8-17-2013

Blanching Optimization and the Effect of Blanching on Functional Components of Yacon (Smallanthus Sonchifoulius) Root Slices

Yu-Ting Chen

Blanching optimization and the effect of blanching on functional components of yacon (*Smallanthus sonchifoulius*) root slices

By

Yu-Ting Chen

A Thesis
Submitted to the Faculty of
Mississippi State University
in Partial Fulfillment of the Requirements
for the Degree of Master of Science
in Food Science, Nutrition and Health Promotion
in the Department of Food Science, Nutrition and Health Promotion

Mississippi State, Mississippi

August 2013

Blanching optimization and the effect of blanching on functional components of yacon (*Smallanthus sonchifoulius*) root slices

By

Yu-Ting Chen

Approved:

Juan L. Silva Professor Food Science, Nutrition and Health Promotion (Major Professor)	Taejo Kim Assistant Research Professor Food Science, Nutrition and Health Promotion (Co-Major Professor)
Diane K. Tidwell Associate Professor Food Science, Nutrition and Health Promotion (Committee Member)	Frank B. Matta Professor Plant and Soil Sciences (Committee Member)
Zahur Z. Haque Professor Food Science, Nutrition and Health Promotion (Graduate Coordinator)	George M. Hopper Dean of the College of Agriculture and Life Sciences

Name: Yu-Ting Chen

Date of Degree: August 17, 2013

Institution: Mississippi State University

Major Field: Food Science, Nutrition and Health Promotion

Major Professor: Dr. Juan L. Silva

Title of Study: Blanching optimization and the effect of blanching on functional components of yacon (*Smallanthus sonchifoulius*) root slices

Pages in Study: 103

Candidate for Degree of Master of Science

Yacon (*Smallanthus sonchifolius*) root products are susceptible to oxidation, reduced quality and functional properties. The optimum water blanching process for yacon root slices was determined through a central composite design with variable temperature (80-100 degrees Celsius), blanching time (2-6 min), and citric acid concentration (0.04-0.20%). Phenolics and fructooligosaccharides of yacon slices were also evaluated after blanching. Yacon slices (3-4 mm) blanched at 90 degrees Celsius, 3.9 to 4.1min, and 0.05% to 0.07% citric acid showed the lowest polyphenol oxidase activity, highest whiteness value, and the highest sensory appearance scores. A second experiment showed that blanching at 100 degrees Celsius with 0.07% citric acid can maintain higher chlorogenic acid (3.52 mg/g more), inulin (5.41% more), and total sugar (34.9%) in yacon slices than blanching without citric acid. Thus, addition of less than 0.1% citric acid to boiling water can minimize loss of functional components of yacon slices during blanching.

Keywords: yacon, polyphenol oxidases (PPOs), inulin, chlorogenic acid

DEDICATION

To my parents,with love

ACKNOWLEDGEMENTS

My sincere appreciation goes to the following people throughout these years. I would like to express my deepest gratitude to my major advisor, Dr. Juan L. Silva, for his patient guidance, consideration and financial support during my study time at Mississippi State University. Sincere gratitude is also extended to my co-major professor, Dr. Taejo Kim, for his teaching, invaluable advice and encouragement. I greatly appreciate my committee members, Dr. Diane K. Tidwell and Dr. Frank B. Matta for their generous time in reviewing the thesis and advice.

Special thanks go to Dr. Rita Moraes for providing the idea, the materials, and her expertise, and Dr. Dja Shin Wang for his expertise and precious time for making this thesis possible. I also appreciate the support of current and past lab colleagues: Dr. Norman N. Arroyo, Weiwei Chen, Qian Gu, Yu-Chen Liao, Mengying Jiang, Amanda Pegg, Dr. Bang-Yuan Chen and Dr. Wei-Lien Weng for their friendship and generous assistance. Thank you all other faculty members and staff in the Department of Food Science, Nutrition and Health Promotion for their cooperation and friendship.

Further appreciation is extended to Dr. Sam Chang and Dr. William Gillis who built my confidence; to all my other friends, especially Jeng-Yang Wu, in Starkville for their love, support and wonderful friendship. Finally, my deepest thanks go to my parents Lin-Kuei Chen and Chin-Chu Tsai and for their endless love and support.

Yu-Ting Chen

TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

CHAPTER I

INTRODUCTION

Yacon is a native plant of the Andean countries in South America. The roots of the yacon plant are known for their sweet taste and are usually eaten raw as a fruit (FAO 2007). Yacon roots are similar to sweet potato roots but have a crunchier texture like nashi, which is also known as the Japanese pear or apple-pear. The yacon root contains nutraceutical properties, high-prebiotic fiber, and phenolic content. Up to 70–80% (dry weight) of the yacon root is composed of fructooligosaccharides (FOS), which are known to have high levels of prebiotic properties (Lachman and others 2003). In addition to medical treatments, dietary interventions have also been shown to be effective in treating insulin resistance and type 2 diabetes (Jenkins and others 2002). To reach a desirable weight and prevent/prolong the onset of diabetes, dietary intake and dietary components play an important role. The intake of a healthy diet that is rich in vegetables and fruits with FOS, such as yacon (*Smallanthus sonchifolius* Poepp. & Endl.), appears to have protective effects (Genta and others 2009). Fructooligosaccharides of inulin type β (2→1), which are mainly oligomers (GF2–GF16), are known for their ability to improve colon health because they are not absorbed in the gastric tract and are a rich source of probiotic microorganisms (Lachman and others 2003).

The yacon root is very susceptible to browning; thus, there is a need for inactivating enzymes. Blanching is to inactivate enzymes by special heat treatment: too

little heat is ineffective, while too much heat causes damages of fruits and vegetables (Bevilacqua and others 2004). Heat treatment at a higher temperature during a shorter period of time can avoid heat damage. For decay control, i.e., hot water rinsing, the temperature is managed at 48 °C to 65 °C for 10 to 25 seconds in combination with prochloraz treatment and fruit waxing for mango (Prusky and others 1999); for litchis, the temperature is managed at 55 °C for 20 seconds (Lichter and others 2000). However, according to previous experiments, these treatments will not work on yacon. The time and temperature of the treatments depend on the fruit, cultivar, cutting size, storage condition, and so on. Therefore, we used a higher temperature (84–100 °C) for a longer time (2.8–6.0 min) with citric acid concentration (0–0.2%) by central composite design (CCD) to optimize the blanching process for yacon slices.

Phenolic compounds produce antioxidant activity in order to protect cell membranes against damage from oxygen radicals. Yacon contains 3.8% phenolic compounds on a dry weight basis (Yan and others 1999). During peeling, when the cell membranes are ruptured, polyphenols and tannins mix with other components (cystoplasmic localizer enzymes) and cause an enzymatic oxidation which causes a bitter and mildly spicy flavor (Butler and Rivera 2004). In addition, green pigment formation occurs in foods such as yacon, sweet potato, and potato burdock as a result of the condensation reaction of two molecules of chlorogenic acid or acffeic acid ester with one molecule of a primary amino compound under aeration in alkaline solution (Yabuta and others 2001). Green pigmentation is also observed during alkali extraction of protein from sunflower meal due to its chlorogenic acid (Yabuta and others 2001; Sabir and others 1974). The oxidation of phenolic compounds through enzymes into o-quinones is the major reason for browning as it causes brown and black pigments. The chlorogenic

acid produces a green color when oxidation occurs or when the solutions of the acid were made slightly alkaline (Barnes and others 1950). Namiki and others (2001) report that the caffeic acid derivatives (such as chlorogenic acid) produce a semiquinone-type free radical; therefore, it also plays an important role in the defense system of plant tissue. The caffeate radicals may polymerize to form brown products with each other. There is also a trihydroxy benzacridine derivative that is reactive with oxygen and is also bound with an amino compound.

Citric acid has been noted for its inhibitory effect on PPO activity through chelation of the copper at the active site of the PPO, making it an anti-browning agent in minimally processed fruits and vegetables (Altunkaya and others 2009; Ahvenaien 1996). Citric acid is a weak organic acid, which is a natural preservative and anti-browning compound. The low pH dip during food processing does not cause changes in flavor like that which occurs in carrots (Altunkaya and others 2009; Boun and others 1991). Acidification of lettuce through citric acid also appeared to stabilize phenolic compounds (Altunkaya and others 2009). Thus, citric acid has been considered as both an anti-browning and antioxidant agent (Wang and others 2003). It can be used to blanch and deactivate the polyphenol oxidase (PPO) activity of the roots with some chemicals.

The objectives of this study were to: 1) determine the optimum water blanching process (time, temperature, and anti-browning agent) for yacon root slices; and 2) compare the effect of the blanching water treatment with citric acid on the quality and functional components of yacon slices.

CHAPTER II

LITERATURE REVIEW

Yacon

Yacon (*Smallanthus sonchifolius* [Poepp.&Endl.] H. Robinson) is a native plant of the Andean countries in South America. It is an herbaceous and dicotyledonous plant with dark-green celery-like leaves that grows to 1.5–2.5 m in height (Manrique and others 2004). The roots of the yacon plant can weigh between 100 g and 1 kg and come in both spherical and oblong shapes. Yacon mainly stores water (860–900 g kg -1) and carbohydrates (90–130 g kg -1) (Hermann and others 1999). Most tubers weigh between 200–500 g and are a light yellow color on the inside and brown on the outside. The flesh color varies by genotype as different genotypes produce different concentrations of yellow pigmentation (Hermann and others 1999). Yacon roots are similar to sweet potato roots but have a crunchier texture like nashi, which is also known as the Japanese pear or apple-pear. The sweet roots of the yacon are usually eaten raw as a fruit. The best soil for growing yacon is high in organic matter with a neutral to slightly acidic pH (Manrique and others 2004).

Yacon belongs to the Asteraceae family along with 21 other *Smallanthus* species (Polreich 2003). Seven *Smallanthus* species are found in Peru, from which yacon is the only domesticated species (Polreich 2003; Brako and Zarucchi 1993). The yacon produces two underground tubers, reddish rhizomes, and large brown roots. The reddish

rhizomes are mainly used for propagation and the larger brown root is often used as food. Researchers have reported significant differences in tuber shape and weight, as well as in the content of FOS and other sugars for various yacon cultivars under field conditions (Hermann and others 1999; Valentova and others 2006). The yacon has been cultivated in the Andean region, Brazil, New Zealand, Japan, Taiwan, and other countries (Genta and others 2005). In South America, yacon tubers can have yellow, orange, red, pink, and even purple flesh with a thin resinous brown, purple, or red bark (Butler and Rivera 2004). However, only a few white-flesh varieties of yacon are available in the U.S. (Weaver 2006). Because of the crunchy texture of the yacon root, South Americans often put it in fruit salads called *salpicón*. Yacon roots can also can be stir-fried, roasted, baked, or made into pies and healthy chips (Weaver 2006).

According to the Centers for Disease Control and Prevention (CDC) (2011), the medical care costs associated with obesity in the U.S. (among adults) were estimated to be as high as $147 billion in 2008. However, FOS can help decrease both the obesity rate and the level of sugar intake by people with diabetes since FOS can be used in place of sugar. In addition to being sweet, the yacon also has a high nutritional value, including nutrients such as FOS and calcium. The juicy and crunchy texture of raw yacon means that it can be served as a fruit. It can also be cooked to increase its sweetness. In addition, the antioxidant phenolic compounds in the yacon make it a functional food.

