carbohydrate, fat and lactose (Barone et al., 2019; Ponchon et al., 2024).

3.1. Physicochemical properties of powders

3.1.1. Particle size

When in a powdered form, WP had a significantly higher proportion of large particles compared with the AL-E and AL-D samples, as shown by the right-hand shift in the curve for the start material (Fig. 2). It is probable that this reflects differences in production during the spray drying stage (highlighted in Fig. 1), as the WP material was industrially agglomerated after spray drying, following standardised commercial practices. Whereas, the AL-E and AL-D samples were spray dried at pilot scale, with a higher particle residence time, correlating with a smaller particle size (Zbicinski et al., 2002). Whilst every effort was made to replicate commercial conditions, differences in equipment size were unavoidable and are likely to have led to a finer powder being produced. This is important commercially as an increase in particle size has been previously linked to improvements in porosity, solubility and wettability (Onwulata et al., 2004; Barone et al., 2019), meaning that the AL-E and AL-D might be more challenging to dissolve in water. This has led to many commercial practices incorporating agglomeration into production lines, but it was not possible to complete this in the current study. Additional details of the full distribution of particle size data have been given in Supplementary Table 1 (ST1).

Table 1
Percentage of protein present in whey protein powder (WP), an α-lactalbumin enriched (AL-E) sample, and an α-lactalbumin deficient (AL-D) sample produced via membrane fractionation of whey protein. Estimates provided for glycomacropeptides (GMP), α-lactalbumin (α-LA), β-lactoglobulin (β-LG), immunoglobulins (IgA and IgG), bovine serum albumin (BSA) and lactoferrin (LF). Adapted from Giles et al. (2026).

Sample	% α-LA	% β-LG	% GMP	%IgA	% IgG	% BSA	% LF
WP	22.70	59.70	14.20	0.23	1.60	1.60	0.04
AL-E	53.20	9.69	37.00	0.00	0.00	0.13	0.00
AL-D	19.00	66.60	11.40	0.22	1.32	1.43	0.04

Table 2
Estimated β-lactoglobulin:α-lactalbumin ratio in whey protein (WP), an α-lactalbumin enriched (AL-E) sample, and an α-lactalbumin deficient (AL-D) sample produced via membrane fractionation of whey protein.

Sample	β-lactoglobulin:α-lactalbumin ratio	α-lactalbumin:β-lactoglobulin ratio
WP	1 : 2.62	0.38 : 1
AL-E	1 : 0.18	5.49 : 1
AL-D	1 : 3.50	0.28 : 1

3.1.2. Microscopy

To better understand the physical structure of the powders, SEM microscopy was completed at two different magnifications (Fig. 3A–C and D-E, respectively). At 200× magnification the WP showed a larger particle size compared with the AL-E and AL-D sample (Fig. 3A–C), which is consistent with the particle size data presented in Fig. 1 and an anticipated effect of agglomeration. When a higher magnification (1000x) was used, differences in the surface characteristics could be seen between AL-E and AL-D. The AL-D sample (Fig. 3E) showed a small spherical shape, with a range of particle sizes. However, the AL-E sample (Fig. 3D) has a complex shape including concave surfaces, with little of the smoothness observed in Fig. 3E. Whilst the difference in spray drying conditions provides an explanation for the difference between WP and AL-E and AL-D, it is noted that AL-E and AL-D were spay dried following the same conditions. Thus, the difference in surface structure between these two samples cannot be explained as artifacts of production methods. A similar observation of fragmented, shell-like structures has been previously reported for α-lactalbumin enriched whey protein, irrespective of production methodology (Barone et al., 2019): here, it was attributed to poor mechanical stability of the powder during spray drying. Differences in the physical appearance of the powders were also noted by researchers: the AL-D sample was the palest in colour, as characterised with colorimetry recordings (Supplementary Fig. 1). This is in line with previous literature, where whiteness has been shown to increase with decreasing particle size (Barone et al., 2019).

3.2. Physicochemical characteristics of suspensions

3.2.1. Particle size

Once prepared as suspensions with bottled water (10% w/v), the particle size results were in direct contrast to that of the powders: WP had the greatest proportion of small particles, whereas AL-E and AL-D had a high proportion of particles larger than 50 μm (Fig. 4). One explanation for the increased particle size in suspension is that the smaller powder particles (AL-E and AL-D) had a greater surface-area to volume ratio, leading to a greater influx of water and swelling when in suspension. Another possibility is that this reflects increased levels of aggregation and turbidity in the suspensions, as a result of altered charges on the particles due to membrane fractionation. To enable

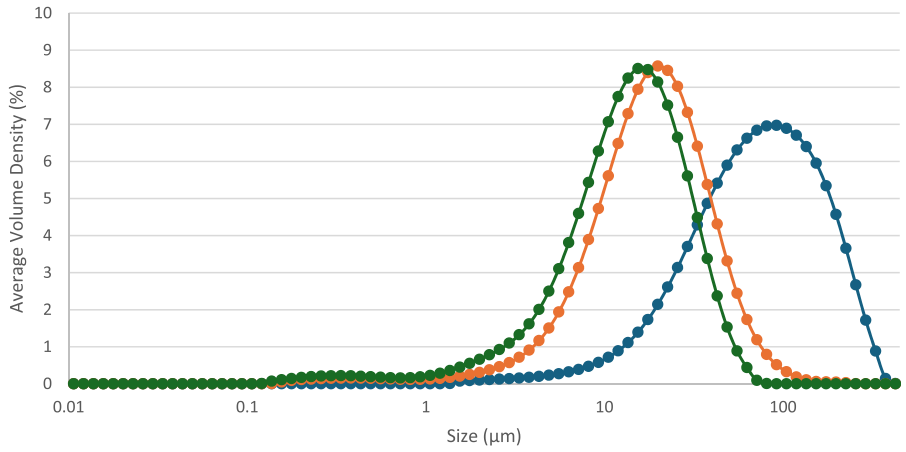

Fig. 2. Average particle size distribution curves from triplicate samples for whey protein (WP) powder (blue), α-lactalbumin enriched (AL-E) protein (orange) and α-lactalbumin deficient (AL-D) protein (green) powders produced during membrane fractionation of whey protein.

