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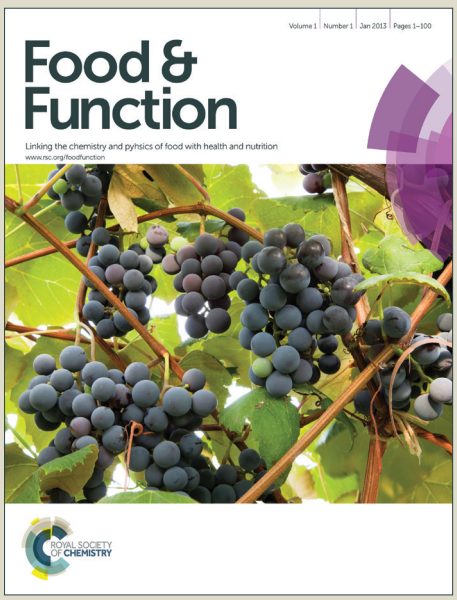

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Influence of hydration and starch digestion on the transient rheology of an aqueous suspension of comminuted potato snack food

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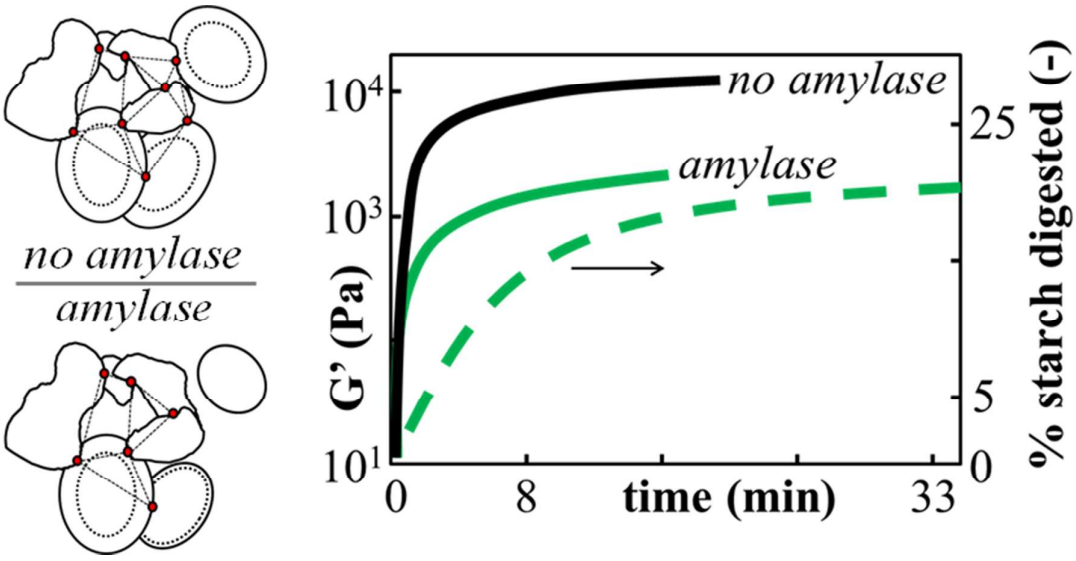
Keywords: Starch, α-amylase, enzyme kinetics, hydration, oral processing, bolus, rheology, structure-function, elasticity, log of slope, percolation

Graphical Abstract

Hydration kinetics controls 'bolus' rheology and starch digestion of comminuted snack foods; *in vitro* rheology provides new insights into oral processing and food design



Abstract

Oral processing of most foods is inherently destructive: solids are broken into particles before reassembly into a hydrated bolus while salivary enzymes degrade food components. In order to investigate the underlying physics driving changes during oral processing, we capture the transient rheological behaviour of a simulated potato chip bolus during hydration by a buffer with or without α-amylase. In the absence of amylase and for all oil contents and solids weight fractions tested, we find a collapse of the transient data when graphed according to simple Fickian diffusion. In the presence of amylase, we find effects on the transient and pseudo steady state bolus rheology. Within the first minute of mixing, the amylase degrades only ≈6% of the starch but that leads to an order of magnitude reduction in the bolus elasticity, as compared to the case without amylase. Thus, for an *in vitro* bolus, only a small amount of starch needs to be digested to have a large impact on the bolus rheology very soon after mixing.

1. Introduction

Oral processing of food creates a bolus whose structure and rheology is continuously altered as it is prepared for swallowing and digestion[1]. The behaviour and properties of the food bolus during oral processing follow a physico-chemical trajectory or breakdown path[1] that impacts the sensory perception during consumption[2, 3]. To reduce complexity when considering this process, we have recently rationalized oral processing as subjecting food to a set of sequential unit operations[4]: first bite, comminution, granulation, bolus formation & processing, swallow and residue. While many of these processes may occur simultaneously, the utility of this approach is that it allows a systematic and controlled investigation into structure-property relationships within any one operation using *in vitro* measurement techniques, e.g., techniques associated with rheology and tribology [4]. An important aspect of oral processing, spanning all unit operations, is the continual secretion—in the mouth of healthy individuals—of saliva, which contains amylase and other components. We focus here on elucidating the impact of hydration and α-amylase on the rheology (transient & pseudo steady state) of a comminuted brittle potato-based snack food.

Oral processing of solid foods involves comminution of the food material to smaller particles that mix with saliva to form a food bolus. The saliva plays a role in aggregation, hydration, dissolution and dilution of the food particles[5-7]. In addition, saliva contains enzymes that are capable of digesting different components in food, e.g., amylase is capable of digesting starch[8]. Amylase lowers the viscosity of gelatinised and hydrated starch suspensions within seconds[9, 10], which is speculated to affect the sensory perception of soft foods such as custard during consumption[11]. However, for foods containing starch with low moisture content, hydration occurs simultaneously with enzymatic degradation. Thus dry solid food that requires longer chewing times before being acceptable for a safe swallow will have longer in-mouth residence times and thereby will be exposed to amylase for longer, compared to soft, well lubricated foods that require little processing before a safe swallow. Remarkably, there is limited knowledge on the effect of salivary α-amylase on the properties of solid-food composites in the context of oral processing.