Antioxidant capacity of phenolics and polyphenol oxidase (PPO) activity in yacon

In a study conducted by Sun and others (2002), cranberries exhibited the highest total antioxidant activity (177.0 ± 4.3 μmol of vitamin C equiv/g of fruit), followed by apples, red grapes, strawberries, peaches, lemons, pears, bananas, oranges, grapefruits,

and pineapples. The sub-layer under the bark of the yacon root (which is 1–2 mm thick) is rich in tannins and polyphenols, including chlorgenic, caffeic, ferric and gallic acid, and quercetin; these account for ~ 200 mg/100 g of the weight of the yacon root (Butler and Rivera 2004; Manrique and others 2004). During peeling, when the cell membranes are ruptured, polyphenols and tannins are mixed with other components (cystoplasmic localizer enzymes such as PPO), which causes enzymatic oxidation. This oxidation produces a bitter and mildly spicy flavor (Butler and Rivera 2004).

PPO is a plastidic enzyme that exists in a latent form in the thylakoid membrane and is not involved in the synthesis of phenolic compounds (Toivonen and Brummell 2008). PPO plays an important role in the beneficial coloration of some fruits and vegetables, such as prunes, dark raisins and teas (Whitaker and others 1995). PPO is the most damaging of enzymes in degradation of color, such as browning and anthocyanins degradation in blueberries, strawberries, plums, and grapes (Whitaker and others 1995; Jiang 2000). The degradation of anthocyanins by PPO increases rapidly when there is an increase in the phenol concentration (Jiang 2000). Yacon flesh contains chlorogenic acid, ferulic acid, and caffeic acid (Simonovska and others 2003). The oxidation of phenolic compounds through enzyme into o-quinones is the major cause of the browning effect. In lychee, although PPO cannot oxidize the degradation of anthocyanina, phenolic extracts stimulate the pigment degradation caused by PPO which results in brown and black pigments (Jiang 2000). The browning rate of anthocyanin degradation is stimulated by chlorogenic acid in blueberries (Kader and others 1997). Yacon enzymatic browning reactions happen after peeling or cutting because of the PPOs (Yoshida and others 2002). The degree of browning depends on the PPO, the amount of endogenous phenolic compounds, the presence of oxygen, the reducing substances, metallic ions, pH, and

temperature (Yoruk and Marshall 2003; Neves and Da Silva 2007). Furthermore, chlorogenic acid causes the green color when oxidation occurs or when solutions of the acid are made slightly alkaline (Barnes and others 1950).

Browning can be inhibited by blanching, as is the case with whole Russet Burbank potatoes. They can be blanched up to 60 min at 50 °C, and the PPO activity ranges from 1.58 ΔABS/min to 0.7 ΔABS/min (Yemenicioglu 2002). The PPO half-life values of various apple cultivars include 25.6–91.2 min at 68 °C and 2.4–4.3 min at 78 °C (Yemenicioglu and others 1997; Unal 2007). The PPO half-life value of a mango kernel is blanching for 18.8 min at 60 °C or 8.5 min at 70 °C (Arogba and others 1998; Unal 2007). The PPO half-life value of Ravat grapes are 4.5 min at 75 °C and 31.6 min at 75 °C for Niagara grapes (Wissemann and Lee 1981; Unal 2007).

Heath benefits of yacon – phenolics, fructooligosaccharides, and inulin

Phenolic compounds contain an aromatic ring with one or more hydroxyl substituents, although a more precise definition is based on metabolic substances (Antolovich and others 2000). These phenolic compounds have antioxidant properties which are reactive with oxygen and help fight against degenerative diseases (Dykes and Rooney 2007). Yacon contains water, carbonhydrates (including fructose, glucose, and sucrose), phenolic compounds, vitamin A, postassium, small amounts of fat, free amino acids such as L-tryptophan, and carotenoids such as carotene and neurosporene (Granato and others 2011; Quinteros 2000; Ohyama and others 1990). Phenolics are present in a wide variety of fruits and vegetables. In apples, the concentration of free phenolics varies from 8 to 45 μg g^{-1}, while the concentration of bound phenolics varies from 50 to 110 μg g^{-1} (Ju and Bramlage 1999). There are 670.9 mg gallic acid equivalent (GAE)/ 100 g in

blueberries, 432.0 mg GAE/100 g in dogwood berries, and 429.5 mg GAE/ 100 g in sour cherries (Marinova and others 2005). In cherries, plums, and raspberries, total phenolics range from 245.7 to 398.5 mg GAE; the antioxidant capacity of these fruits range from 354.8 to 692.3 mg /100 g vitamin C equivalent antioxidant capacity (Kim and Padilla-Zakour 2004). Phenolic compounds have antioxidative activities which help protect cell membranes against damage from oxygen radicals. Yacon contains 3.8% phenolic compounds on a dry weight basis (Yan and others 1999). The main antioxidant activity and phenolic compound in yacon roots is chlorogenic acid (Park and others 2009). Chlorogenic acid is the major cinnamic derivative found in blueberries, but it increases the rate of anthocyanin degradation (Kader and others 1997). Among the components in yacon, the beneficial components of yacon are phenolic compounds and FOS which have antioxidants, antidiabetics, and prebiotics (Ojansivu and others 2011; Campos and others 2012).

It is widely known that oligosaccharides are FOS and inulin. According to previous research studies, inulin is a fructose polymer consisting of 8 to 10 fructose units with a terminal glucose unit (Roberfroid and Slavin 2000; Stewart and others 2008). Chains with 2 to 10 fructose units that are produced by hydrolysis of inulin using endoinulinase are categorized as oligofructose or FOS (Stewart and others 2008; Niness 1999). In other studies, inulin and oligofructose are fructans with a degree of polymerization ranging from 2 to 60 and 2 to 20, respectively (Roberfroid 1993). However, inulin and FOS appear in a wide variety of fruits and vegetables, including pineapples, mangoes, bananas, plums, onions, shallots, chicories, and artichokes. FOS can be described as a caloric prebiotic (Lhomme and others 2003; Renuka and others 2009). There are different amounts of FOS in a fully ripe banana, ranging between 297 to

1600 µg/g of dry matter (Agopian and others 2008); in the dry root of yacon it ranges between 22–77 g/100g (Hermann and others 1999). In addition to being a prebiotic, FOS also provides the sweetness in the yacon root.

As the preceding data illustrates, the yacon provides many health benefits to consumers. In a previous study, yacon FOS were selectively fermented and promoted the growth of bifidobacteria and lactobacilli (Campos and others 2012). A diet rich in yacon FOS enhances short chain fatty acids in the cecal material and increases cell density and crypt formation in caecum tissue (Campos and others 2012; Pedreschi and others 2003). Therefore, studies have demonstrated that the intake of yacon can enhance the colon health of the consumer.

Obesity increases the risk of coronary heart disease, high blood pressure, type 2 diabetes, high total cholesterol, and many other diseases. More than 33% of adults and 17% of children in the U.S. are obese (CDC, 2011). Yacon consumption is recommended to be included in the diets of both obese people and people with diabetes because the fructose in yacon is not absorbed by the human small intestine (Hallfrish 1990). Moreover, the fructose in yakon is sweeter, more soluble, and less glucoenic than glucose and sucrose (Hallfrish 1990). The inulin types FOS are non-reducing sugars that are present in wheat, barley, onions, leeks, garlic, asparagus, and bananas (Van Loo and others 1995). Inulin is also present in Jerusalem artichokes and chicory (Campos and others 2012). Yacon has a high concentration (21.17 ± 0.83% d.m.) of non-reducing sugars (Hermann and others 1999; Scher and others 2009). The prebiotic FOS-inulin has the ability to modulate the growth of bacteria in the gastrointestinal tract and modulate the innate immune system (Lomax and Calder 2009). The FOS-inulin can also enhance the killing of *Salmonella enteritidis* (SE) and decrease inflammasome activation by

increasing the macrophage population (Babu and others 2012). The intake of yacon flour through the femoral midshaft of Wistar rats fed ad libitum was shown to stimulate the intestinal absorption of calcium and mineral retention, and to strengthen the structural properties of bones (Lobo 2007).

Yacon preservation and blanching

Although yacon is an easy crop to grow, it is not an easy food to preserve. Spoilage and browning are both preservation problems associated with the yacon root and yacon slices. During peeling, when the cell membranes are ruptured, polyphenols and tannins are mixed with other components (cystoplasmic localizer enzymes), causing enzymatic oxidation (Butler and Riverera 2004). The color of food is important to customers as it often reflects the quality of the food. For example, in post-harvest and processing, enzymatic browning of yacon causes quality losses (Neves and Da Silva 2007). Blanching is to inactivate enzymes by special heat treatment: too little heat is ineffective, while too much heat causes damages of fruits and vegetables (Bevilacqua and others 2004). Heat treatments at high temperatures and short times can decrease heat damage. Scher and others (2012) showed the color changes included L^*, a^*, b^*, hue, chrome, and total color difference (TCD) values in 100°C steam blanching yacon for 1, 2, 4, 6, 8 and 10 min. For decay control, the treatment used involves hot water rinsing ranging from 48 °C to 65 °C for 10 to 25 s (Prusky and others 1999). However, the time and temperature of treatments depends on the fruit, cultivar, cutting size, storage condition, and other factors. The optimum conditions for water blanching to retain color were 80 °C, 3 min with 0.25% citric acid (Xin and others 2010). Steam blanching 1.75 ±0.35 mm yacon slices at 100 °C for 4 min by autoclave shows the best condition (Fante

and others 2011). Butler and Rivera (2004) used boiling water to remove the yacon skin through the process of blanching. The PPO in both Bosc and Red pears was completely inactivated at 75°C for 30 min (Siddiq and others 1993). Heat treatment of pre-cut peach slices at 50°C for 10 min significantly prolonged their shelf-life (Koukounaras and others 2008). If the temperature of hot water is not high enough, it causes browning quicker than the un-blanched yacon slices because the temperature can increase the PPO activity and promote chlorogenic acid oxidation (Butler and Rivera 2004). In addition, blanching is a unit operation for elimination of air and deactivates enzymes that are present in plant tissue (Gonazlez-Martinez and others 2004) and is widely used in the processing industry.

Central composite design

The central composite design (CCD) is an experimental design for studying the interactive effects of three or more operating variables with a minimum number of experiments. The CCD is both a widely used design framework for response surface methodology and a building model for the response variables (Nasirizadeh and others 2012). For a three-factor response surface methods study, an orthogonal CCD would require 20 runs (Gardiner and Gettinby 1998). A CCD was used to study the effect of ozone treatment for acid dye effluents and to optimize the variables which influence the efficiency of the color of dye effluents, such as salt concentration, pH, and time (Muthukumar and others 2004). A CCD was also used to study the effect on enzyme inactivation in strawberry and orange products and to optimize the variables, such as peroxidase (POD), PPO, and pectin methylesterase (PME) activities under high hydrostatic pressure treatment (50–400 MPa) combined with heat treatment (20–60 °C)

(Cano and others 1996). Response surface analysis predicted that the crispiest banana chips should be produced by blanching at 69 °C for 22 min (Jackson and others 1996).