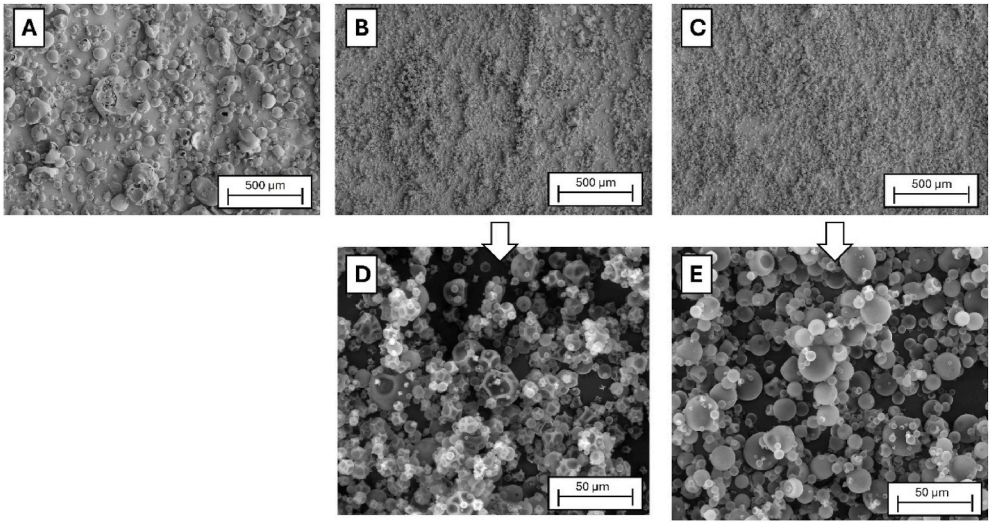

Fig. 3. SEM of powders at 200× magnification; [A]: whey protein powder (WP); [B]: α-lactalbumin enriched protein (AL-E) powder; [C]: α-lactalbumin deficient protein (AL-D) powder. SEM of powders at 1000× magnification; [D]: α-lactalbumin enriched protein (AL-E) powder; [E]: α-lactalbumin deficient protein (AL-D) powder. Scale bars represent 500 µm [A-C] and 50 µm [D and E].

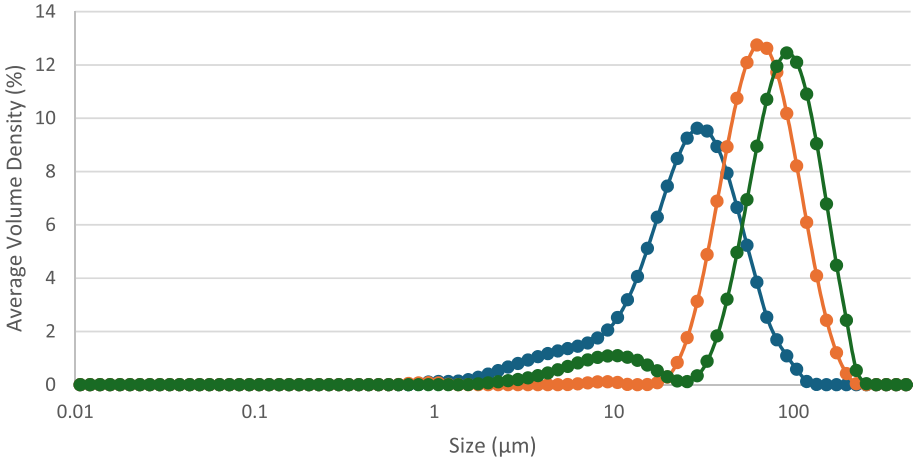

Fig. 4. Average particle size distribution curves from triplicate samples for 10% w/v suspensions of whey protein (WP) (blue), α-lactalbumin enriched protein (AL-E) (orange) and α-lactalbumin deficient protein (AL-D) (green) produced during membrane fractionation of whey protein.

representative recordings, suspensions were prepared in a consistent manner for all methodologies (on a magnetic stirrer for 60 min). Whilst

all suspensions were visibly dissolved after this time and no separation was observed in a settling test (data not reported), it is possible that non-dissolved larger particles present in AL-E and AL-D are skewing the data towards a larger particle size distribution (with a shift to the right) in Fig. 4. This is a potential limitation of particle size recordings, which the researchers have tried to minimise through controlled preparation protocols. This preparation protocol has previously been shown to be sufficient for these products; thus, whilst acknowledging the limitations, we theorise that this is a result of inherent differences in reaction with the water and hydration process, rather than an artifact of non-dissolved powder. Thus, we also suggest that whilst recognising the limitation in our study that different spray drying practices were used for WP compared to the other two samples, we do not anticipate that this will have affected results as all suspensions were visibly dissolved following an established protocol. The association between particle size and sensory perception in whey protein beverages is unclear meaning it is unknown if this difference will be perceivable to consumers, which could be an avenue for further research. Additional details of the full distribution of particle size data have been given in Supplementary Table 2 (ST2).

3.2.2. *Rheology profiles of suspensions*

The rheology of 10% w/v suspensions was recorded across a range of shear rates. Minimal shear thinning was observed at lower shear rates, indicated by the near-horizontal line shown for all three samples (Fig. 5). This remained horizontal until shear rates in excess of 100 s^{-1} where shear thickening was seen (Fig. 5). This is in keeping with previous literature on whey protein, where shear thickening has been attributed to both particle clustering (Sağlam et al., 2013) and instrumental limitations associated with the selected geometry leading to secondary flow effects (Ewoldt et al., 2015). Viscosity at 50 s^{-1} is commonly used to represent shear rates experienced during swallowing and thickness perception (Wood, 1968; Chojnicka et al., 2008; Chojnicka-Paszun et al., 2014). In the current study viscosity at 51.8 s^{-1} has been used, as this is the closest value recorded by the instrument. At this shear rate, the difference in viscosity between the samples was <0.5 mPa. Research into just-noticeable-differences (JND) in viscosity has shown that an increase from 45 to 83 mPas was the average JND (Withers et al., 2013). In addition, a correlation between viscosity and thickness perception has been reported for highly viscous drinks: in a study using tara gum solutions, viscosity was strongly related to perceived thickness, however it is noted that the solutions ranged from 10 to 320 mPa at 50s^{-1} (McCrickerd et al., 2012), whereas the samples in the current study were 1.5-2.5 mPa at the same shear rate. Therefore, it is unlikely that the small viscosity differences between the samples would have been perceivable.

