DAIRY PRODUCTS RESEARCH CENTRE

MOOREPARK

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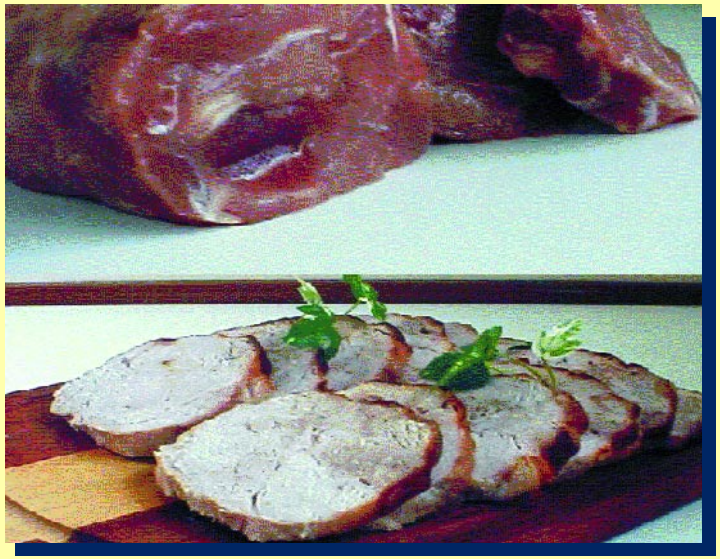
Use of Novel Dairy Ingredients in Processed Meats

Dr. Brendan O' Kennedy

TGase, a commercially available enzyme, when used with sodium caseinate can successfully replace salt as a binding agent in processed meats.



Optimisation of Ingredient Formulation in Processed Meat Products

(Use of Novel Dairy Ingredients in Processed Meats)

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Summary and Conclusions

Reformed and restructured meat are two major categories of processed meat products. Reformed meat products require intact meat pieces to bind together while restructured meat products are extensively minced prior to restructuring. Salts such as sodium chloride and phosphates together with mechanical treatment and heat, have been used to bind meat pieces together. In the process the proteins in muscle become soluble, bind large amounts of water and gel on heating.

While heat-induced gelation of soluble meat protein provides binding in reformed meat products and reduces cook losses in restructured meat products, no binding occurs in raw meat systems. Non-meat proteins, especially soya protein, are routinely used in processed meat products, often in conjunction with salts, to increase water and fat binding during the cooking process. However, such proteins do not bind intact meat pieces in either the raw or cooked state.

Transglutaminase (TGase) is a food-grade commercially available enzyme which can crosslink suitable proteins leading to the formation of a protein matrix (gel) and immobilisation of large quantities of water. This property could improve the water-binding properties of non-meat proteins in restructured meat products. The prospect of crosslinking native meat proteins and non-meat proteins or native meat proteins on adjacent meat pieces would make salt-free reformed meat products a realistic objective.

Hence, the main objective of this project was to study protein-protein interactions in reformed and restructured meats, especially between meat proteins and added non-meat proteins in the absence of salts but in the presence of a protein crosslinking enzyme.

The main conclusions were as follows:

* ***Reformed meats:*** *the introduction of sodium caseinate and TGase into reformed meat systems resulted in binding of the individual meat pieces together after storage at 5°C overnight. The resulting crosslinked protein, as well as promoting cohesiveness between meat pieces at 5°C, was also stable to heat (cooking). This ensured that the cooked meat maintained its integrity and sliceability.*

It was found that TGase alone did not induce crosslinking reactions between intact raw meat pieces, but did so in the presence of added sodium caseinate. Increasing the whey protein (WPC-75) concentration decreased the binding strength of the meat.

Optimum binding strength was obtained at 84:16 (casein:whey) in raw meat and 80:20 in cooked at a TGase concentration of 500 EU/kg meat.

* ***Restructured meats:*** *a model meat system was used to assess the effects of TGase in*

the presence of a range of non-meat proteins but in the absence of salt, to improve the water-binding of restructured meats following cooking.

