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# EFFECT OF THE DIFFERENCES IN PROTEIN STRUCTURE ON DIGESTIBILITY

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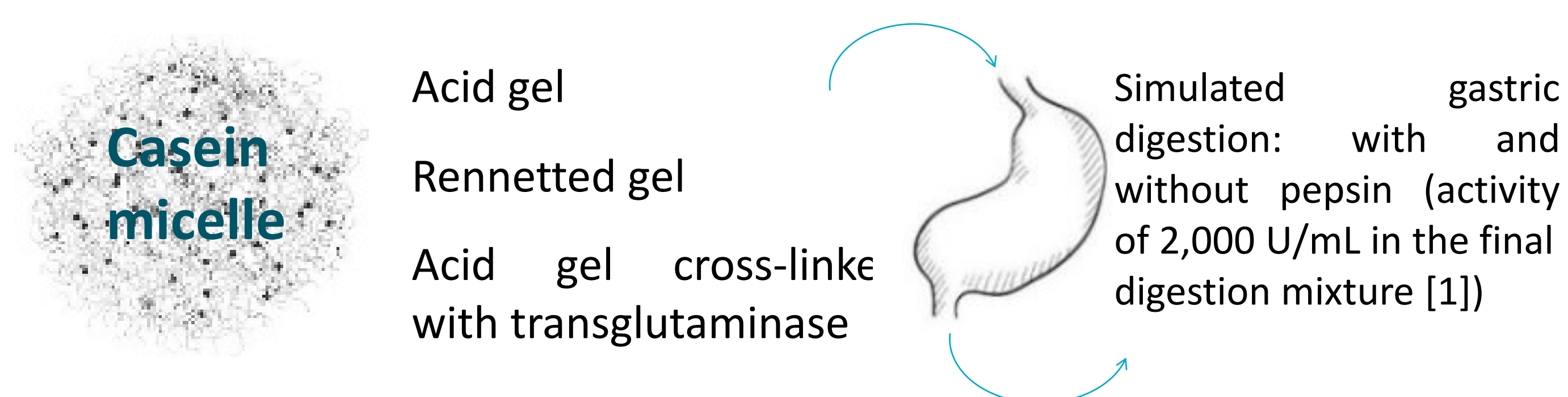
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Casein protein networks provide the underlying structure of most dairy-based products, resulting in the formation of a range of gel structures such as cheese and yoghurt. Despite increasing interest to modulate food structure as a potential way of controlling food breakdown, the underlying mechanisms are not still fully understood. Protein digestion begins in the stomach due to the action of pepsin (an enzyme) under acidic conditions of the gastric fluids, in combination with mechanical mixing and shear from the movement in the stomach. Little is known about the influence of the protein network on pepsin kinetics, including the mass transfer of the enzyme molecules into the gel particles. In the present work, the devolution of micellar casein dairy gels upon gastric digestion was investigated at the nanoscale, highlighting the role of food structure on digestibility.

