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Mass transport processes in orange-fleshed sweet potatoes leading to structural changes during in vitro gastric digestion

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ABSTRACT

During cooking, food undergoes structural modifications, which may impact its behavior during digestion. The objective of this study was to determine the macro- and micro-structural changes, moisture uptake, and acid uptake into sweet potatoes during simulated gastric digestion as influenced by cooking method. Sweet potatoes were cut and cooked (boiled, steamed, microwave steamed or fried), followed by in vitro gastric digestion (up to 240 min). Acidity, moisture content, and hardness were measured during the digestion period. Light microscopy was completed on cooked and digested samples to observe microstructural changes. Effective diffusivity of acid and moisture was modeled following Fick's second law in MATLAB. Acid and moisture uptake were significantly influenced by cooking method and digestion time ($p < 0.0001$). Hardness was significantly influenced by cooking method, digestion time, and their interaction ($p < 0.0001$). Microstructural changes were observed both as a result of cooking and after in vitro gastric digestion.

1. Introduction

Orange fleshed sweet potatoes (referred to sweet potatoes, *Ipomoea batatas* L.) are nutritious tubers, rich in carbohydrates and dietary fiber, vitamins, minerals, and antioxidants, such as β-carotene (Burri, 2011; Teow et al., 2007). Orange fleshed sweet potatoes can be cooked many different ways prior to consumption, such as boiled, steamed, roasted, deep fried, baked, and microwaved (Bengtsson et al., 2008; Burri, 2011). These cooking methods involve differing mechanisms of heat transfer and environmental conditions (i.e. cooking in air vs. water vs. steam), which may result in varying microstructural changes in the food matrix depending on the cooking method (Aguilera, 2005; Parada and Aguilera, 2007). These structural changes may also influence nutrient absorption, as it has been shown that structural changes induced by processing and thermal treatments influence β-carotene bioaccessibility in carrots and sweet potatoes (Bengtsson et al., 2009; Tumuhimbise et al., 2009; Tydeman et al., 2010).

During digestion, the first physical change is in the mouth; ingested food is broken down and mixed with saliva during mastication, forming a bolus. When the food particles reach a certain size they are transported through the esophagus to the stomach, where gastric digestion occurs. During gastric digestion, mechanical and chemical breakdown occur due to both stomach contractions and gastric secretions, respectively (Bornhorst and Singh, 2014). Gastric secretions contain enzymes (i.e. pepsin and lipase), electrolytes (i.e. sodium chloride), and are acidic (pH ≈ 2). These characteristics of the gastric fluid modify the food structure due to enzymatic and acid hydrolysis, resulting in softening of the food matrix. The rate of diffusion of gastric fluids into food matrices in the gastric environment may have implications in the overall gastric breakdown as well as absorption of nutrients in the small intestine. Aside from the digestion fluid composition, there are other factors that may influence the gastric acid diffusion rate, which include food composition, food properties, and processing of food, among other factors (Mennah-Govela et al., 2015).

It has been previously shown that there is a link between acid diffusion and food softening in canning, curing, and pickling processing of foods. Softening in foods occurs when the pectic material in the plant cell wall is hydrolyzed or broken down (Demain and Phaff, 1957). It has been demonstrated that pH and temperature are some of the factors that may influence cell wall hydrolysis (Krall and McFeeters, 1998; McFeeters and Fleming, 1989, 1990). Depending on the type of acid, the rate of softening may be faster or may prevent the tissue from softening. For example brine

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containing gluconic acid helped retained texture in carrots (Heil and McCarthy, 1989), compared to brine containing acetic acid, which was shown to facilitate the softening of cucumbers (McFeeters et al., 1995). Additionally, previous studies have shown that the addition of salts, i.e. sodium chloride, may interact as a firming agent preventing the softening of the plant tissue (Gabaldón-Leyva et al., 2007; Heil and McCarthy, 1989; Howard et al., 1994; McFeeters and Fleming, 1991; Widjaja, 2010).

Food softening during digestion may be correlated to the food breakdown rate as well as the gastric emptying rate. These two processes are associated with transport to the small intestine, meaning that rate of food softening during gastric digestion may influence absorption of nutrients (Bornhorst et al., 2015). Previous studies have shown that in order for nutrients to be absorbed by the small intestine, they have to be released from the cell wall. The disruption of cell walls occurring as a result of processing or during gastric digestion (Failla et al., 2009; Lemmens et al., 2010; Parada and Aguilera, 2007; Van Buggenhout et al., 2010).

It was hypothesized that the cooking methods examined (boiling, steaming, frying, and microwave steaming) would induce structural changes in sweet potatoes, which would modify their uptake of acid and water as well as their propensity for additional structural changes during simulated gastric digestion.

2. Materials & methods

2.1. Raw materials

Sweet potatoes were purchased from a local supermarket (Lansing, MI, U.S.A.), and stored at 4 °C for use within 4 weeks.

2.2. Sweet potato cooking procedure

Sweet potatoes were cut into cubes (approx. 0.012 × 0.012 × 0.012 m) by first using a potato cutter to obtain long strips and followed by manually cutting each strip into cubes of uniform size. Samples were taken only from the interior of the sweet potatoes, and any pieces containing peel were discarded.

The cooking methods selected were boiling, steaming, microwave steaming, and frying. The total cooking time for each method was selected based on preliminary trials to ensure similar hardness after cooking (data not shown). For all cooking methods, 20 cubes were cooked together in one batch to ensure comparable sample heating conditions for each experiment. For boiled sweet potatoes, cubes were immersed in boiling water (100 °C) for 15 min. For steamed sweet potatoes, cubes were put in a metal steamer that was placed above a pot of with boiling water. Cubes were heated with the lid on the pot for 20 min. For microwave steamed sweet potatoes, cubes were placed in the top compartment of a microwave steamer and were cooked for 8 min and kept covered for an additional 2 min. For fried sweet potatoes, cubes were fried in soybean oil (Meijer Inc., Grand Rapids, MI, U.S.A.) at 180 °C for 5 min. In all cooking methods, with the exception of microwave steamed, a thermocouple was kept inside one cube during cooking to record the temperature profile (data not shown) over time to ensure similar cooking conditions for each batch.

2.3. Volume change after cooking

Sweet potato volume was measured using a 5000S electronic digital caliper (Chicago Brand, Medford, OR, U.S.A.). Three sides of the cubes were measured before and after cooking times to estimate the volume. Ten cubes were measured before and after cooking for each cooking method.

2.4. Simulated digestion

2.4.1. Simulated saliva formulation

Saliva was prepared following Bornhorst and Singh (2013). All components were mixed in deionized water: mucin (1 g/L, Sigma-Aldrich, MO, U.S.A.), α-amylase (from *Bacillus subtilis*, 1.18 g/L, MP Biomedicals, Catalog Number 100447, activity of 160,000 BAU/g, Santa Ana, CA, U.S.A.), NaCl (0.117 g/L, Avantor Performance Materials, PA, U.S.A.), KCl (0.149 g/L, Fisher Science Education, IL, U.S.A.), and $NaHCO_3$ (0.21 g/L, Fisher Science Education, IL, U.S.A.). After addition of all ingredients, the pH was adjusted to 7 with 0.01 N NaOH (Bornhorst and Singh, 2013).

2.4.2. Gastric juice formulation

Gastric juice was prepared by mixing the following ingredients in deionized water: mucin (1.5 g/L, Sigma-Aldrich, MO, U.S.A.), NaCl (8.78 g/L, Avantor Performance Materials, PA, U.S.A.), and pepsin from porcine pancreas (1.0 g/L, Sigma-Aldrich, MO, U.S.A.). After addition of all ingredients, the pH was adjusted to 1.8 using 0.1 N HCl (Bornhorst and Singh, 2013).