Salivary α-amylase is capable of hydrolysing starch, but the extent to which this occurs in the mouth and stomach is not well understood[8]. The amylolytic breakdown of various starches using α-amylases from porcine, human and bacterial sources has been investigated extensively (see for example[9, 10, 12-16]). It has been proposed that salivary amylase "initiates" amylolysis of starch in the mouth[5, 16-18] and "during the first few minutes that the food is in the stomach"[19]. Carpenter[18] suggests that salivary amylases are less important to digestion and that their main role is to clear the oral cavity of substances that would otherwise be available for microbial growth, e.g., by aiding removal of starchy foods that may pack in the teeth during and following mastication. We note here that this action is also important to mouthfeel as tooth-packing is an undesirable sensory percept associated with many snack foods. In addition, we also note that this clearance activity highlights that oral processing time scales are much longer than those associated with just the presence of the food bolus in the mouth, i.e., the residue phase is important to overall product perception. In the stomach environment, the review of Carpenter[18] highlights that the salivary α-amylase activity is "greatly reduced", but we infer from others work[20] that salivary amylase is potentially

protected within the bolus so that it can be active in the stomach for a significant amount of time[14]. To support the notion that salivary amylase assists stomach digestion, Bornhorst *et al.*[14] show that the effective modulus of rice boluses in the presence of a bacterial α-amylase in simulated gastric juices decreases over a 3 hour period. In addition, bolus rheology is considered to play a role in the glucose release from foods as it passes through the digestive tract to the stomach[21].

In considering the evolution of brittle snack food bolus rheology during oral processing and along the digestive pathway, we have previously shown that comminution of potato chips (PCs) containing varying oil levels and mixed with physiological buffer leads to an increase in dispersion modulus with time[22]. This increase is proposed to be predominately associated with the hydration and swelling of the food particles, including starches and plant cell wall material, and dissolution of any soluble components. Over time scales much longer than oral processing times, we demonstrated[22] that the dispersion rheology can be predicted from knowledge of the solids and oil content. In particular, we showed that oil can alter the *in vitro* formed food bolus in a predictable manner, and we suggested that this effect may be relevant to how the food bolus is perceived during oral processing[22].

We now address the temporal development of the rheology of comminuted PCs during the hydration process and during coupled hydration & starch digestion. The impact of oil content on the rheology of the bolus is also investigated. We aim to show the development of appropriate experimental protocols that address in a systematic way the effect of liquid (hydration, dissolution, dilution) and enzymatic degradation on food boluses at time scales appropriate for oral processing. The goal of this research is develop a framework that can be used to model the behaviour of PCs—and other foods—undergoing oral processing, which we can then use, in combination with knowledge of the food microstructure, to predict their behaviour *in vivo*. Such insight will assist in the rational design of snack foods that are optimised for both health & nutrition and consumer acceptability.

2. Materials and methods

Unless otherwise stated, all reagents were obtained from Sigma Aldrich (Castle Hill, NSW, Australia) and were of the highest available grade.

2.1 Buffers: A sodium acetate buffer (pH 6.0) is used. The buffer is prepared by adding 11.8 mL of glacial acetic acid (SG 1.05 g/mL) to 900 mL of deionised water, adjusting the pH to 6.0 with 2 M NaOH. 4 mL of 1 M $CaCl_2$ solution and 100 µL of 4.9 M $MgCl_2$ solution are added, and then the solution is made up to 1000 mL with pure water and refrigerated (≈ 4°C) until use. All buffers are fridge stored until the day of testing. On the day of testing, buffers are equilibrated to 23 °C +/- 0.5 °C before use.

2.2 Potato chips: Unsalted and thinly cut potato chips (PC) with controlled oil content are used: 22.9% w/w oil (LF) and 33.5% w/w oil (FF), where FF is the unsalted equivalent of store-bought Lay's® Classic Potato Chips. The PCs are experimental chips provided by PepsiCo., USA; the production date and oil content are the only variable properties. All chips

are freezer stored (≈ -18 °C) in sealed bags until use. As previously reported, the PC thickness is ≈ 1 mm, which does not depend upon oil content[22].

2.3 α-amylase: Porcine pancreatic α-amylase (powdered, 150,000 U/g, Megazyme, Bray, Ireland) is added to the sodium acetate buffer. For the majority of the work detailed here, we use 200 U/mL for porcine pancreatic α-amylase, based on prior experience and literature values for the activity of amylase in human saliva[23] (where a unit of activity is defined as the amount of enzyme that will liberate 1.0 mg of maltose from starch in 3 min at pH 6.9 at 20 °C). From literature, we note that, despite minor structural differences between human salivary α-amylase and porcine pancreatic α-amylase[24], the activity and action pattern of the two enzymes are comparable[8]. Saliva is complex chemically, structurally and rheologically, and it also alters significantly with time after expectoration[4, 25]. It is challenge to control its properties and challenging to associate 'cause and effect' when dealing with whole saliva. For this reason, we isolate key aspects individually and in this case focus on hydration and amylase degradation of starches.

2.4 Comminution of PC samples: Each PC sample is removed from the refrigerator, immediately added to a Bosch food processor (MCM4100AU, Bosch) and comminuted using a periodic method comprising 5 seconds 'on' and 5 seconds 'off,' for 5 sets. Comminuted PCs are then transferred into individual metal cups and sealed with Parafilm (Labtek, Parafilm) to limit softening of the PC *via* moisture ingress. All samples are left to equilibrate to room temperature, i.e., 23 °C +/- 0.5 °C, for one hour before testing, and all crushed samples are used on the day of comminution. The comminution process results in a mixture of plant cells (with free cavities and cavities containing starch granules), cell clusters, free starch granules and the associated oil.

2.5 Mixing of crushed PC and buffer or amylase solution: The standard procedure consists of mixing the comminuted PC and buffer or amylase solution using a plastic spatula: the metal cup in which the crushed PC are stored is set in place on the rheometer, the buffer is poured into the metal cup and the spatula is used to mix the sample for 5 seconds. The vane tool is then brought into place, and the rheological testing is started; the loading of the vane takes approximately 15 seconds.

For a subset of samples, i*n situ* mixing was performed (in place of spatula mixing) to avoid the time taken to load the vane into place prior to measurement. *In situ* mixing involves lowering the vane tool into the solid comminuted PCs, adding buffer or amylase solution while shearing for 5 seconds at 50 s^{-1}. The rheological measurements are then started on cessation of shear. In this case, time zero corresponds to the beginning of mixing.