Sodium caseinate proved to be best of the non-meat proteins assessed, but the ratio between the caseinate and TGase concentrations needed to be adjusted accurately.

At a sodium caseinate concentration of 3.3% of the total meat dispersion and a TGase concentration of 1500 EU/kg, cook losses were reduced to a level similar to meat dispersions cooked in the presence of salts.

Research and Results

The major reasons for incorporating non-meat proteins into restructured or reformed meat products are to provide cohesion between intact meat pieces and/or to bind water. The normal range of non-meat proteins (dairy, soya, etc.) are often particularly bad at achieving these aims when compared to the effects of NaCl/phosphate. The use of *TGase* as a means of improving both the cohesive power and the water-holding capacity of added non-meat protein is one way of improving their functionality.

Preliminary Investigations with TGase *(see Formulation 1 - page 10)*

Sodium caseinate dispersions formed strong gels in the presence of *TGase* at 5°C after 24 h. Whey protein solutions remained liquid under the same conditions. However, following prior heating of the whey protein solution (85°C, 20 min) strong gels were obtained. The requirement for unfolding of the whey protein molecules underlines the protein conformational requirement necessary for efficient protein crosslinking using *TGase*. In this regard, reconstituted skim milk powder and soya protein isolate did not form stand-up gels under the same conditions.
An ECHIP™ statistically designed experiment was undertaken to determine whether pre-heated whey protein could be used to replace caseinate as the substrate for *TGase* and still maintain the integrity of the gel. Increasing *TGase* concentration gel **increased** gel strength and the force required to break the gel at different caseinate/whey protein ratios. The final protein concentration was maintained at 10% (w/w).

The force required to break the whey or casein gels was similar at equal concentrations of protein and *TGase*. Casein gels were more brittle and whey protein gels more elastic. The *TGase* concentration directly affected the gel firmness and the force required to break the gel at equal concentrations of protein.

Reformed Meat Studies *(see Formulation 2 - page 11)*

Both caseinate and denatured whey protein were susceptible to *TGase* induced gelation at 5°C. However, their success in binding intact meat pieces was variable. A general view of

the process and final meat product is shown in *Fig.1*. Sodium caseinate and WPC-75 (12.5% w/w) solutions were mixed to give ratios from 100/0 to 0/100 casein/whey protein.

It was previously established that *TGase* would not crosslink whey proteins unless they were denatured. It was therefore necessary to heat all solutions (85°C, 10 min) prior to the introduction of the enzyme. Due to the instability of WPC-75 to heat, the pH of the formulation was raised to 7.35 and citrate was included (to chelate calcium) at a rate of 40 mg/g whey protein.

Increasing the *TGase* concentration increased binding strength of the meat as did increasing the caseinate concentration. Increasing WPC-75 concentration led to a decreased binding. It is suggested that prior denaturation of WPC-75 may have led to excessive aggregation of the protein, even in the presence of caseinate, thus limiting or altering its gelation or cohesive properties in the presence of *TGase*.

Optimum binding strength was obtained at 84:16 (casein:whey) in raw meat and 80:20 in cooked meats at a *TGase* level of 500 EU/kg meat. *TGase* alone did not induce crosslinking reactions between intact raw meat pieces, indicating that intact muscle protein was not conformationally suitable for the enzyme-mediated crosslinking reaction.

In the absence of sodium chloride, the addition of caseinate and *TGase* provided cohesion between intact meat pieces in reformed meat systems. The crosslinked protein induced by *TGase*, as well as promoting cohesiveness at the junction between raw meat pieces, was also stable to heat. This ensured that the subsequently cooked meat retained its integrity and sliceability.

The innovative or commercial application of these results is in the production of raw reformed meat systems, which resemble large muscle joints. They would obviously be produced from trimmings or low-value meat cuts. This value-added product is capable of being sliced both in the raw or cooked state. The reformed meat is subjected to minimal processing and contains no added salt or polyphosphates to induce binding. Muscle integrity has not been altered and, therefore, flavour and texture are unaffected.

Restructured Meat Studies

A model meat system (developed at University College Cork) was used to assess the effects of *TGase* in the presence of a range of non-meat proteins but in the absence of salt, to improve the water-binding of restructured meats following cooking.