2.4.3. Oral and gastric digestion conditions

Oral and gastric digestion conditions were performed following Mennah-Govela and Bornhorst (in press) digestion model in a shaking water bath; which was slightly modified from previous studies (Bornhorst and Singh, 2013; Hedren et al., 2002; Mennah-Govela et al., 2015; Minekus et al., 2014). Ten sweet potato cubes (initial mass of 12–20 g, depending on the cooking method) were weighed, placed in a 250 mL glass bottle, and hand-mixed with 0.2 mL/g of saliva for 30 s. One bottle was used for each replicate of each digestion time. Immediately after mixing, 100 mL of preheated gastric juice at 37 °C was added to the bottle, and the bottles were placed inside a shaking water bath (37 °C, 100 rpm). Samples were taken after oral digestion (0.5 min) and after 15, 30, 45, 60, 90, 120, 180, and 240 min of gastric digestion. Sampling was conducted by removing an individual bottle from the shaking water bath and the 10 cubes were analyzed from that digestion time. After the samples were removed, the sweet potato cubes were separated from the gastric juice with a sieve. The cubes were weighed and both the sweet potato cubes and gastric juice were retained for further analysis. From the 10 sweet potato cubes in one digestion, 6 cubes were used for duplicate measurements of acidity (3 cubes per replicate) and 4 cubes were used for duplicate measurements of moisture content (2 cubes per replicate). Separate digestions were completed for both texture and fat content measurements. Simulated digestions were performed in triplicate for each cooking method and digestion time point.

2.5. Sweet potato behavior during simulated gastric digestion

2.5.1. Acidity and pH measurement

Briefly, from the 6 cubes of the glass bottle, acidity measurements were done in duplicates, where 3 cubes (approx. 6 g) were weighed and homogenized with 20 mL of deionized water using a Polytron model PT 10/35 homogenizer (Brinkmann Instruments Co., Switzerland) for 30 s. The initial pH was measured using a HI 99161 portable pH meter (HANNA instruments, Woonsocket, RI, U.S.A.). Acidity measurements where completed via potentiometric titrations. Sodium hydroxide (0.01 N NaOH) was added to each sample until the pH reached a value of 8.2 ± 0.05.

2.5.2. Moisture content

The 4 cubes left in the glass bottle were used to determine duplicate measurements of moisture content. Moisture was determined gravimetrically by drying in a convection oven for

16 h at 100 °C until constant weight.

2.5.3. Fat content analysis

Separate digestions were prepared to measure fat content of fried sweet potatoes by Soxhlet extraction following the AOAC 945.16 Official Method (AOAC, 2012). Fat content was measured before digestion and after 60, 120, and 240 min of simulated gastric digestion. An average of three replicates per digestion time were measured.

2.5.4. Texture analysis

Separate digestions were done for hardness measurements of sweet potato cubes during simulated digestion. Hardness change of individual cubes was quantified as the maximum force during compression to 6 mm using a 5 cm diameter cylinder probe with a test speed of 2 mm/s following the method for potatoes by Kaur et al. (2002) with minor modifications. All analyses were completed using a TA.HD *Plus* Texture Analyzer (Texture Technologies Corp., Hamilton, MA, U.S.A.). Hardness was measured before digestion and after 15, 30, 60, 120, 180, and 240 min of simulated gastric digestion. Eight cubes were measured from each gastric digestion time (n = 24 measurements for each cooking method x digestion time combination).

2.5.5. Microstructural changes

Microstructural analysis was done before and after 240 min of digestion for all cooking methods. Samples were fixed in 10% Neutral Buffered Formalin for four days. After this time, samples were placed in 30% ethanol and processed on a Tissue Tek II Vacuum Infiltration Processor (Sakura Finetek, Alphen aan den Rijn, Netherlands) followed by embedding with paraffin using the ThermoFisher HistoCenter III embedding station (ThermoFisher Scientific, Waltham, MA, U.S.A.). Samples were cut in half and embedded with the cut surface down. Once samples were cooled, the excess of paraffin was removed from the edges. Samples were placed on a Reichert Jung 2030 rotary microtome (Reichert Technologies, Depew, NY, U.S.A.) and were finely sectioned at 5–6 μm. Sections were air-dried overnight and then were placed in a 56 °C slide incubator to ensure adherence to the slides for 2–24 h. Sections were stained with both Toluidine Blue (Mandalari et al., 2008), and Periodic Acid Schiff (Tumuhimbise et al., 2009). Samples were examined using a light microscope with 10 and 20× objectives, and images were taken with a Nikon Digital Camera DXM1200 (Nikon Instruments Inc., Melville, NY, U.S.A.).

2.6. Diffusion modeling

The effective diffusivity (D_{eff}) both gastric acid and water into the sweet potato cubes was estimated using Fick's second law for an infinite slab (Crank, 1975). The equation was modified to account for the cubic shape, using Neumann's rule as a product of three infinite and perpendicular slabs, similar to what has previously been done in rehydration of potato cubes (Markowski et al., 2009):

$$\frac{C-C_e}{C_0-C_e} = \frac{8^3}{\pi^6}\left[\sum_{n=1}^{\infty}\frac{1}{(2n+1)^2}\exp\left(-(2n+1)^2\frac{D_{eff}\pi^2 t}{L^2}\right)\right]^3 \quad (1)$$

where C is the concentration of acid or water (mg HCl/g of dry matter (DM) or g H_2O/g DM), C_0 is the initial concentration (mg HCl/g DM or g H_2O/g DM), C_e is the equilibrium concentration (mg HCl/g DM or g H_2O/g DM), L is the length of the edge of the sweet potato cube (m), and t is the digestion time (s). The initial conditions (initial acid concentration and initial moisture content) for each cooking method were measured experimentally as the acidity and moisture content after cooking, respectively (Table 1). The average length of the cubes after cooking varied within each treatment ($p < 0.05$); therefore, the cube size in the model was adjusted for each cooking method (Table 1). It was assumed that cubes were symmetrical.

The diffusion model is for well-mixed conditions since the pH of the gastric environment is the same around different locations in the simulated digestion system, and the Biot number is estimated to be on the order of magnitude of 10^2-10^3. The model was developed by making the following assumptions: 1) the system is isothermal (37 °C), 2) the sample size and cubic shape remains constant during the digestion period, 3) the acid concentration of the gastric environment is homogenous and isotropic, 4) effective diffusivity is independent of time, 5) mass transfer was by diffusion only, and 6) the surface of the cube is instantaneously at the concentration of acid or water in the gastric juice. In addition to the D_{eff}, the equilibrium concentration (C_{eq}) was calculated. The model was fitted using *nlinfit,* a nonlinear regression function based in ordinary least squares in MATLAB R2013b (MathWorks, Natick, MA, U.S.A).

2.7. Statistical analysis

SAS Enterprise 4.3 (SAS, Cary, NC, U.S.A.) was used for statistical analysis. An analysis of variance was conducted using a 2-factor factorial design to determine differences in acid uptake, moisture content, and texture during simulated gastric digestion. The factors were cooking treatment (boiled, steamed, microwaved steamed, and fried), and digestion time (0–240 min). The Tukey-Kramer test was used to analyze the differences between means when main effects were significant. A one-way analysis of variance was used to assess differences between the changes in volume during cooking, and differences in initial length, moisture content, and acidity. Statistical significance was assessed at a level of $p < 0.05$. Values are given as averages with error bars representing the standard error of the mean.

3. Results

3.1. Volume change after cooking

The change in volume in the sweet potatoes cubes as a result of cooking is given in Fig. 1. In average, initial sweet potatoes cube volume (before cooking) was $1.9 \pm 0.07 \times 10^{-6}$ m^3. Sweet potato cube volume was significantly influenced by the cooking method ($p < 0.001$). It can be seen in Fig. 1 that for each cooking methods, the volume was significantly different before and after cooking for all cooking methods. After cooking, steamed and boiled sweet potatoes had similar volume ($p > 0.05$). Fried and microwave steamed had significantly ($p < 0.05$) lower volume, and were different from each other as well as from the rest of the cooking methods. Microwave steamed cubes had the greatest volume change during cooking (65%), followed by fried (53%). Boiled sweet potatoes volume changed the least, decreasing 22% after cooking.

