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MASSEY UNIVERSITY

# Colloidal interactions in an alternate make cheese

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## Abstract

The role of emulsion structure and interactions on the material and technical functionality of an alternate make cheese (AMC) was investigated. Lab scale cheese samples (25 g comprising 23 wt.% fat and 20 wt.%) were prepared by recombining model emulsions with a separate protein phase under controlled temperature, shear speed and residence time in a rapid visco analyser (RVA). Sodium caseinate and Tween 20 were used respectively to stabilize fat globules for the model emulsions. Preliminary experiments were carried out for samples prepared using either calcium caseinate or sodium caseinate as protein phase. Structural characterisation of samples showed emulsion structure and distribution within these phases to be dependent on protein type. It was inferred that the calcium from calcium caseinate matrix modified the interfacial layer of the emulsions stabilised by sodium caseinate, as indicated by the increased fat globule size distribution after cheese making. In comparison, the size of fat globules covered with sodium caseinate appeared relatively stable in cheese produced from cheese curd. Based on these observations, caseinates were subsequently replaced by cheese curd as the protein phase for the remainder of the study.

For cheese samples prepared with low fat cheese curd, fat droplets stabilised with sodium caseinate were hypothesised as binding with the surrounding protein matrix, and thereby these fat globules could be considered as 'active fillers'. Confocal laser scanning microscopy supported this hypothesis showing homogeneously dispersed fat droplets within the protein network. This emulsion system did not show fat-protein phase separation in baking (170 °C 10 minutes) as droplets were prevented from coalescing as a consequence of entrapment within the protein phase.

Fat globules covered with Tween 20 were hypothesised as behaving as 'inactive fillers', with the adsorbed layer not anticipated to form bonds with the surrounding protein network. Confocal and scanning electron microscopy instead showed localised domains of fat droplets within the protein structure that underwent partial coalescence on cooling of the cheese after manufacture. Cheeses comprising Tween stabilised droplets exhibited phase separation on baking and visible oil-off on the surface of cheese arising

from extensive coalescence taking place within the localised regions of fat due to melting of the partially coalesced structures. Additional rheological analysis of cheeses was carried out to determine the effect of droplet-protein interactions on the material properties of the cheese samples. Notably, findings were presented in relation to a non-fat control cheese. Findings showed that, at temperatures below 30 °C when fat was crystallized, both inactive and active fillers had a higher relative modulus to the non-fat sample. However, at elevated temperature without fat crystals, inactive fillers resulted in a relative reduction in storage modulus when compared to the non-fat cheese, while active fillers increased relative storage modulus.

Model cheeses prepared with either sodium caseinate or Tween 20 stabilised emulsions were then compared to cheese samples comprising non-homogenised cream as the emulsion phase. Structural analysis of samples determined that cheeses comprising fat globules stabilized with native milk fat globule membrane behaved in a manner analogous to samples prepared with the Tween stabilised emulsion, indicating the presence of inactive droplets. However, it was also observed that increasing the residence time of cheese production within the RVA caused a transition of the interaction behaviour of the emulsion from inactive to active, as evidenced by corresponding changes to structural, material and functional properties of the cheese.

Further exploration of this transition determined that the mechanical work applied during cheese preparation was sufficient to homogenise fat droplets during extended shearing, resulting in a reduction to fat droplet size. Droplet homogenisation during shearing was also found to have disrupted the native milk fat globule membrane, allowing protein adsorption to take place. It was also determined that whey proteins were the predominant interfacial fraction adsorbed as a consequence of extended shearing, and were considered responsible for the transition of droplets from inactive to active. Combined findings have shown that the material and functional properties of an alternate make cheese composition could be strongly influenced by the interactions of the emulsion phase with the surrounding protein network. These interactions could, in turn, be manipulated through formulation and/or process design, providing greater control over product properties.

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## Acronyms

AMC	Alternate make cheese
AMC_NaCas	Alternate make cheese produced from emulsified fat fully covered with sodium caseinate
AMC_Tween	Alternate make cheese produced from emulsified fat fully covered with Tween 20
AMC_NC	Alternate make cheese produced from fat globules with native milk fat globule membrane
AMC_AMF	Alternate make cheese produced from anhydrous milk fat without emulsifiers
AMF	Anhydrous milk fat
$\beta$ -ME	$\beta$ -Mercaptoethanol
Ca	Capillary number
CaCas	Calcium caseinate
CLSM	Confocal laser scanning microscopy
cm	Centimetre
d(0.1)	Volume-weighted diameter of 10 % smallest droplets
d(0.9)	Volume-weighted diameter of 10 % largest droplets
d <sub>4,3</sub>	Volume-weighted average mean diameter
EDTA	Ethylene diamine tetra acetic acid
FG	Fast Green
FO	Free oil
g	Gram
G'	Storage modulus
G''	Loss modulus
G <sub>m</sub> '	Storage modulus of non-fat cheese
Hz	Hertz
LFCC	Low fat cheese curd
m	Mass (in equations); or meter (after numbers)

min	minutes
mm	Millimetre
mg	Microgram
nm	Nanometre
NaCas	Sodium caseinate
NaOH	Sodium hydroxide
NR	Nile Red
PEG	Polyethylene glycol
Re	Reynold number
RhPe	1,2-dimyristoyl-sn-glycero-3-phosphoethanolamine-N-(lissamine rhodamine B sulfonyl)
RVA	Rapid visco analyzer
rpm	Revolve per minute
SDS-PAGE	Sodium dodecyl sulfate-poly acrylamide electrophoresis
vol	volume
Tween	Tween 20
Tris	Tris(hydroxymethyl)aminomethane
μl	Microliter
WPI	Whey protein isolate
wt	Weight
w/v	Weight/ volume