3.2.3. *Tribology profiles of suspensions*

Tribology, when combined with rheology, can be used to more accurately portray the experience of oral processing, giving a better representation of the system properties of products (Pradal & Stokes, 2016). It has been previously shown to correlate well with sensory perception of whey protein beverages (Chen & Stokes, 2012; Giles et al., 2025b). When comparing the tribology profiles of the suspensions, large differences were seen (Fig. 6). At all sliding velocities measured, the AL-E was associated with lower oral friction compared with WP and AL-D (Fig. 6). To account for the small differences in viscosity between the samples (discussed above), friction was also presented against sliding velocity multiplied by the viscosity at 1000 s^{-1} (Fig. 6B). This practice enables the comparison of relative lubrication between suspensions with diferrent viscosities, as discussed previously by this research group and others (Giles et al., 2025b; Kew et al., 2021). When corrected for viscosity, the relationship between the suspensions was still seen (Fig. 6), suggesting that the difference in friction was not a result of changes in viscosity. Thus, it is likely to be the result of microstructural differences in the composition of suspensions. Previous research has shown a negative correlation between instrumental friction and slipperiness perception (Giles et al., 2025b; Chojnicka-Paszun et al., 2014), but the relationship with other sensory attributes remains unclear. There is limited research suggesting a correlation between friction coefficient and astringency perception for tea (Rossetti et al., 2009), however more research is needed to understand what size of difference can be perceived by consumers. Thus it is unclear if the differences seen between the samples will be below the level of detection. This highlights the need for further research into the real-world applications of tribological recordings, as tribological understanding is yet to reach standalone predictive capabilities. The authors suggest that instead tribology data should be considered alongside other physicochemcial parameters, as has been demonstrated in this study.

3.3. *Sensory profile of α-lactalbumin-enriched and -deficient whey protein beverages*

3.3.1. *Descriptive sensory profiling*

Descriptive profiling was completed by a trained sensory panel to understand the impact of increasing the α-lactalbumin: β-lactoglobulin ratio in whey protein on the sensory profile. This was elucidated through comparisons of WP, AL-E and AL-D in single sip sensory analysis. The panel assessed 24 attributes developed by a consensus vocabulary for aroma (n = 4), taste and flavour (n = 7), mouthfeel (n = 5), and after-

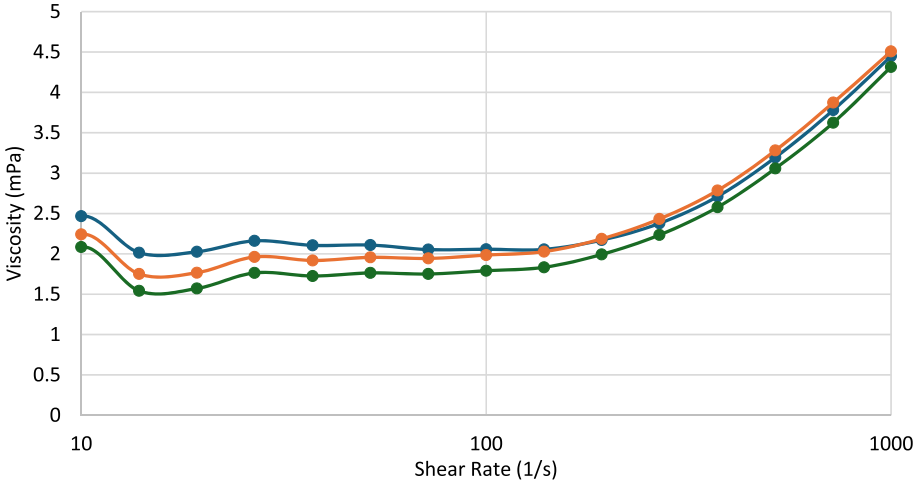

Fig. 5. Viscosity (mPa) across a range of shear rates (10-1000 s^{-1}) for 10% whey protein suspensions of the whey protein (WP) (blue), α-lactalbumin enriched protein (AL-E) (orange) and α-lactalbumin deficient protein (AL-D) (green) produced during membrane fractionation of whey protein.