3.2. Acidity and pH measurements

pH both of the gastric juice and the digested samples were measured using a pH meter for boiled (Fig. 2), steamed, microwaved steamed, and fried sweet potatoes (data not shown for steamed, microwave steamed, or fried). For all the cooking methods the pH profiles followed the same trend as shown in Fig. 2, where the pH of the gastric juice increased from 1.8 to 3.1 ± 0.5, and the pH of the food sample decreased (from 7 ± 0.3 to 3.7 ± 0.4) during the 240 min gastric digestion period.

Table 1
Average initial conditions and cube side length of sweet potatoes after cooking that were used in the diffusion model. Values were determined experimentally and are given as the average of 10 or 6 replicates for length or initial conditions (acidity and moisture content), respectively. Different letters within each column represent significantly different means ($p < 0.05$).

Cooking method	Length (m)	Acidity (mg HCl/g dry matter)	Moisture content (g H_2O/g dry matter)
Boiled	0.0116[a]	1.8[a]	1.8[a]
Steamed	0.0111[b]	2.9[a]	1.5[b]
Microwaved Steamed	0.0087[d]	3.2[a]	0.6[c]
Fried	0.0098[c]	3.0[a]	0.6[c]

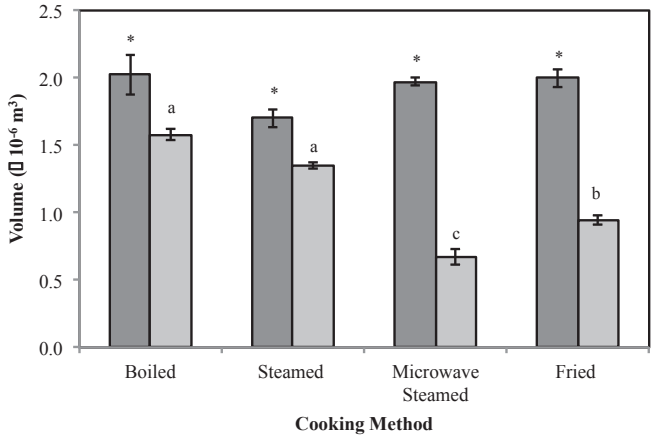

Fig. 1. Volume before (■) and after (■) cooking of sweet potato cubes. Values are given as an average (n = 10) with error bars representing the standard error of the mean. Values before cooking were not statistically different. Different letters in each cooking method represent statistically different means in the volume values after cooking; stars (*) represent significant differences in volume before and after cooking within each cooking method ($p < 0.05$).

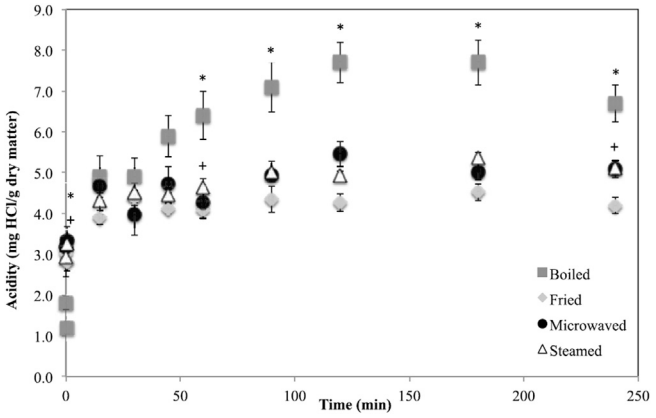

Fig. 3. Acidity changes in boiled (■), steamed (Δ), microwave steamed (•), and fried (◆) orange fleshed sweet potatoes over 240 min of simulated gastric digestion. Initial points represent before and after oral digestion (0 and 0.5 min), which may be hard to distinguish due to the scale. Values represent averages (n = 6 replicates) with error bars representing the standard error of the mean. Stars (*) show that the means are statistically significant from all the other treatments ($p < 0.05$), plus symbols (+) represent not statistically differences from the other cooking methods ($p > 0.05$).

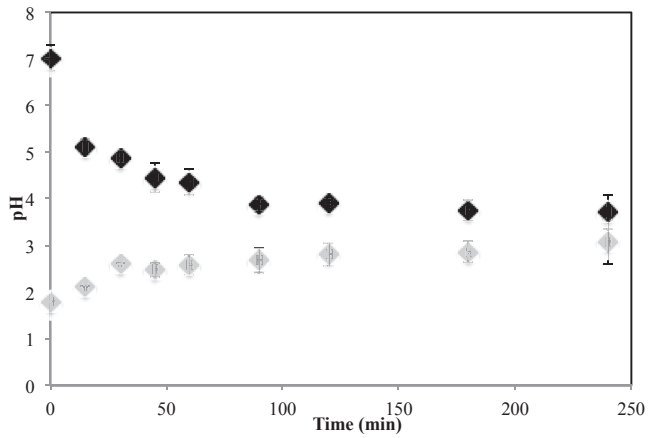

Fig. 2. pH change of both gastric juice (◆) and boiled orange fleshed sweet potatoes (◆) over 240 min of simulated gastric digestion. Values represent averages (n = 6 for sweet potatoes, and n = 3 for gastric juice) with error bars representing the standard error of the mean.

Acidity values before digestion, after oral digestion (0.5 min), and up to 240 min of incubation in gastric juice are shown in Fig. 3. The acidity of the sweet potatoes cubes was significantly influenced by the cooking method and gastric digestion time ($p < 0.0001$) as well as their interaction ($p < 0.0001$). As seen in Fig. 3, in most of the digestion time points, acid uptake of boiled sweet potatoes was significantly higher than the rest of the cooking methods ($p < 0.05$). After 240 min, boiled sweet potatoes had the greatest acidity increase, going from 1.8 ± 0.2 to 6.7 ± 0.5 mg HCl/g dry matter. In most of the digestion times steamed, microwave steamed, and fried were not significantly different ($p > 0.05$). However, fried sweet potatoes showed a trend of the lowest acid increase, with acidity values ranging from 3.0 ± 0.2 to 4.2 ± 0.2 mg HCl/g dry matter after 240 min of simulated gastric digestion. Steamed and microwave steamed sweet potatoes followed a similar pattern increasing from 2.9 ± 0.5 to 5.1 ± 0.2 mg HCl/g dry matter (steamed), and 3.2 ± 0.3 to 5.1 ± 0.2 mg HCl/g dry matter (microwave steamed).

3.3. Moisture content

Moisture content (dry basis) evolution during simulated gastric digestion for the four cooking methods is shown in Fig. 4. Moisture content was significantly influenced by the cooking method and the digestion time ($p < 0.0001$) as well as the interaction between cooking method and digestion time ($p < 0.0001$). Overall, fried and microwave steamed sweet potatoes had a greater increase in moisture over time, and lowest initial dry basis moisture content, going from 0.59 ± 0.03 to 1.59 ± 0.05 g H_2O/g dry matter (DM), and from 0.62 ± 0.03 to 1.60 ± 0.06 g H_2O/g DM, respectively (±standard error of the mean). Boiled and steamed sweet potatoes had similar dry basis moisture uptake, from 1.83 ± 0.04 to 2.20 ± 0.02 for boiled, and from 1.49 ± 0.05 to 2.11 ± 0.02 for steamed, during the 240 min digestion period.

