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**Goat and cow casein derived
ingredients and their interactions with
iron**

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Abstract

The objective of this study was to gain a fundamental understanding of how goat casein micelles and the products of casein proteins behave when fortified with iron.

Iron fortified skim milk was characterised by analysing the mass balance of micellar/non micellar fractions, chemical changes, micellar size changes and internal structure. Two treatments were examined to determine where in the processing line the addition of iron might best be added to a milk system. On average, at least 72% of the iron is bound to the micellar phase across the treatments and iron concentrations. Small angle X-ray scattering (SAXS) indicated that internal changes, mainly at the location of the colloidal calcium phosphate, occurred with iron addition.

Casein was extracted from goat milk using isoelectric precipitation however the extraction was more difficult than using cow milk. Iron fortification of the caseinates resulted in a tendency for oxidation and precipitation of the proteins to occur causing the formation of large aggregates. The caseinates could not stabilise the same amounts of iron to that of an intact casein micelle.

Phosphopeptides were isolated by adding calcium and ethanol to caseinate digests. There was an increase in serine, glutamic acid and isoleucine residues compared to caseinate. There was an increase in phosphorus from 7.8 ± 0.3 mg P/ g solids to 45.4 ± 2.4 mg P/ g solids in the isolate. The phosphopeptides were composed of smaller, more hydrophilic peptides compared to the full digest prior to precipitation. Ferrous sulfate was then investigated for use as the precipitant, instead of calcium. The peptides produced similar trends in terms of amino acid profile changes, phosphorus concentration increase and yield. Immobilised metal affinity chromatography was also investigated however this had a low throughput that may not be effective at process scale.

The effect of heating, cooling, ionic strength of the solution, holding time, iron loading, processing order and use in a model milk system were investigated to simulate potential industrial processing conditions using the calcium - extracted phosphopeptides. It was found that goat peptide isolates were able to bind 54.4 ± 0.5 mg Fe/ g protein compared to goat milk of 4.3 ± 0.1 mg Fe/ g protein. The optimal conditions for binding were found to be at pH 6.7 in a low ionic strength solution,

around 37 °C. There was a potential synergistic effect of adding the peptides to milk in terms of iron binding capacity. There were few differences in the amount of iron that could be bound comparing cow and goat derived phosphopeptides under the tested conditions.

The oxidation potential of ingredients was determined using malondialdehyde (MDA) as an oxidation product marker. There was a reduction in oxidation when iron was bound to milk or peptides compared to free ferrous sulfate in solution with intact goat milk performing the best producing 0.46 ± 0.04 μg MDA/mL after 3 days at 30 °C compared to the blank of 1.25 ± 0.16 μg MDA/mL. The goat peptides produced non-significantly different levels of MDA compared to the blank containing no ferrous sulfate.

Caco-2 cell lines are a way of approximating how systems may function in an intestine in terms of nutrient absorption. Iron absorption was improved in the order of casein hydrolysates > caseinate > skim milk for goat milk. In contrast, cow milk appeared to perform better without any modifications to the proteins. On an equal iron filtrate basis after the digestion and intestinal phase, calcium- precipitated goat phosphopeptides produced a response of 9.64 ± 0.94 ng ferritin/ nM iron. This response was greater than all other treatments with the exception of goat milk fortified with 5 mM iron and ascorbic acid with 12.30 ± 1.23 ng ferritin/ nM iron.

This work covers a wide range of milk products and iron interactions and has helped to build a fundamental understanding of goat milk protein functionality. The underpinning considerations to a manufacturing setting may allow further development of large scale ingredient production for the improved stability of iron fortified systems.

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