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The effect of fibre and gelatinised starch type on amylolysis and apparent viscosity during *in vitro* digestion at a physiological shear rate

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ABSTRACT

An *in vitro* system was used to determine if the addition of insoluble or soluble fibre to aqueous suspensions of gelatinised starch affected the rate at which the starch was digested. Pre-gelatinised potato or corn starch suspensions were digested with porcine pancreatic amylase in the presence of either finely milled insoluble fibres from various sources or with guar gum. *In vitro* digestion was conducted at 37 °C in a rheometer at a low and constant shear rate of 10 s^{-1} and the quantity of glucose released measured. The rates of starch digestion and suspension viscosity declined asymptotically and were unaffected by the addition of wheat fibre, but were considerably reduced by the addition of wood and AllBran® fibre and to a much greater extent (60%) by the addition of guar. The latter effect may be due to inhibition of amylase activity by non starch polysaccharide sequences.

1. Introduction

The presence of significant quantities of soluble or insoluble fibres in the diet increases the apparent viscosity of digesta and reduces convective mixing within the gut lumen (Lentle & Janssen, 2008; Blackburn & Johnson, 1981; Jenkins, Marchie, Augustin, Ros, & Kendall, 2004). This may hinder the movement of enzymatic secretions into substrates (Tester & Sommerville, 2003) and is likely to reduce rate of transfer of liberated nutrients to sites of absorption at the intestinal mucosa.

The effects of fibre in reducing the overall rate at which carbohydrate is digested, and hence the rate at which glucose is assimilated (Braaten et al., 1991, 1994; Jenkins et al., 1987), may be useful in the treatment of type II diabetes. However, control of post prandial blood glucose levels by such means is inconsistent and varies with the type and dose of dietary fibre (Bjorck, Granfeldt, Liljeberg, Tovar, & Asp, 1994; Samra & Anderson, 2007) and the size of the fibre particles (Granfeldt, Hagander, & Bjorck, 1995).

The physical form of starch is known to influence its digestibility for example, fully gelatinised starch is digested up to 8 times faster than non gelatinised starch (Holm, Lundquist, Bjorck, Eliasson, & Asp, 1988) and the digestibilities of various fractions within gelatinised starch differ significantly (Englyst, Englyst, Hudson, Cole, & Cummings, 1999; Butterworth, Warren, Grassby, Patel, & Ellis, 2012). The fraction of gelatinised starch that is digested within 20 min is arbitrarily termed rapidly digestible starch (RDS) is thought to account for the initial postprandial rise in blood glucose and insulin (Noda et al., 2008). Correspondingly, the fraction of starch that is digested between 20 and 120 min is termed slowly digested starch (SDS) and that remaining after 120 min, resistant starch (RS) appears to have little influence on the post prandial levels of glucose and insulin. These divisions are considered to have little relevance in representing physico-chemical properties of the starch (Butterworth et al., 2012). The rate of starch digestion varies with the type of starch (Biliaderis, 1991; Naguleswaran, Vasanthan, Hoover, & Bressler, 2014), the surface to volume ratio of

Abbreviations: RDS, rapidly digested starch; SDS, slowly digestible starch; RS, resistant starch; P, potato starch; C, corn starch; WF600, WF600 wheat fibre; Pr, Prolux wheat fibre; W, wood fibre; Ab, Kellogg's AllBran® fibre; G, guar gum.

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ungelatinised granules, the presence of other plant chemicals (Nebesny, Rosicka, & Tkaczyk, 2004; Noda et al., 2008; Snow & Odea, 1981), the surface porosity of ungelatinised granules (Sujka & Jamroz, 2007) and the degree to which the starch is gelatinised (Wang & Copeland, 2013). The addition of fibre during cooking of the food is reported to limit the extent to which starch is gelatinised, increasing the proportion of RS (Bjorck et al., 1987) and "masking" the starch granules (Parada & Aguilera, 2011). The rate of digestion of gelatinised starch can be modelled in various ways although during the early stages of digestion, regressions of L_n transformed starch concentration with time were shown to accurately model the data for the digestion of gelatinised starch from various sources (Butterworth et al., 2012).

However, it is noteworthy that while effects of the above factors have been reported for the *in vitro* digestion of starch, few of these studies have related differences in systemic levels of blood glucose *in vivo* to the rate of glucose production during *in vitro* digestion. One such study, showed that the rate of increase in the concentration of glucose in venous blood in the hepatic portal system of pigs fed a range of partially purified cereal starches was generally lower than that during *in vitro* digestion but that the correlation improved when the effects of physiological processes such as gastric emptying were incorporated into the model (van Kempen, Regmi, Matte, & Zijlstra, 2010). Similarly, the adjustment of *in vitro* data for rates of apparent glucose disposal in humans increased the accuracy of predicting the glycaemic response to human foods (Monro, Mishra, & Venn, 2010). Hence it appears that digestion *in vitro* can be used to estimate the effects of starch structure on digestive processes *in vivo* if allowance is made for the *in vivo* factors involved in glucose homeostasis.