In restructured meat systems (see *Fig. 2* for make procedure) the major emphasis is on the ability of the non-meat protein ingredient to reduce cook losses. Retorting of 70% meat dispersions in the absence of additives (protein, *TGase*, salt) resulted in cook losses of 39 - 40%.

Fig. 1 *Production of Reformed Gammon Pork Meat*

(a) Meat cut into irregular pieces prior to protein/enzyme addition.
(b) After the addition of the aqueous protein/enzyme mixture and vacuum extrusion and
(c) After cooking in a convection oven at 180°C for 90 minutes.

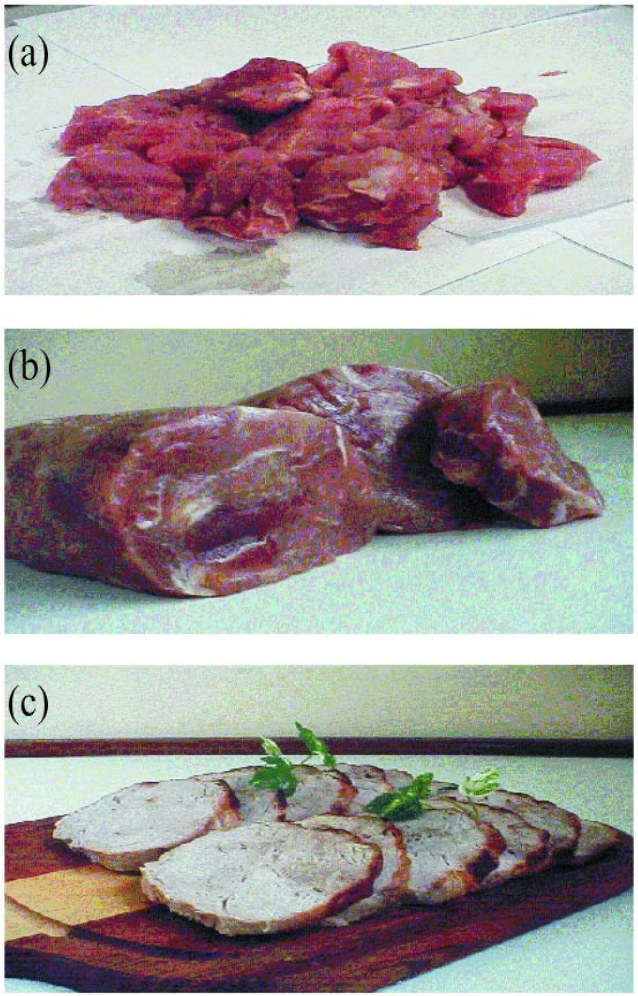
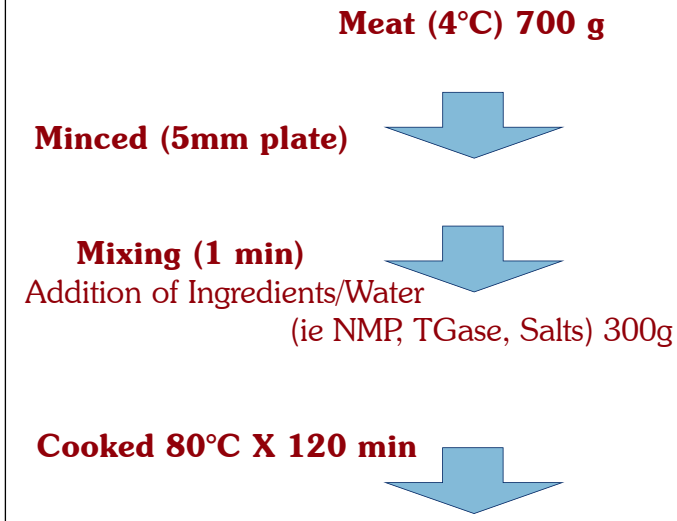


Fig. 2: ***Protocol for the production of Restructured Meat***



The inclusion of TGase alone in the meat dispersion had no effect on cook losses following retorting, irrespective of the concentration used. The inclusion of 3% salt alone reduced cook losses from the meat dispersion to less than 5%, following retorting. This would be expected as both NaCl and phosphate solubilise meat proteins which subsequently gel on heating to immobilise water and reduce cook loss.