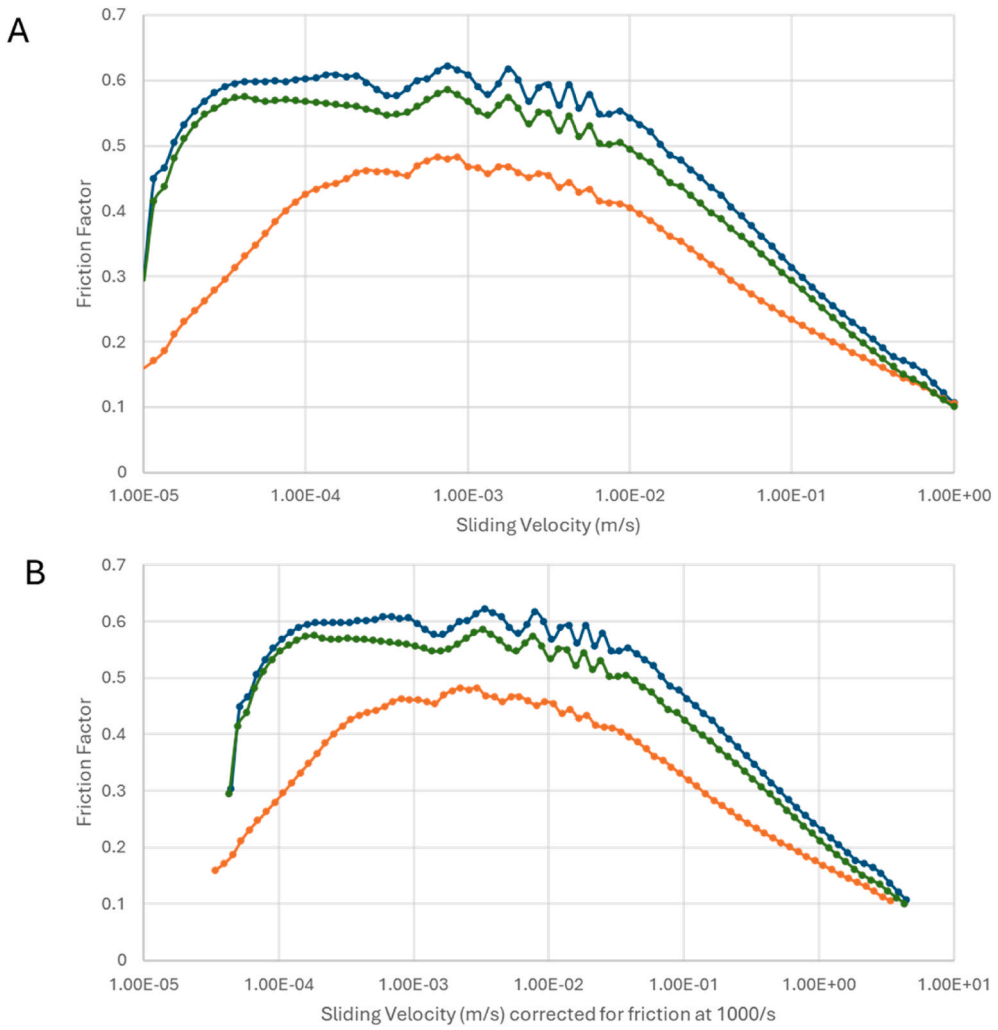

Fig. 6. Lubrication curves for 10% whey protein suspensions of whey protein (WP) (blue), α-lactalbumin enriched protein (AL-E) (orange) and α-lactalbumin deficient protein (AL-D) (green) powders produced during membrane fractionation of whey protein. Created using data collected on the second instrumental run; [A]: as a function of sliding speed; [B]: as a function of the frictional parameter (speed * viscosity at 1000 s^{-1}).

effect (n = 8). The references used for these attributes are detailed in Supplementary Table 3 (ST3) and the mean scores for each are given in Supplementary Table 4 (ST4). Of the four aroma attributes investigated, WP scored significantly higher for dairy, mushroom and cheesey aromas compared with AL-E and AL-D samples, whereas sweet aroma did not vary significantly between the samples.

With regards to taste, significant differences between the three samples were seen for all taste and flavour attributes investigated, with the exception of cheesey perception. The only attribute where AL-E was significantly higher than both other samples was bitterness (Fig. 7A): AL-E had a mean score of 28.5, which was approximately double that of WP and AL-D. The AL-E sample also had a distinct tingly/peppery mouthfeel that was virtually absent in the WP and AL-D samples (Fig. 7B). Both bitterness and the tingly mouthfeel sensation remained significantly elevated in AL-E post swallowing, seen in the after-effects (Fig. 7C). Although the authors are not aware of previous literature characterising the sensory profile of α-lactalbumin, it is known that α-lactalbumin has a high leucine content, which has been previously profiled as bitter (Delompré et al., 2019). It is, therefore, possible that the higher leucine content may contribute to the increased bitterness of the AL-E sample. An alternative possibility is that membrane fractionation may have enriched the mineral content as well as the α-lactalbumin in the AL-E sample which may be contributing to the bitterness. This was investigated in Section 3.4. Future research could further investigate this by characterising pure α-lactalbumin samples, however pure samples are not generated using membrane fractionation, and it is known that the production method will alter the profile of the product generated (Barone et al., 2019): thus analysis of a pure sample was outside the scope of the current study.

Both AL-E and AL-D samples scored significantly lower than WP for powdered milk and mushroom flavour, as well as powdered milk aftertaste (Fig. 7A and B). Whilst this may appear to indicate inherent characteristics of the proteins, a likely interpretation of these results is that the volatile compounds responsible for powdered milk flavour may have been reduced in the fractionation process. Alternatively, this may reflect changes to the mineral profile as a result of ultrafiltration (Section 3.4). This finding is of consequence for commercial applications where a dairy note can either be desired (such as in a pudding or custard) or disliked (as in many protein drinks). Future research could use these samples in a consumer context to investigate any association between these flavour changes and liking, leading to the optimisation of production processes to either enhance or diminish this flavour.

Mouthfeel was of particular interest in these samples as mouthdrying is a known driver of disliking for whey protein-fortified products (Norton et al., 2020; Zhang et al., 2020). Perception of mouthcoating and body were both significantly higher in the WP and AL-E compared with AL-D (Fig. 7B). This aligns with viscosity recordings where WP and AL-E were both of higher viscosity than AL-D at the shear rate of 51.8 s^{-1}