In situ[26] mixing using the vane is desirable over *ex situ* mixing for two reasons: (1) mixing is potentially more controlled; and (2) there is potential to capture the dynamics at shorter time scales following mixing.

For convenience, we will refer to the aqueous suspension of comminuted PC particles as a 'simulated bolus,' noting, in particular, that 'simulated' is used in the context of the bolus formation & processing stage of oral processing. This is not to imply that our

comminution method simulates *in vivo* chewing; for simulated chewing, we direct the reader elsewhere[27].

2.6 Amylase Digestion studies: Samples are prepared in an identical fashion as for rheological testing. Only the lowest PC solids content, i.e., 11 g of PC in 50 g of amylase solution, is used for digestion experiments, as higher PC solids weight fractions do not have enough free liquid to allow sampling of the digesta. Following mixing of the amylase solution and PCs, 300 µL aliquots are removed immediately upon mixing, i.e., time zero, and at a series of time points between 1 and 35 min post enzyme addition. Aliquots are immediately mixed with 300 µL of 0.3 M $NaCO_3$ in a 1.5 mL microfuge tube in order to halt the enzyme reaction[28]. Aliquots are then centrifuged to remove unreacted insoluble PC solids, and 5 µL of the supernatant is removed to a clean 1.5 mL microfuge tube, to which is added 95 µL of deionised water. The PAHBAH reducing sugar assay [29] is employed to quantify the reducing sugar products of starch digestion, using maltose as a standard.

2.7 Rheological testing: Small amplitude oscillatory shear rheology measurements are performed on the PC particle dispersions at stresses and strains that are within the linear viscoelastic regime. A TA AR1500 stress-controlled rheometer is used with a 22 mm diameter titanium vane tool in a 44 mm diameter aluminum cup. The vane is lowered into the sample to a depth of 11 mm above the cup bottom.

The time dependence of the linear viscoelasticity (storage and loss moduli) of the sample following dispersion of the PC into buffer or amylase solution is characterised at a constant frequency of 6.28 rad/s. We denote the time as an apparent time, t_{app}, to acknowledge that there is a finite time between the start of the measurement and when the sample is mixed with buffer or amylase solution (note, this time is typically of the order of 15 seconds). The viscoelasticity of the suspensions are monitored with time for up to 20 minutes.

3. Results and Discussion

3.1 Digestion of potato chips by α-amylase: Starch digestion by α-amylase has been shown to follow pseudo first order kinetics and can be described using the familiar first order rate equation[30]:

$$C_t = C_\infty(1 - e^{-kt}) \qquad [1]$$

C_t is the proportion of starch digested at a given time, C_∞ is the product concentration at the end point of the reaction, t is time and k is the first order rate constant. Under the conditions employed in the present experiment, the digestion of the starch in comminuted PCs by amylase follows first order kinetics, as shown by the open squares in figure 1; this is consistent with a previous study on potato chips [31]. The rate of starch digestion here is faster than that reported in Butterworth *et al.*[30] for PCs because we have used higher enzyme and substrate concentrations; the conditions here more closely match the environment in the oral cavity while the previous study was designed to mimic duodenal starch digestion (discussed further below).

The influence of starch digestion by amylase on the rheology of comminuted PC suspensions with time, following *in situ* mixing in physiological buffer, is shown in figure 1. G' increases rapidly with time within the first two minutes in the buffer (with or without amylase), as the PC solids hydrate. The insert in figure 1 highlights that reliable measurements are only achieved after about 15 seconds of first exposure to buffer or amylase solution (note, time zero is the start of mixing). Amylase affects the rheology within 30 seconds, which corresponds to the digestion of only 5-6% of starch. These results demonstrate that the digestion of starch by amylase markedly affects the rheological properties of comminuted PC suspensions, but we observe that less than a fifth of the total starch is digested even after 20 minutes. The G' and the amount of digested starch in the amylase buffer approach a plateau with time, which suggests that a majority of the starch in this sample is protected from amylase digestion under the conditions explored. This hypothesis is supported by a recent paper showing that starch structures can be intrinsically resistant to enzyme hydrolysis[32].

[figure 1 here]

Common approaches to solving equation 1 for a given data set include direct fitting through non-linear regression and taking logarithms of the equation and plotting the data in semi-logarithmic form. These approaches, however, require that the value of C_∞ be known to a high degree of accuracy, which is not always possible. Here, we analyze amylase digestion data using the logarithm of slope (LOS) modification of the Guggenheim method: we plot the log of the first derivative of the starch digestion curve, yielding a straight line with a slope of $-k$ and a y-intercept equal to $\ln(C_\infty k)$:

$$\ln\left(\frac{dC}{dt}\right) = \ln(C_\infty k) - kt \qquad [2]$$

An additional advantage of plotting digestion data in the form of equation 2 is that the resultant plot is sensitive to the presence of multiple-rate processes and may reveal whether digestion progresses as a single first order process or through more than one digestion process[30].

The rate of starch digestion observed in the present study, as determined by the LOS plot method (see figure 2), is k =0.0022 s^{-1} (+/- 0.0003 s^{-1}), with a half-life for the reaction of 5.3 min. The terminal extent of digestion in the present study was 24.3 % of the total starch in the system. It seems that the total available starch for digestion was limited by the hydration of the sample when the PC was simultaneously hydrated and hydrolysed. When the PC was pre-hydrated for 3 hours, as shown by the open triangles in figure 2, an initial rapid digestion phase with a half-life of 16 min (k = 0.007 s^{-1} +/- 0.002 s^{-1}) occurs during which ≈15% of the total starch in the system is digested, followed by a slower digestion regime with a half-life of 30 min (k = 0.0004 s^{-1} +/- 0.0002 s^{-1}) that accounts for a further ≈35% of the starch digestion.

[figure 2 here]

During the initial phase (up to ≈500 s), the percentage of starch digested for both systems is similar, suggesting the presence of a rapidly hydrating fraction of starch that is readily available for enzyme action. After 500 s, the data for pre-hydrated and hydrating PCs diverge; the hydrating sample reaches an asymptotic limit, whereas the pre-hydrated sample continues to be digested. This suggests the existence of a fraction of starch that is rapidly hydrated and readily accessible to enzyme digestion, whereas the majority of the starch is much slower to hydrate and is digested at a much slower rate. In the pre-hydrated PC, an overall starch availability of ≈50% for digestion is well in line with the value of 40% found in the study by Goni *et al.* [31] .