CHAPTER III

MATERIALS AND METHODS

Experiment I: Optimization of the Blanching Process for Yacon (*Smallanthus Sonchifolius*) Root Slices

Yacon Samples

Fresh yacon (*Smallanthus sonchifolius*) roots were grown and harvested at the University of Mississippi, Oxford, MS, USA (provided by Dr. Rita Maria Moraes), and transported to the Food Science, Nutrition, and Health Promotion Department of Mississippi State University, Oktibbeha County, MS, USA, within four hours. Yacon is a root of Ecuadorian origin with brown skin and yellowish flesh, measuring around 15 cm × 6 cm × 6 cm. The roots were cleaned and selected considering a uniform size and the absence of visible injury and infection, and stored at room temperature (25°C) for one week. When ready to be processed, each yacon root was cut into 3–5 mm slices with a knife and randomly selected for the different treatments and experiments (Jeong and others 2008; Scher and others 2009).

Treatments (Blanching)

The experiment was designed according to a central composite design (CCD). For each treatment, three slices of yacon (approx. 15 g) were blanched in a water bath (1 L). The treatments were chosen based on the CCD with three variables, temperature (X_1, 84–100°C), time (X_2, 2.8–6.0 min), and citric acid concentration (X_3, 0–0.2%) (Table 3.1).

The citric acid was purchased from Fisher Scientific Inc., (Pittsburgh, PA, USA). Blanched slices were blot-dried with a napkin and stored in plastic plates in the dark at 4°C for one week. The samples were analyzed for polyphenol oxidase (PPO) activity, color (whiteness and total color difference (TCD)), and image sensory appearance (color score and browning percentage) after 2, 6, and 10 days of storage.

Yacon Polyphenol Oxidase Activity

Blanched yacon slices were analyzed for PPO activity after 2, 6, and 10 days of storage according to the method by Neves and Da Silva (2007). One yacon slice (5 g) was homogenized (Polytron® Brinkmann Homogenizer, Switzerland) with 10 mL of 0.05 M phosphate buffer (pH 6.0) (Sigma-Aldrich Inc. St. Louis, MO, USA). The homogenate was filtered through a 0.45 µm syringe filter (Millipore, Bedford, MA, USA). The PPO-active fraction (supernatant) was collected and used for further analysis.

PPO produces o-quinine compounds that turn into brown pigments after reacting with catechol (Sigma-Aldrich Inc. St. Louis, MO, USA). The reaction mixture contained 0.2 mL of PPO extracts (supernatant) and 1.8 mL of 0.1 M catechol solution in 0.1 M phosphate buffer, pH 7.0. The mixture was incubated at 37°C for 30 min. The blank contained 2 mL of substrate solution. PPO activity was measured as the rate of increase in absorbance at 420 nm on a Spectronic Genesys 5 UV-Vis Spectrophotometer (Fisher Scientific Inc., Pittsburgh, PA, USA), as reported by Kumar and others (2008). The linear portion of the activity curve was used to express PPO enzyme activity (Wuyts and others 2006). One unit of PPO activity was defined as the amount of enzyme causing a change in absorbance of 0.001 min^{-1} (Galeazzi and others 1981).

Hunter Color

Color (Hunter 'L', 'a', and 'b') were measured using a Hunter Color Labscan (Labscan 60,000°/45° Spectrocolorimeter, LabScan SpecWare V1.01 software, HunterLab, Reston, VA, USA). Color was measured on one yacon slice for each treatment. Higher 'L' values (up to 100) correspond to white and low 'L' values (down to zero) correspond to black; positive 'a' values correspond to red and negative 'a' values correspond to green; positive 'b' values correspond to yellow and negative 'b' values correspond to blue (CIE, 1986). Numerical values of 'L', 'a', and 'b' were converted into hue angle (H° = tan-1 (b/a)), chroma or color intensity (chroma = $(a^2 + b^2)^{1/2}$) (Ozoglu and Bayind 2002), whiteness index (WI) (WI = 100-[(100-L∗)2+a∗2+b∗2]1/2) (Hsu and others 2003), and TCD (∆E = [(L-L0)² +(a-a0)²+(b-b0)²)]1/2) (Hill and others 1997); where Lo, ao, and bo corresponded to fresh raw yacon slices. There is a linear correlation between image quality and naturalness of the product color, and both deteriorate as soon as hues start to deviate from those in the original image (Ridder and others 1995). In addition, chroma variation affects the impression of quality and natural colors less than hue variation (Ridder and others 1995). Cortes and others (1999) analyzed 'L', hue, chroma, and total color difference for different treatments in banana slices, and found that the conditions when analyzing color changes were the same in comparison to color parameters of raw ripe fruit chips.

Digital Image Sensory Analysis

Yacon slice images were captured through a digital IXUS 60 camera (Cannon Corporation, Tokyo, Japan) mounted on an adjustable stand positioned 10 cm above a white cardboard base where the yacon slices were placed to avoid shadow and glare

(Manzocco and others 2012). The camera settings were set to shutter time at 1/40 s, F-number at f/2.8, and focal length at 6 mm. The images were saved in a .jpeg file format and pictures were 2816 × 2112 pixels, 180 dpi. The digital image samples were labeled with 3-digit random codes and shown to trained participants through a computer for sensory appearance rating: color scores and color browning percentage. The 13 panelists were asked to practice visual assessment of the image samples during training, before the experiment commenced. The attention was focused on color and browning percentage, since browning of yacon slices is an early indicator of quality decay. Color scores (Figure A.1) ranged from 1 to 5 according to browning intensity (1- dark brown; 2- red brown; 3- medium brown; 4- brown; 5- yellowish, original color) to correspond to the general color of the yacon slices (Figure A.1). During training, examples of each sensory color score of yacon slice images were shown to the 13 panelists. Sensory browning percentage was also scored for each yacon slice image; scores lower than four were termed brown. A number between 0%, 25%, 50%, 75%, and 100% sensory browning percentage was chosen on the score sheet for each yacon slice observed through the images (Figure A.1). Examples of these five kinds of sensory browning percentage for yacon slice images were also shown to the 13 panelists during training (Figure A.1). The percentage of browning was the percentage ratio between the sum of pixels of yacon slices in the brown color range and the overall sum of pixels relevant to yacon slices (Manzocco and others 2012).

Experimental Design and Statistical Analysis

The CCD was used to investigate the significance of temperature, time, and citric acid concentration on yacon blanching. PPO activity, color change (whiteness and TCD),

and digital image sensory ratings (browning intensity and browning percentage) for each treatment were analyzed statistically by a CCD. A three-factor, five-level CCD leading to 20 runs was employed for the optimization of the blanching process of yacon root slices. The three independent factors were temperature (X1), time (X2) and citric acid concentration (X3). Each independent variable had five levels, -1.682, -1, 0, 1, and 1.682 (Table 3.1). The results were analyzed to generate three-dimensional response surface graphs and contour plots with Minitab 5.1 (Minitab Inc., State College, PA, USA).

Experiment II: Phenolic Compounds and Fructooligosaccharides Content of Yacon (*Smallanthus Sonchifolius*) Root Slices after Blanching

Chemicals and Reagents

Chlorogenic acid (95% purity), inulin, glucose, fructose, sucrose, and Folin-Ciocalteu's phenol reagent were purchased from Sigma (St. Louis, MO, USA). Sulfuric acid, sodium carbonate, methanol, and high-performance liquid chromatography (HPLC) grade water were purchased from Fisher Scientific (Pittsburgh, PA, USA), unless otherwise noted.

Sample Preparation

The yacon root variety "Sweet Crisp" was purchased from Sacred Succulents (Sebastopol, CA, USA). The roots were washed and blot dried at room temperature, peeled and cut into 3-4 mm slices, and then blanched in boiling water with or without 0.07% citric acid. Hunter color, PPO activity, sugars (including inulin), and total phenolic content (including chlorogenic acid) were measured.

Yacon extraction and polyphenol oxidase (PPO) activity

As described in experiment I, one yacon slice (5 g) was homogenized (Polytron® Brinkmann Homogenizer, Switzerland) with 10 mL of 0.05 M phosphate buffer (pH 6.0). The homogenate was filtered through a 0.45 µm syringe filter (Millipore, Bedford, MA, USA). The PPO-active fractions (supernatant) were collected and used for further analysis.

PPO produces o-quinine compounds that turn into brown pigments after reacting with catechol. The reaction mixture containing 0.2 mL of PPO extracts (supernatant) and 1.8 mL of 0.1 M catechol solution in 0.1 M phosphate buffer, pH 7.0. The mixture was incubated at 37°C for 30 min. The blank contained 2 mL of substrate solution. PPO activity was measured as the rate of increase in absorbance at 420 nm on a Spectronic Genesys 5 UV-Vis Spectrophotometer (Fisher Scientific Inc., Pittsburgh, PA, USA), as reported by Kumar and others (2008). The linear portion of the activity curve was used to express PPO enzyme activity (Wuyts and others 2006). One unit of PPO activity was defined as the amount of enzyme causing a change in absorbance of 0.001 min-1 (Galeazzi and others 1981).

Hunter Color

Color (Hunter L, a and b) were measured using a Hunter Color Labscan (Labscan 60000°/45° Spectrocolorimeter, LabScaan SpecWare V1.01 software). Color was measured on one yacon slice for each treatment. Higher 'L' values (up to 100) correspond to white and low values (down to zero) correspond to black; positive 'a' values correspond to red and negative values correspond to green; positive 'b' values correspond to yellow, and negative values correspond to blue (CIE, 1986). Numerical values of 'L', 'a' and 'b' were convered into hue angle (H°= tan-1 (b/a)), chroma or color

intensity (chroma = $(a^2 + b^2)^{1/2}$) (Ozoglu and Bayind 2002), whiteness index (WI) (WI = 100-[(100-L∗)2+a∗2+b∗2]1/2 (Hsu and others 2003), and total color difference (∆E = [(L-L0)² +(a-a0)²+(b-b0)²)]1/2 (Hill and others 1997) , the Lo, ao, bo are correspond to fresh raw yacon slices. A linear relation was found between image quality and naturalness of the product color, and both of them deteriorate as soon as hues start to deviate from the ones in original image (Ridder and others 1995). In addition, chroma variation affected the impression of quality and natural colors to a lesser extent than did hue variation (Ridder and others 1995). Cortes and others (1999) analyzed L, hue, chroma and total color difference for different treatments in banana slices. They reported the conditions when analyzing color changes were not different in comparison to color parameters of raw ripe fruit chips (Cortes and others 1999).

Phenolic Compounds and Sugars

Extraction of phenolic compounds and sugars was performed according to the method by Campos et al. (2012), with some modifications. One slice of yacon (5 g) was homogenized (Polytron® Brinkmann Homogenizer, Switzerland) in 10 mL of acidified 80% methanol (v/v) and vortexed for 30 seconds. After 60 min incubation in a rotary mixer (Dynal® Inc., NY, USA) at 200 rpm, the mixture was centrifuged at 6,000 g for 10 min (Eppendorf centrifuge 5415C; Brinkmann Instruments, NY, USA) and the supernatant was obtained.

Total phenolic content in yacon was determined according to the Folin-Ciocalteu method (Waterhouse 2001) using a standard curve of chlorogenic acid (Figure A.2) (Singleton 1965). The results show the units in part per million (ppm). Twenty microliters of each yacon extract were diluted with 1,580 µL deionized water and mixed

with 100 µL of Folin-Ciocalteu's phenol reagent. After 2 min of reaction, 300 µL of sodium carbonate solution was added and let stand for 2 hours. The absorbance of the sample was measured at 765 nm using a Spectronic Genesys 5 UV-Vis Spectrophotometer (Fisher Scientific Inc, Pittsburgh, PA). Measurements for each extract were done in triplicate and mean values were recorded.