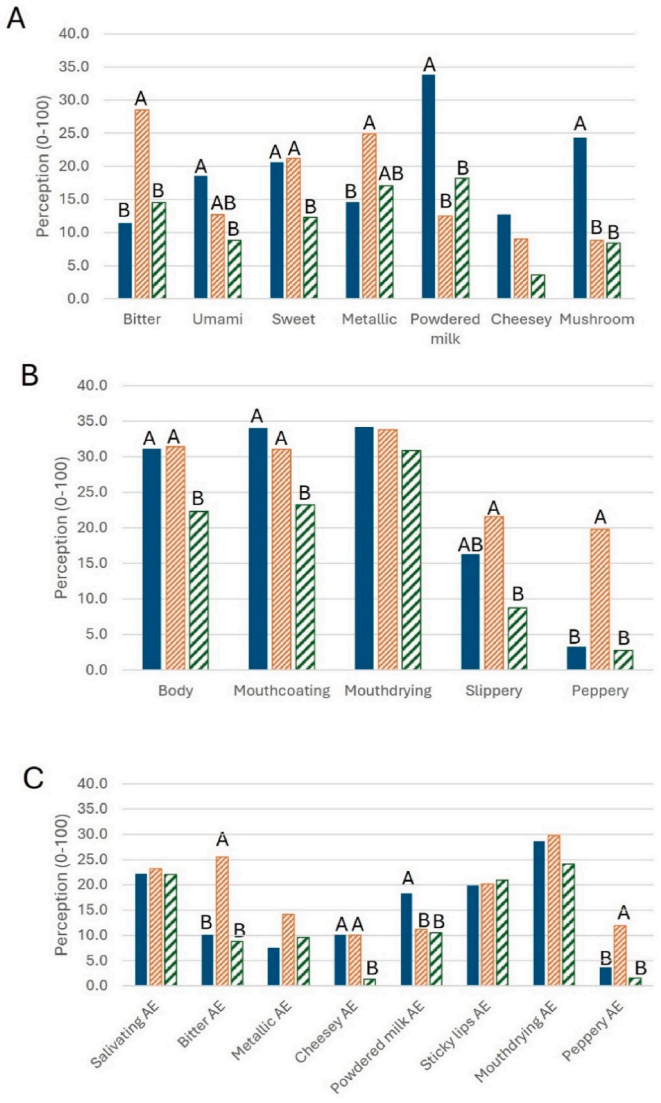

Fig. 7. Mean scores for sensory perception of 10% whey protein (blue), α-lactalbumin enriched protein (orange, thin stripes), and α-lactalubmin defficient protein (green, thick stripes). Samples were scored by a trained sensory panel for [A]: taste and flavour; [B]: mouthfeel; [C]: after-effects scored 30 s after consumption. Attributes which showed significant differences ($p < 0.05$) were compared using Tukey to perform multiple pair-wise comparisons.

(Fig. 5), showing good translation between instrumental and sensory findings for this measurement. There was also a significant difference in slippery perception with AL-E scoring significantly higher than AL-D (Fig. 7C). This is in alignment with tribology profiles where AL-E was associated with significantly lower friction levels across all sliding speeds investigated (Fig. 6). This difference could not be explained by changes in viscosity and instead is thought to represent microstructural differences in the movement of proteins within the liquid. This relationship with slipperiness perception suggests that this attribute sufficiently represents tribology profiles for these beverages, as suggested in Giles et al. (2025). No significant differences were seen between the samples for mouthdrying perception (Fig. 7B). However, mouthdrying has been previously shown to display temporal variability (Bull et al., 2017; Methven et al., 2010), hence required further investigation through sequential profiling before conclusions could be drawn about the effect of protein profile on this attribute.

3.3.2. *Sequential profiling*

As previously discussed, some sensory attributes are known to display temporal variability in whey beverages (Methven et al., 2010), meaning that their intensity significantly changes with repeated consumption. An example of this is whey protein-associated mouthdrying (Bull et al., 2017; Methven et al., 2010). This is important to consider in best representing the sensory experience of consumers where more than one sip of a product is anticipated (Giles et al., 2025a). To investigate this, sequential profiling was completed. The aim of the study was to understand the contribution of α-lactalbumin to the sensory and physicochemical profile of whey. Thus, it was of interest to understand the effect of increasing the β-lactoglobulin: α-lactalbumin ratio, as in the AL-D sample (1 : 3.50 compared with 1 : 2.62 in WP). These values are highlighted in Table 2. Due to the stark difference in sensory profile shown in the AL-E sample compared with the other samples (Section 3.3.1) this was not included in the sequential profile, to allow differences between the WP and the AL-D sample to be fully investigated. As a temporal method, sequential profiling is ideally suited to discriminate small differences between samples, the inclusion of vastly different samples can skew the data and supress smaller differences. There were no significant differences due to repeated consumption for any of the three flavour attributes assessed: sweetness, bitterness, and powdered milk perception (Fig. 8A, B, and 8C, respectively). However, powdered milk flavour was significantly greater in the WP than in the AL-D sample, which was consistent with the single-sip profiling.

Of the four mouthfeel attributes assessed with sequential profiling (mouthcoating, mouthdrying, slippery, and salivating), only mouthcoating increased significantly with repeated consumption (Fig. 9). This is in keeping with the literature on similar products (Giles et al., 2025a) and highlights the need for temporal investigations to fully understand the sensory profile of these beverages. However, both samples displayed similar build-up and there was no significant difference between them ($p = 0.131$). Mouthdrying was also anticipated to build-up with repeated consumption, as demonstrated previously (Bull et al., 2017; Methven et al., 2010). Contrary to expectations, in the current study the build-up in mouthdrying perception with repeated consumption did not reach statistical significance. The average perception score increased by > 15 points from the first to last sip for both products indicating an increasing trend (Fig. 9C) but significance was not reached ($p = 0.315$). It is possible that this omission of a significant difference was due in part to high levels of individual variability in mouthdrying perception, as previously discussed by the authors: it has been suggested that confounding factors such as hydration, salivatory reflex speed and age may influence results (Giles et al., 2025a; Norton et al., 2021). Thus, conclusions on the effect of α-lactalbumin on mouthdrying are made with caution: these results indicate that the α-lactalbumin: β-lactoglobulin ratio is not responsible for whey protein-associated mouthdrying, as its increase in AL-D compared with WP did not influence mouthdrying perception in the present study. However, with further research to understand the factors affecting mouthdrying perception, more definitive conclusions may be able to be drawn in the future regarding this attribute and the contribution of individual proteins.