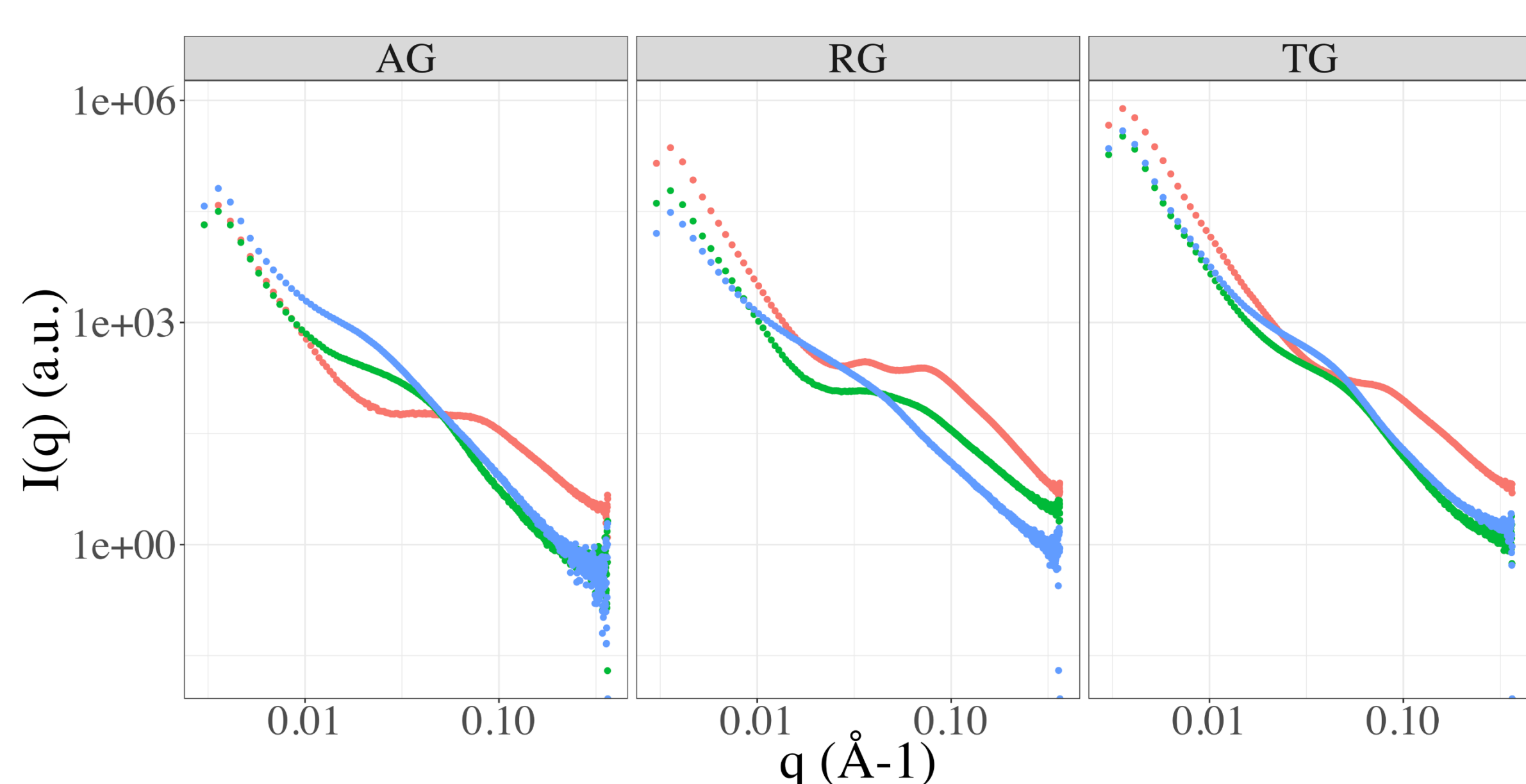
## Materials and Methods



The physical characteristics of each gel and its digesta were evaluated using small angled X-ray scattering (SAXS). The devolution of casein protein networks as gastric digestion proceeded were examined by monitoring the release of soluble proteins into the supernatant of the digesta.

