Interactions of Whey Protein Isolate and Human Saliva – as related to the Astringency of Whey Protein Beverages

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Abstract

Interactions between 3 different proteins (lactoferrin, β-lactoglobulin and Whey Protein Isolate) and human saliva were determined. Lactoferrin and whey proteins are known to be astringent at low pH. Astringency is defined as the tactile sensation, mainly on the tongue, caused by astringent compounds when in contact with human saliva. Proline-rich proteins are already known to be directly involved in the astringency of polyphenols. Whey proteins do not contain polyphenols. However, because whey proteins at low pH develop an astringent sensation when consumed, it was expected to detect proline-rich proteins in the interaction between Whey Protein Isolate (WPI) and saliva as well.

The protein solutions were adjusted to different pH-levels, ranging from neutral to high acidic, where a part of each protein solution was heat-treated. All solutions were mixed with human saliva in the same ratio (w/w). One part of all mixtures was pH-readjusted. Additionally, WPI model solutions were prepared, adjusted to different pH-levels, heat-treated and then consumed by voluntary participants, who swirled each solution in their mouth for at least 10 seconds. These mixtures of WPI and saliva were collected for further analysis. After consuming the WPI model solutions, followed by rinsing the mouth with water, tongue swabs were taken to determine the particle sizes and ζ-potentials of the remaining material on the tongue. Control tongue swabs of the clean tongue were taken by the participants before any consumption of the WPI model solutions.

All mixtures as well as lactoferrin, β-lactoglobulin (β-lg), WPI and saliva on their own, were analysed for particle size, ζ-potential and turbidity, which may give an indication for possible aggregation/precipitation of the proteins as well as the analysis of the SDS-PAGE profile of the sediments of the sample mixtures.

Saliva is negatively charged between neutral pH and 3.0, whereas lactoferrin has a positive charge below pH 8.0. WPI has a positive charge below pH 5.1; the same applies to β-lg. None of the proteins themselves showed aggregation/precipitation at

pH-levels 6.8, 3.6, 3.4, 3.0, 2.5 or 2.0. However, after the proteins were mixed with saliva, the pH of mixtures shifted towards neutral pH.

The mixtures of lactoferrin (unheated/heat-treated) and saliva neither showed any significant increases in particle size nor the presence of turbidity. Salivary proteins were not detected in any mixtures at any observed pH either, despite the known fact that lactoferrin causes astringency. The mixtures of β-lg (unheated/heated) and saliva displayed high particle sizes below final pH 3.6, whereas the high turbidities of both mixtures were measured between final pH 3.6 and 3.4. Furthermore, only at final pH 2.8 were salivary proteins (mainly glycosylated proline-rich proteins and α-amylase) detected. However, higher concentrations of salivary proteins were measured when heat-treated β-lg was mixed with saliva. The mixtures of WPI and saliva presented the strongest interaction compared to lactoferrin and β-lg. High aggregation/precipitation occurred in the mixtures between pH 4.3 and 3.0, where significantly high particle sizes and turbidities were detected.

The pH-readjusted mixtures of lactoferrin/β-lactoglobulin/WPI and saliva showed similar values in particle size and turbidity as the mixtures of the proteins and saliva without pH-readjustment at similar pH-values. Furthermore, the pH-readjusted mixtures of the proteins and saliva showed in their sediments the presence of α-amylase and glycosylated proline-rich proteins.

The mixtures of heat-treated WPI and saliva, collected from the mouth after taking a sip (ratio unknown), revealed that the strongest interactions occurred when WPI-solutions were adjusted to pH 3.6 and 3.4. Similar observations were made for heat-treated WPI-solutions, which were adjusted to pH 3.6 and 3.4, when mixed with saliva 1:1 (w/w). However, additionally to the glycosylated proline-rich proteins and α-amylase, faint bands of mucin as well as basic proline-rich proteins were detected in the mixtures collected from the mouth.

The proteins of the material remaining on the tongue followed the consumption of WPI-solutions and rinsing with water showed that the particle size measurements

were not reliable. However, pH-levels between 6.8 and 5.7 occurred and negative charges were measured on the tongue after rinsing the mouth twice with water.

The strongest interactions between the proteins and human saliva occurred when the proteins, in particular β-lg and WPI, were positively charged and then mixed with saliva (negative charge). Concluding from that it is suggested that electrostatic interactions may cause the astringent sensations. However, since no evidence could be found that salivary proteins were involved in the interaction between lactoferrin and saliva (without pH-readjustment), it is suggested that other interactions than electrostatic interactions cause the astringent sensation of lactoferrin.

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