3.4. *Demineralisation of products and the contribution of minerals to the sensory profile*

As discussed in Section 3.31, it was hypothesised that the high bitterness and peppery sensation seen in AL-E may be the result of increased mineral composition present in this sample as an artifact of the production processes. Analysis confirmed that zinc, iron, potassium and phosphorus were all substantially higher in the AL-E sample than in WP (Table 3). Calcium and magnesium were comparable between the two samples. To investigate the contribution of minerals to the sensory profile of WP, and to further understanding of the sensory effect of enriching α-lactalbumin levels, both samples were treated with an additional ultrafiltration membrane. It was anticipated that this small

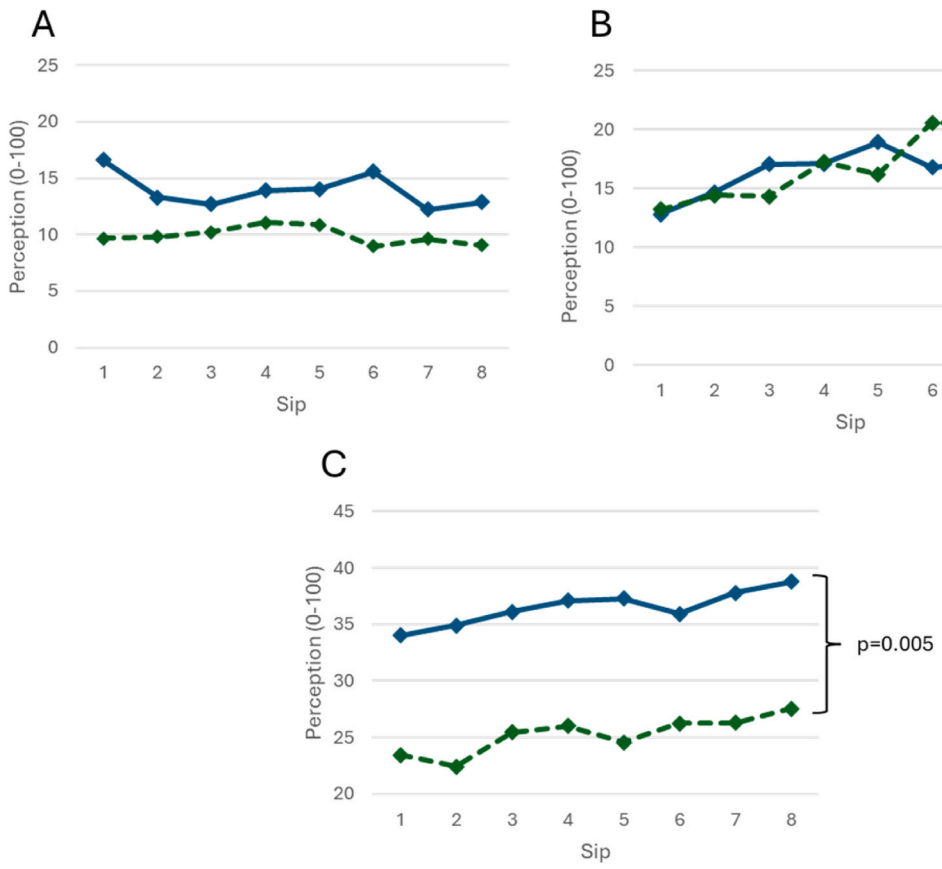

Fig. 8. Average taste and flavour perception score during sequential profiling over 8 consecutive sips of 10% suspensions of whey protein (blue, solid line) and α-lactalbumin deficient protein (green, dashed line). Attributes shown are: [A] sweetness; [B] bitterness; and [C] powdered milk. Overall significance value included when $p < 0.05$.

pore size would remove small peptides and minerals as they would pass through the membrane, whereas larger proteins would not. However, this treatment only reduced the potassium concentration (Table 3). One interpretation may be that the minerals are bound to the whey proteins and, hence, did not pass through the small pore size of the membrane. Contrary to our predictions, iron, zinc, and calcium increased in concentration after ultrafiltration; this may be a result of filtering out other small molecules such as peptides, meaning the relative concentration of the remaining minerals increased.

WP with and without ultrafiltration was compared to understand the impact of this process on the sensory profile. Ultrafiltration was associated with a significant reduction in all aroma attributes, as well as powdered milk flavour and umami perception. It is possible that this additional processing step led to the loss of volatiles responsible for these flavours. Whilst it was outside of scope to analyse and identify these volatiles, future research could perform this analysis to better attribute the compounds responsible. Additionality, ultrafiltration may have altered the molecular structures present, leading to fewer aroma compounds being available for release. More research would be needed to understand the potential sensory implications of additional processing steps prior to commercialisation to avoid unforeseen consequences. However, in cases where the flavour of a product needs to be minimised, this additional step may present a novel opportunity to reduce flavour intensity without the need for added ingredients.

Comparisons were also made between the AL-E samples with and without ultrafiltration. There was a significant reduction in bitter and metallic intensity of the AL-E sample following ultrafiltration (Fig. 10). Whilst this reduction is in line with the reduction in potassium concentration, the levels present in the AL-E-ultrafiltration sample remained significantly higher than previously published detection thresholds (Schiffman et al., 1995). It is noted that the authors were not able to find literature on noticeable difference thresholds for potassium: this could be addressed through spiking experiments in the future to better understand detection levels and concentrations likely to negatively impact sensory perception. Another possible explanation is that the ultrafiltration influenced the binding of potassium and other minerals present in the AL-E sample, leading to reduced interactions between minerals and tastebuds, resulting in a lower bitterness and metallic perception. The analysis only reported total mineral content, as opposed to comparing bound and free mineral content, so it was not possible to quantify this change. It was also noted that the "tingly/-peppery" attribute previously noted in the AL-E sample, reduced to similar levels as that of WP as a result of the ultrafiltration (Fig. 10). Thus, this sensation may be a response to the increased levels of free minerals, rather than an intrinsic sensation association with α-lactalbumin: again, spiking experiments could address this mechanism in the future to better understand the contribution of minerals to the sensory profile. Finally, as discussed for WP, it is possible that the additional processing step of ultrafiltration led to a loss of volatiles and small peptides influencing the sensory perception of samples, irrespective of mineral levels. Previous research into α-lactalbumin enrichment has shown that the composition and microstructure of powders can be influenced by the choice of enrichment approach (Barone et al., 2019), supporting the suggestion that further processing will influence the sensory profile. With regards to other mouthfeel attributes, there was no significant difference in body or mouthcoating perception which is in keeping with the previous sensory profile (Section 3.3.1). There was no significant difference in slippery perception post-ultrafiltration, which was low for all samples.