3.2 Mass transfer, hydration and implications for transient rheology: In the context of PC solids in amylase solution, hydration acts concurrently with enzymatic digestion. It is therefore necessary to consider how these kinetic processes operate together to determine their effect on the evolution of bolus rheology. To rationalise the connection between hydration and time dependent rheology, we suggest here that the suspension modulus is dependent upon the movement of water through the packed PC particles and the movement of water into the dehydrated, [partially] gelatinized starch granules. Because the particles are loosely packed and the suspension is mixed prior to rheological testing, we assume that water movement to the particles is a zero order process. Therefore, we hypothesize that the percolated-like network develops according to mass transfer of water into the starch granules and plant cellular matter.

The increase in G' with time following *ex situ* mixing of PC solids in buffer or amylase solution was measured for the LF and FF PCs at several different solid weight fractions. All samples tested are soft solids, i.e., G' > G" at all times tested. An example of typical data is shown in figure 3 for LF PC at two PC weight fractions, 18% and 29.5% w/w. It is noted that at equivalent PC weight fraction in buffer only, the LF simulated PC bolus has a larger G' than the FF simulated PC bolus.

[figure 3 here]

Considering the process of hydration, the kinetics of water adsorption into cereal grains, nuts and starch has been interpreted using empirical models[33-35] and quasi-analytical solutions to Fick's Laws[36-38] . It is found that the hydration process can follow either a square-root[39-41] ($\sqrt{t}$) or exponential[42] (e^{-t}) dependence on time, and it is commonly observed that $\sqrt{t}$ is followed at short times and e^{-t} is followed over long times. We propose that the evolution of G' with time follows similar kinetics to hydration processes. The lines in figure 3 are fits to the data using the exponential series shown by equation 3. By graphing the raw data as ln(1-G'/G'$_\infty$) versus time, whereby a straight line would indicate a single rate controlling process, we find at least two linear regions, indicating a sequence of dominant rate controlling processes (*data not shown*). Therefore, we show in figure 3 a single exponential term and two exponential terms fits to the raw data; we find that two exponential terms provide a good fit to the experimental data. Two stages are also observed by plotting G'$_r$ = G'/G'$_\infty$ against $\sqrt{t}$ (see figure 4, open symbols). The observation of two first order rate processes is also found for the hydration of pasta, by Cunningham *et al.*[36]. Note that G'$_\infty$ in equation 3 is defined here as the pseudo-steady state G' values taken at a time of 20 minutes.

$$G'_r = G'/G'_\infty = a_0 + a_1 e^{-k_1 t} + a_2 e^{-k_2 t} + \ldots \quad [3]$$

[figure 4 here]

We would like to make one note on the choice of $\sqrt{t}$ or e^{-t}. For calculating an approximate diffusion coefficient (which we do below), for modelling the transient data over the full 20 minutes (figure 3) and for identifying the controlling kinetic processes over 20 minutes, we feel the e^{-t} form is best suited because, as we stated earlier, e^{-t} better fits long-time data. The $\sqrt{t}$ form is well suited to short time behaviour, and it is in the short time that we see differences from PC weight fraction & oil content on the transient behaviour. As we will show below, the pseudo steady state elastic modulus of the matrix phase does not depend upon oil content, but, as we show in figure 5, the transient elastic modulus is affected by oil and solids content. Either form is a valid solution to Fick's second law of diffusion; which form one uses depends on what transient behaviour one wishes to highlight.

[figure 5 here]

We take the approach that the bolus rheology develops simultaneously with hydration and enzyme action and investigate how the full collection of data scales with $\sqrt{t}$ by graphing G'$_r$ vs. $\sqrt{t}$ in figures 4 and 5 for LF and FF PCs at PC weight fractions ranging from 18-29.5% w/w. When no amylase is present (open symbols, figure 4), we see good collapse of all the G' data to essentially a single curve; the magnitude of differences observed between samples are in line with individual sample variations. We suggest this result arises because the kinetics of water absorption is not a function of solids or the fat content. We would like to stress,

however, that the magnitude of G' at any time (including G'_{∞}) is a function of both solids content and oil content.

Remarkably, the rate constant associated with the growth in modulus over the duration of the experiment (k_1 = 0.003 +/- 0.001 s^{-1}), in the absence of amylase, is similar to the enzyme digestion rate constant (k = 0.0022 +/- 0.0003 s^{-1}) calculated using equation 1 and the data in figures 1 & 2. We suggest that the rate determining step for starch digestion is water absorption, which is also what drives the growth in modulus with time for the PC suspensions in buffer when amylase is not present.

When amylase is present in the PC suspensions (closed symbols in figure 4 and all data in figure 5), we find a noticeable effect on the elasticity from the solids content and potentially the oil content. We also found for the LF samples at 25-29.5% w/w PC that there was no apparent linear region at short times (closed symbols in figure 4). The lack of a single relationship across the full sample set is because hydration of the PC particles and enzymatic degradation of starch are coupled. G' is strongly dependent on solids fraction, as shown in figure 3. In simplistic terms, if we consider the system to be a packed suspension of soft particles, hydration of dispersed particles promotes an increase in G' due to an increase in effective volume fraction of solids while starch digestion serves to decrease the effective particle size and volume leading to a decrease in the G' of the PC suspension. Thus the solids fraction and effective volume occupied by the solids continually changes during the measurement of G' until pseudo steady state is reached. As noted in figure 1, the plateau in G' corresponds to when starch is no longer significantly digested.

