# Application of Acid Solubilization Isoelectric Precipitation to Recover Protein from Low Value Red Meat

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#### **Story in Brief**

Three experiments were conducted to determine the composition and gel attributes of protein recovered using acid solubilization isoelectric precipitation (Acid-SIP) on low value red meat. In the studies, the Acid-SIP process was used to recover a protein concentrate from beef heart, pork heart, and whole ground pork picnic shoulder. Half of each protein concentrate was treated with salt (NaCl) to determine its effect on cooked gel attributes. Raw protein concentrates were evaluated for proximate analysis, color, collagen, and cholesterol. They were also cooked and evaluated for textural properties, proximate analysis, and color. Raw protein concentrates showed a significant reduction in fat, ash, collagen, and cholesterol compared to the original, untreated product (control). Color was slightly affected by treatment. Cooked gels showed similar improvements in proximate composition as observed in the raw protein concentrates. Color differences in cooked treated product versus untreated product were also observed. Cook yield and water holding ability (WHA) were significantly improved when the Acid-SIP process was applied. Texture profile analysis (TPA) demonstrated improvement in gel attributes of the Acid-SIP treated cooked gels. The addition of salt had minimal effect on the gel strength of the Acid-SIP gels. This data suggests that the Acid-SIP process improved nutritional composition of low value red meat while maintaining its protein functionality with regard to cooked gel attributes.

Key Words: Beef Heart, Pork Heart, Whole Ground Pork Picnic Shoulder, Acid Solubilization Isoelectric Precipitation, Composition, Gel Attributes

#### Introduction

Acid solubilization isoelectric precipitation (SIP) is a meat recovery process that takes advantage of the protein solubility differences to separate myofibrillar proteins from collagen, fat, and bone. The process extracts proteins that are still functional as they have not been heat-treated. Thus, the process could be applied to recover a stable, inexpensive and functional protein concentrate from materials that otherwise might normally just be collectively rendered. Beef heart is considered poor-binding meat due to the high amounts of collagen present in the muscle (Pearson and Gillett, 1996). Therefore, beef heart was chosen as the initial model to determine the applicability and process parameters of the Acid-SIP process on red meat (Mireles DeWitt et al., 2002). Due to the age when harvested, lower collagen levels give pork heart a higher binding potential than beef heart, but it is still sold as a low-value meat byproduct. By testing the process on a complex matrix of whole ground pork picnic shoulder, the process could be tested for its ability to recover a functional protein concentrate from skeletal muscle as well as smooth muscle protein. In addition, proximate analysis on whole ground pork picnic shoulder would verify Acid-SIP's ability to separate the protein from bone as well as collagen and fat. The objectives of these studies were to determine the composition and textural properties of protein concentrates recovered using Acid-SIP.

## **Materials and Methods**

# **Preparation of Acid-SIP Proteins**

All samples were collected and maintained at 5°C or less throughout preparation and treatment.

In experiment 1 and 2, beef hearts (8-10) from 1 local packing company and pork hearts (6-8) from the Oklahoma Food & Agricultural Product Center were collected and held overnight (5°C) prior to trimming off the cap, valves, and external fat. The hearts were ground and used to prepare meat blocks.

In experiment 3, fresh skinned picnic shoulders (8) were ground with a whole body grinder to determine if the process could recover a functional protein from a more complex protein, bone, collagen, and fat matrix. The whole ground pork picnic shoulder was used to prepare meat blocks.

A portion of all the meat blocks were processed using the Acid-SIP process according to Mireles DeWitt et al (2002) with the following modifications. A 1:9 (w/v) mixture of beef heart, pork heart, or whole ground picnic shoulder to 2mM citrate buffer was blended for 1 min. The pH was lowered to 2.5 with 2N HCl and then centrifuged at 3300 x g for 30 min. The forces for the initial separation used in these experiments were reduced (from 10000 x g to 3300 x g) in order to facilitate recovery of sufficient product in a timely manner to conduct texture studies. Previous work by Kelleher and Hultin (2000) suggested higher forces were needed to reduce the amounts of phospholipids in the product to control lipid oxidation over long-term storage. However, preliminary experiments in our laboratory demonstrated that lipid oxidation would not be an issue in the shorter time frame (2-3 d) covered in these studies. The supernatant was collected and myofibrillar proteins were precipitated by raising the pH 5.5 with 2N NaOH. Precipitated proteins were recovered by centrifugation at 3300 x g.

## **Preparation of the Treatments**

Precipitated protein from meat blocks and their respective untreated controls were centrifuged at 10000 x g for 15 min to dewater the sample. Cryoprotectants (4% sucrose, 4% sorbitol, and 0.3% sodium tripolyphosphate) were added, and the mixture was adjusted to pH 7 with 5% NaHCO<sub>3</sub>(Kelleher and Hultin, 2000). Initial moisture was determined (AOAC, 1995), and the samples were blast frozen (-80 °C). The following morning the samples were tempered to 4 °C and equilibrated to 78% moisture using a jacketed Stephan UMC 5 silent cutter with vacuum. Each treatment was split into two groups, no NaCl (O) and 2% NaCl (N).

## Analysis of Raw Treatment

Proximate Analysis was performed for moisture, crude fat, crude protein, and ash (AOAC, 1995). Collagen and cholesterol were also run (AOAC, 1995). Differential scanning calorimetry (Fernandez-Martin et al 1997) and color (Minolta CR-300) were analyzed.