A reversed-phase HPLC column was used for separation and quantification of major phenolic compounds. Each extract sample was mixed with 4 N HCl at 1:9 (v/v) and placed in a water bath at 95°C for one hour to separate the free phenolic compounds from their conjugated forms (Lee 2000). The acid hydrolyzed samples were cooled for 30 min to 25°C, and were centrifuged at 12,000 rpm (17,000 g) for 5 min in an Eppendorf 5414 microcentrifuge (Eppendorf, Brinkmann Instruments, NY, USA). The supernatant of each sample was filtered through a 0.45 µm syringe filter (Millipore, Bedford, MA), and individual phenolic compounds were separated using a Gemini C18 column (250×4.6 mm, Phenomenex®, Torrance, CA, USA) at 40°C in an Agilent HPLC 1100 (Agilent Technologies Inc., Santa Clara, CA, USA) equipped with a diode array detector. The two mobile phases were 80% acetonitrile (solvent A) and 20% 0.01 N H2SO4 (solvent B) with a flow of 0.6 mL/min; 25 µL of the sample was injected. A linear gradient to separate phenolic compounds was used as follows: at 0 min, 5% solvent A, 95% solvent B; at 1 min 10% solvent A, 90% solvent B; at 30 min, 70% solvent A, 30% solvent B; at 31 min, 10% solvent A, 90% solvent B; at 32 min, 5% solvent A, 95% solvent B with a 5 min post-run (Kim and others 2009).

Individual phenolic compounds were detected at 320 nm. Each sample and the standard were analyzed in duplicate. Peaks for individual phenolic compounds were integrated and analyzed through ChemStation software (Agilent Technologies Inc.). The

column was calibrated for 5 min between injections with the mobile phase. Individual phenolic compounds were identified and quantified through comparison of their retention time and peak area to the standards. All the results were expressed as milligrams of chlorogenic acid equivalent per milliliter of yacon extract sample, using a standard curve generated with 50, 100, 250, and 500 ppm of chlorogenic acid (Figure A.3).

To separate inulin, sucrose, glucose, and fructose, the yacon extract was centrifuged at 16,000 g for 5 min (Eppendorf, Brinkmann Instruments, NY, USA). Each supernatant was filtered through a 0.45 μm syringe filter (Millipore, Bedford, MA). Sugars were separated using a HPLC ion exclusion column (300 × 7.8 mm, Aminex®, Richmond, CA, USA) at 40°C in an Agilent HPLC 1100 (Agilent Technologies Inc., Santa Clara, CA). The Agilent HPLC was equipped with a refractive index signal (RID) detector (Agilent Technologies Inc., Santa Clara, CA). The mobile phase was 0.01N H2SO4 with a flow rate of 6 mL/min. Peaks for inulin, glucose, fructose, and sucrose were analyzed through ChemStation software (Agilent Technologies). Sugars were identified based on the retention time of the standards and quantified through calibration curves built from standards (Kim and others 2009). All the results of inulin, sucrose, glucose, and fructose were expressed as g per 100 g of extract, using a standard curve generated with 0.5, 1, 1.5, and 2 g per 100 g of each sugar (Figures A.4 to A.7).

Table 3.1 Five-level central composite design (CCD) used in this experiment.

Factor	Code	-1.682	-1	0	1	1.682
Boiling water temperature (°C)	X_1	80	84	90	96	100
Time (min)	X_2	2.0	2.8	4.0	5.2	6.0
Citric acid concentration (%)	X_3	0.00	0.04	0.10	0.16	0.20

CHAPTER IV

RESULTS AND DISCUSSION

Experiment I: Optimization of the Blanching Process for Yacon (*Smallanthus Sonchifolius*) Root Slices

Statistical Results of Experimental Design

Experimental data from the CCD (Table A.1) were analyzed and fit to a polynomial regression model. The adequacy of the model was evaluated by lack of fit, coefficient of determination (R2), and Fisher's test ratio (F-value) obtained from the analysis of variance generated by Minitab 5.1 (Minitab Inc, State College, PA). Three-dimensional response surface plots and contour plots were generated by keeping one response variable at its optimal level and plotting it against the remaining factors. The effect of independent variables on each of the response variables is shown in Table 4.1. Response surface and contour plots were generated to show the effects of blanching temperature, time, and citric acid concentration on each of the responses (Figures 4.1 to 4.15).

Effects of Process Variables on Polyphenol Oxidase (PPO) Activity

The 20 experimental runs proposed by the CCD, with the three factors (X1 = temperature, X2 = time, and X3= citric acid concentration), five levels (Table 3.1), and six replicates at the centered point were used for fitting a second-order response surface

model. The model obtained for PPO activity of processed yacon slices as affected by the three variables was:

$$\begin{aligned}\text{PPO activity} = {} & (1.1849 \pm 0.228229) - [(0.0213 \pm 0.004858) * X_1] - \\ & [(0.0622 \pm 0.016843 * X_2)] - [(0.2969 \pm 0.329689) * X_3] + \\ & [(0.0001 \pm 0.000027) * X_1 * X_1] + [(0.0056 \pm 0.000665) * X_2 * X_2] + \\ & [(0.2559 \pm 0.266169) * X_3 * X_3] + [(0.0001 \pm 0.000176) * X_1 * X_2] + \\ & [(0.0032 \pm 0.003525) * X_1 * X_3] - [(0.0312 \pm 0.017623) * X_2 * X_3]\end{aligned} \quad (4.1)$$

Analysis of variance (ANOVA) was conducted to determine the significance ($P < 0.05$) of the model. The variance was partitioned into linear (X1, X2, and X3), quadratic (X1*X1, X2*X2, and X3*X3), and interaction (X1*X2, X1*X3, and X2*X3) components to assess the adequacy of the second-order polynomial function and the relative significance of the terms for each variable. This model explained 84.4% (R2 of 0.844) of the variation in PPO activity.

The effect of temperature, time, and citric acid concentration on PPO activity is depicted by response surface and contour plots (Figures 4.1 to 4.3). In the response surface plot, when citric acid was held constant at 0.1%, PPO activity as affected ($P < 0.05$) by blanching time more than blanching temperature (Table A.2 and Figure 4.1). In the contour plot, when citric acid was held constant at 0.1%, the lowest PPO activity was at approx. 93°C for 5.0 min (Figure 4.1). In the response surface plot, when blanching time was held constant at 4.0 min, PPO activity as affected by citric acid concentration more than as affected by blanching time (Figure 4.2). In the contour plot, when blanching time was held constant at 4.0 min, the lowest PPO activity was at 95°C with 0.3% citric acid (Figure 4.2). In the response surface plot, when blanching temperature

was held constant at 90°C, PPO activity as affected by citric acid concentration more than as affected by blanching time (Figure 4.3). In the contour plots, the lowest PPO activity was at approx. 5.0 min with 0.2% citric acid concentration (Figure 4.3). Phenolic endogenous compounds oxidize to unstable o-quinones that are later polymerized to brown pigments; this is the primary reason for browning (Yoruk and Marshell 2003). In Table A.1, the control, unblanched yacon slices showed the highest PPO value (0.18). Longer time (run 12; 90°C, 6.0 min, 0.1%) and higher temperature (run 10; 100°C, 4.0 min at 0.1% citric acid concentration) showed the lowest PPO activity. However, citric acid concentration higher than 0.1% did not affect PPO activity (P >0.05). Thus, the optimization of the blanching process for yacon root slices was obtained by looking at response surface data and contour plots for PPO activity. The lowest PPO activity was obtained by blanching at 90°C for 4.0 min and 0.1% citric acid concentration (Figures 4.1 to 4.3).

Phenolic compounds and PPO are directly responsible for the enzymatic browning reaction in damaged fruits when processed (Lee and others 1990). PPO (EC: 1.10.3.1) is a copper-containing enzyme causing a browning reaction after peeling and cutting of many fruits and vegetables (Ayaz and others 2008). It oxidizes o-phenolic substrates to o-quinones that can further synthesize melanin and cause dark coloring (Ayaz and others 2008; Granato and others 2011). PPO has been reported to exist in most plants and is present in particularly high amounts in yacon, bananas, apples, pears, potatoes, avocados, and peaches (Koukounaras and others 2008; Lee and others 1990; Garcia and others 2002). Catechin and chlorogenic acid are well known polyphenol substrates of PPO in many fruits and vegetables, such as yacon, apples, pears, peaches, plums, cherries, and apricots (Oszmianski and others 1990; Rish and others 1988). In

apples, the maximum PPO activity on chlorogenic acid was obtained at 25–35°C (Cho and Ahn 1999). High temperatures are necessary to inactivate PPO and peroxidase activity in plant products (Bizzarri and others 1981; Muftugil 1985). For example, PPO in eggplant was completely inactivated at 75°C for 30 min or 80°C for 5.0 min (Fujita and Tono 1988). The half-activity of the PPO in mango was lost at 85°C for 2.1 min or 80°C for 4.0 min (Park and others 1980). The PPO in both Bosc and Red pears was completely inactivated at 75°C for 30 min (Siddiq and others 1993). Heat treatment of pre-cut peach slices at 50°C for 10 min significantly prolonged their shelf-life (Koukounaras and others 2008). However, blanching at 75°C for 30 min was not sufficient for complete inactivation of peroxidase enzyme in green beans, potatoes, and squash (Muftugil, 1985). The different in phenolic compounds and PPO activity among a variety of fruits and vegetables cause the varying degrees of browning (Lee and others 1990).

Effects of process variables on Hunter color

After analysis of various color values of yacon slices as affected by time, temperature, and citric acid, it was concluded that Hunter 'L', 'a', 'b', hue, and chrome were not affected (data not shown). Whiteness and total color difference (TCD) values were affected by the three factors studied ($P < 0.05$) (Table A.2). The three parameter combinations used for the blanching of yacon slices were important in optimizing for highest whiteness, and lowest TCD. The 20 experimental runs proposed by the CCD, with three factors (X1 = temperature, X2 = time, and X3= citric acid concentration), five levels (Table 3.1), and six replicates at the center point were used for fitting a second-order response surface model. The models obtained for whiteness and TCD of processed yacon slices were:

$$\text{Whiteness} = (164.3\pm84.086) - [(3.8\pm1.79)*X_1] + [(7.4\pm6.205*X_2)] + [(119.9\pm121.466)*X_3] + [(0\pm0.01)*X_1*X_1] - [(1.1\pm0.245)*X_2*X_2] - [(325.8\pm98.064)*X_3*X_3] + [(0\pm0.065)*X_1*X_2] - [(0.5\pm1.299)*X_1*X_3] + [(12.7\pm6.493)*X_2*X_3] \quad (4.2)$$

$$\text{TCD} = [-(164.3\pm84.086)] - [(1.3\pm1.2923)*X_1] + [-(0.6\pm4.4801*X_2)] + [-(244.7\pm87.6975)*X_3] + [(0\pm0.0071)*X_1*X_1] - [(0.5\pm0.177)*X_2*X_2] - [(344.2\pm70.8012)*X_3*X_3] + [(0\pm0.0469)*X_1*X_2] - [(1.9\pm0.9376)*X_1*X_3] + [- (5.8\pm4.6878)*X_2*X_3] \quad (4.3)$$

ANOVA was performed to determine the significance ($P < 0.05$) of the model. The variance was partitioned into linear (X1, X2, and X3), quadratic (X1*X1, X2*X2, and X3*X3), and interaction (X1*X2, X1*X3, and X2*X3) components to assess the adequacy of the second-order polynomial function and the relative significance of the terms for each experiment. The coefficients of the citric acid concentration factor (X3) for whiteness and TCD were 119.9±121.466 (X3) and 244.7±87.6975 (X3), respectively, indicating that this factor had a strong effect on the color of yacon slices ($P < 0.05$) (Table 4.1).