3.4. Fat content analysis

Fat content was not significantly influenced by time of simulated gastric digestion ($P = 0.1762$). The initial fat content of fried sweet potatoes was 5.45 ± 1.08%. After 60, 120, and 240 min of simulated gastric digestion, the fat content decreased to 4.14 ± 0.05%,

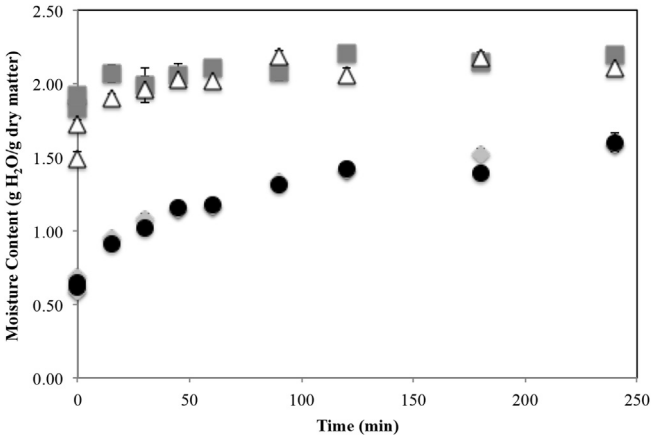

Fig. 4. Dry basis moisture content of boiled (■), steamed (Δ), microwave steamed (•), and fried (◆) orange fleshed sweet potatoes during a 240 min gastric digestion period. Values are given as averages (n = 6) ± standard error of the mean; error bars are included but are too small to be seen for many data points. At all digestion times (from 0.5 to 240 min) individual means from boiled and steamed were significantly different from fried and microwaved steamed sweet potatoes ($p < 0.0001$). The difference in individual means for initial moisture content is given in Table 1.

3.90 ± 0.21%, and 3.63 ± 0.13%, respectively (±standard error of the mean).

3.5. Texture analysis

The hardness (N) for each cooking method during the 240 min digestion period in gastric juice is shown in Table 2. Hardness of the sweet potatoes cubes was significantly influenced by the cooking method, digestion time, and their interaction ($p < 0.0001$). Fried sweet potatoes exhibited the greatest change (58%) from its initial hardness value. The initial hardness of fried sweet potatoes was significantly higher ($p < 0.0001$) than the other cooking methods. Steamed sweet potatoes had the lowest change in hardness after 240 min of digestion with 31% change. As seen in Table 2, the hardness in boiled, steamed, and microwave steamed sweet potatoes significantly decreased during digestion time ($p < 0.0001$). However, hardness at time 0 in fried sweet potatoes was significantly different from the other digestion times ($p < 0.0001$), but did not change significantly between 15 and 240 min of digestion. The hardness between cooking methods at most of the times during digestion were similar, with the exception of a few time points. After 30 min of digestion, hardness in boiled sweet potatoes was significantly different than steamed ($p < 0.05$), but hardness in microwave steamed and fried were not different from both steamed and boiled. Similarly, after 180 min of digestion, hardness in fried sweet potatoes was significantly higher than boiled ($p < 0.05$), but steamed and microwave steamed did not differ significantly. After 240 min of digestion, the final hardness values of all the cooking methods were not significantly different.

3.6. Microstructural changes

Light microscopy was used to observe microstructural changes after 240 min of simulated gastric digestion using Toluidine Blue (Fig. 5) and Periodic Acid Schiff (Fig. 6) stains, with 10× and 20× magnification. Toluidine blue is frequently used to observe the morphology of cell walls (O'Brien et al., 1964), in this case it was selected to detect cell wall variations before and after digestion. Fig. 5A and B shows steamed samples; boiled and microwave steamed sweet potatoes followed a similar trend after digestion (images not shown). These samples show that before digestion the cells are complete and without breakage, however after 240 min of digestion cell wall breakage can be observed (marked with arrows). Fried sweet potatoes did not show cell wall breakdown after 240 min of simulated gastric digestion (Fig. 5C and D). However, gaps between cells can be observed after 4 h of digestion in fried sweet potatoes (marked with arrows). Samples with Periodic Acid Schiff (PAS; Fig. 6) complement the observations made in samples stained with Toluidine Blue. Since PAS stains starch and some complex polysaccharides (Feder and O'Brien, 1968), it can be seen that there may have been some breakdown of the starch in steamed (Fig. 6A and B), and in fried sweet potatoes (Fig. 6C and D). Similar results as in steamed were seen for boiled and microwave steamed (data not shown). Black arrows in Fig. 6B and D shows some examples of starch degradation observed after 240 min of simulated gastric digestion.

3.7. Diffusion modeling

The effective diffusion coefficients (D_{eff}) and equilibrium concentration (C_{eq}) of both gastric acid and water were estimated for each cooking method and are shown in Table 3. The coefficient of determination (R^2) and the confidence intervals of the model are also given in Table 3. The diffusion model provided a good fit for almost all of the data sets ($R^2 \geq 0.90$), with the exception of microwave steamed ($R^2 = 0.76$), and boiled ($R^2 = 0.85$), in the acid and water effective diffusivity models, respectively.

It can be observed that for each cooking method, with the exception of boiled, the D_{eff} of acid and water were not equivalent, but there was not a consistent trend across cooking methods to describe the differences between acid and water diffusion. For example, in fried sweet potatoes the D_{eff} of gastric acid was 1.64×10^{-9} m^2/s compared to a value of 0.22×10^{-9} m^2/s for water.

The confidence intervals shown in Table 3 demonstrate that there is not a significant difference between acid effective diffusivity from the different cooking methods. However, there is a trend of faster acid effective diffusivity in fried and slower diffusivity in boiled and microwave steamed sweet potatoes. Based on the

Table 2
Hardness during simulated gastric digestion for boiled, steamed, microwave steamed and fried. Values are represented as averages (n = 24) ± standard error of the mean. Different letters within each column (abc) and within each row (zyx) represent statistically different means ($p < 0.05$).

Digestion time (min)	Sweet potato hardness (N)			
	Boiled	Steamed	Microwave steamed	Fried
0	$5.8 \pm 0.2^{a,z}$	$6.0 \pm 0.2^{a,z}$	$6.3 \pm 0.3^{a,z}$	$11.2 \pm 0.9^{a,y}$
15	5.1 ± 0.6^{ab}	6.1 ± 0.5^{a}	5.3 ± 0.3^{ab}	5.2 ± 0.3^{b}
30	$4.5 \pm 0.6^{abc,y}$	$6.4 \pm 0.4^{a,z}$	$5.2 \pm 0.2^{ab,zy}$	$5.6 \pm 0.5^{b,zy}$
60	4.0 ± 0.4^{bc}	4.8 ± 0.3^{ab}	4.4 ± 0.3^{b}	4.6 ± 0.3^{b}
120	3.9 ± 0.7^{bc}	5.3 ± 0.4^{ab}	4.3 ± 0.2^{b}	5.1 ± 0.4^{b}
180	$3.2 \pm 0.4^{c,y}$	$3.3 \pm 0.2^{b,zy}$	$4.0 \pm 0.2^{b,zy}$	$5.1 \pm 0.3^{b,z}$
240	3.1 ± 0.3^{c}	4.1 ± 0.5^{b}	3.8 ± 0.1^{b}	4.7 ± 0.3^{b}