## GEL STRUCTURE

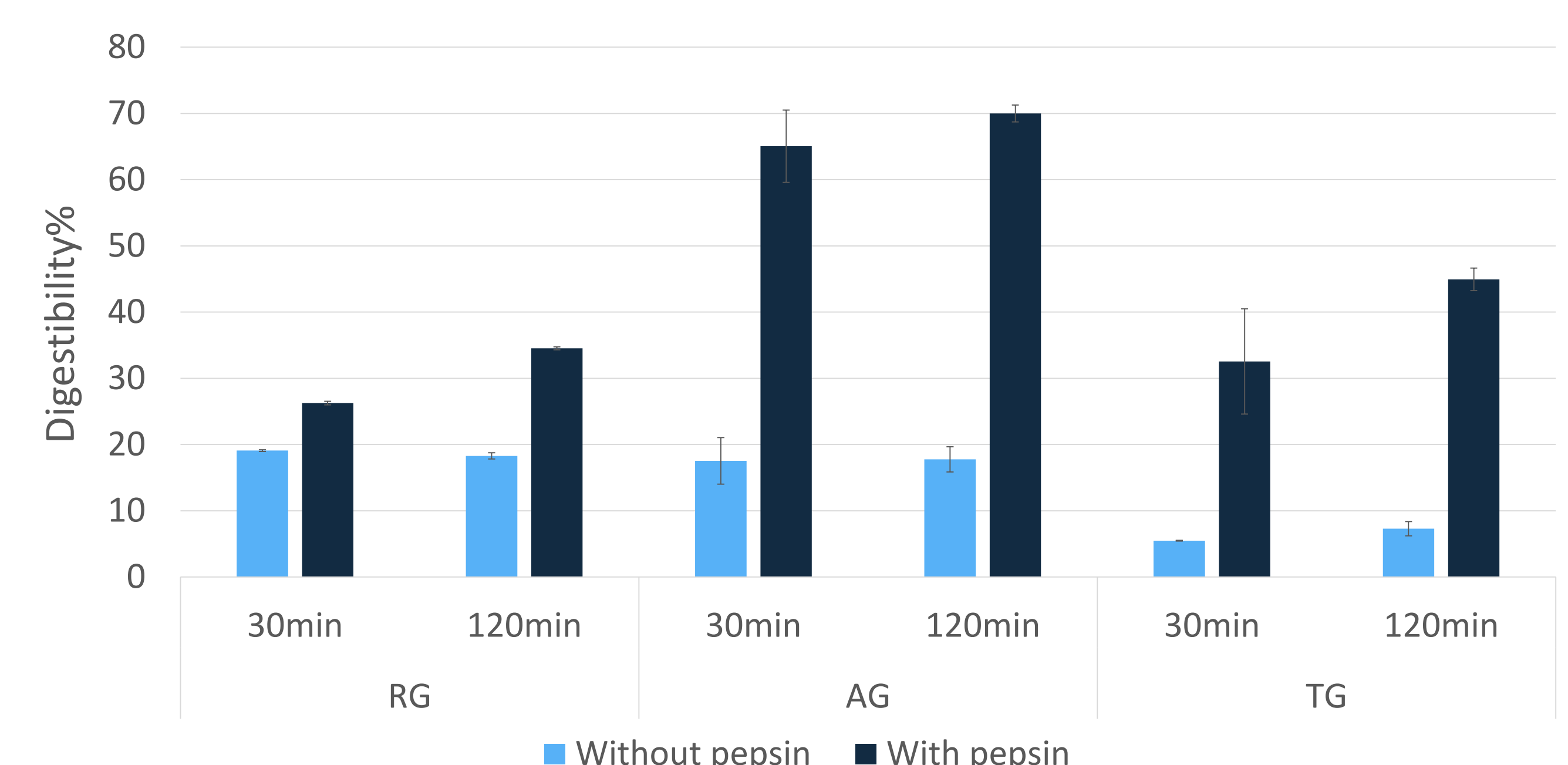
- The response detected for 5% and 10% (w/w) protein gels were similar, and the SAXS spectra for 10% gels are shown in Fig. 1.
- A clear shoulder was observed in the SAXS spectra for each gel at  $\sim 0.080\text{\AA}^{-1}$ , attributed to internal spacing of casein micelles. After 2-hrs of gastric digestion, a shift was observed in this shoulder to lower  $q$  in response to the disruption in the casein micellar structure.
- A more pronounced shift was observed for the samples subjected to gastric digestion with pepsin present, compared to those digested without.
- The second inflexion observed for the rennetted gel at  $0.035\text{\AA}^{-1}$  is due to a reduction in spacing of CCP nanoclusters likely as a result of difference in the gel pH [3].



**Figure 1** SAXS profiles of 10% casein gel digesta of the rennetted gel (RG), acid gel (AG), and transglutaminase cross-linked gel (TG) before (pink) and after 2 hours digestion with (blue) and without pepsin (green).

## GEL DIGESTIBILITY

- The rennetted gel and acid gel broke down and released more protein from exposure to digestive fluids and mechanical shear alone without pepsin, compared to the transglutaminase cross-linked gel indicating a stronger network.
- The addition of the hydrolytic enzyme pepsin showed a significant increase in the amount of soluble protein for all samples, especially the acid gel as a result of its weak structure caused by the solubilization of colloidal calcium phosphate from within the micelles at pH values below 6.0. A much larger increase in the rate of gel degradation was noted for the transglutaminase cross-linked gel with the addition of pepsin.
- In the presence of pepsin, the acid gel was shown to be more digestible than the other gel samples. However, none were completely (100%) digested after 120mins under simulated gastric conditions.



**Figure 2** Final protein concentration (%w/w) in supernatant for three different gels (rennet gel (RG), acid gel (AG), transglutaminase cross-linked gel (TG)) after 30min and 2 hours of digestion with (dark blue) and without pepsin (light blue), relative to total nitrogen in the sample was expressed as digestibility percentage as an indicator of the extent of gastric digestion.

## Conclusion

The results confirm that structural differences between acid, cross-linked and rennetted protein gels will influence digestibility.

The acidic environment of the simulated gastric fluids and the mechanical shear applied to the sample within the stomach led to some devolution of the gel protein network. However this was increased in the presence of pepsin, allowing us to systematically examine the influence of pepsin during the gastric digestion process.