# Analysis of Cooked Gels

Remaining raw sample from each treatment was stuffed into two 21-mm cellulose casings, cooked in a 90 °C water bath for 30 min, and chilled overnight. Cook yield and water holding ability (WHA) were determined according to Daum-Thunberg, et al. (1992). Color was evaluated using a Minolta CR-300 and proximate analyses were run.

### Evaluation of Data

All tests were run in at least duplicate for all three replications of each experiment. The data for each experiment were analyzed for a completely randomized design using generalized least squares (PROC Mixed, SAS Institute, Cary, NC). The models included treatment and NaCl levels as main effects. The interactions between treatment and NaCl were included in the models. Mean separation was accomplished using Least Significant Difference.

### **Results and Discussion**

The application of Acid-SIP to each of the meat blocks resulted in products having significantly reduced fat and ash (Table 1). Collagen and cholesterol were also reduced, which demonstrated that the product had improved nutritional benefits. As expected, color was influenced by treatment as sarcoplasmic proteins (hemoglobin and myoglobin) were removed during the process. Therefore, treated samples had a higher L\* value (lighter) and a lower a\* value (not as red) compared to the control.

Table 1. Composition of raw, no NaCl treatments (dry wt basis)						
Sample <sup>1</sup>	Fat %	Ash %	Protein %	Collagen (g/100g)	Cholesterol (mg/100g)	
BH	$2.48 \pm 1.28$	$4.79 \pm .42$	$59.67 \pm 2.11$	$2.05 \pm .35$	$394.16 \pm 13.48$	
BH A-SIP	$.01 \pm .02$	$2.49 \pm .11$	$66.20\pm3.51$	.39 ± .41	$188.69\pm28.95$	
PH	$6.47 \pm 1.03$	$4.64 \pm .20$	$55.50\pm2.88$	$.36 \pm .02$	550.65 <sup>2</sup>	
PH A-SIP	<.01 <sup>3</sup>	$2.82 \pm .23$	$65.98 \pm 1.06$	N/A <sup>4</sup>	251.14 ± 41.89	
WGPPS	$31.58\pm2.36$	$10.30\pm2.64$	$54.23 \pm 2.70$	$1.13 \pm .12$	$198.91 \pm 26.89$	
WGPPS A-SIP	<.01 <sup>3</sup>	$2.96 \pm .19$	$64.43 \pm 2.28$	N/A <sup>4</sup>	$174.84 \pm 10.10$	

<sup>1</sup>BH: Beef Heart, Acid-SIP: Acid Solubilization Isoelectric Precipitation, PH: Pork Heart, WGPS: Whole Ground Pork Picnic Shoulder

<sup>2</sup>Reference value reported for raw pork heart by Agricultural Research Service (2001) in the USDA Nutrient Database

<sup>3</sup>Value too low to obtain accurate results

#### <sup>4</sup>Results not available

Acid-SIP had significantly higher cook yield and WHA compared to the control (Table 2). The addition of salt had minimal effect on the functional characteristics of protein recovered using the Acid-SIP process. In contrast, there was significant improvement in cook yield and WHA in the untreated control when salt was added. Improvement in protein functional characteristics such as cook yield and WHA in salt-treated controls was due to the salt's ability to create a protein surface matrix by opening up the myofibrillar proteins, which allows for improved

binding (Pearson and Gillett, 1996). However, the results from proteins recovered by the Acid-SIP process indicated the myofibrillar proteins are already in a state to bind. The binding ability of protein recovered by Acid-SIP without any added NaCl was comparable to salt-treated controls, and the addition of salt only minimally enhanced binding ability. The TPA results further demonstrated the improved gel attributes. It also revealed that the Acid-SIP process itself does not degrade the functionality of the proteins.

Table 2. Binding ability of gels					
Sample <sup>1</sup>	COOK YIELD %	WHA (g water/g protein)			
BH O	$67.52 \pm 8.16$	$1.17 \pm .22$			
BH N	87.85 ± 3.96	$1.59 \pm .31$			
BH Acid-SIP O	$99.57 \pm 3.71$	$2.18 \pm .49$			
BH Acid-SIP N	$98.17 \pm .81$	$2.40 \pm .44$			
PH O	$69.03 \pm 1.69$	N/A <sup>2</sup>			
PH N	85.41 ± 4.73	N/A <sup>2</sup>			
PH Acid-SIP O	99.78 ± 2.85	2.24 ± .53			
PH Acid-SIP N	97.45 ± 1.75	2.59 ± .55			
WGPPS O	$60.23 \pm .91$	N/A			
WGPPS N	$78.00 \pm 3.97$	N/A			
WGPPS Acid-SIP O	$92.94 \pm 3.20$	2.25 ± .37			
WGPPS Acid-SIP N	96.36 ± 1.35	2.49 ± .53			

<sup>1</sup>BH: Beef Heart, Acid-SIP: Acid Solubilization Isoelectric Precipitation, PH: Pork Heart, WGPPS: Whole Ground Pork Picnic Shoulder, O: No NaCl, N: 2% NaCl

#### <sup>2</sup>Results not available

Based on results, proteins obtained from the Acid-SIP process have improved nutritional and textural properties compared to the source from where they originated. In addition, the studies demonstrated that while salt plays a large role in gel formation of untreated controls, gel formation of protein recovered using the Acid-SIP process did not require salt. Further work is needed to determine if the proteins recovered from this process would be economical for use in formulation of lower fat, cholesterol, and salt foods or pet foods.

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