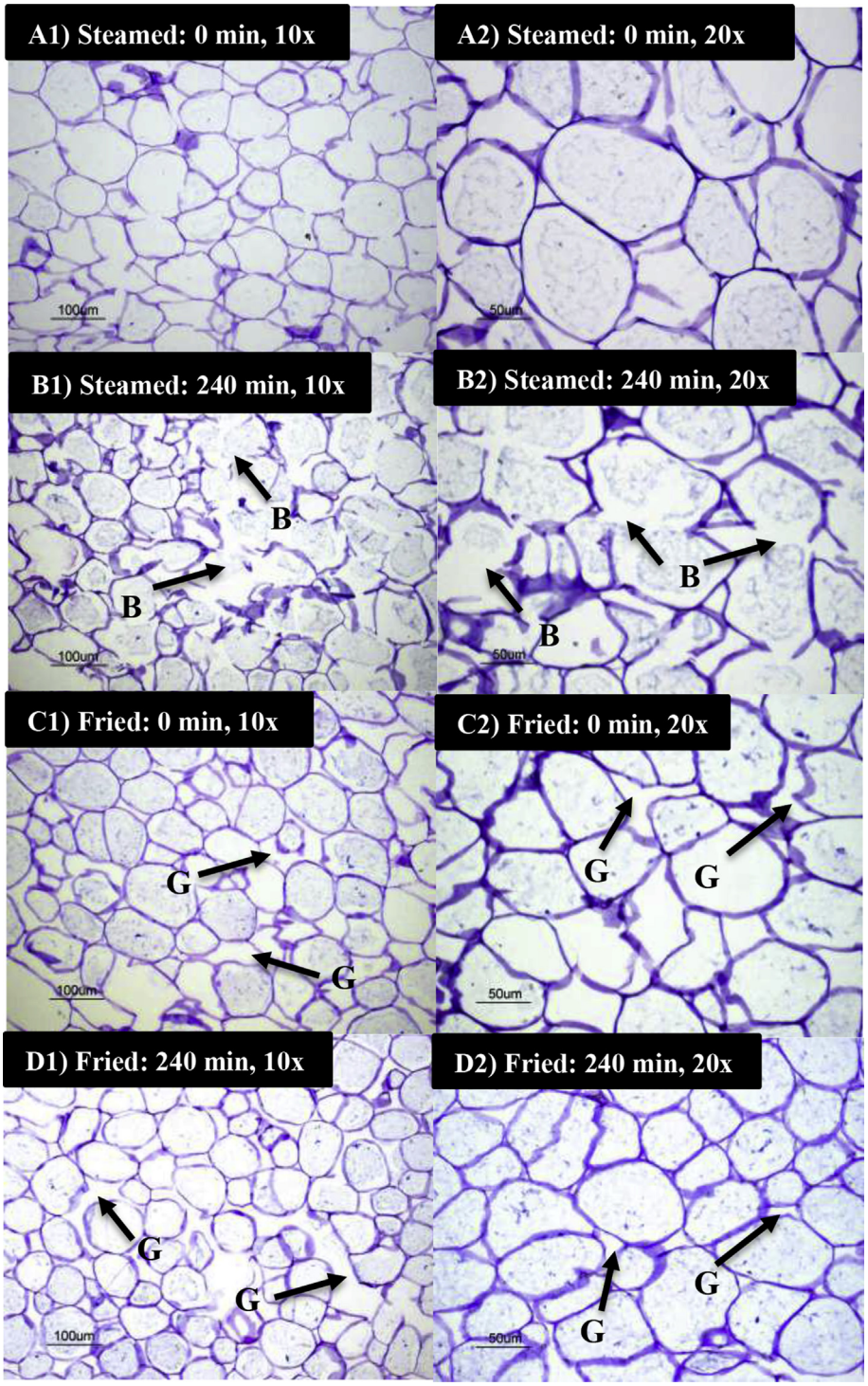

Fig. 5. Light microscopy images of sweet potato samples stained with Toluidine Blue for steamed (A and B) and fried (C and D) before (A and C) and after 240 min of simulated gastric digestion (B and D). The images are shown with a magnification of 10× (1) and 20× (2). B: breakage of cell walls; G: gaps between cells. (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

confidence intervals, water diffusivity in steamed sweet potatoes is significantly different from microwave steamed and fried. Even though no differences between boiled and microwave steamed and fried were seen, there is a trend that may show a higher effective diffusivity in boiled sweet potatoes. Although trends were present, the confidence intervals did not show significant differences between different cooking methods. In future experiments, it is recommended to perform more replicates to lower the confidence intervals and be able to show significant differences. In addition, some of the confidence intervals were negative due to variability in the data. Normalization of data prior to run the mathematical model is recommended to avoid negative confidence intervals in future analyses.

When the equilibrium concentration is considered, it can be

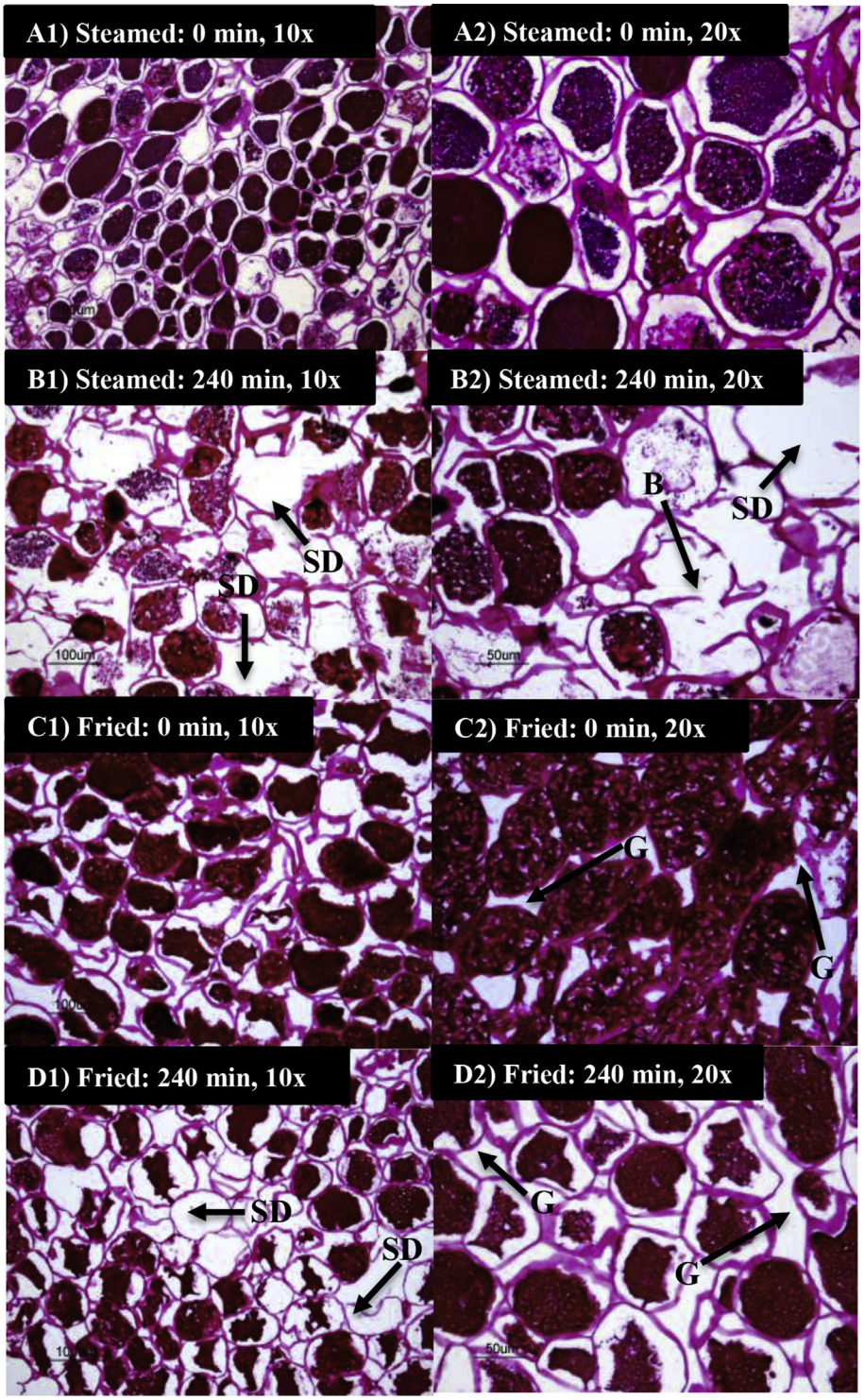

Fig. 6. Light microscopy images of sweet potatoes stained with PAS for steamed (A and B) and fried (C and D) sweet potatoes before (A and C) and after 240 min of digestion (B and D). The images are shown with a magnification of 10× (1) and 20× (2). SD: starch degradation; G: gaps between cells; B: breakage of cell walls.

observed that boiled samples had significantly higher acid equilibrium concentration (7.8 mL HCl/g dry matter) than fried, steamed, and microwave steamed. Although confidence intervals of moisture equilibrium concentration show there is not significant differences between boiled, steamed, and microwave steamed, similar trends were seen in the equilibrium concentration of water. However, boiled sweet potatoes had significantly higher equilibrium concentration (2.2 mL H_2O/g dry matter) than fried sweet potatoes (1.9 mL H_2O/g dry matter).

