Investigating Astringency Mechanism of WPI8855 in Acidic Condition

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Abstract

Whey protein isolate is used as a functional ingredient in acidic whey protein beverages, but the associated astringency is a big hurdle to introduce these beverages into the mainstream market. If we can solve the astringency issue, Fonterra would have big advantages over their competitors. Our hypothesis is that whey protein interacts with human saliva proteins and the subsequent precipitation causes astringency.

In the present study, ion exchange whey protein isolates (WPI) 8855, and solutions of pure α-lac and β-lg were used to determine which whey protein fractions are responsible for sedimentation in artificial or human saliva. It has been shown that sedimentation correlates to the level of astringency. Therefore only the level of sedimentation was investigated. The human saliva and artificial saliva were also compared in the astringency titration model in order to determine whether artificial saliva is representative of human saliva.

Heat treatment (85°C, 30s) of whey protein solution was performed to mimic commercial beverage manufacture. The heated and non-heated whey protein solutions were titrated with artificial saliva, human saliva or sodium bicarbonate buffer in the range of pH 3 to 6. The sediment was recovered by centrifugation of the titrated samples, and analysed using liquid chromatography-mass spectrometry (LC-MS/MS) or one and two dimensional polyacrylamide gel electrophoresis (PAGE) with amido black and periodic acid Schiff stain.

This study showed that β-lg is the key sedimentation component in heated acidic WPI8855 beverages due to the heat aggregation, pH change through the isoelectric point and interaction with human saliva proteins, including mucin, proline-rich proteins (PRPS) and α-amylase. BSA also interacted with artificial and human saliva, whereas α-lac did not interact with either artificial or human saliva. Heat treatment caused extensive whey protein aggregation and precipitation. Artificial saliva and human saliva behaved differently in this astringency titration model, therefore it is not

recommended to use artificial saliva in an *in vitro* model to predict astringency *in vivo*. Artificial saliva interacted with whey protein and caused additional precipitation compared to titration with sodium bicarbonate, whereas human saliva was able to hinder some whey protein sedimentation caused by titration with sodium bicarbonate. If astringency is caused by the amount of precipitation of protein, heat treatment would be a major factor in the astringency of whey proteins.

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µl = microlitre
2-ME = 2-mercaptoethanol

BSA= bovine serum albumin

g = gram

GMP = glycomacropeptide

HCl = hydrochloric acid
Histidine-rich proteins = HRP

l = litre
M = molar
mg = milligram
min = minutes
ml = millilitre
mM = millimolar
NaCl = sodium chloride
$NaHCO_3$ = sodium bicarbonate
pH = measure of acidity
PRP = proline-rich protein

RP-HPLC = reverse phase-high performance liquid chromatography
rpm = revolutions per minute
SDS = Sodium dodecyl sulfate

SDS PAGE = sodium dodecyl sulphate polyacrylamide gel electrophoresis

WPC = whey protein concentrate

WPI = whey protein isolate
α-lac = alpha-lactalbumin

β-lg = beta-lactoglobulin