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Does viscosity or structure govern the rate at which starch granules are digested?

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ABSTRACT

The rates of in vitro digestion of incompletely or fully gelatinised potato and corn starch were measured at 37° C over 20 min in a rheometer fitted with cup and vane geometry at shear rates of 0.1, 1 and 10 s^{-1}. Shear rate did not influence the rate of starch digestion provided it was close to physiological levels. However, rates of digestion were significantly reduced when shear rates were below the physiological range (0.1 s^{-1}) or when gelatinisation was incomplete. At physiological shear rates the relationship between starch digestion and viscosity was sigmoid in form and following a short initial slow phase a rapid decline in viscosity occurred as starch was digested and the structural integrity of the granules was lost. Conversely, when shear rate was reduced below physiological levels or gelatinisation was incomplete, digestion was hindered, granule integrity was maintained and the relationship between starch and viscosity became linear.

1. Introduction

Starch contributes around 60% of the total energy in the human diet (Bjorck, Granfeldt, Liljeberg, Tovar, & Asp, 1994; Copeland, Blazek, Salman, & Tang, 2009). However, the rapid digestion of starch can lead to the sustained elevation of blood sugar and associated development of illnesses such as metabolic syndrome (Riccardi & Rivellese, 2000). Hence strategies to control the rate at which starch is digested in processed foods may be important for the maintenance of metabolic health (Nuttall, 1993).

A number of in vivo and in vitro studies have shown that the rates at which starchy foods are digested vary (Holm, Lundquist, Bjorck, Eliasson, & Asp, 1988) with the extent to which the starch granules are gelatinised (Englyst, Kingman, & Cummings, 1992). Hence the rate of digestion of gelatinised starch by amylase is 3–10 times greater than that of native un-gelatinised starch (Holm et al., 1988; Noda et al., 2008; Snow & Odea, 1981). This effect results from disruption of the physical structures of amylose and amylopectin within the starch granules as they are hydrated during gelatinisation (Gallant, Bouchet, & Baldwin, 1997) and hence access to the starch molecules by digestive enzymes. The temperature at which starch granules are cooked (Holm et al., 1988), their water content (Gunaratne, Ranaweera, & Corke, 2007) and the duration of cooking (Li & Yeh, 2001) can also affect the extent of gelatinisation and hence digestibility. Thus, post prandial blood glucose levels do not rise consistently in proportion to the quantity of starch that is consumed (Ells, Seal, Kettlitz, Bal, & Mathers, 2005; Parada & Aguilera, 2009).

It is known that agents that increase the viscosity of digesta, such as inert particles, guar or beta-glucan can also reduce the rate at which starch is digested, possibly by impeding the admixture of enzyme with substrate (Hardacre, Yap, Lentle, & Monro, 2015; Smits, Veldman, Verstegen, & Beynen, 1997; Wood, Braaten, Scott, Riedel, & Poste, 1990). Shear stress developed by the gut has rarely been estimated but values up to 1.2 Pa have been reported (Jeffrey, Udaykumar, & Schulze, 2003). The apparent viscosity of digesta measured at physiological shear rates of between 0.1 s^{-1} and 10 s^{-1} has been reported to range between that of water (~0.0008 Pa s) to 0.1 Pa s in the small intestine (de Loubens, Lentle, Love, Hulls, & Janssen, 2013), almost 10 Pa s in the small intestine of pigs (Takahashi & Sakata, 2004) and 73 Pa s in the caecum of chickens (Takahashi, Goto, & Sakata, 2004). At such high viscosities the mixing of food with digestive enzymes is likely to be hindered (Holm et al., 1988; Svihus, Uhlen, & Harstad, 2005) as would be the rates of digestion and subsequent absorption of digestive products. Therefore, any in vitro method that is used to gain insight into the

Abbreviations: P, potato; C, corn; G%, degree of gelatinisation; Ck, cooking time.

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dynamics of the digestion of particular substrates must replicate the viscosity and shear rate and hence shear stress conditions that occur in the gut (Lee, Bailey, & Cartwright, 2003; Lentle & Janssen, 2008; Shelat et al., 2015).

The aim of the current work was to determine relationships between the rates of enzymatic digestion of aqueous suspensions of starch granules from two representative but diverse food plant species that have undergone various degrees of gelatinisation and the viscosity of the suspension at shear rates within the physiological range (0.1 s^{-1}, 1 s^{-1} and 10 s^{-1}) using an in vitro system that replicated the gastric and intestinal phases of digestion. Changes in the physical states of the starch granules during digestion were inferred from the relationship between the proportion of undigested starch and viscosity. The rates at which the starches were digested were determined indirectly from the rates at which glucose was liberated during amylolysis while the rates at which the physical state of the starch changed during digestion were determined indirectly from the rate of reduction in apparent viscosity of the digestate.