4. Discussion

Sweet potatoes are a common type of tuber, which is normally cooked and consumed in a variety of ways including boiled, fried,

Table 3
Effective diffusivity and equilibrium concentration for acid and water in sweet potatoes prepared with different cooking methods. The coefficient of determination (R^2) and the 95% confidence intervals are given to show the goodness of fit of the model and the model uncertainty.

Cooking method	D_{eff}		C_{eq}		R^2		95% confidence intervals D_{eff}		95% confidence intervals C_{eq}	
	Acid ($\cdot 10^9$ m^2/s)	Water ($\cdot 10^9$ m^2/s)	Acid (mg HCl/g DM)	Water (g H_2O/g DM)	Acid	Water	Acid ($\cdot 10^9$ m^2/s)	Water ($\cdot 10^9$ m^2/s)	Acid (mg HCl/g DM)	Water (g H_2O/g DM)
Boiled	0.9	1.0	7.8	2.2	0.93	0.85	[−0.2 1.9]	[−0.3 2.4]	[6.0 9.5]	[2.1 2.3]
Steamed	1.3	2.1	5.2	2.1	0.94	0.91	[0.3 2.4]	[0.3 4.0]	[4.9 5.6]	[2.0 2.2]
Microwave steamed	0.9	0.1	5.2	1.9	0.76	0.98	[−0.7 2.5]	[0.0 0.3]	[4.5 5.9]	[1.4 2.5]
Fried	1.6	0.2	4.4	1.9	0.90	0.99	[−0.4 3.6]	[0.1 0.3]	[4.1 4.7]	[1.6 2.1]

steamed, and microwave steamed. These four cooking methods were chosen as thermal treatments that may be used by a typical consumer. The objective of this study was to determine the influence of cooking method on the behavior of sweet potato during simulated gastric digestion.

To gain a better understanding of sweet potato physical changes after cooking, the cube volume, moisture content, and hardness were measured before and after cooking for each cooking treatment. As seen in Fig. 1, for all cooking methods, the sweet potato cube volume decreased after cooking. Boiled sweet potato showed the smallest decrease after cooking (i.e. the largest volume), and microwave steamed showed the greatest volume decrease (i.e. the smallest volume). The sweet potato cube volume was related to the moisture uptake, with the exception of fried sweet potato. For example, boiled and steamed sweet potatoes had similar size after cooking (average size 1.45×10^{-6} m^3). Fried sweet potatoes had a lower volume after cooking (0.9×10^{-6} m^3), and microwave steamed sweet potatoes had the lowest volume (0.7×10^{-6} m^3). As seen in Fig. 4, these follow a similar trend as the moisture uptake. Boiled and steamed moisture uptake were not statistically significant, but were greater than microwave steamed and fried. The trends in these results indicate that the quantity of water absorption or loss during cooking may have an impact on the volume change of the sweet potato cubes during cooking and the moisture uptake during digestion.

Cooking methods of sweet potatoes also influenced its macro- and micro-structure. The hardness after cooking was the same for boiled, steamed, and microwave steamed (average 6.0 ± 0.1 N), but after frying the hardness of the sweet potatoes was higher (11.2 ± 0.9 N) (Table 2). The higher initial hardness of fried sweet potatoes is likely due to the crust formation during frying. Crust formation during potato frying involves cellular changes in the outer layer of the potato. Some of these changes include: starch gelatinization, denaturation of proteins, dehydration, expansion and browning of the tissue, oil uptake, among other physico-chemical changes (Bouchon and Aguilera, 2001; Bouchon et al., 2001). Variations in sweet potato tissue between fried and all other cooking methods were also observed in light microscopy images (Figs. 5 and 6).

Further analyses were done to examine both macro- and micro-structural changes caused by simulated gastric digestion (i.e. texture and light microscopy). Texture results showed that fried sweet potato had a greatest decrease in hardness (59%) during simulated gastric digestion. It is hypothesized that this decrease was mainly due to the rapid softening of the crust formed during frying. During the first 15 min of gastric digestion, the hardness of the fried sweet potatoes decreased drastically from 11.2 ± 0.9 N to 5.2 ± 0.3 N after 15 min of digestion (Table 2). Similar rapid decreases in hardness were not observed in the sweet potatoes from the other cooking methods. However, if the crust is not taken into account and the initial hardness is considered at 15 min of digestion, the hardness decrease after 240 min of digestion was only 10% (the hardness evolution from 15 min to 240 min was not significant different). After 240 min of simulated gastric digestion, the hardness in all cooking methods was not significantly different (average 3.8 ± 0.2 N). However, the hardness at different digestion time points was different across cooking treatments. Specifically, hardness was statistically significant at 30 min of digestion in steamed (6.4 ± 0.4 N) and boiled sweet potatoes (4.5 ± 0.6 N). Similarly, at 180 min of digestion the hardness in boiled sweet potatoes (3.2 ± 0.4 N) was significantly different than the hardness in fried sweet potatoes (5.1 ± 0.3 N). These trends in hardness decrease may correspond to the microstructural changes observed through light microscopy.

Microscopy images show that the microstructure of sweet potatoes is influenced by both cooking and simulated digestion. Light microscopy images were obtained from the center of the sweet potato cubes before and after 240 min of simulated gastric digestion. Even though the sweet potato cubes remained visually intact (i.e. cubes were still intact and had not completely disintegrated during simulated digestion, as seen in Fig. 7), microstructural changes after 240 min of digestion were observed in the center of the cube (Figs. 5 and 6).

Fried sweet potatoes showed intracellular spaces after simulated digestion, but not cell wall breakage. This observation is in agreement with fried samples having the lowest change in texture (when not considering the softening of the crust) and the fastest effective diffusivity of acid compared to the four cooking methods. It is hypothesized that as intercellular spaces that were formed, this allowed acid go around the cell walls more easily, increasing its effective diffusivity in fried sweet potatoes. Evidence of intracellular gaps is shown in Fig. 5 D1, where fried samples did not show rupture of cell walls, but gaps between them, which might be due to the oil absorption during frying. It is hypothesized that during frying, the vegetable oil taken up by the sweet potatoes cube surrounds the cell walls, which may inhibit their breakdown by gastric acid. The images in fried samples show that the cells are more spread out after cooking compared to the steamed sweet potatoes (Fig. 5 C1 (with arrows) vs. A1). Since the effective diffusivity of the acid in fried samples was also greater than that of steamed, it suggests that in the fried samples, gastric acid is going into the food matrix, but not going inside the cells. These results are consistent with the hardness decrease in fried sweet potatoes, without taking into account the crust layer, which had the least hardness decrease compared to the other cooking methods. Since lipid digestion is not the main focus of this study, lipase was not added in the formulation of the gastric juice (Bornhorst and Singh, 2013; Minekus et al., 2014). Although the fat content of the fried sweet potatoes did not show a statistically significant decrease over digestion time, it followed a decreasing trend from 5.5 ± 1.1%, before digestion to 3.6 ± 0.1%. This decrease in oil, coupled with the increase in acid during digestion, suggests that there could be a possible exchange of oil leaving the food matrix with gastric acid coming into the sweet potato matrix.