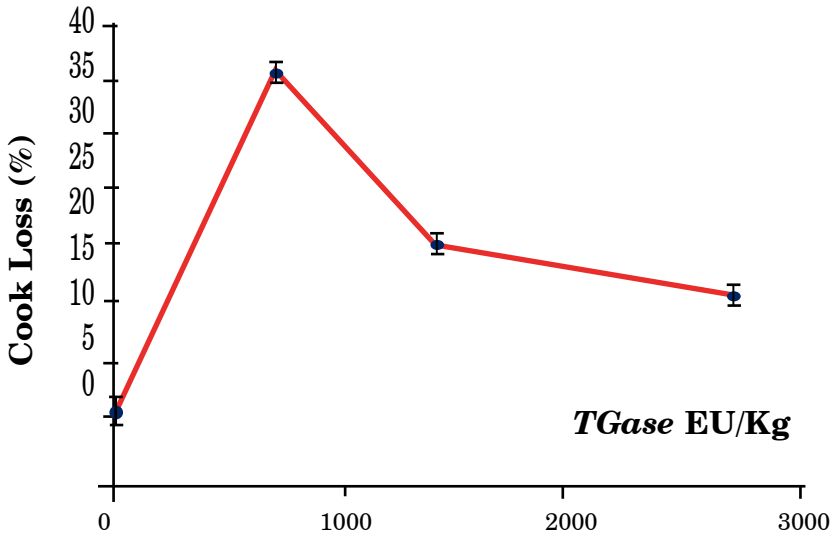
A combined addition of *TGase* and salt to meat dispersions gave rise to a marked increase in cook losses following heating (*Fig. 3*). Cook losses increased from 5% in the absence of *TGase* to 37% when *TGase* was present in high concentrations. Reducing the *TGase* concentration resulted in a reduction in cook losses. *It was concluded that the effect of TGase was to reduce the water-holding capacity of the salt solubilised meat proteins through crosslinking reactions.*

The inclusion of increasing quantities of sodium caseinate/*TGase* combinations in the meat dispersions in the absence of salt led to a decrease in cook losses (40% to 27%). Addition of increasing quantities of sodium caseinate alone to the meat dispersions, in the absence of salt, had little effect on cook losses (38%). Later, the added protein concentration was standardised to 3.375% of the total meat dispersion.

Using this concentration of added protein, in the form of sodium caseinate, it was observed that as the *TGase* concentration increased from zero to 530 EU/kg meat dispersion, cook losses were reduced from 29% to 11.6% (*Fig. 4*) and the strength of the raw meat system increased from 46 g to 111 g using a ball probe.

Increasing the *TGase* concentration to 1500 EU/kg meat dispersion reduced further the cook losses to 7.85% and increased the strength of the raw meat to 612 g. *However, further increases in TGase concentration (3000 EU/kg meat dispersion) resulted in a dramatic increase in cook loss (28%) and a reduction in raw meat strength (547 g).* For the final screening of protein substrates the formulation conditions were therefore standardised to 70% meat, 3.375% protein, 1500 *TGase* EU/kg final dispersion.

Fig. 3 *A combined addition of TGase and salt to meat dispersions gave rise to a marked increase in cook losses following heating.*



Six different protein sources were compared with the control sodium caseinate substrate and a meat system containing 3% salt for their ability to immobilise water both in the raw state and following cooking. These included soya protein isolate, soluble wheat protein, whey protein isolate (pre-heated), milk proteinate, pea protein isolate and blood plasma (*Table 1*). *Cook losses for soya protein isolate (29.65%), pea protein isolate (31.2%) and whey protein isolate (33.4%) were high compared to both the salt and caseinate standards.*