3.3 Estimating the diffusion coefficient: The diffusion coefficient can be a function of moisture and time[43], and mass transfer may not follow Fickian dynamics[44]. For instance, the higher PC weight fractions with amylase and all the non-amylase samples followed sigmoidal behaviour (figure 4), which Crank[44] hypothesised could be explained, theoretically, by either "slow structural changes" of the polymers through which liquid diffuses or "differential swelling" within the bulk sample, where "polymer" refers to a [solid] sheet. In the case of a starch granule, the latter case would result in heterogeneity of the starch granule modulus in addition to a position dependent diffusion coefficient. This behaviour, we hypothesize, would be more pronounced for granules in an amylase buffer because the hydrating granule will likely have three regions with different moduli and diffusion: (1) the interior dry region, (2) a middle region approximating a soft solid, and (3) an outer region approximating a soft solid but with reaction products and free polymer present. There was some scepticism regarding Crank's original formulation[45] and there have been other similar theories suggested[45, 46]. Interestingly for sigmoidal behaviour, the inflection point is hypothesized to occur around 50% absorption[46], which is approximately the case for our data (figure 4). We can conclude that oil and PC weight fraction impact the hydration + enzyme digestion process.

As an approximation, we estimate the diffusion coefficient according to the exponential form of Fick's second law, solved assuming unsteady state diffusion in spherical coordinates and a constant surface concentration. The rate constant multiplied against time can be re-arranged to give $D = k_1 r^2/\pi^2$, where D is the diffusion coefficient, k_1 the rate constant found by fitting the data and r the granule radius. For LF at 18% w/w PC in a buffer without amylase, the diffusion coefficient is estimated to be $D \approx 10^{-12}$ m^2/s, assuming a

granule radius of 50 μm. This value agrees well with data in literature[33, 34, 36, 38-42, 47]. This supports the notion that hydration kinetics plays a dominant role in the evolution of the PC suspension rheology with time.

3.4 Influence of fat and PC solids on comminuted PC bolus rheology: Figure 6 shows G'_m for the comminuted PC suspensions as a function of PC mass in buffer and amylase solution, measured at pseudo steady state; G'_m is the matrix elastic modulus, calculated by applying the van der Poel equation to the composite, or measured, bulk elastic modulus and representing the hydrated solids without the impact from the oil droplets (*see figure 6 and Boehm et al.[22] for more details*). G'_m is found to be an order of magnitude lower when amylase is present, and LF PCs have a higher G'_m than FF PCs for the same PC mass content. We demonstrated previously[22] that this apparent dependence on fat content is due to a higher overall solids content per PC mass as the fat content is decreased, and its contribution is accounted for by considering it to be present as dispersed droplets. Using this approach, we observe similar behaviour when amylase is present.

Conclusions

The data presented above shows a clear effect from the inclusion of α-amylase on the rheology of a simulated PC bolus during concurrent hydration and enzyme digestion. A significantly lower bolus modulus is observed in the presence of α-amylase, even over times for which the extent of starch digestion is less than 5%. Based on these findings, we suggest that the functional consequence of salivary α-amylase is not only to initiate the digestion process but also to control bolus formation and oral processing of starchy foods, at least for low moisture foods. For our simulated bolus, α-amylase impacts the bolus rheology within the first 30 seconds after *in situ* mixing and this is distinguished from hydration effects. In the absence of α-amylase and regardless of solids and fat content, a unified increase in PC bolus modulus with time is observed that is ascribed to be due to particle hydration kinetics. Unified behaviour is not observed when α-amylase is present because the rheology is affected by both hydration and digestion. We find that the hydration kinetics is similar to the kinetics of starch digestion by amylase, indicating that the rate determining step for starch in low moisture foods is hydration. We conclude that *in situ* mixing coupled to rheological measurements is a powerful means in which to investigate system variables on the dynamics of bolus formation and as *in vitro* means in which to investigate oral processing and the first stages of digestion.

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Acknowledgment
Stefan K. Baier is an employee of PepsiCo Inc. The views expressed in this article are those of the authors and do not necessarily reflect the position or policy of PepsiCo Inc.

Author contribution
All authors contributed an equal portion to the research and manuscript, and all authors have approved the final version of the manuscript.

Abbreviations: PC: Potato chip/s; LF: Low fat PC (22.9% w/w oil); FF: Full fat PC (33.5% w/w oil)

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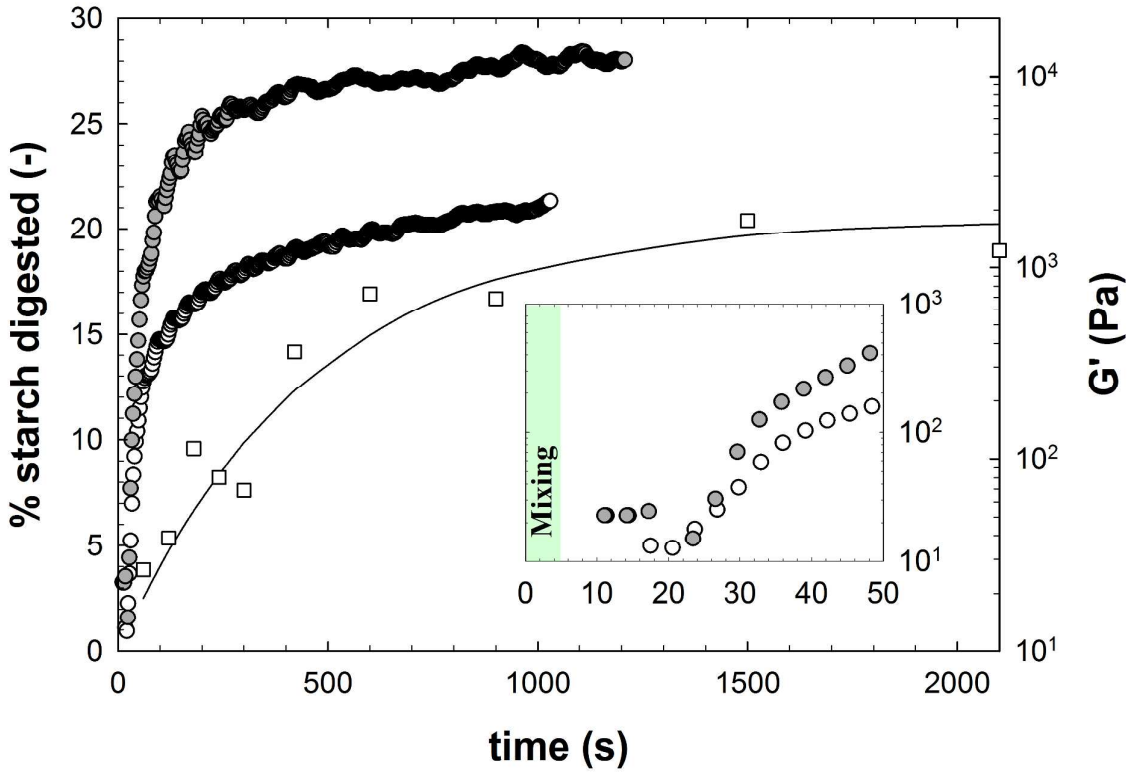

Figure 1. On the left y-axis, the percent starch digested (□) is graphed versus time, showing a fit to first order kinetics with C_∞ = 24.3% & k = 0.0022 s^{-1}. On the right y-axis, the transient bolus elastic modulus without amylase (●) or with amylase (○) in the buffer is graphed versus time, for a bolus of 25% w/w LF PC; the bolus was mixed for 5 seconds *in situ* using the vane tool. The inset graph shows G' in the region t: 0-50 seconds, with the time for mixing highlighted. G' was measured at an angular frequency of 6.28 rad/s.