Whiteness of yacon slices was affected by a second-order relation of the three variables according to the results of the CCD (Table A.1 and Figures 4.4 to 4.6). In the raw data, the highest whiteness value was 31.7 for run 9 (80°C for 4.0 min with 0.1% citric acid), and the lowest whiteness value was 20.8 for run 1 (84°C for 2.8 min with 0.04% citric acid) (Table A.1). In the response surface plot, when citric acid in blanching water was held constant at 0.1%, whiteness as affected by blanching time more than as affected by blanching temperature (Figure 4.4). In the response surface plot, when

blanching time was held constant at 4.0 min, whiteness as affected by citric acid concentration more than as affected by blanching temperature (Figure 4.5). In the response surface plot, when blanching temperature was held at 90°C, the whiteness as affected by citric acid concentration more than as affected by blanching time (Figure 4.6). In the contour plots, when blanching temperature was held at 90 °C, the whiteness shows the optimum (highest whiteness) to be at 5.0 min with 0.18% citric acid concentration (Figure 4.6). In the raw data, the lowest TCD value was 4.62 for run 14 (90°C for 4.0 min with 0.2% citric acid), and the highest TCD value was 8.73 for run 8 (96°C for 5.2 min with 0.16% citric acid) (Table A.1). In the response surface plot, when citric acid in blanching water was held constant at 0.1%, TCD as affected by blanching time more than as affected by blanching temperature (Figure 4.7). In the response surface plot, when blanching time was held at 4.0 min, TCD as affected by citric acid concentration more than as affected by blanching temperature (Figure 4.8). In the response surface plot, when blanching temperature was held at 90°C, TCD as affected by citric acid concentration morn than as affected by blanching time (Figure 4.9). In the contour plot, when blanching temperature was held at 90°C, the optimum (lowest TCD) was at 4.5 min with 0.13% citric acid concentration (Figure 4.9). The response results for whiteness and TCD after 2, 6, and 10 days were similar ($P > 0.05$), so only the 10 day data analysis are discussed in this manuscript. Based on the above results, optimization of the blanching process for yacon slices for the highest whiteness value was 90°C at 5.0 min with a 0.18% citric acid concentration (Figure 4.4 to 4.6); while for the lowest TCD it was 90°C at 4.5 min with a 0.13% citric acid concentration (Figure 4.7 to 4.9).

Fruits and vegetables change their color during storage undergoing degreening and browning. Heat treatment can inhibit the enzymes that cause such a color change.

Short-time treatments at high temperatures (≥45°C) have been effectively used to reduce browning in fresh-cut products such as lettuce, celery, and Chinese water chestnuts (Koukounaras and others 2008; Saltveit 2000; Loaiza-Velarde and others 2003; Peng and Jiang 2004). Nevertheless, heat treatment leads to an enhanced rate of degreening in apples (Liu 1978; Klein and Lurie 1990; Klein and Lurie 1992). Cucumbers turn yellow through immersion in water at 45°C for 30–60 min (Chan and Linse, 1989), which is similar to yellowing of zucchini by forced vapor heat for 30 min at 45°C (Jacobi and others 1996). Moreover, Klein and Lurie (1992) used heating to enhance yellow color development of Golden Delicious apples while retaining their firmness. However, in papaya and banana skins and flesh, the color changes were not affected by immersion in water at 42°C for 30 min followed by 49°C for 90 min (Lurie 1998; Paull and Chen 1990; Seymour and others 1987). Thus, the blanching treatment was used in the current study to inhibit degreening or browning. In addition, the heat treatment itself caused the color of fruits and vegetables to change. Blanching yacon slices change its color from chalky to a translucent by visual in experiments (data not shown). The regular or total transmission measurement could measure a transparent product because the transparent product allowed some light to pass trough, but specularly and diffusely reflect light as well (Anonymous 2008). Color was seen in diffuse reflection when the light is in front of the product and was seen in diffuse transmission when the light is behind the product (Anonymous 2008). Thus, translucent objects are not easy to measure because any variations in their thickness and background will affect their color (Anonymous 2008). During the blanching, the factors that caused the color change were the dilution of pigment or intercellular air (Mark 1949). Mark (1949) illustrated that after blanching the carotenoids in carrot cells they become dissolved in fat droplets, and the color of the

carotenoids of oil droplets is a bright yellow; the color of the carotenoids of the chromoplast is a deep red or orange. Moreover, Mark and Lurie (1949) wrote that the proportion of the presence of air filled intercellular spaces changed the general appearance of the products and its storage life. Pre-storage heating also increases the color change rate (Mark and Lurie 1992).

PPO is the major enzyme causing the browning of fresh cut fruits and vegetables through the oxidation of phenolics such as chlorogenic acid. Chlorogenic acid and its derivatives are antioxidants present in fruits and vegetables, such as apples, tomatoes, papaya, bananas, and blueberries (Ojansivu and others 2011; Ohnishi and others 1994; Cano and other 1996; Murata and others 1993; Kader and others 1997). Chlorogenic acid is also present in leaves and tissues of dicotyledonous plants, such as yacon, potato, and coffee beans. PPO causes the brown color due to its reaction with o-phenolic substrates, and chlorogenic acid causes greening when oxidized or made into a slightly alkaline solution (Barnes and others 1950). The pigment is formed by the condensation reaction of two molecules of chlorogenic acid, or caffeic acid ester, with one molecule of primary amino compound in the air and alkali solution (Namiki and others 2001). In homogenized fresh blueberry, Kader and others (1997) showed that polymeric color increased rapidly for the first 10 min, and decreased gradually until the end of the treatment due to the insolubility of the polymers and chlorogenic acid in blueberry. Therefore, different pigment chemicals are inhibited through various treatments. Chlorophyll in apple peel, plantain peel, and tomato pericarp, decreased at 35–40°C in hot air treatment (Lurie 1998; Lurie and others 1990; Seymour and other 1987). Anthocyanin pigments in blanched blueberry juice were bluer and less red than traditional blueberry juice through thermal treatment (Rossi 2003).

The best blanching treatment for color of yacon slices was similar to that for lowest PPO activity. Thus, the color measurements can be considered as an indirect index of PPO activity and chlorogenic acid oxidation on yacon slices, and samples without browning or anomalous colors are reasonably PPO-free (Severini and others 2003). Color is the first and most important parameter for food quality assessment evaluated by consumers and the food industry (Loiselle and others 1990); it is also an easy and fast measurement when compared to enzymatic activity measurement (Reis and others 2008).

Effect of Process Variables on Sensory Rating

Response surface and contour plots for digital image sensory (rating score and browning percentage) as affected by temperature (X1), time (X2), and citric acid concentration (X3) are shown in Table 4.1. The models obtained for sensory rating and sensory browning percentage of processed yacon slices were:

$$\begin{aligned}\text{Sensory Rating} = {} & (20.79\pm16.9681) - [(0.43\pm0.3611)*X_1] + [(1.01\pm1.2337*X_2)] - \\ & [(28.24\pm24.1196)*X_3] + [(0\pm0.002)*X_1*X_1] - [(0.1\pm0.0495)*X_2*X_2] - \\ & [(76.43\pm19.7948)*X_3*X_3] + [(0\pm0.0129)*X_1*X_2] - \\ & [(0.5\pm0.2577)*X_1*X_3] + [(1.79\pm1.2887)*X_2*X_3]\end{aligned} \tag{4.4}$$

$$\begin{aligned}\text{Sensory Browning Percentage} = {} & (69.49\pm777.1) - [(0.47\pm16.54)*X_1] + \\ & [(2.69\pm56.5*X_2)] + [(1097.07\pm1104.63)*X_3] + \\ & [(0.01\pm0.09)*X1*X_1] - [(4.4\pm2.27)*X_2*X_2] - \\ & [(893.02\pm906.56)*X_3*X_3] - [(0.45\pm0.59)*X_1*X_2] \\ & - [(15.59\pm11.8)*X_1*X_3] - [(22.05\pm59.02)*X_2*X_3]\end{aligned} \tag{4.5}$$

ANOVA was conducted to determine the significance of the model. The variance was partitioned into linear (X1, X2, and X3), quadratic (X1*X1, X2*X2, and X3*X3), and interaction (X1*X2, X1*X3, and X2*X3) components to assess the adequacy of the

second-order polynomial function and the relative significance of the terms for each experiment. The coefficient of the citric acid concentration factor (X3) of sensory browning percentage was highest at 1097.07±1104.63 (X3), indicating that this factor was highly significant (Table 4.1).

The higher the sensory score, the closer the color to that of the original fresh yacon slices. In the raw data, the highest sensory rating score was 4.1 for run 12 (90°C for 6.0 min with 0.1% citric acid), and the lowest sensory rating score was 1.8 for run 13 (90°C for 4.0 min with no citric acid) (Table A.1). In the response surface plot, when citric acid concentration in blanching water was held at 0.1%, sensory rating score as affected by blanching time more than as affected by blanching temperature (Figure 4.10). In the response surface plot, when blanching time was held at 4.0 min, sensory rating score as affected by citric acid concentration more than as affected by blanching temperature (Figure 4.11). In the response surface plot, when blanching temperature was held at 90°C, sensory rating score was affected by citric acid concentration more than was affected by blanching temperature (Figure 4.12). In the contour plot, when blanching temperature was held at 90°C, the optimum (the highest sensory rating score) was at 5.5 min with 0.18% citric acid concentration (Figure 4.12).

In the raw data, the lowest sensory browning percentage value was 21 for run 14 (90°C for 4.0 min with 0.2% citric acid), and the highest sensory browning percentage value was 98 for run 13 (90°C for 4.0 min with 0.2% citric acid) (Table A.1). In the responds surface plot, when citric acid in blanching water held at 0.1%, sensory browning percentage as affected by blanching time more than as affected by blanching temperature (Figure 4.13). In the contour plots, the optimum (lowest sensory browning percentage) was at 103°C for 5.0 min (Figure 4.13). In the responds surface plot, when blanching

time was held at 4.0 min, sensory browning percentage as affected by citric acid concentration more than as affected by blanching temperature (Figure 4.14) In the responds surface plot, when blanching temperature was held at 90°C, sensory browning percentage as affected by both citric acid concentration and blanching time (Figure 4.15) In the contour plot, when blanching temperature was held at 90°C, the optimum (lowest sensory browning percentage) was at 5.5 min with 0.18% citric acid concentration (Figure 4.15). However, there was no effect of linear and square factors on sensory browning percentage ($P > 0.05$) (Figures 4.13 to 4.15), so the highest image sensory rating score was chosen at 90°C, 5.5 min, and 0.18% citric acid concentration.

