Food Chemistry 134 (2012) 1374–1384

Contents lists available at SciVerse ScienceDirect

Food Chemistry

journal homepage: www.elsevier.com/locate/foodchem

# Influence of oxidative browning inhibitors and isolation techniques on sweet potato protein recovery and composition

Lawrence A. Arogundade [a,b], Tai-Hua Mu [a,*]

[a] Food Chemistry Laboratory, Department of Fruit and Vegetable Processing, Key Laboratory of Agro-Product Processing and Quality Control, Chinese Academy of Agricultural Sciences, No. 2, Yuan Ming Yuan West Road, Haidian District, PO Box 5109, Beijing 100193, PR China
[b] Chemistry Department, College of Natural Sciences, Federal University of Agriculture, Box 28, UNAAB Post Office, Abeokuta, Ogun State, Nigeria

ARTICLE INFO

*Article history:*
Received 5 December 2011
Received in revised form 31 January 2012
Accepted 6 March 2012
Available online 19 March 2012

*Keywords:*
Sweet potato protein
Oxidative browning inhibitor
Protein recovery
Isolation technique
Chemical composition
Thermal stability

ABSTRACT

Effects of oxidative browning inhibitors on sweet potato protein (SPP) recovery and quality were studied. Oxidative browning inhibitors successfully decreased sweet potato oxidative browning, but reduced both SPP extractability and recovery. Ultrafiltration/diafiltration processed sweet potato (UDSP) protein (at pH 4, 6 and 7) showed significantly ($p < 0.05$) higher yield, purity, solubility, thermal stability and amino acid constituents than that of isoelectrically precipitated sweet potato (IPSP) protein (at pH 4). The yield of UDSP proteins was more than twice that of IPSP protein. Denaturation temperature ($T_d$), enthalpy change ($\Delta H$) and solubility (at pH 3 and 8) of UDSP proteins were in the ranges 82.89–90.29 °C, 6.34–11.35 (J/g) and 71.4–94.2%, respectively, while that of IPSP protein were 85.27 °C, 2.35 (J/g) 31.2% and 55.5%, respectively. Ratio of SPP essential amino acid to the total amino acid ratio ranged from 0.49 to 0.51. SPP *in vitro* digestibility and digestibility-corrected amino acid score (PDCAAS) ranged 70–80.7% and 44.79–51.08%, respectively.



## 1. Introduction

Sweet potato remains one of the worldwide cheap sources of raw material for starch industries. In China, the main industrial utilization of sweet potato is starch production, which goes with attendant wastewater generation problem. Apart from the economic burden of transporting this starch liquid waste to repository, it also constitutes ecological problem. Protein constitutes one of the major resourceful constituents of this starch liquid waste. In view of the high dependence of some starch industries on sweet potato, its ease of propagation, high crop yield and postharvest storage problem, exploiting sweet potato as an alternative protein source for human and animal feeding will give room for effective utilization of this starch wastewater and the tuberous root resource at large. Attempts have been made to produce protein from sweet potato or recover protein and other valuable constituents from starch waste-water (Cheng, Xu, & Wang, 2004; Mu, Tan, & Xue, 2009). Sweet potato protein is of comparable with or superior nutritional quality to most vegetable proteins (Mu et al., 2009), but its characteristic discolouration stigma constitutes a major setback in its utilization in the food system. This discolouration problem arises from enzymatic oxidative browning caused by polyphenol oxidase (PPO). PPO is a copper containing enzyme, which catalyzes the hydroxylation of certain phenols, especially mono and di-phenols in the *o*-position adjacent to an existing –OH group to *o*-diphenols, which further oxidises to *o*-quinones. These *o*-quinones condense and react non-enzymatically with amino acids and proteins, resulting in the formation of brown/dark melanin pigments (Severini, Baiano, De Pilli, Romaniello, & Derossi, 2003). These reactions are undesirable in food systems because of their negative effect on food appearance, development of off-flavours and losses in nutritional quality (Severini et al., 2003).