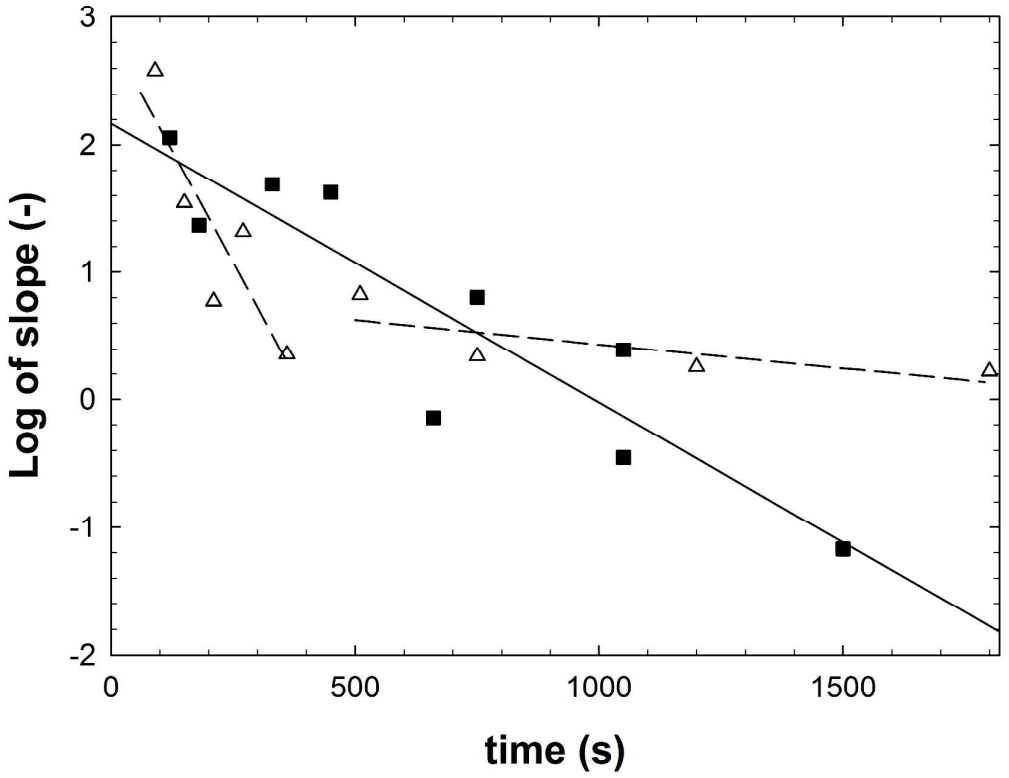

Figure 2. Log of slope, LOS, graphed versus time for a sample of 18% w/w PC which was dry upon mixing with amylase solution (■), or an 18% w/w PC that was pre-hydrated for 3 hours in a buffer without amylase prior to mixing with amylase solution (Δ). For the pre-hydrated samples: $k_1 = 0.0070\ s^{-1}$ with ≈15% starch digestion, and $k_2 = 0.00038\ s^{-1}$ with another ≈35% starch digestion.