There is a linear correlation between image quality and naturalness of the product color (Ridder and others 1995). Image sensory analysis proved to be effective in the image data quality, commercially available computational power, and data storage capabilities (Foca and others 2011). Using raw images at higher resolutions can enhance the quality of analyzed data and calibration models. According to the experimental design, the lower TCD of yacon slices would have higher sensory color score of yacon slice images (Figure 4.7 to 4.12). A significant relationship between phenol content and the brown percentage of explants has been shown (Roussos and others 2001). In addition, enzymatic browning deactivation is affected by the type and size of fruit and vegetable pieces (Bizzarri and others 1981; Muftugil 1985). The one of sensory rating score and sensory browning percentage is similar as the discussion in color change of yacon slices.

Overlapped Contour Plots

Although the optimum conditions for each of the dependent variable responses (PPO activity, whiteness, TCD, sensory rating, sensory browning percentage) were

slightly different, the graphical approach of overlapping the different response surfaces can serve to determine the optimum value of the responses (Reis and others 2008). When blanching temperature was held constant at 90°C, PPO activity and TCD decreased ($P < 0.05$) with blanching time and citric acid concentration, and the optimum blanching treatment was determined at 90°C for 4.1–5.2 min with 0.02–0.03% citric acid concentration. This yielded a value for PPO activity of 0.040–0.042 (ABS change/min), and a TCD of 9.4–10.0 (Figure 4.16). The overlapped contour plot on PPO activity and whiteness shows the optimum blanching treatment to be at 90°C for 3.8–4.2 min with 0.035%–0.05% citric acid concentration. This gave a PPO activity value of 0.040–0.042 (ABS) and a whiteness value of 22–24 (Figure 4.17). The overlapped contour plot on PPO activity and the sensory rating score shows the optimum blanching treatment at 90°C for 3.8–5.5 min with a 0.025%–0.045% citric acid concentration. This yielded a PPO activity value of 0.039–0.042 (ABS) for PPO activity and a sensory rating score of 2–3 (Figure 4.18). The overlapped contour plot on whiteness and sensory rating score shows the optimum blanching treatment at 90°C for 3.0–4.5 min with 0.020%–0.035% citric acid concentration. This yielded a value for whiteness of 20–25 and a sensory rating score of 2–3 (Figure 4.19). The contour plots for PPO activity, whiteness, and sensory rating score were overlapped to investigate the optimal conditions for blanching treatment (Figure 4.20). The area created by the intersection of the overlapped contour plots of PPO activity, whiteness, and sensory rating score showed the optimal conditions for blanching treatment to be at 0.040–0.042 (ABS change/min) for PPO activity, 23–24 for whiteness, and a sensory rating score of 3. This area lies between 3.9–4.1 min, 0.05%–0.07% citric acid concentration, and 90°C blanching temperature (Figure 4.20).

The above results show that heating treatments can be effective in preventing browning of yacon slices. Walter and Giesbrecht (1982) shows that 30 min pre soak in 80°C of sweet potato was enough to inactivate its PPO system. Citric acid is a weak organic acid that acts as a natural preservative and anti-browning compound (Altunkaya and others 2009). It has been reported that citric acid inhibits PPO activity through the chelation of copper at the active site of the PPO, and thus is an anti-browning agent in minimally processed fruits and vegetables (Altunkaya and Gokman 2009; Ahvenaien 1996). Eshtiaghi and Knorr (1993) showed that PPO can be completely inactivated at 20°C when potato cubes were immersed in 0.5% or 1.0% citric acid solution Acidification of lettuce with citric acid also appeared to stabilize phenolic compounds (Altunkaya and Gokman 2009). Therefore, citric acid is considered as both an anti-browning and an anti-oxidizing agent. The low pH dip during food processing does not cause changes in flavor like that which occurs in carrots (Altunkaya and others 2009; Boun and Huxsoll 1991); dipping peeled carrots for 30s at 70°C in citric acid solution delayed the formation of a white lignin-type material on the abraded carrot surface during storage (Boun and Huxsoll 1991). In addition, sulfite can also inhibit browning on the reaction of PPO with o-diphenols. The gradual inhibition of the enzyme causing mushroom browning was observed through pre-incubation of PPO and sulfite (Embs and markakis 1965). Blanching at high temperatures for a short time prevents the loss of nutrition from fruits and vegetables. Mayer-Miebach and Spieß (2003) showed a high availability and stability of lycopene in carrot products after blanching at 90°C in oxygen-free conditions. More functions of critic acid and nutrition and high temperature for the short time treatments is discussed in experiment II.

Table 4.1 Three way ANOVA on the effects of linear, quadratic, and interaction on dependent variable responses: PPO activity, whiteness, TCD, sensory rating, sensory browning percentage.

Sorce	PPO activity		Whiteness		Total Color Difference		Sensory Rating		Sensory Browning %	
	Coef	SE	Coef	SE	Coef	SE	Coef	SE	Coef	SE
Constant	1.1849	0.228229	164.3	84.086	-35	60.7091	20.79	16.9681	69.49	777.1
X_1	-0.0213	0.004858	-3.8	1.79	1.3	1.2923	-0.43	0.3611	0.47	16.54
X_2	-0.0622	0.016843	7.4	6.205	-0.6	4.4801	1.01	1.2337	2.69	56.5
X_3	-0.2969	0.329689	119.9	121.466	-244.7	87.6975	-28.24	24.1196	1097.07	1104.63
X_1*X_1	0.0001	0.000027	0	0.01	0	0.0071	0	0.002	0.01	0.09
X_2*X_2	0.0056	0.000665	-1.1	0.245	0.5	0.177	-0.1	0.0495	4.4	2.27
X_3*X_3	0.2559	0.266169	-325.8	98.064	344.2	70.8012	-76.43	19.7948	893.02	906.56
X_1*X_2	0.0001	0.000176	0	0.065	0	0.0469	0	0.0129	-0.45	0.59
X_1*X_3	0.0032	0.003525	-0.5	1.299	1.9	0.9376	0.5	0.2577	-15.59	11.8
X_2*X_3	-0.0312	0.017623	12.7	6.493	-5.8	4.6878	1.79	1.2887	-22.05	59.02

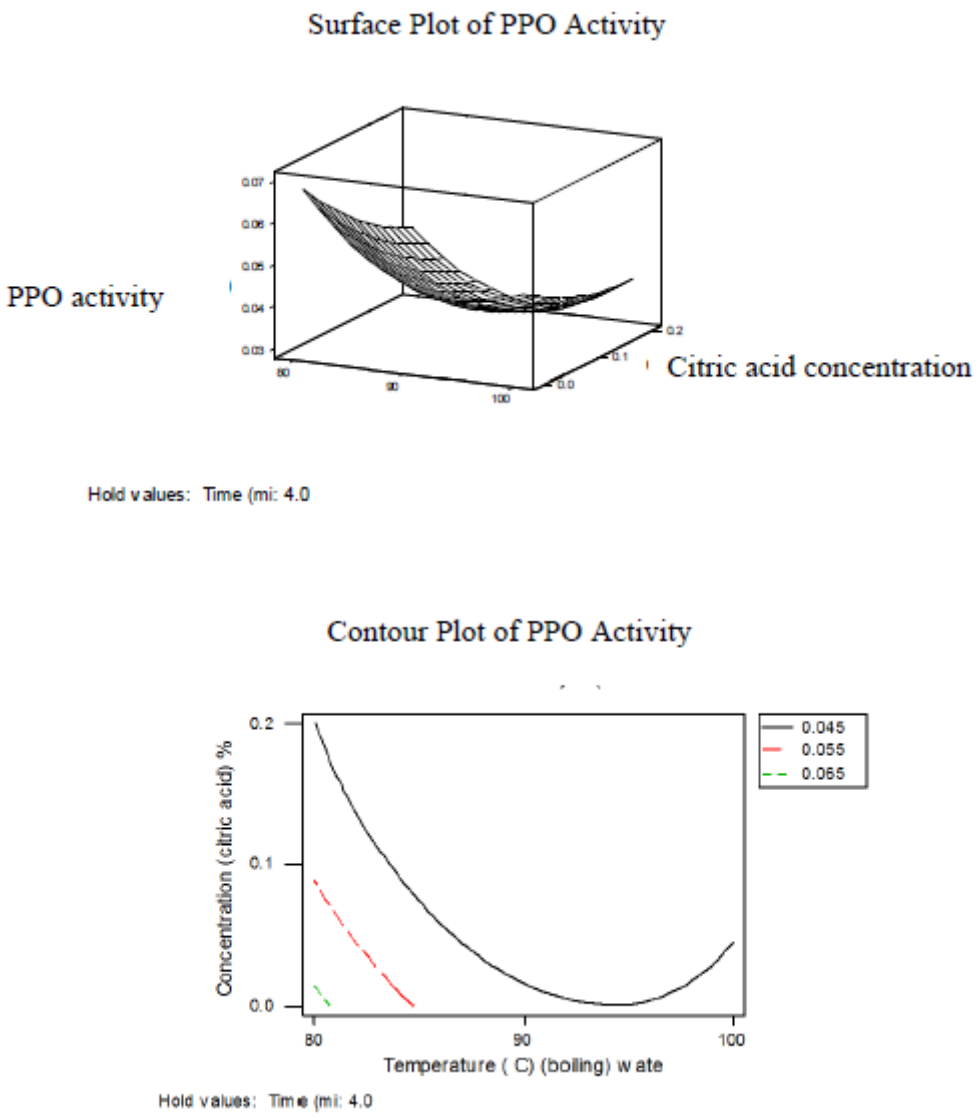
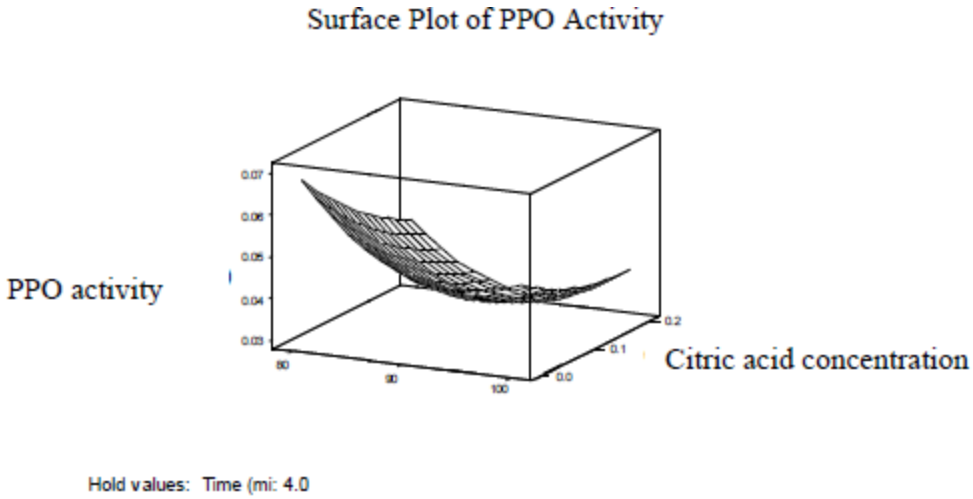


Figure 4.1 Surface and contour plots for PPO activity on yacon slices blanched with 0.1% citric acid as affected by temperature (°C) and time (min).