Surface treatment involving dipping fresh-cut, peeled sweet potato into aqueous solution containing antioxidants is one of the common methods of controlling food browning phenomenon. Examples of such antibrowning agents are sulphites, ascorbic acid and citric acid (Buta, Moline, Spaulding, & Wang, 1999; Sapers, 1993). These antibrowning agents inhibit oxidative browning through different mechanisms. For instance, antibrowning activity of sulphites occurs by inhibiting PPO, citric acid by decreasing pH of the aqueous medium below the range of PPO activity and by acting as chelating agent, while ascorbic acid controls browning through its ability to reduce the *o*-quinones back to their phenolic substrates (McEvily, Iyengar, & Otwell, 1992). These antibrowning agents may have negative or positive effect on extractable protein and are capable of co-extracting undesirable food components, such as phytic acid and trypsin inhibitor. Phytic acid has been associated with reduction in mineral and protein bioavailability through its mineral chelation and protein complexation tendencies, while the trypsin inhibitor reduces protein digestibility. These undesirable components must be removed or reduced in order to

* Corresponding author. Tel./fax: +86 10 62815541.
*E-mail addresses:* mutaihuacaas@126.com, mutaihua@126.com (T.-H. Mu).

0308-8146/$ - see front matter © 2012 Elsevier Ltd. All rights reserved.
http://dx.doi.org/10.1016/j.foodchem.2012.03.035

increase such protein's usefulness as food supplement and functional agent. Traditional protein processing techniques involving extraction, heat/acid coagulation of protein and separation by centrifugation partially solve these problems. Ultrafiltration membrane technology has been suggested as a good alternative method for producing better quality protein isolates or concentrates. Since most of these undesirable components are of smaller molecular weight than proteins, careful selection of membrane and operating parameters, can sieve them off.

Some studies on the use oxidative browning inhibitors on sweet potato to prevent darkening during processing have been reported (Ahmed, Akter, & Eun, 2010; Mu et al., 2009), but their effects on sweet potato protein recovery and composition have not been reported. Moreover, limited information is available on the relative merit of traditional (isoelectric) and membrane sweet potato protein processing techniques on its nutrition viability. Therefore the aims of this study were to determine the effect of antibrowning agents on sweet potato protein solubilization and recovery. In addition, the effects of protein isolation techniques on nutritional and antinutritional properties of sweet potato protein have been studied.

## 2. Materials and methods

### 2.1. Raw materials

*Long shu* (No. 9) cultivar of sweet potatoes (*Ipomoea batatas*) weighing about 25 kg were purchased from the retail outlet in Beijing, China and stored at room temperature for our immediate use. Phytic acid, pepsin, albumin bovine serum V, trypsin and Folin–Ciocalteu's phenol reagents were purchased from Beijing Biodes Biotechnology Co. Ltd. (Beijing, China). Gallic acid monohydrate and 5-sulfosalicylic acid dehydrate were purchased from Sinopharm Chemical Reagent Co. Ltd. (Shanghai, China). Pancreatin, *N*α-benzoyl-DL-arginine-4-nitroanilide hydrochloride and (±)-catechin were purchased from Sigma–Aldrich (St. Louis, Mo). All other chemicals used were of analytical grade.

### 2.2. Determination of sweet potato tuberous root crude protein and moisture content

The crude protein ($N\% \times 6.25$) content of fresh sweet potato tuberous root was determined by Kjeldahl method in triplicates, using Foss Tecator automatic protein analyser (2300 Kjeltec analyser unit) after digestion. The moisture content of the fresh tuberous root was determined by oven drying known weight of protein sample for 23 h, in triplicates.