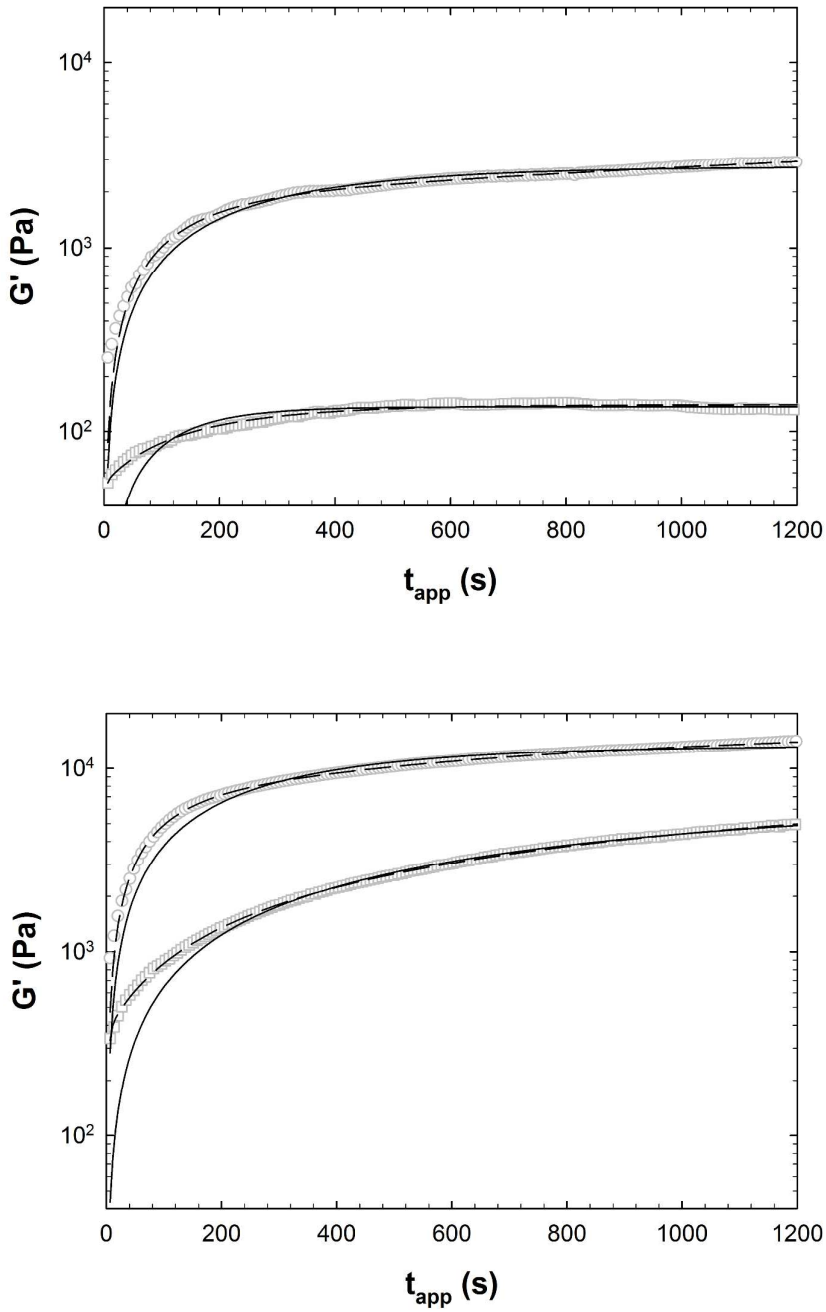

Figure 3. (a) Transient data for 18% w/w LF in a buffer without (○) or with amylase (□). (b) Transient data for 29.5% w/w LF in a buffer without (○) or with amylase (□). A single exponential term (—) or two exponential terms (— —) were fit to the G' versus time data. G' was measured at an angular frequency of 6.28 rad/s.

Table 1. Parameters for single exponential term or two exponential terms fit to transient G' for LF in buffer without and with amylase.

$G' = A_1*(1-e^{-k_1 t})$

		k_1 (s^{-1})	A_1 (Pa)
18% w/w LF PC	*no amylase*	0.004 +/- 0.00008	2700 +/- 20
	amylase	0.009 +/- 0.0004	140 +/- 1
29% w/w LF PC	*no amylase*	0.003 +/- 0.00008	13000 +/- 100
	amylase	0.0009 +/- 0.00003	7300 +/- 100

$G' = A_1*(1-e^{-k_1 t}) + A_2*(1-e^{-k_2 t})$

		k_1 (s^{-1})	k_2 (s^{-1})	A_1 (Pa)	A_2 (Pa)
18% w/w LF PC	*no amylase*	0.007 +/- 0.0002	0.0000002 +/- 0.0001	1800 +/- 50	$4.9*10^6$ +/- $3.5*10^9$
	amylase	0.5 +/- 0.4	0.005 +/- 0.0002	52 +/- 2	87 +/- 2
29% w/w LF PC	*no amylase*	0.01 +/- 0.0002	0.001 +/- 0.00004	5600 +/- 90	12000 +/- 200
	amylase	0.3 +/- 0.06	0.0005 +/- 0.000009	360 +/- 6	9600 +/- 100

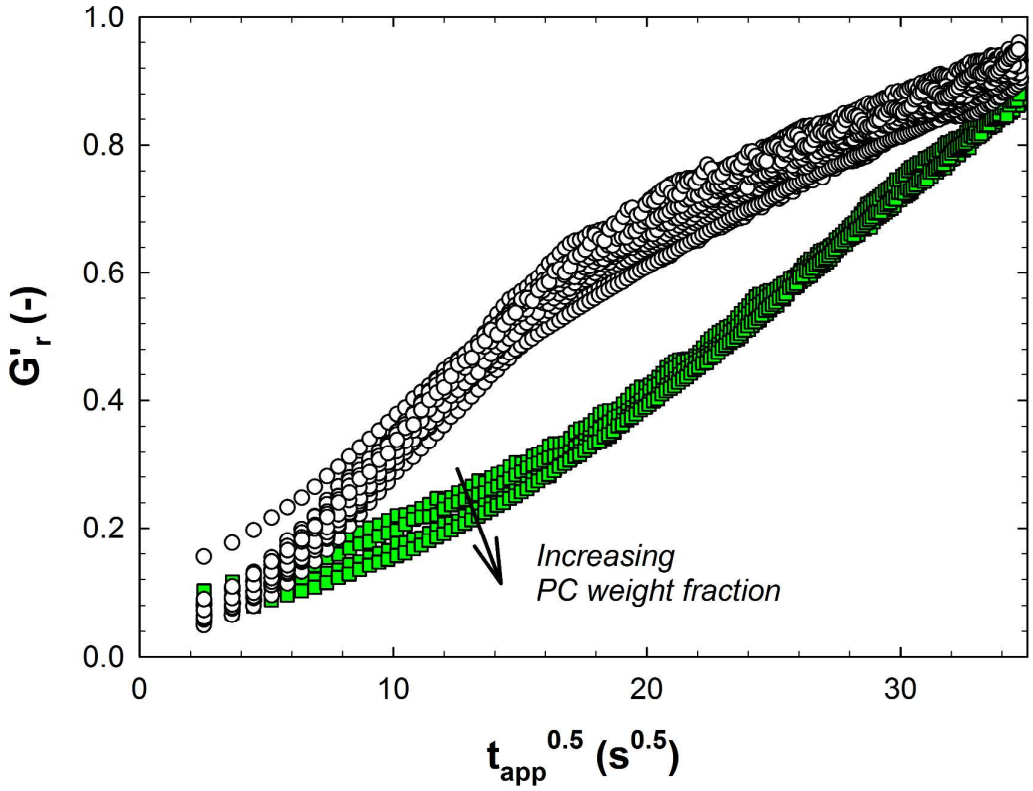

Figure 4. G'_r graphed versus $t_{app}^{0.5}$ for LF in amylase solution at 25-29.5% w/w PC (■) and LF & FF in buffer without amylase at 18-29.5% w/w PC (○).$G'_r = G'/G'_\infty$ and G'_∞ is the pseudo-steady state value measured after 20 minutes. The arrow representing increasing solids weight fraction only applies to the amylase data (■). (*color version available online*)