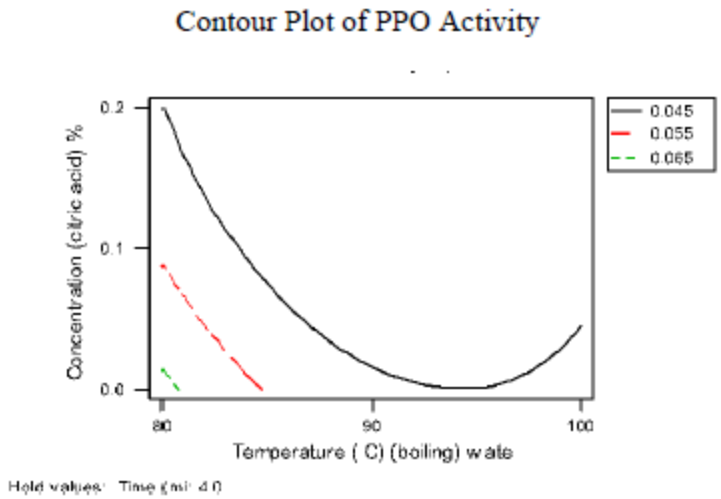


Figure 4.2 Surface and contour plots for PPO activity on yacon slices blanched after 4.0 min as affected by temperature (°C) and citric acid (%)

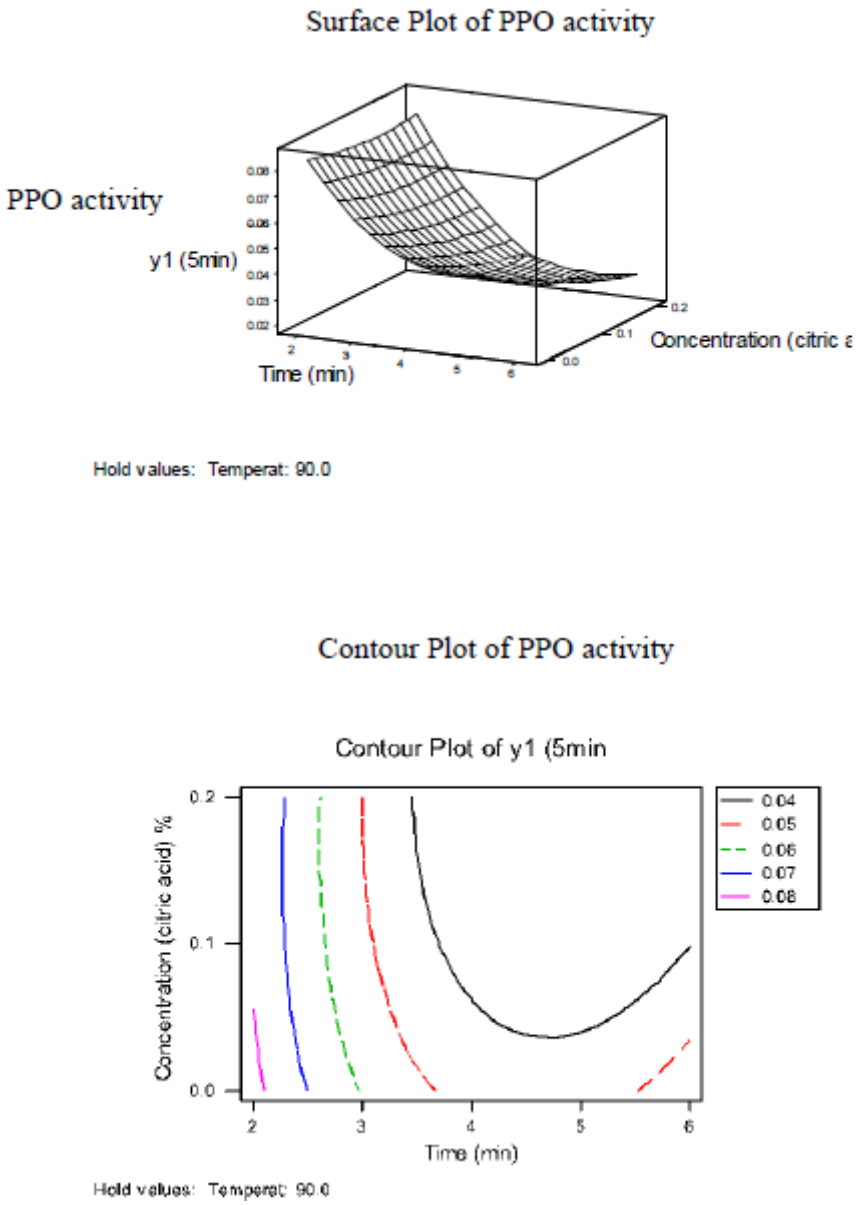
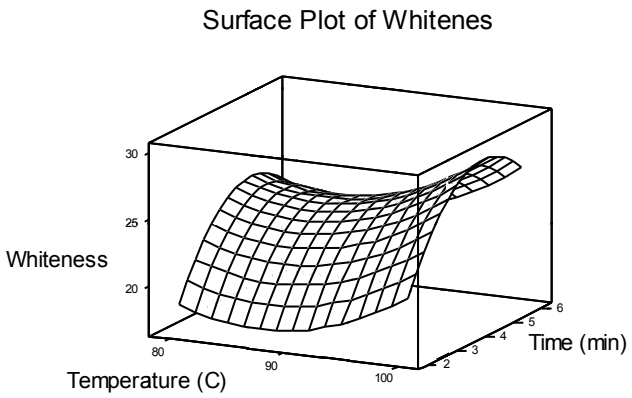


Figure 4.3 Surface and contour plots for PPO activity on yacon slices blanched at 90°C as affected by time (min) and citric acid (%).



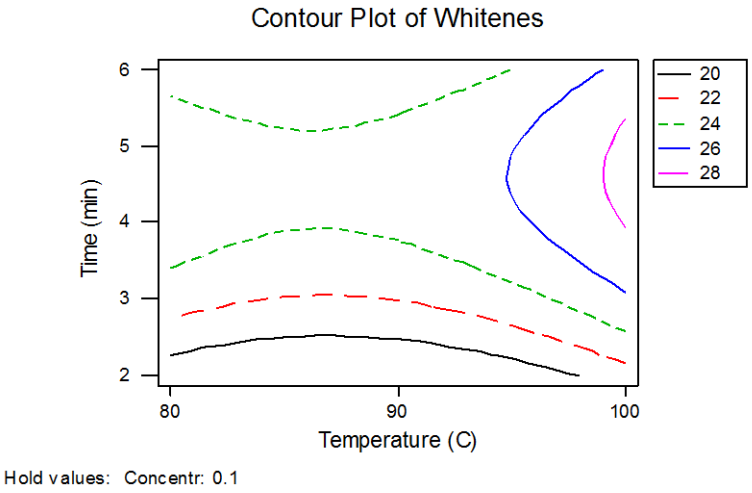
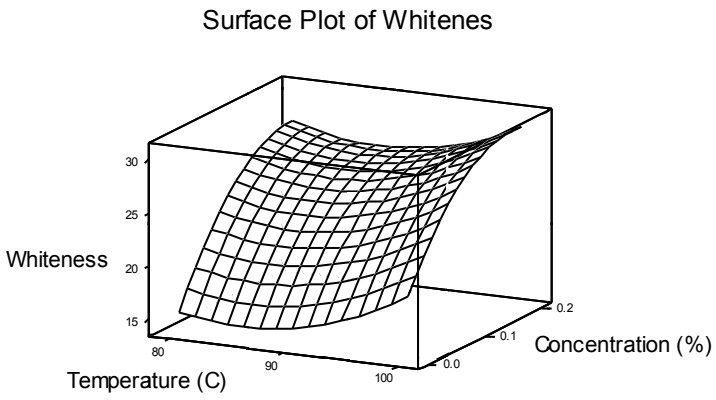


Figure 4.4 Surface and contour plots for whiteness on yacon slices blanched with 0.1% citric acid as affected by temperature (°C) and time (min).



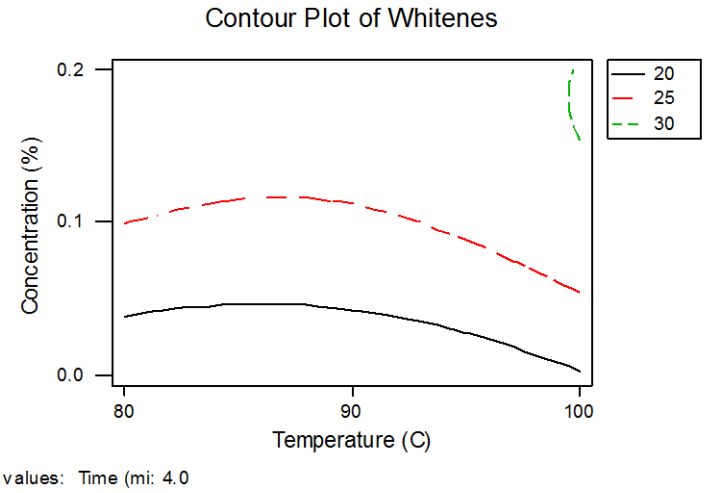


Figure 4.5 Surface and contour plots for whiteness on yacon slices blanched for 4.0 min as affected by temperature (°C) and citric acid (%).

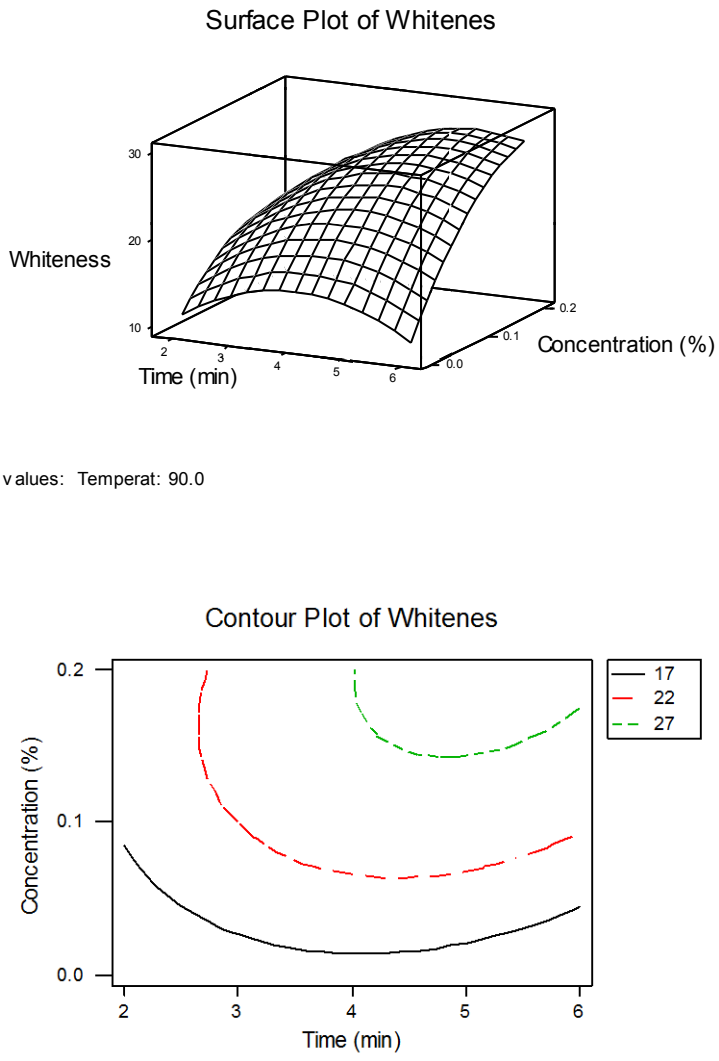
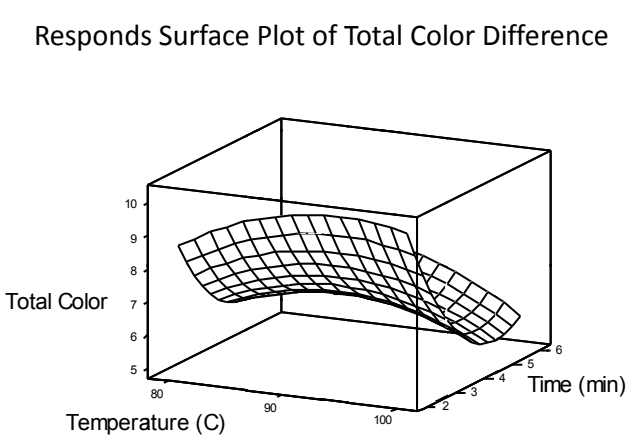


