



## Short communication

## Resistant starch analysis of commonly consumed potatoes: Content varies by cooking method and service temperature but not by variety

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## ABSTRACT

Resistant starch (RS) has unique digestive and absorptive properties which may provide health benefits. We conducted a study to determine the contributions of cultivar, cooking method and service temperature on the RS contents of potatoes (*Solanum tuberosum* L.). We hypothesized that the RS content would vary by variety, cooking method and service temperature. Potatoes of three common commercial varieties (Yukon Gold, Dark Red Norland, and Russet Burbank) were subjected to two methods of cooking (baking or boiling) and three service temperatures: hot (65 °C), chilled (4 °C) and reheated (4 °C for 6d; reheated to 65 °C) and analyzed the starch content by modification of a commercially available assay. Results showed that RS content (g/100 g) varied by cooking method and service temperature but not variety. Baked potatoes had higher RS contents than boiled; chilled potatoes had more RS than either hot or reheated. These results may assist in dietary choices for reducing chronic disease risk.

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## 1. Introduction

Obesity, diabetes, and cardiovascular disease are major health concerns facing Americans today that are linked by diet (Barclay et al., 2008; McCullough et al., 2002). Importantly, factors such as resistant starch (RS) have been shown to be beneficial in the risk reduction of chronic cardiometabolic disease (Bindels, Walter, & Ramer-Tait, 2015). RS has digestive and absorptive properties which may provide health benefit. Dietary RS is unique in that it is not rapidly digested like ordinary starch and provides important health benefits, some that are similar to those provided by dietary fiber, and some that are unique to RS (Haralampu, 2000). Demonstrated physiological effects of RS include increased satiety (Bodinham, Frost, & Robertson, 2010; Raben et al., 1994), improved insulin sensitivity and glycemic control (Keenan et al., 2015; Robertson, Bickerton, Dennis, Vidal, & Frayn, 2005), and enhanced gut health (Keenan et al., 2015; Phillips et al., 1995).

White potatoes (*Solanum tuberosum* L.) are a commonly consumed food with an average yearly consumption of ~128 lb per

capita in the US (USDA, 2014). In addition to carbohydrates, potatoes of all varieties provides important nutrients such as protein, vitamins (vitamin C, thiamin, riboflavin, niacin), and minerals (potassium and phosphorus) (Camire, Kubow, & Donnelly, 2009; Freedman & Keast, 2011; Weaver & Marr, 2013). The majority of the carbohydrate content of potatoes is starch, some of which is RS that is relatively slowly hydrolyzed in the gut (Jansen, Flamme, Schüler, & Vandrey, 2001).

In plant tissues, starch is packaged in granules and is comprised of varying amounts of amylose and amylopectin. Amylose is a straight chain polyglucan made of  $\alpha$ -D-glucose units bound to each other through  $\alpha$ 1–4 glycosidic bonds. Amylopectin is a highly branched polymer of glucose with linear glucose units linked with  $\alpha$ 1–4 and branched glucose units linked with  $\alpha$ 1–6 glycosidic bonds (Zobel, 1988). Cooking disrupts starch granules rendering the starch accessible to digestive enzymes. RS is comprised of starch and starch degradation products that are not absorbed in the small intestine but, instead, pass to the large intestine (Berry, 1986).

RS, like dietary fiber, can be fermented by commensal gut microbiota. Fermentation yields short-chain fatty acids, including propionate, a substrate for hepatic gluconeogenesis, and butyrate, the major source of energy for colonocytes (Topping & Clifton, 2001). The molar yield of butyrate from RS is greater than that observed for non-starch polysaccharide prebiotics such as pectin (Pryde, Duncan, Hold, Stewart, & Flint, 2002). Butyrate is thought to protect against colonic mutagenesis and tumorigenesis (Hamer et al., 2008). The RS content of potatoes can be modified

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by the cooking preparation and service temperature. RS3, or retrograde RS, is the most resistant starch fraction and is primarily formed by the retrogradation of amylose during cooling (Sajilata, Singhal, & Kulkarni, 2006).

The consumption of high RS potatoes may therefore provide health related benefits for obesity prevention, glycemic control and colonic health (Jenkins et al., 1998).

Relatively little work has been performed to characterize the RS composition of potatoes as commonly consumed. The starch content of potatoes is highly variable, ranging from 11% to 30% of freshly cultivated potatoes to 4–40% in fresh wild cultivars (Jansen et al., 2001). In general, fresh potatoes contain about 20 percent dry matter, 60–80% of which is starch. The starch in potatoes is generally 70–80% amylopectin with the remainder being amylose (Zeeman, Kossmann, & Smith, 2010).

