

**Interim Report – SBOE Incubation Grant Program****Generation of Potato-Based Resistant Starch Ingredients for Testing within Commercial Product Prototypes by an Industrial Partner****A Project of the University of Idaho's College of Agricultural and Life Science (CALs)****March 12, 2012****Principle Investigator:**

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**“Gap” Project Objective:**

“Gap” funds are requested to *generate sufficient quantities of novel potato resistant starch (RS) ingredients (UI role) for an industrial partner (J.R. Simplot Co.) to test within their existing commercial potato product lines (i.e., mashed potatoes, potato croquettes, extruded potato products, etc.) to validate processability and product quality.*

**Phase I: Bench-top Level Testing**

- Prepare a range of RS ingredients with variable molar substitution levels (0.1-0.2) and/or cross-linking levels (0.01-0.4% phosphorus) for testing in bench-top commercial product trials conducted by Simplot
- Verify initial RS/SDS levels within the various modified RS ingredients

**Phase II: Pilot- Level Testing**

- Prepare greater quantities of select RS ingredients (those identified in the previous bullet above) for use in larger-scale (pilot) commercial product tests conducted by Simplot

**Summary:**

Initial bench-top testing of potato resistant starch (RS) ingredients was completed by Simplot using an instant mashed potato product format. At 50% incorporation within an agglomerated instant mash mix, potato RS ingredients (modified with appropriate levels of propylene oxide and/or phosphorus oxychloride) appeared to contribute favorable characteristics and physical properties to mashed potato products. Based on discussions with Simplot, a decision was made to move forward with pilot scale-processing to efficiently generate additional amounts of potato RS materials needed for testing within other product applications (e.g., croquettes, restructured hash browns, French-fries, etc.). In preparation for production scale-up of RS materials, modification reaction conditions are being fine-tuned and validated to determine which potato RS materials will ultimately be selected for pilot-scale processing. RS levels, Digestibility Index and *in vitro* estimated Glycemic Index (eGI) values for various potato RS ingredients modified with sodium trimetaphosphate/sodium

tripolyphosphate (STMP/STPP) and octenyl succinic anhydride (OSA) have been verified, and are outlined below.

Simplot potato granules were dual-modified with STMP (11.9%, potato granule weight basis) and STPP (0.12%, potato granule weight basis) based on conditions reported by Yeo and Seib (2009). Reaction condition variables included pH (11.5 or 12.8), the concentration of isopropanol within the reaction medium (40, 45, or 100%), and the concentration of potato granules within the reaction system (32, 36, or 41%). Modified and unmodified potato granule materials were analyzed for percent of RDS/SDS/RS, with select results reported in Table 1 below. In short, all modification conditions tested developed significant levels of RS, with variable levels of SDS.

**Table 1. *In vitro* rapidly-digestible (RDS), slowly-digestible (SDS), and resistant (RS) starch contents for potato granules (PG) modified with sodium trimetaphosphate (STMP)<sup>1</sup> and sodium tripolyphosphate (STPP)<sup>1</sup> under varied reaction system conditions.**

Treatments	RDS	SDS	RS
Condition 1 (100% isopropanol, 41%PG, pH 12.8)	39.8	13.8	30.1
Condition 2 (45% isopropanol, 41%PG, pH 11.5)	21.5	26.5	34.8
Condition 3 (45% isopropanol, 36%PG, pH 11.5)	37.1	6.5	38.6
Condition 4 (45% isopropanol, 32%PG, pH 11.5)	35.4	10.9	36.6
Condition 5 (40% isopropanol, 32%PG, pH 11.5)	33.5	9.2	39.5
Unmodified Potato Granule, No Treatment	77.5	2.3	4.3

<sup>1</sup> Modified with STMP and STPP at 11.9% and 0.12% (based on potato granule weight), respectively.

Modified potato granules and an unmodified control were further analyzed for digestibility index (HI90), which is the amount of digested starch (i.e. glucose) expressed as a percentage (%) of the total starch content after 90 minutes of *in vitro* digestion (Table 2). In addition, *in vitro* Glycemic Index (eGI) was estimated according to the method of Goni et al. (1997) (Table 2). The eGI values for all modified granules (Table 2) were reduced 42-48% by modification (relative to the unmodified control), corresponding to a medium glycemic category. Degree of substitution values will be determined to ensure that extents of modification fall within allowable ranges.

