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

A thesis presented in partial fulfilment of the requirements for
the degree of

Doctor of Philosophy

in

Food Technology

Massey University, Palmerston North, New Zealand

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2017



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.

Acknowledgments

I would firstly like to thank my supervisor Dr. Alistair Carr for allowing me to pursue a PhD. His discussions and ideas have made for a very interesting and exciting project. This sense of humour made the long stints in the Synchrotron labs and during the whole project a lot more colourful. Thank you for carefully reading my thesis and asking valid questions which have enriched the discussion.

To my co-supervisor Dr. Lara Matia-Merino, thank you for your hard work and support during my studies. She has provided focus to the project and kept me motivated when I was struggling. Her directness helped move work along which I am grateful for while her advice and personal support showed her true caring nature.

Dr. Bridget Ingham: my co-supervisor who had her work cut out for her. I am thankful that I was able to incorporate techniques like SAXS in my work as it allowed me to experience a whole different world in science, not to mention travelling to the Synchrotrons. It has to be mentioned that she has great patience in explaining the theory, equations and models on numerous occasions and assessing my fitted models many times over.

I would like to acknowledge Dairy Goat Co-operative, specifically Dr. Colin Prosser for giving me the opportunity to undertake a PhD by providing the funding for the whole project.

Thanks to Bob (Dr. Robin Stewart) for firstly performing the Caco-2 cell assays for the part of the project and also showing me how the method works (and making me thankful that none of my work was as temperamental and demanding). Also, for being a great office buddy and sharing a few good yarns.

Thank you to the Laboratory Technicians in the School: Steve Glasgow, Micelle Tamehana, Janiene Gilliland, Warwick Johnson, Garry Radford and John Edwards (SEAT) for help using equipment, making orders and running methods (as well as some amusing chats). Trevor Loo from the Fundamental Sciences department taught me how to carry out digestions and gave some good pointers in this area so I must acknowledge how much this helped me. Along the same track, thanks to Don Otter for helping me with mass spectrometry, if anything, I have learnt how to read the

output even if results didn't eventuate. Finally, thank you to Maggie Zou who helped me run the HPLC and showing me the ins and outs of this technique.

Thank you to Prof. Dick FitzGerald for the useful discussions regarding IMAC; this helped me immensely to achieve results for this section of work.

Thank you to the Australian Synchrotron and the Lawrence Berkeley Advanced Light Source, specifically Nigel Kirby and Cheng Wang for helping our team in running the equipment and input in analysis.

Thank you to Circo Acrofit Studios and the people there that make it so much fun. My time in Palmerston North has been made more fulfilling when I knew class was coming up. This place was great for exercise and humour and I have made some awesome friends here.

Thank you to Kate Donohue who initially convinced me to carry out a postgraduate degree and being an excellent support through the shared experience of doing a PhD. Also, for turning me to the 'R side' of statistics, making me pretty confident in plot coding but also giving insight into statistical analysis. Our time together as flatmates was super fun as evidenced by us being inseparable during weekend shenanigans.

I need to also thank my parents for motivating and encouraging me (along with financial support) during this time. Thanks for being proud of me and being okay with me delaying becoming a 'real adult' and stretching out student life for as long as possible.

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