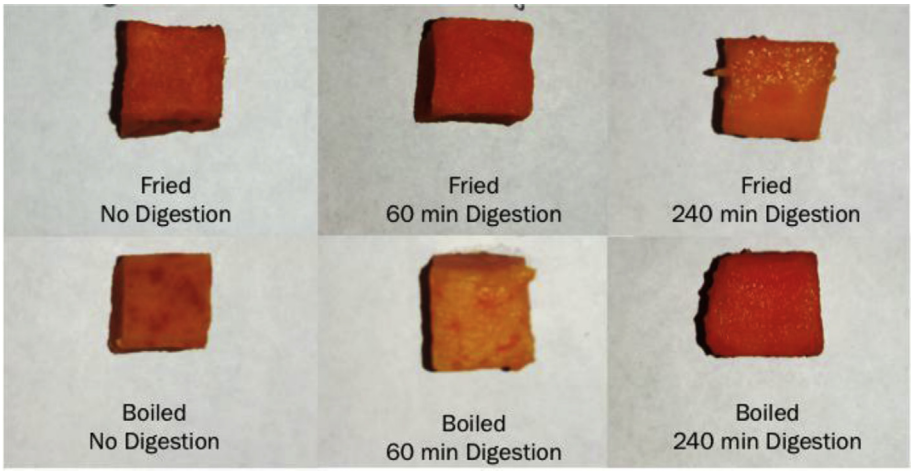

Fig. 7. Examples of sweet potato cubes after boiled and fried, and after 60 and 240 min of simulated gastric digestion.

Cell wall breakage was observed in steamed sweet potatoes after 240 min of simulated digestion suggesting that the gastric acid was capable of breaking the sweet potato cell walls (Fig. 5 B1 and B2). This is also seen in samples with stained with PAS (Fig. 6), where starch and other polysaccharides in the compound middle lamella of the cell wall are stained in red (Feder and O'Brien, 1968), which allows for visualization of starch breakdown (Fig. 6 B1 and B2). Starch breakdown may have occurred due to the α-amylase from the saliva during the oral digestion (Bornhorst et al., 2014), and cell wall breakdown may be related to the low pH of the gastric environment. Previous studies have shown that fruit cell walls are more prone to break down in an acidic environment than in an environment with a pH greater than 4.0, due to acid hydrolysis of pectin in the cell wall (Knee, 1982; McFeeters and Fleming, 1991).

Structure results suggest that the effects of cooking are evident in microstructure and texture at certain digestion times. However, it is important to consider that the microscopy images only represent a very small area of a sweet potato cube (approx. 5.7×10^{-7} m^2).

Previous studies have shown that, in addition to physical and structural changes in the food during simulated digestion, biochemical changes (i.e. acidity and moisture content) may occur (Mennah-Govela et al., 2015). In the present study, the cooking treatment influenced the effective diffusivity and equilibrium concentration of acid and water in sweet potatoes. Interestingly, it has been found that acid and moisture uptake over digestion time do not follow the same trend (Figs. 3 and 4), and they should be estimated separately. For example, steamed sweet potatoes had a similar acid uptake to microwave steamed (both going from 3.0 ± 0.3 to 5.1 ± 0.2 mg HCl/g DM). Alternatively, steamed had a moisture uptake similar to boiled sweet potatoes, and microwaved steamed and fried had a similar moisture uptake. These values are consistent with the equilibrium concentrations (C_e) of both water and acid of all the cooking methods that were estimated in the diffusion model (Table 3). It can be seen that boiled sweet potatoes reached the highest C_e of acid (7.8 mg HCl/g DM), and fried sweet potatoes had the lowest C_e (4.4 mg HCl/g DM). However, smaller differences were seen in the water C_e, were boiled and steamed reached 2.2 and 2.1 g H_2O/g DM, respectively and microwave steamed and fried reached 1.9 g H_2O/g DM. It is hypothesized that these differences are due to porosity and water holding capacity differences after cooking; an investigation into these factors merits future investigation.

As a result of the acid and moisture uptake, the D_{eff} of acid and water in each cooking method were similar (Table 3). Large confidence intervals of the effective diffusivities might be due to the inherent variations in the agricultural products (i.e. sweet potatoes) prior to cooking and simulating digestion. Although not statistically significant, the trends in effective diffusivity indicated that fried and microwave steamed sweet potatoes may have a higher effective diffusivity of acid compared to water. Rastogi et al., 2004 reported similar trends in rehydration of carrots with different pretreatments. They found that carrots pretreated with water prior to rehydration had a higher water diffusion coefficient than solute diffusion coefficient. These findings may support the results in the current study since boiled and steamed sweet potatoes, with water in the cooking process and higher initial moisture content, might have greater D_{eff} of water compared to microwave steamed and fried, which had lower initial moisture content. The volume decrease results (Fig. 1) suggest that during microwave steaming and frying, significant water loss and subsequent structural modifications occurred, resulting in a drier and more porous material compared to boiled and steamed sweet potatoes.

Limited information was found related to the D_{eff} of acid during simulated gastric digestion. Kong and Singh (2011) found that boiled carrots (cooked for 2 min) had a D_{eff} of gastric acid (pH of 1.8) of 8.72×10^{-11} m^2/s (Kong and Singh, 2011). Another study done using boiled white potatoes estimated that the D_{eff} of the acid was 1.03×10^{-9} m^2/s (Widjaja, 2010). However, these methods used either pH or a colored marker to estimate acid diffusion instead of a direct measurement of acidity, which may not accurately estimate the true diffusivity of gastric acid. By using experimental measurements of titratable acidity, the effective diffusivity of gastric acid was estimated in different types and varieties of rice and ranged from 0.59×10^{-9} m^2/s to 3.73×10^{-8} m^2/s (Mennah-Govela et al., 2015). Overall, previous studies have reported acid effective diffusivity values that range from 10^{-8} to 10^{-11}. As seen in Table 3, the current study values fall in between this range of order of magnitude (10^{-9}), however the differences might be due to several factors including processing method, macro and microstructural changes, and density differences between matrices. From the previous studies, the results reported by Kong and Singh (2011) for boiled carrots are the closest to the diffusivity values estimated in the current study for sweet potatoes, possibly due to similar structure and properties of the two tubers. Mennah-Govela et al. (2015) showed higher acid diffusion compared to the present study, most likely due to the porous structure the rice bolus, which had some spaces between the grains, allowing the acid to diffuse at

a faster rate. In contrast to a rice bolus, sweet potatoes have a less porous structure. Finally, carrots had a smaller D_{eff}, possibly due to the lower cooking time of the carrots (2 min), having a harder texture and a more rigid structure. These results demonstrate the importance of microstructure on acid and water diffusion into foods during digestion. Elucidation on the specific microstructural and physical properties that control diffusion into differing food matrices is an area that merits future investigation.

5. Conclusions

This study shows the relationship between cooking method and the physico-chemical changes of sweet potato during simulated gastric digestion. The current findings indicate that the effective diffusivity of acid does not follow the same trend as the moisture effective diffusivity during simulated gastric digestion of cooked sweet potatoes and should be estimated separately. Overall, mass transport processes in sweet potato were influenced by cooking method and simulated gastric digestion. Textural and microstructural changes of sweet potatoes after 240 min of simulated gastric digestion were seen in all cooking methods. The results from this study are important to develop a better understanding of the influence of cooking and the physical and chemical changes in foods during simulated gastric digestion.

References

Aguilera, J.M., 2005. Why food microstructure? J. Food Eng. 67 (1), 3–11.

AOAC, 2012. Official Methods of Analysis of AOAC International, nineteenth ed.

Bengtsson, A., Alminguer, M.L., Svanberg, U., 2009. In vitro bioaccessibility of beta-carotene from heat-processed orange-fleshed sweet potato. J. Agric. Food Chem. 57 (20), 9693–9698.

Bengtsson, A., Namutebi, A., Alminger, M.L., Svanberg, U., 2008. Effects of various traditional processing methods on the all-trans-β-carotene content of orange-fleshed sweet potato. J. Food Compos. Anal. 21 (2), 134–143.