In this study, an *in vitro* system was used to determine if the addition of soluble or insoluble fibre to suspensions of gelatinised starch influenced the rate at which the starch was digested. Hence we compared changes in the apparent viscosities and digestion of fully gelatinised potato and corn starches alone and when mixed with either of two finely divided wheat fibre types, a finely ground wood fibre, a modified commercial high fibre breakfast cereal (Kellogg's AllBran®) or guar gum. The four types of insoluble fibre were chosen on the grounds that they differed chemically, for solubility and in particle size distribution.

2. Materials and methods

The gelatinised starches and mixtures of these starches with each of the four fibre types were subjected to an *in vitro* digestive regime that simulated an initial gastric and subsequent small intestinal environment. Digestion was carried out at a constant shear rate of 10 s^{-1} using a stress-controlled rheometer (Rheometrics SR500; Rheometrics Instruments, Piscataway, NJ, USA) fitted with cup and vane geometry maintained at 37 °C throughout each experiment. A shear rate of 10 s^{-1}, was used to keep the fibre and starch in suspension and was close to reported physiological shear rates (Lentle et al., 2007). The viscosity of the digestate was continuously recorded throughout the experiment and subsamples of digestate (250 μL) taken at intervals over 120 min to determine the rate at which starch was hydrolysed. Each treatment was replicated twice.

2.1. Fibres

The insoluble fibres were chosen on the basis of differences in their physical morphology and chemical composition (Fig. 1). They comprised two commercially available preparations of insoluble, cellulosic, food grade fibre products derived from wheat 'WF600' (J. Rettenmaier & Söhne, Rosenberg, Germany) and 'Prolux' (Oppenheimer Pty Ltd., NSW, Australia); a finely milled food grade wood fibre (Lignocel® Type C120, J. Rettenmaier & Söhne, Rosenberg, Germany) with high lignin content (Table 1), derived from a species of *Pinus* and supplied by Plant and Food Research Ltd and the fibre component of Kellogg's AllBran® that was derived from wheat bran. The proportion of crude insoluble fibre in the particulate materials was determined by accredited methods (Robertson & van Soest, 1981) in the Nutrition Laboratory at Massey University. Guar gum, a soluble, non-starch, food grade fibre containing 85% of hydrateable galactomannans; was supplied by Formula Foods Corporation Ltd (Christchurch, NZ). Guar was chosen as it is recognised by the FDA (US Food and Drug Administration) as being effective in controlling hyperglycaemia (Finley et al., 2013).

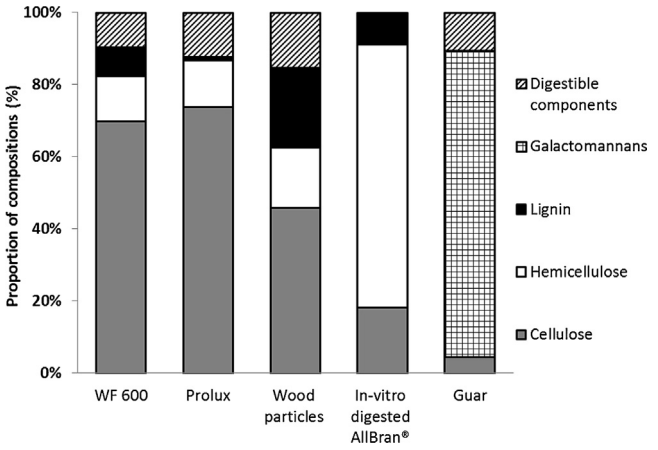

Fig. 1. Chemical composition of the five fibre types.

2.2. Preparation of AllBran® fibre

The commercial AllBran® product comprised about 70% of digestible material, probably in the form of starch and sugars that were used to formulate the breakfast food. The insoluble fraction of the AllBran® fibre was obtained by *in vitro* digestion (Section 2.7) using simulated gastric (pepsin at ~pH 2 for 30 min) and small intestinal (pancreatin/amyloglucosidase at pH 6 for 2 h) digestion at 37 °C. The residue (Fig. 1) was subsequently washed and dried at 40 °C for 12 h (Hardacre, Yap, Lentle, Janssen, & Monro, 2014) before use.

2.3. The concentration of fibres used

Since the objective of this work was to approximate conditions in the small intestine, the suspension viscosity was adjusted to be similar to the mean viscosity of the digesta removed at autopsy from the small intestine of six slaughtered pigs. The concentration of 'as supplied' fibres used was between 6.8 and 9.8% w/w depending on the moisture content and bulk density of the fibre. Similarly a 0.4% (w/w) concentration of guar was used as it had an apparent viscosity similar to that of the suspensions of hydrated insoluble fibre.

2.4. Starch

Two commercial unmodified starches commonly used by the food industry were used for this work. Potato starch (Wind Mill, Holland) was supplied by National Starch (NZ) and corn starch was supplied by the NZ Starch Company (NZ). The particle size of potato starch was between 25 and 50 μm and that of corn between 10 and 30 μm. Both contained between 20 and 25% of amylose and 80–75% amylopectin. The protein content of the starches was assayed by the