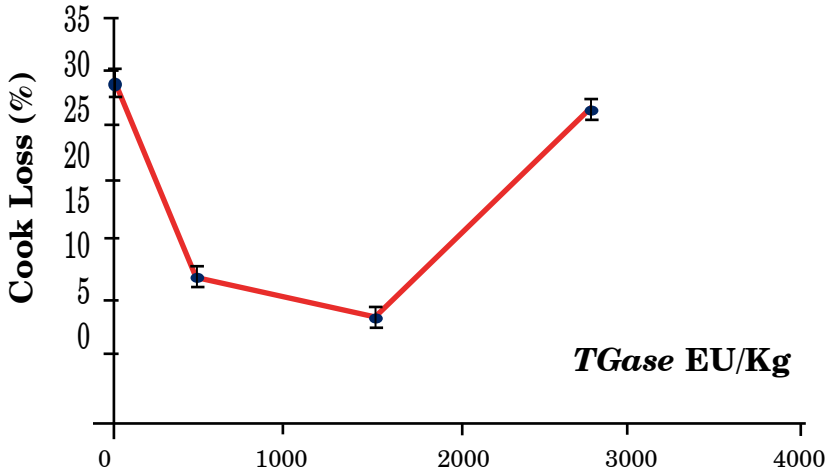
The milk proteinate (20%), blood plasma (23.6%) and soluble wheat protein (15.1%) showed improved ability to control water loss during cooking. *However, none of the protein sources showed water immobilisation properties as good as sodium caseinate in the presence of TGase.*

It was concluded that correct adjustment of the stoichiometry between caseinate and TGase in meat dispersions with no added salt can rival salt-containing meat systems as regards achieving low cook losses.

Table 1. *The effect of TGase addition, in the presence of a range of protein ingredients, on cook loss and consistency of restructured meat compared to meat restructured by using salts*

Treatment	Cook Loss (%)	Meat Consistency (g Force)	
		Raw	Cooked
Salt	3.4 + -0.8	117 + -3.2	236 + -8.6
Sodium Caseinate	8.0 + -2.3	595 + -19.7	346 + -14.3
Soya Protein Isolate	29.7 + -0.94	416 + -12.8	419 + -9.6
Soluble Wheat Protein	15.1 + -0.53	214 + -4	286 + -5.7
Whey Protein Isolate	33.4 +- 0.76	623 + -16.4	658 + -25.9
Milk Proteinate	20.0 + -1.85	711 + -34.3	447 + -22.4
Pea Protein Isolate	31.2 + -0.72	340 + -11.9	401 + -16.5
Blood Plasma	23.6 + -0.78	238 + -11.4	432 + -37.7

Fig. 4 *With added protein (as sodium caseinate) concentration standardised to 3.375% of the total meat dispersion, cook losses were reduced from 29% to 11.6% as the TGase concentration increased from zero to 530 EU/kg .*



Formulation 1

Gel formation using TGase

12.5% (w/w) Caseinate and WPI
pH adjusted to 7.0
Mixed to give various ratios
Heated to 80°C for 30 min
Cooled to 5°C
TGase added (0 - 0.5%)
Final protein concentration 10%
Stored at 5°C for 24 h
Gel firmness at 25°C (Instron)

Formulation 2

Production of Reformed Meat

12.5% (w/w) Caseinate and WPC-75
Mixed to give various ratios
Addition of 40 mg tri-sodium citrate/g protein
pH adjusted to 7.35
Heated to 80°C for 30 min
Cooled to 5°C
TGase added (0 - 0.5%)
Final protein concentration 10%
Gammon pork meat cut into irregular pieces
50 g protein/enzyme mixture added to 1 kg meat
Each batch contained 5 kg meat
Extruded into 10 cm casings
Stored at 5°C for 24 h
Both raw and cooked meats sliced (180 x 31 x 9 mm)
Meat cohesion determined on a TA Instruments Texture Analyser

Publications

O'Kennedy, B.T., Neville, D.P. and Kelly, P.M. (1998). Texture improvement in restructured meat products using Transglutaminase in combination with milk proteins. *Proceedings of 1st International Symposium on Enzymatic Protein Processing.*

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