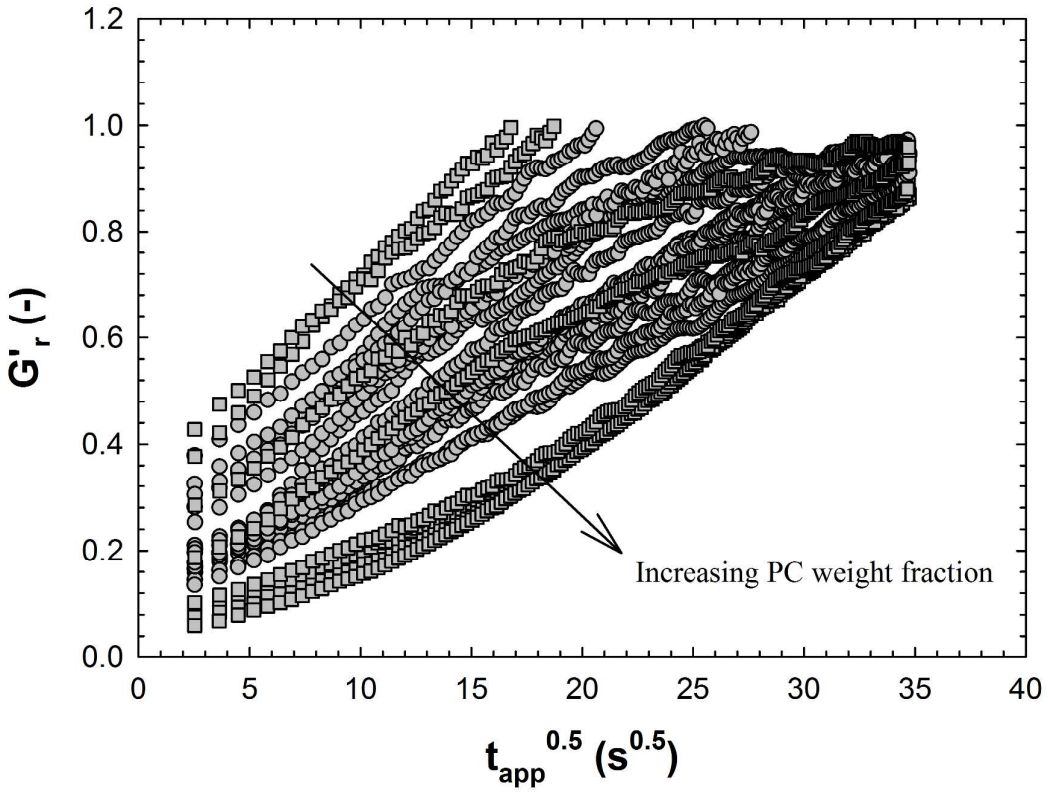

Figure 5. For a buffer with amylase, G'_r is graphed versus $t^{0.5}$, where $G'_r = G'/G'_\infty$ and G'_∞ is the pseudo-steady state value measured after 20 minutes. LF (□) & FF (○) were used at PC weight fractions ranging from 18% to 29.5% w/w. The arrow represents increasing PC weight fraction. G' was measured at an angular frequency of 6.28 rad/s.

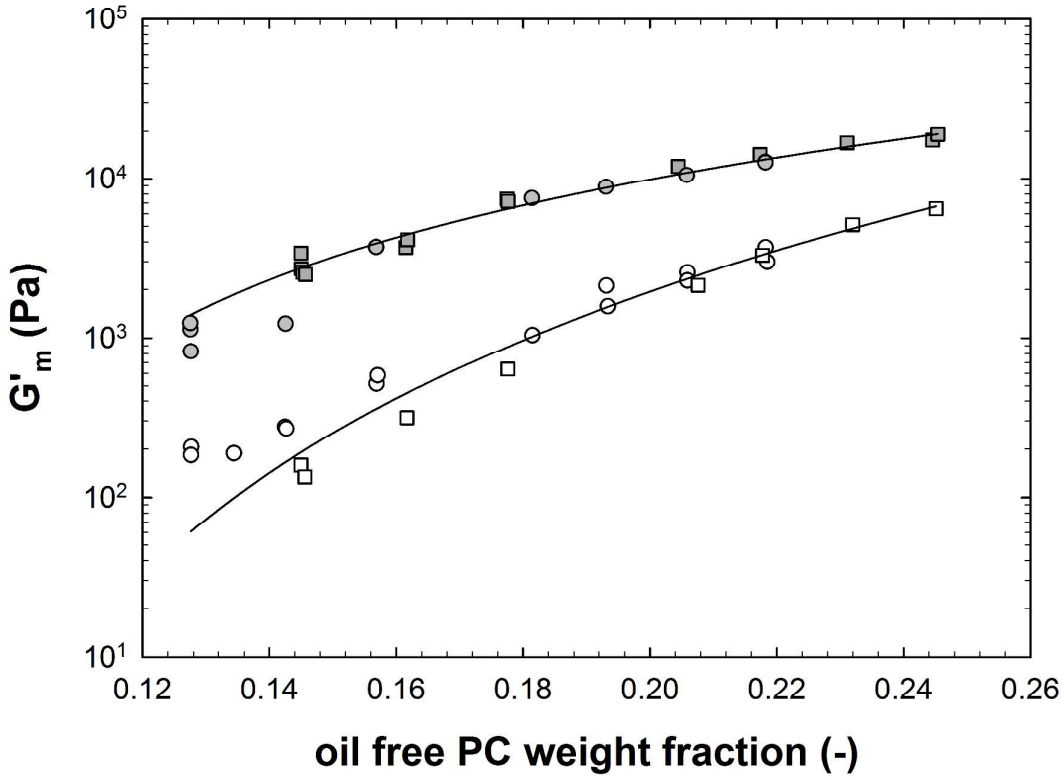

Figure 6. $G'_m=G'_c/(1-[(15/6)*\phi_f/(1.5+\phi_f)])$ graphed versus oil free PC weight fraction for LF (■,□) & FF (●,○) in a buffer without amylase (■,●) and with amylase (□,○). The data is fit with the percolation equation with free fitting parameters (—). G' was measured at an angular frequency of 6.28 rad/s.

Table 2. Fitting parameters for the percolation equation applied to pseudo-steady state G' for LF & FF in buffer without and with amylase. Percolation equation: $G' = a*(c-c_o)^b$

	No amylase	Amylase
a	735 kPa	7.96 MPa
c_0	0.084	0.075
b	2	4