**Table 2. *In vitro* digestibility index and estimated glycemic index (eGI) values for potato granules (PG) modified with sodium trimetaphosphate (STMP)<sup>1</sup> and sodium tripolyphosphate (STPP)<sup>1</sup> under varied reaction system conditions.**

Treatment	Digested Starch Content (HI90) <sup>2</sup>	eGI <sup>3</sup>	GI Category <sup>4</sup>
Condition 3 (45% isopropanol, 36%PG, pH 11.5)	24.0	58.5	Medium
Condition 4 (45% isopropanol, 32%PG, pH 11.5)	30.0	63.3	Medium
Condition 5 (40% isopropanol, 32%PG, pH 11.5)	31.9	64.8	Medium
Unmodified Potato Granule, No Treatment	90.0	111.5	High

<sup>1</sup> Modified with STMP and STPP at 11.9% and 0.12% (based on potato granule weight), respectively

<sup>2</sup> Digestibility index (HI90) was determined by the amount of digested starch (i.e., glucose) after 90 minutes of *in vitro* digestion expressed as a percentage (%) of the total starch content.

<sup>3</sup> *In vitro* eGI was estimated by the method of Goni et al. (1997).

<sup>4</sup> GI categories are defined as follows: low GI = less than 55; medium GI = 55-69; high GI = greater than 69.

Potato granules were also modified with reduced levels of STMP and STPP (36-75% lower than original modification levels depicted in Table 1) within a 40% aqueous isopropanol medium using a 32% (w/v) potato granule slurry. Modified and unmodified samples were then analyzed for percent of RDS/SDS/RS (Table 3). As anticipated, RS levels decreased in direct proportion to reducing levels of reagent addition, and were not sufficiently high to warrant further analysis of eGI.

**Table 3. *In vitro* rapidly-digestible (RDS), slowly-digestible (SDS), and resistant (RS) starch contents for potato granules (PG) modified with reduced levels of sodium trimetaphosphate (STMP)<sup>1</sup> and sodium tripolyphosphate (STPP)<sup>1</sup> within a 40% aqueous isopropanol medium using a 32% (w/v) potato granule slurry.**

Treatments	RDS	SDS	RS
Condition 6 (8.8% STMP, 0.09% STPP)	72.2	0	18.0
Condition 7 (6.5% STMP, 0.07% STPP)	72.0	8.2	10.1
Condition 8 (5.4% STMP, 0.05% STPP)	75.4	2.0	6.6
Condition 9 (4.2% STMP, 0.04% STPP)	77.2	0.0	7.9
Condition 10 (3.0% STMP, 0.03% STPP)	74.8	0.1	9.1
Unmodified Potato Granule, No Treatment	77.5	2.3	4.3

Potato granules were also modified with 3% octenyl succinic anhydride (OSA, based on potato granule weight), using either an aqueous or 40% aqueous isopropanol reaction medium. For modifications conducted strictly within an aqueous medium, potato granule concentration was limited to 17% (w/v) in order to avoid excessive material swelling during modification. In contrast, potato granule concentration within the aqueous isopropanol could be increased to 32% (w/v) potato granules without incurring problems with excessive viscosity. All modifications were conducted at pH 8.5-9.0 for 3 hours at 25°C. Modified and unmodified samples were then analyzed for RDS/SDS/RS (Table 4). Though the water (relative to the aqueous isopropanol) reaction medium generally yielded higher RS levels, neither of the reactions generated sufficient levels of RS/SDS to warrant further analysis of eGI.