Ultrafiltration of the AL-E sample was associated with a small but

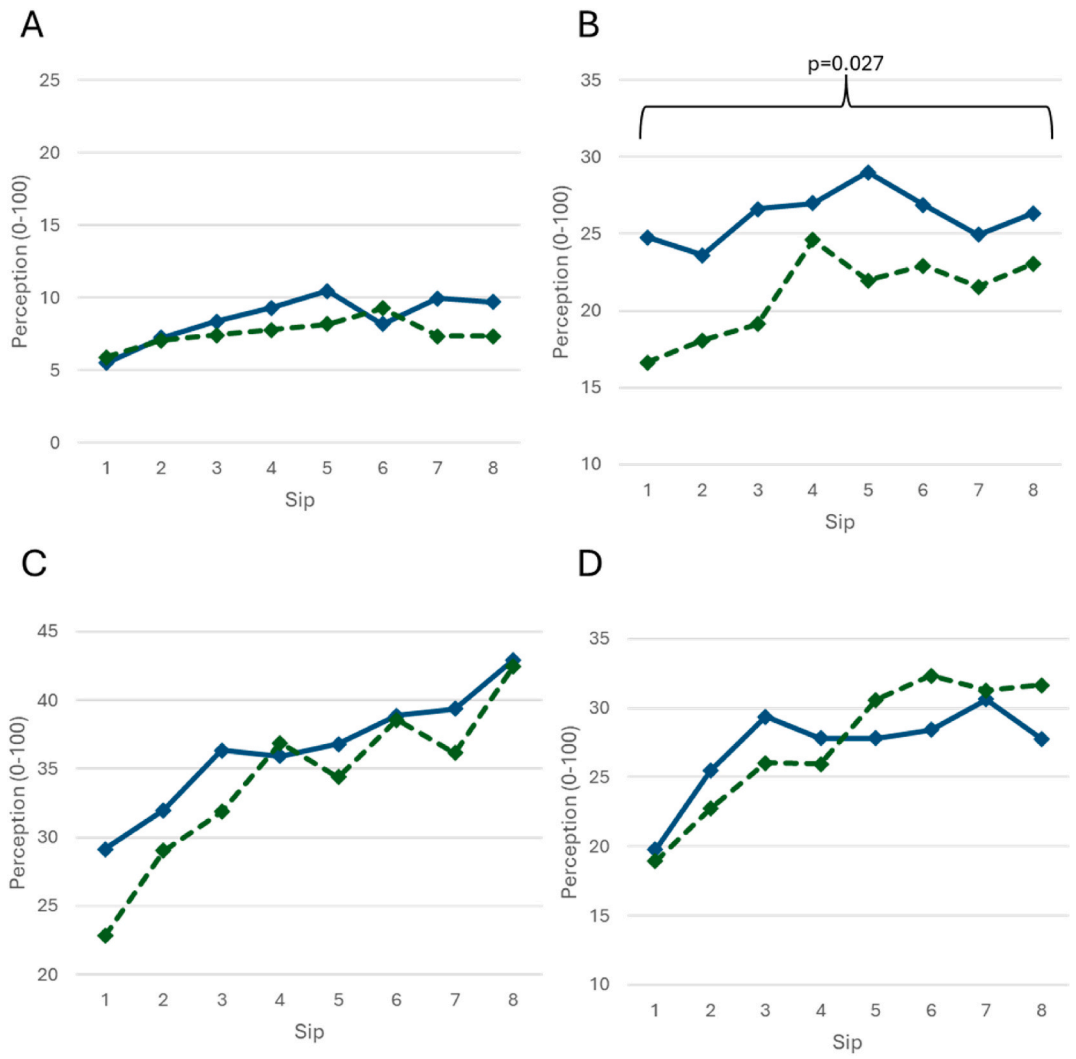

Fig. 9. Average mouthfeel perception score during sequential profiling over eight consecutive sips of 10% suspensions of whey protein (blue, solid line) and α-lactalbumin deficient protein (green, dashed line). Attributes shown are: [A] slippery; [B] mouthcoating; [C] mouthdrying; and [D] salivating. Overall significance value included when $p < 0.05$.

Table 3
Mineral composition of whey protein (WP) and an α-lactalbumin enriched (AL-E) sample treated with and without ultrafiltration (-UF). This was determined by ALS (Cambridgeshire, UK) following a UKAS-accredited standardised protocol.

Sample	Iron (mg/100g)	Potassium (mg/100g)	Magnesium (mg/100g)	Phosphorus (mg/100g)	Zinc (mg/100g)	Chloride (g/100g)	Calcium (mg/100g)
WP	0.49	534	66.1	204	0.339	<0.03	473
WP-UF	0.74	404	75.8	186	1.399	<0.03	535
AL-E	1.42	1100	57.7	408	0.565	0.04	463
AL-E-UF	1.86	847	69.7	402	0.732	<0.03	646

significant reduction in mouthdrying perception (Fig. 10), however the ultrafiltration of WP was not associated with a difference for this attribute. No significant difference was seen between WP and AL-E with regards to mouthdrying, further indicating that α-lactalbumin is not solely responsible for whey-protein associated mouthdrying, but may contribute. More research is needed into the sensory profile of all protein components present in whey protein in order to conclude on the individual contributions to this sensation.