### 2.3. Preparation of sweet potato protein extract

Sweet potato extracts used in the first phase of this study were prepared by grating 30 g of fresh peeled sweet potato tuberous roots into 150 ml of appropriate solvent or solution at room temperature (~25 °C). Whenever antibrowning solution was used as extractant, the grated sweet potato was left in the extractant for at least 20 min for proper equilibration and thereafter homogenised with Philip kitchen blender (350 W, setting 2), for 2 min at room temperature. The homogenate was then filtered through four layered cheese cloth and the filtrate centrifuged at 10,000*g* for 20 min at 15 °C. For each of the experiment, fresh extracts were prepared for the intended experiment. Before carrying out aqueous extraction of sweet potato protein in the presence of oxidative browning inhibitors, solubilization of the sweet potato protein in distilled water or alkaline aqueous medium was first carried out to determine the suitability of either medium for optimum sweet potato protein extractability. Extractable protein, determined by Kjeldahl method, showed that distilled water had same extractable protein as obtained with alkaline treatment. Thus distilled water containing appropriate oxidative browning inhibitor was thereafter used as extractant without pH adjustment in the subsequent experiments, i.e. oxidative browning inhibition, total phenolic and protein recovery assay.

### 2.4. Sweet potato oxidative browning inhibition assay

Effect of oxidative browning inhibitors on sweet potato protein browning indices was assessed on the protein extracts (Section 2.3). Solutions of sodium bisulphite, sodium metabisulphite, ascorbic acid or citric acid of different concentrations (0.01, 0.05, 0.1, 0.2 and 0.3 mol/l) were used as oxidative browning inhibitor. The browning indices of the various extracts were determined by the method of Youn and Choi (1996), after standing for 2 h at 420 nm, with UV–visible spectrophotometer (UV-1101 Techcomp, China), in triplicates. Distilled water was used as blank.

### 2.5. Determination of phenolics in the aqueous extract

Phenolics in the aqueous extracts were obtained in the presence of various oxidative browning inhibitors (Section 2.3) were determined by the spectrophotometric method, as described by Altunkaya and Gökmen (2008) with slight modification, using Folin–Ciocalteu's phenol reagent. Triplicate samples of 0.5 ml extract (clear supernatant) were taken at 2 h and mixed with 5 ml of 0.2 N Folin–Ciocalteu's phenol reagent. The mixture was allowed to react for about 3 min, 4 ml of saturated sodium carbonate (75 g/l) was added and the mixture incubated at 50 °C for 30 min. The absorbance was measured at 760 nm. Gallic acid (0.02–0.4 mg/ml) having linearity range with $r = 0.99$ was used as standard. The phenolic contents were calculated on the basis of gallic acid standard curve and expressed as mg gallic acid equivalent (GAE) per gram protein (dry weight basis).

### 2.6. Sweet potato protein recovery in the presence of oxidative browning inhibitors

Sweet potato protein extracts were obtained as indicated above (Section 2.3), with different antibrowning agent treatments. They were clarified at 5000*g* for 20 min and set aside in two portions for the determination of protein recovery by isoelectric precipitation and ultrafiltration techniques. The protein content of the first portion was isoelectrically precipitated at pH 4 by adjusting the extract pH to 4 with either 2 mol/l HCl, or NaOH, in triplicate and was then left for 1 h at room temperature. It was thereafter centrifuged at 10,000*g* for 20 min at 15 °C to obtain clear supernatant. Total protein in extract and soluble (*unprecipitated*) protein in the clear supernatants were determined by Lowry method using bovine serum albumin V as standard. Protein recovery was calculated as the percentage ratio of total protein in extract minus soluble protein to total protein in extract. Corrections were made for the volume of HCl and NaOH added during pH adjustment. Protein recovery by membrane method was done by ultrafiltering the second portion of the extract with a normal flow filtration membrane, equipped with molecular weight cut-off of 10,000 Da membrane at room temperature. The ultrafiltration was carried out under a nitrogen gas pressure of 0.16 ± 0.03 MPa. The soluble protein in the permeate was determined, in triplicates, by Lowry method and recovery calculated as percentage ratio of total extract protein, minus protein in permeate, to total protein in extract.