**Table 4. *In vitro* rapidly-digestible (RDS), slowly-digestible (SDS), and resistant (RS) starch contents for potato granules (PG) modified with 3% octenyl succinic anhydride (based on potato granule weight) within either an aqueous or 42% aqueous isopropanol reaction medium for 3 hours at 25°C.**

Treatments	RDS	SDS	RS
Condition 11 (water medium)	81.8	0.0	10.9
Condition 12 (42% aqueous isopropanol)	81.0	1.8	5.4
Unmodified control-isopropanol <sup>6</sup>	78.4	2.4	7.4

In summary, the reagents that will be considered for industrial scale-up purposes will be propylene oxide, dual modification with propylene oxide and phosphorus oxychloride, and the STMP/STPP combination (provided that adequate RS can be generated using allowable addition levels). Penford Food Ingredients (Denver, CO) has been identified as the preferred industrial partner for conducting processing scale-up of the potato RS ingredients. Penford has an existing business relationship with J.R. Simplot, and has industrial capacity to generate modified potato granule materials. A three-way non-disclosure agreement is in place between the University of Idaho, J.R. Simplot, and Penford, and two separate discussions have taken place with Penford in planning for the process scale-up, which will occur later this Spring at the Penford Cedar Rapids, Iowa facility. Both University of Idaho and J.R. Simplot personnel will be present scale-up activities.

**Budgetary Updates:**

Total SBOE funding received: \$50,000.00  
 Total spent as of February 29, 2012: \$38,785.96  
 \$11,214.04

Funds remaining by category:  
 Salary/fringe benefits: \$ 5,970.49  
 Travel: \$ 2,100.00  
 Operating expense: \$ 3,143.55  
 \$11,214.04

<b><i>Detailed Allocations</i></b>	
<b><i>Library Support</i></b>	None
<b><i>Graduate Research Assistantships/Research Associates</i></b>	None
<b><i>Post Doctoral Fellows</i></b>	A total of \$12,600 requested for salary and fringe benefits. To date \$10,319 has been expensed with the remainder to be paid out during the spring completion phase of the project.
<b><i>Technician Support</i></b>	A total of \$26,100 requested for salary and fringe benefits. To date \$22,410 has been expensed with the remainder to be paid out during the spring completion phase of the project.
<b><i>Maintenance Contracts</i></b>	None
<b><i>Research Equipment</i></b>	None
<b><i>Competitively Awarded Summer Research Support</i></b>	None

<b>Start-Up Funds for New Hires</b>	None
<b>Incentives to Reward Faculty for Research Achievements</b>	In October 2011, this technology was recognized with the Idaho Early-Stage Innovation of the Year Award by the Idaho Technology Council.
<b>Other</b>	
<b>Total Allocation</b>	

<b>Detailed Allocations</b>	<b>University of Idaho</b>
<b>Publications in refereed journals</b>	None
<b>Presentations at professional meetings and conferences</b>	None
<b>Grants Received as a result</b>	None
<b>Grants Pending</b>	Our industrial partner, J.R. Simplot, has agreed to help fund the scale-up activities to be conducted by Penford Food Ingredients at their Cedar Rapids, IA facility in regard to plant, personnel and material expenses. Simplot has also agreed to pay the fees for US and world patent applications.
<b>Student Participation</b>	None
<b>Faculty Participation</b>	None
<b>Other Participation</b>	None

<b>Patents Awarded</b>	None
<b>Patents Pending</b>	<p>U.S. Patent Application of Kerry C. Huber, Serial No. 13/366,900, Title: Methods of Preparing Potato Food Products with Enhanced Resistant Starch Content, Feb 9, 2012</p> <p>U.S. Patent Application of K. C. Huber and W. C. Yu, Title: Potato Products with Enhanced Resistant Starch Content and Moderated Glycemic Response and Methods thereof. Filed October 2, 2010 (pending U.S. Application No: 12896542; International Application No: PTC/US10/51164).</p>
<b>Manuscripts Submitted</b>	<p>In Preparation - Anantachote, A., Huber, K. C. and Nelson, J. E. (2012). "Separation and Isolation of Intact Parenchyma Cells from Raw (uncooked) Potato (<i>Solanum tuberosum</i>) Tissue".</p> <p>In Preparation – Yu, W.C., Huber, K. C. and Nelson J. E. (2012). "Development of a Multifunctional Potato Ingredient with Enhanced Resistant Starch Content and a Moderated Glycemic Response".</p>