Bornhorst, G.M., Ferrua, M.J., Singh, R.P., 2015. A proposed food breakdown classification system to predict food behavior during gastric digestion. J. Food Sci. 80 (5), R924–R934.

Bornhorst, G.M., Hivert, H., Singh, R.P., 2014. Rice bolus texture changes due to α-amylase. LWT—Food Sci. Technol. 55 (1), 27–33.

Bornhorst, G.M., Singh, R.P., 2013. Kinetics of in vitro bread bolus digestion with varying oral and gastric digestion parameters. Food Biophys. 8 (1), 50–59.

Bornhorst, G.M., Singh, R.P., 2014. Gastric digestion in vivo and in vitro: how the structural aspects of food influence the digestion process. Annu. Rev. Food Sci. Technol. 5, 111–132.

Bouchon, P., Aguilera, J.M., 2001. Microstructural analysis of frying potatoes. Int. J. Food Sci. Technol. 36 (6), 669–676.

Bouchon, P., Hollins, P., Pearson, M., Pyle, D., Tobin, M., 2001. Oil distribution in fried potatoes monitored by infrared microspectroscopy. J. Food Sci. 66 (7), 918–923.

Burri, B.J., 2011. Evaluating sweet potato as an intervention food to prevent vitamin A deficiency. Compr. Rev. Food Sci. Food Saf. 10 (2), 118–130.

Crank, J., 1975. The Mathematics of Diffusion.

Demain, A., Phaff, H., 1957. Cucumber curing, softening of cucumbers during curing. J. Agric. Food Chem. 5 (1), 60–64.

Failla, M.L., Thakkar, S.K., Kim, J.Y., 2009. In vitro bioaccessibility of beta-carotene in orange fleshed sweet potato (Ipomoea batatas, Lam.). J. Agric. Food Chem. 57 (22), 10922–10927.

Feder, N., O'Brien, T., 1968. Plant microtechnique: some principles and new methods. Am. J. Bot. 123–142.

Gabaldón-Leyva, C.A., Quintero-Ramos, A., Barnard, J., Balandrán-Quintana, R.R., Talamás-Abbud, R., Jiménez-Castro, J., 2007. Effect of ultrasound on the mass transfer and physical changes in brine bell pepper at different temperatures. J. Food Eng. 81 (2), 374–379.

Hedren, E., Diaz, V., Svanberg, U., 2002. Estimation of carotenoid accessibility from carrots determined by an in vitro digestion method. Eur. J. Clin. Nutr. 56 (5), 425–430.

Heil, J., McCarthy, M., 1989. Influence of acidification on texture of canned carrots. J. Food Sci. 54 (4), 1092–1093.

Howard, L., Burma, P., Wagner, A., 1994. Firmness and cell wall characteristics of pasteurized jalapeno pepper rings affected by calcium chloride and acetic acid. J. Food Sci. 59 (6), 1184–1186.

Kaur, L., Singh, N., Sodhi, N.S., Gujral, H.S., 2002. Some properties of potatoes and their starches I. Cooking, textural and rheological properties of potatoes. Food Chem. 79 (2), 177–181.

Knee, M., 1982. Fruit softening III. Requirement for oxygen and pH effects. J. Exp. Bot. 33 (6), 1263–1269.

Kong, F., Singh, R.P., 2011. Solid loss of carrots during simulated gastric digestion. Food Biophys. 6 (1), 84–93.

Krall, S.M., McFeeters, R.F., 1998. Pectin hydrolysis: effect of temperature, degree of methylation, pH, and calcium on hydrolysis rates. J. Agric. Food Chem. 46 (4), 1311–1315.

Lemmens, L., Van Buggenhout, S., Van Loey, A.M., Hendrickx, M.E., 2010. Particle size reduction leading to cell wall rupture is more important for the β-carotene bioaccessibility of raw compared to thermally processed carrots. J. Agric. Food Chem. 58 (24), 12769–12776.

Mandalari, G., Faulks, R.M., Rich, G.T., Lo Turco, V., Picout, D.R., Lo Curto, R.B., Bisignano, G., Dugo, P., Dugo, G., Waldron, K.W., 2008. Release of protein, lipid, and vitamin E from almond seeds during digestion. J. Agric. Food Chem. 56 (9), 3409–3416.

Markowski, M., Bondaruk, J., Błaszczak, W., 2009. Rehydration behavior of vacuum-microwave-dried potato cubes. Dry. Technol. 27 (2), 296–305.

McFeeters, R., Brenes Balbuena, M., Fleming, H., 1995. Softening rates of fermented cucumber tissue: effects of pH, calcium, and temperature. J. Food Sci. 60 (4), 786–788.

McFeeters, R., Fleming, H., 1990. Effect of calcium ions on the thermodynamics of cucumber tissue softening. J. Food Sci. 55 (2), 446–449.

McFeeters, R., Fleming, H., 1991. pH effect on calcium inhibition of softening of cucumber mesocarp tissue. J. Food Sci. 56 (3), 730–732.

McFeeters, R.F., Fleming, H.P., 1989. Inhibition of cucumber tissue softening in acid brines by multivalent cations: inadequacy of the pectin“ egg box” model to explain textural effects. J. Agric. Food Chem. 37 (4), 1053–1059.

Mennah-Govela, Y.A., Bornhorst, G.M., 2016. Acid and moisture uptake in steamed and boiled sweet potatoes and associated structural changes during in vitro gastric digestion. Food Res. Int. (in press).

Mennah-Govela, Y.A., Bornhorst, G.M., Singh, R.P., 2015. Acid diffusion into rice boluses is influenced by rice type, variety, and presence of α-amylase. J. Food Sci. 80 (2), E316–E325.

Minekus, M., Alminger, M., Alvito, P., Ballance, S., Bohn, T., Bourlieu, C., Carrière, F., Boutrou, R., Corredig, M., Dupont, D., 2014. A standardised static in vitro digestion method suitable for food—an international consensus. Food & Funct. 5 (6), 1113–1124.

O'Brien, T., Feder, N., McCully, M.E., 1964. Polychromatic staining of plant cell walls by toluidine blue O. Protoplasma 59 (2), 368–373.

Parada, J., Aguilera, J.M., 2007. Food microstructure affects the bioavailability of several nutrients. J. Food Sci. 72 (2), R21–R32.

Rastogi, N., Nayak, C., Raghavarao, K., 2004. Influence of osmotic pre-treatments on rehydration characteristics of carrots. J. Food Eng. 65 (2), 287–292.

Teow, C.C., Truong, V.-D., McFeeters, R.F., Thompson, R.L., Pecota, K.V., Yencho, G.C., 2007. Antioxidant activities, phenolic and β-carotene contents of sweet potato genotypes with varying flesh colours. Food Chem. 103 (3), 829–838.

Tumuhimbise, G.A., Namutebi, A., Muyonga, J.H., 2009. Microstructure and in vitro beta carotene bioaccessibility of heat processed orange fleshed sweet potato. Plant Foods Hum. Nutr. 64 (4), 312–318.

Tydeman, E.A., Parker, M.L., Wickham, M.S., Rich, G.T., Faulks, R.M., Gidley, M.J., Fillery-Travis, A., Waldron, K.W., 2010. Effect of carrot (Daucus carota) microstructure on carotene bioaccessibility in the upper gastrointestinal tract. 1. In vitro simulations of carrot digestion. J. Agric. Food Chem. 58 (17), 9847–9854.

Van Buggenhout, S., Alminger, M., Lemmens, L., Colle, I., Knockaert, G., Moelants, K., Van Loey, A., Hendrickx, M., 2010. In vitro approaches to estimate the effect of food processing on carotenoid bioavailability need thorough understanding of process induced microstructural changes. Trends Food Sci. Technol. 21 (12), 607–618.

Widjaja, J.M., 2010. Kinetics of Gastric Juice Diffusion to Solid Food during Digestion. University of California, Davis.