The total starch content of potato tubers is significantly affected by their genotype, the environment, and condition under which they are grown (Bach, Yada, Bizimungu, Fan, & Sullivan, 2013). Not all of the starch in potatoes is made bioavailable by cooking as some of it remains ungelatinized and is poorly digested. When potatoes are chilled they form retrograde starch, or RS3, which is not available for digestion in the small intestine (Haralampu, 2000). It is likely that these components, collectively referred to as RS, vary among potato varieties, and are affected by cooking method and service temperature. We hypothesized that the RS content of the potatoes tested would vary by variety, cooking method and service temperature. To test this, we conducted the following study to determine the contributions of variety, cooking method and service temperature on the RS content.

## 2. Methods and materials

### 2.1. Materials

Potatoes of three varieties grown in the Red River Valley of North Dakota and Minnesota (Yukon Gold, Dark Red Norland, and Russet Burbank) were provided by the Northern Plains Potato Growers Association, East Grand Forks, MN, from the 2013 growing season immediately after harvest. All of the potatoes used in these experiments were not treated with a sprout inhibitor.

### 2.2. Potato preparation

The tubers were prepared for analysis in the Metabolic Kitchen of the Grand Forks Human Nutrition Research Center, Grand Forks, ND. All three varieties were handled in an identical fashion. Uniformly sized potatoes were prepared in each of two ways: (1) washed, wrapped in aluminum foil and baked at 177 °C (350 °F) for 65–80 min (baked); or (2) washed, peeled, eyes and bruises removed and cut into small pieces (2.5 cm<sup>2</sup>) and boiled at 100 °C (212 °F) until they reached the same degree of tenderness (~10–11 min) (boiled). Cooked potatoes from each cooking treatment were analyzed under three temperatures representing common conditions of food service: (1) maintained at 65 °C (150 °F) (hot); (2) held at 4 °C (40 °F) for six days (chilled); and (3) held at 4 °C (40 °F) for six days followed by reheating to 65 °C (150 °F) (reheated). All individual potato samples were prepared for analysis with the use of a food preparation style potato ricer prior to analysis by placing a sample into the ricer and forcing it through the with hand pressure.

### 2.3. Resistant starch determination

Resistant starch was determined utilizing the commercially available Megazyme Resistant Starch Assay (K-RSTAR, Megazyme International Ireland Ltd, Co. Wicklow, Ireland) kit. This method

was modified by miniaturization of the assay and inclusion of a bacteriostatic agent (sodium azide) to prevent microbial alteration of the carbohydrate profile during analysis. These changes facilitated the development of quantitative standard curves for amylose (A8515, Sigma-Aldrich, St. Louis, MO, USA) and amylopectin (A0512, Sigma-Aldrich, St. Louis, MO, USA) over the range of 0–5 µg/mL. Potato samples were analyzed in triplicate. Riced potato samples (0.50 g) were weighed into 16 × 125 mm screw-cap test tubes; 4 mL pancreatic amylase solution (10 mg/mL) (3 U/mL amyloglucosidase)/sodium azide (0.03%) was added to each sample. The tubes were capped, incubated at 37 °C with continuous shaking at 100 rpm for exactly 16 h. to allow for solubilization of the non-resistant starch and hydrolyzation to D-glucose.

The reaction was terminated by addition of 4 mL of absolute ethanol. Resistant starch was recovered by centrifugation (2000g, 10 min at room temperature). Pellets were washed twice with 8 mL of 50% ethanol at 4 °C and centrifuged (2000g, 10 min at room temperature). The pellet was dissolved by addition of 2 mL of 2 M KOH with vigorously stirring. The solution was then neutralized with 8 mL of 1.2 M sodium acetate buffer (pH 3.8) followed immediately by adding 0.1 mL amyloglucosidase and incubating the samples at 50 °C with shaking for 60 min. Contents of the tubes were diluted 1:10 with distilled water. An aliquot of each solution was centrifuged (1500g, 10 min), and 40 µL of the supernate was mixed with 1.2 mL of the glucose oxidase-peroxidase-4-aminoantipyrene reagent (Megazyme Resistant Starch Assay, Megazyme International Ireland Ltd, Co. Wicklow, Ireland) and incubated at 50 °C for 20 min. An aliquot of the reaction mixtures was then transferred to a 96-well plate and absorbance was measured at 510 nm against a reagent blank using a microplate reader (SpectraMax 190, Molecular Devices, Sunnyvale, CA, USA).

Three replicates of each potato sample were analyzed. Results were corrected for moisture content; data are presented as mean ± standard deviation (SD).

### 2.4. Statistical analysis

The effects of potato variety (Yukon Gold, Dark Red Norland, and Russet Burbank), method of preparation (baked or boiled), service temperature (hot, chilled or reheated) and their interactions on the potato RS content was analyzed using 3-way analysis of variance (ANOVA). Tukey's contrasts were used for post hoc comparisons of means. SAS V9.4 (SAS Institute, Inc., Cary NC) was used for all statistical analyses.