2. Materials and methods

2.1. General methodology

The degree of gelatinisation of starch (DG) can be determined by thermodynamic methods, notably differential scanning calorimetry (DSC) (Holm et al., 1988; Parada & Aguilera, 2009, 2011). However, it can also be determined indirectly by changes in viscosity of an aqueous starch suspension as it is heated to around 95 °C (Gunaratne et al., 2007; Kaur, Singh, McCarthy, & Singh, 2007). Suspensions of gelatinised starch are stable and do not settle, however, the density of non gelatinised granules is such that they must be suspended in a viscous medium to reduce their rate of settling. In this work we used a 70% fructose solution with a Newtonian viscosity of 0.04 Pa s. This was useful in determining the behaviour of suspensions of starch granules but the high concentration of fructose reduced water activity (A_w = 0.74) and largely inhibited gelatinisation. Exploitation of these properties allowed the continuous determination of changes in viscosity during digestion within the cup and bob geometry of a rheometer.

Three replicates each of either ungelatinised potato (*Solanum tuberosum* L.) or ungelatinised corn (maize, *Zea mays* L.) starch were each suspended in 70% fructose solutions. Similarly, three replicates of suspensions of potato (P) or corn (C) starch were each suspended in MilliQ water, and heated for either 10 or 30 min to partially or fully gelatinise the starch, stirring was maintained during gelatinisation. All suspensions were subsequently mixed with enzymes representative of the gastric and small intestinal phases of digestion and maintained under these conditions for 20 min during which they were continuously subjected to shear rates of 0.1, 1 or 10 s^{-1}. The apparent viscosity of each of the suspensions was continuously recorded and 250 μL aliquots withdrawn for determination of glucose at 0, 1, 2, 5, 10, 15 and 20 min after the addition of amylase that represented the commencement of the small intestinal phase of digestion.

2.2. Starches

Two commercially available unmodified (native) starches, commonly used in food industry, were used in this work. Potato starch (WindMill, Holland) was supplied by National starch (NZ) and corn (maize) starch supplied by the NZ starch company (Auckland, NZ). The moisture contents of the potato and corn starches determined by oven drying to constant weight at 108 °C were 15.5% and 11.3% respectively.

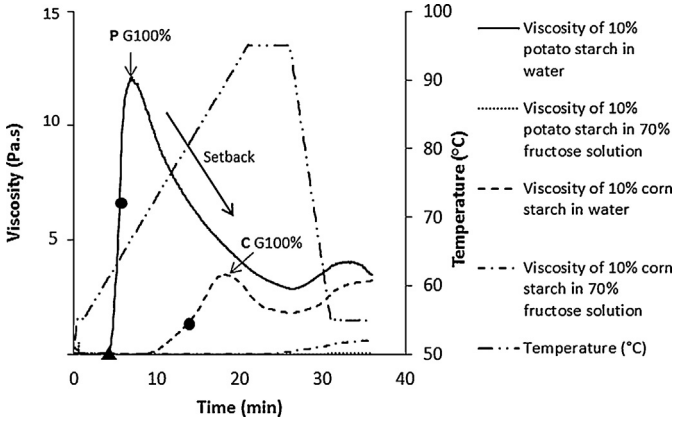

Fig. 1. Rheology of starches during gelatinisation: RVA pasting profile for aqueous suspensions of 10% (w/w) potato (P) or corn starch (C), ▲ = G0%, ● = G50%, G100% marked on graph.

2.3. Determination of the degree of gelatinisation

The degree of gelatinisation (G) was determined indirectly from changes in the apparent viscosity of aqueous suspensions incorporating 10% (w/w) of potato or corn starches in MilliQ water during heating in an RVA (Rapid Visco Analyser, RVA-4, Newport Scientific, Warriewood, Australia). The starch suspensions were initially stirred at 900 rpm for 60 s to suspend the starch granules, subsequently reducing to 160 rpm which was maintained throughout the remainder of the procedure. The samples were then heated from 55 °C to 95 °C at 2 °C/min and the temperature held at 95 °C for 5 min before cooling over 5 min to 55 °C. The apparent viscosity was recorded throughout this procedure (Fig. 1). The maximum viscosity during gelatinisation is termed the "pasting viscosity" (PV) and the temperature at which it occurred, the "pasting temperature" (PT) were both recorded. These estimations were repeated for three replicates of the potato and corn starch suspensions and the mean PV and PT for each determined. The temperatures at which 50% and 100% of the PV was developed were recorded for the potato and corn starches and the degree of gelatinisation at each temperature nominally defined as G50% or G100% (Fig. 1).

2.4. Preparation of starch samples for digestion

Gelatinised starch suspensions of 50 mL each for the digestion experiments were prepared from 10% (w/w) suspensions of native P or C starches in MilliQ water in 150 mL beakers according to the conditions determined in Section 2.3. Each starch suspension was first stirred at 150 rpm for 5 min using a magnetic stir-bar to thoroughly disperse the component granules, the stirring speed was then reduced to 50 rpm to minimise damage to the granules as they were gelatinised. Gelatinisation was carried out in a water bath for either 10 or 30 min at the temperature previously determined to achieve the desired G%. Temperatures appropriate for G100% were applied for 30 min only to the P and C starches suspended in the 70% fructose solution.

2.5. Hydration properties

The swelling factor (Q) for each of the starch granule types was determined as the ratio of their wet weight, after either soaking or gelatinisation, to their dry weight (Bagley & Christianson, 1982). Where the dry weight of the starch added (5 g) was calculated using the previously determined moisture content of the 'as supplied' starch.