Monitoring reaction kinetics of starch by NMR spectroscopy

A.C. Dona, School of Molecular and Microbial Biosciences, University of Sydney, NSW 2006, Australia.

The glycemic index (GI) is a measure of the rate at which glucose sugar is absorbed into the circulatory system after a meal, thus providing body fuel (Brand-Mille *et al.*, 2002). The GI of a food is related to a person's physical disposition and satiety after a meal and is used as a measure of health-status including obesity and diabetes (Jenkins *et al.*, 2002). Hypothetically, starch's structure in foods, determines its rate of digestion. However, correlating the glucose digestion process with the characteristics of the starch in food remains to be studied. Characterisation of starch samples by techniques including nuclear magnetic resonance (NMR) (Le Botlan & Desbois, 1995) and size exclusion chromatography (SEC) require a stable starch solution.

NMR is one technique that can be used to study the dissolution kinetics of complex mixtures, including starch in various solvents (DMSO:H₂O) at different temperatures. The kinetics of dissolution were studied using the intensity of peaks in NMR spectra that increased as starch dissolved. Rate constants for dissolution were determined in the temperature range 30°C-80°C. Arrhenius analysis was used to estimate the activation energy of dissolution. Complete dissolution was confirmed by using dynamic light scattering (DLS).

The kinetics of the reactions involved in digestion of starch

Starch + Glucoamylase $\rightarrow \beta$ -glucose $\leftrightarrow \alpha$ -glucose

in aqueous solutions were monitored. ¹H NMR generated progress curves in a more direct way than conventional glucose assay methods (Wang *et al.*, 2006). In addition the amount of α and β glucose released and that then underwent mutarotation (Figure) were quantified. Estimates of rate constants that describe the kinetics of the dissolution process and the mutarotation of the anomers were obtained by nonlinear regression in *Mathematica*. The kinetic model will be used to correlate starch-digestion rates measured *in vitro* with the GI measured *in vivo*



The figure shows ¹H NMR (400.13MHz) time course of spectra showing the mutarotation of α -D glucose to β -D glucose. Temperature was 25°C, Acetate buffer pH 5.2. NMR parameters: $\pi/2$ pulse duration 8 μ s, number of transients per spectra 8, a line broadening of 0.3Hz was applied to each fid before Fourier transformation. *Inset:* Progress curves showing the conversion of the α -anomer to the β -anomer.

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