## 3. Results and discussion

We were able to adapt a widely used starch analysis kit for use with a smaller sample size. Using this modification we determined the RS content of potatoes by variety, cooking technique and service method. The RS content (mean ± SD) of the potatoes is presented in Table 1. Results show that the RS composition (g/100 g) was significantly affected by both method of cooking preparation ( $p < 0.0001$ ) and service temperature ( $p < 0.0001$ ), but not by potato variety. We expected that the potatoes would vary by variety but were unable to detect any differences.

The RS difference observed by preparation technique may be due, in part, to the hydration level of the cooked potato and the nature of the starch granule. Baked potatoes had an increased level of RS compared to boiled potatoes, regardless of variety. Baking is known to increase RS (Sajilata et al., 2006) as the use of heat treatment with moisture has been shown to increase RS formation. The use of 18% moisture with heat treatment has been shown to increase RS formation while at 27% moisture, starch degrades to some extent making it more available to enzymatic action

**Table 1**  
Resistant starch content of potatoes by variety, preparation technique, and service method.<sup>1</sup>

Preparation:	Baked <sup>a</sup>			Boiled		
	Hot	Chilled <sup>b</sup>	Reheated	Hot	Chilled <sup>b</sup>	Reheated
Variety:						
Red Norland	3.8 ± 0.6	4.8 ± 0.7	3.8 ± 1.1	2.5 ± 0.9	3.1 ± 0.2	2.6 ± 0.4
Russet Burbank	3.5 ± 0.2	4.7 ± 0.5	4.2 ± 0.7	2.6 ± 0.4	3.8 ± 0.2	3.6 ± 0.6
Yukon Gold	3.5 ± 0.4	5.4 ± 0.5	4.3 ± 0.4	2.3 ± 0.8	3.9 ± 0.3	2.2 ± 0.5

<sup>1</sup> Mean and SD.

<sup>2</sup> Data were analyzed by 3-way ANOVA: Variety (V),  $p = 0.40$ ; Service Method (S),  $p < 0.0001$ ; Preparation Technique (P),  $p < 0.0001$ ;  $V \times S$ ,  $p = 0.17$ ;  $V \times P$ ,  $p = 0.12$ ;  $S \times P$ ,  $p = 0.88$ ;  $V \times S \times P$ ,  $p = 0.72$ .

<sup>a</sup> Resistant starch was greater in baked (4.22 g/100 g) than boiled (2.93 g/100 g) potatoes,  $p < 0.0001$ , by ANOVA.

<sup>b</sup> Chilled potatoes contained more resistant starch (4.27 g/100 g) than hot (3.00 g/100 g) or reheated potatoes (3.45 g/100 g),  $p < 0.001$ , by Tukey contrasts.

(Sievert & Pomeranz, 1989). This may explain the difference we observed with baking vs. boiling of the potatoes.

When prepared to be served hot, the potatoes had less RS than either chilled or reheated potatoes. We observed that when the service method was chilled, potatoes contained more RS (4.3 ± 0.9 g/100 g) than hot (3.0 ± 0.8 g/100 g) or reheated potatoes (3.4 ± 1.0 g/100 g),  $p < 0.001$ , by Tukey contrasts. As described previously, this is because amylose goes through retrogradation increasing the RS3 content when potatoes are chilled (Tahvonon, Hietanen, Sihvonen, & Salminen, 2006).

Preparing commonly used potatoes by baking and serving chilled or reheated increases the RS content. Intake of increased RS from the inclusion of potatoes in the diet may be helpful in the primary prevention of chronic metabolic disease. The slow digestion of high RS foods is associated with improved lipids, glycemia and reduced insulinemia (Haub, Louk, & Lopez, 2012). RS that is not absorbed in the small intestine may behave in a manner associated with dietary fibers (Topping & Clifton, 2001) and result in increased fecal bulk (Phillips et al., 1995), and alteration of colonic microbiota (Martinez, Kim, Duffy, Schlegel, & Walter, 2010). Clinical trials demonstrate improvements in cholesterol levels (Dodevska et al., 2015; Gentile et al., 2015) and insulin sensitivity (Maki et al., 2012) in response to RS intake.

#### 4. Conclusions

No significant differences were found in the RS contents of three potato varieties grown in the Red River Valley of North Dakota and Minnesota: Dark Red Norland, Russet Burbank, and Yukon Gold. However, in all potatoes RS was significantly affected by both cooking and service methods: baked potatoes > boiled potatoes; and, regardless of cooking method, chilled > reheated > hot potatoes. This demonstrates that, while all potatoes contained RS, the methods of cooking and service do modify RS levels. Potatoes are a source of dietary RS, which can be increased by baking and serving chilled or reheated. Choosing potatoes prepared in these ways will increase dietary RS intake and may be helpful in the prevention of common chronic metabolic disease states. Future efforts in breeding to increase the RS content of different potato varieties will help to increase dietary RS levels.

#### 5. Conflict of interest

All authors declare no conflict of interest.

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