4. Conclusion

The majority of the literature has focused on the contribution of β-lactoglobulin to the sensory profile of whey protein, with limited exploration into the effects of other proteins. To address this, the current study compared an α-lactalbumin enriched (AL-E) protein stream, an α-lactalbumin deficient protein stream (AL-D), and a commercial whey protein. When assessed by a trained sensory panel it was shown that the AL-E sample was associated with a significantly higher slipperiness perception, which was correlated with reduced instrumental friction and an altered tribology profile. In addition, this sample scored significantly higher for bitterness taste and peppery mouthfeel: additional investigations suggested that these attributes were a result of the high free mineral content and small peptides present, rather than intrinsic characteristics of α-lactalbumin. Other findings of note were that the dairy flavour and aroma was significantly reduced through the removal of α-lactalbumin from commercial whey protein, suggesting that

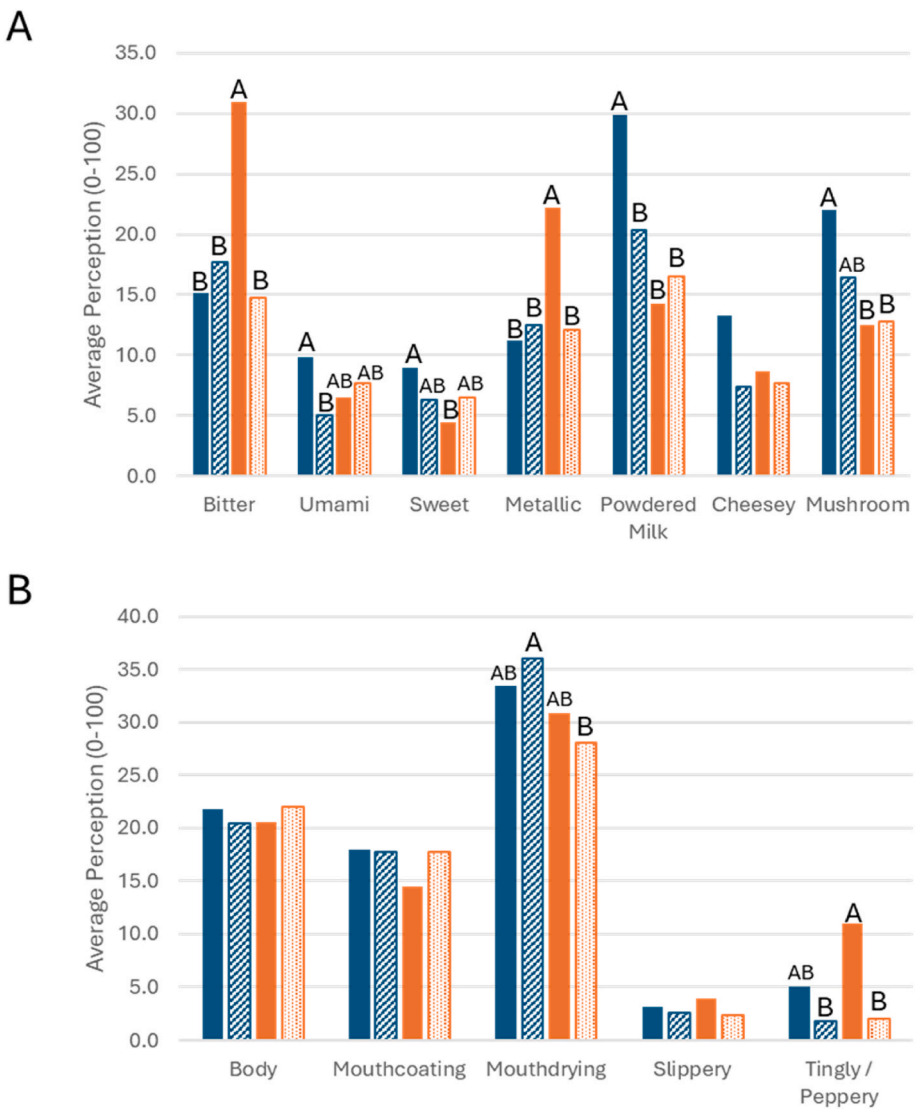

Fig. 10. Mean scores for sensory perception of 10% whey protein suspensions with and without ultrafiltration (blue solid bar and blue lined bar, respectively) and an α-lactalbumin enriched protein sample generated through membrane fractionation of whey protein, with and without ultrafiltration (orange solid bar and orange botted bar, respectively). Samples were scored by a trained sensory panel for [A]: taste and flavour; [B]: mouthfeel. Attributes which showed significant differences ($p < 0.05$) were compared using Tukey to perform multiple pair-wise comparisons.

ultrafiltration influenced the presence of volatiles and small peptides responsible for this attribute. There was also a decrease in mouthcoating and body perception in both samples, compared with commercial whey protein. Palatability and preference were outside the scope of this study but it is possible that the milder flavour and altered texture of the AL-D stream may lead to different applications and impact consumer acceptance. This could be investigated in future research with consumers. This study has combined physicochemical and sensory data to increase understanding of the effect of membrane fractionation to increase the levels of α-lactalbumin in a whey protein beverage, supporting ingredient development and optimising production processes.

CRediT authorship contribution statement

Holly Giles: Writing – original draft, Methodology, Formal analysis, Data curation, Conceptualization. **Stephanie P. Bull:** Writing – review & editing, Supervision, Methodology, Formal analysis, Data curation, Conceptualization. **Stella Lignou:** Writing – review & editing, Supervision, Methodology, Formal analysis, Data curation, Conceptualization. **Alun Hughes:** Writing – review & editing, Methodology, Conceptualization. **David Warren-Walker:** Writing – review & editing, Methodology, Conceptualization. **Joe Gallagher:** Writing – review & editing, Supervision, Methodology, Conceptualization. **Marianthi Faka:** Writing – review & editing, Supervision, Conceptualization. **Lisa Methven:** Writing – review & editing, Supervision, Methodology, Data curation, Conceptualization.

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Declaration of competing interest

The authors declare the following financial interests/personal relationships which may be considered as potential competing interests: This work was funded as part of a BBSRC CASE studentship (BB/T008776/1). This studentship is partly funded by Arla Foods Ingredients, a manufacturer with a commercial interest in whey protein. Arla Foods Ingredients were not involved in the evaluation or interpretation of results to ensure impartiality and took a supervisory reviewing role. Additional funding was granted by the 2024 Aber-Innovation Solutions Catalyst programme (supporting the membrane fractionation of whey and production of samples) and the 2024 IAA Aberystwyth University grant (involved in the demineralisation of samples).

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Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.idairyj.2026.106616.

Data availability

Data will be made available on request.

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