Figure 4.6 Surface and contour plots for whiteness on yacon slices blanched at 90℃ as affected by time (min) and citric acid (%)



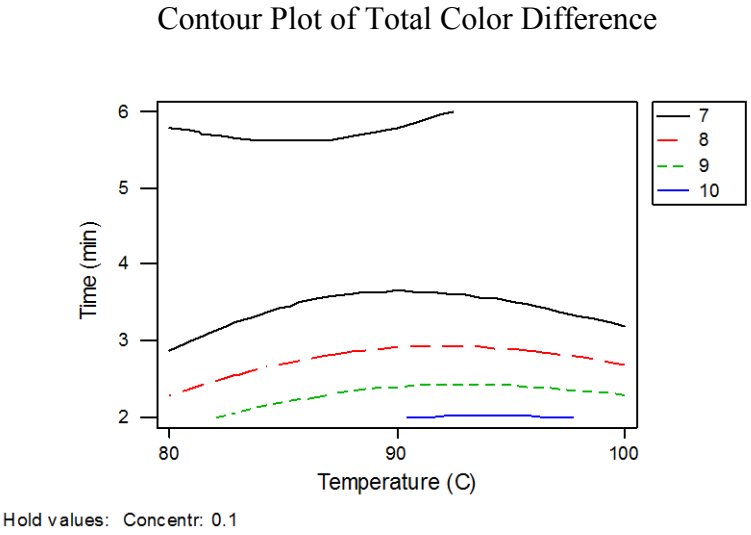
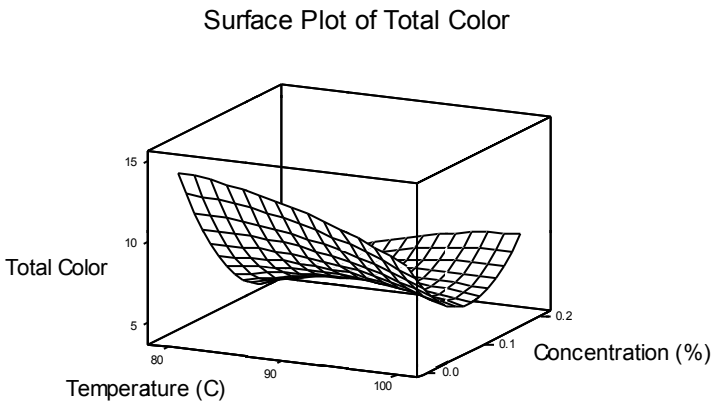


Figure 4.7 Surface and contour plots for TCD on yacon slices blanched with 0.1% citric acid as affected by temperature (°C) and time (min)



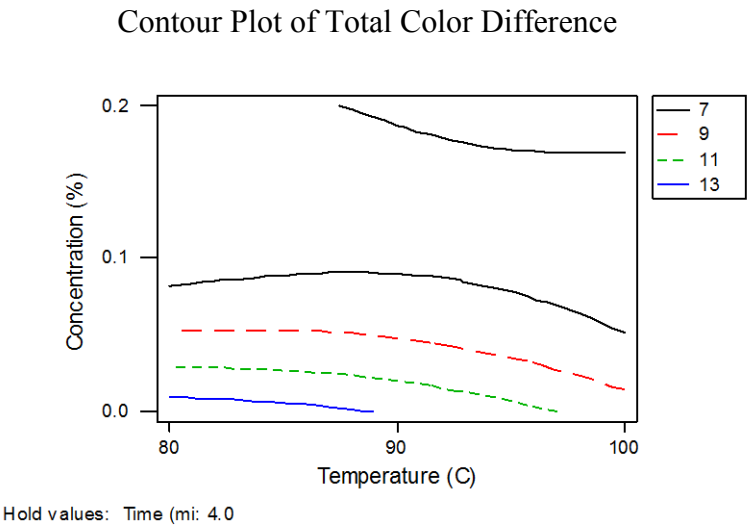
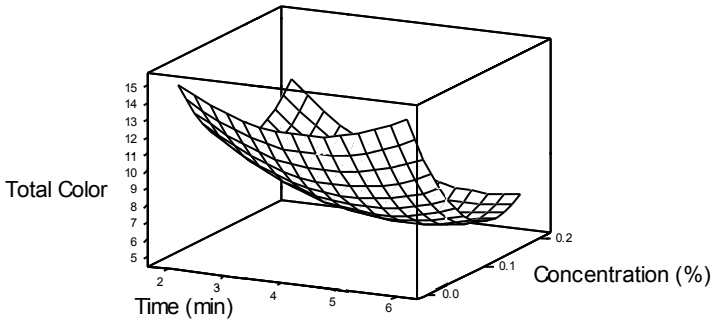


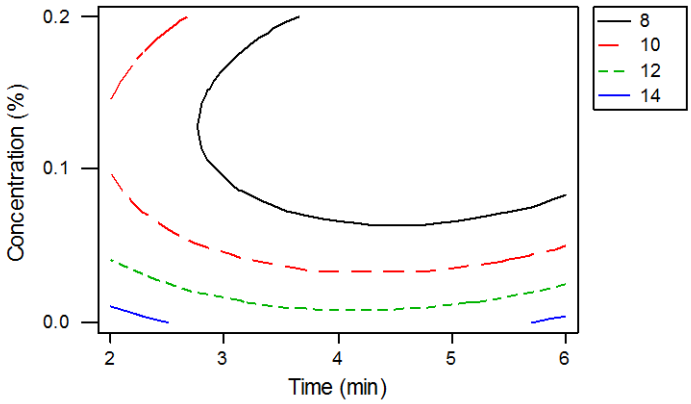
Figure 4.8 Surface and contour plots for TCD on yacon slices blanched for 4 min as affected by temperature (°C) and citric acid (%)

Responds Surface Plot of Total Color Difference



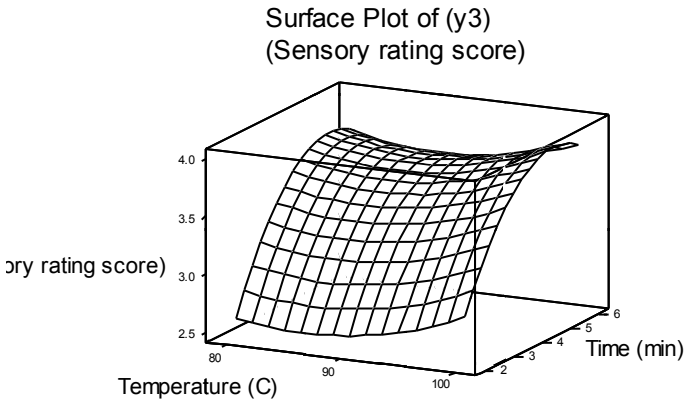
Hold values: Temperat: 90.0

Contour Plot of Total Color Difference



Hold values: Temperat: 90.0

Figure 4.9 Surface and contour plots for total color difference on yacon slices blanched at 90℃ as affected by time (min) and citric acid (%).



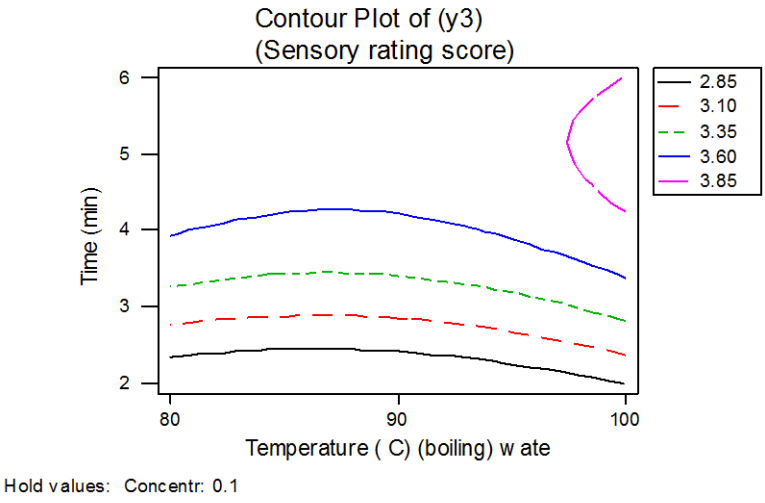
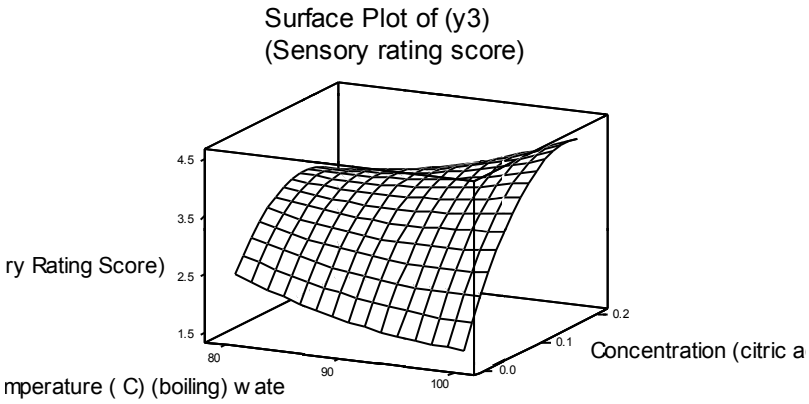


Figure 4.10 Surface and contour plots for sensory rating score on yacon slices blanched with 0.1% citric acid as affected by temperature (°C) and time (min).



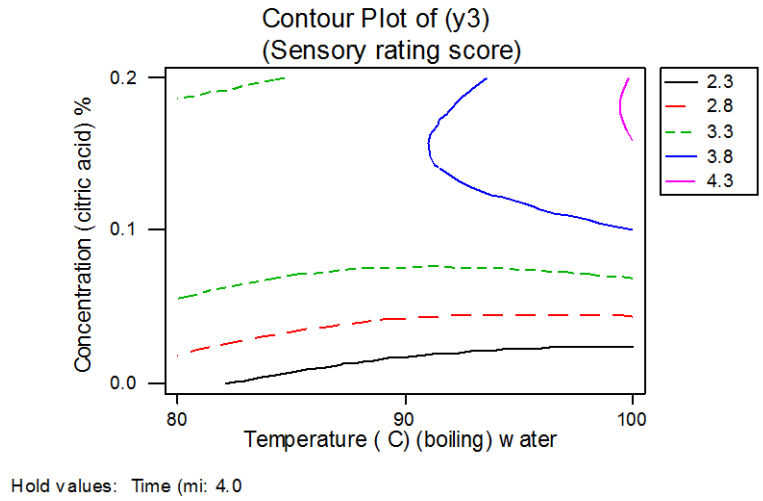


Figure 4.11 Surface and contour plots for sensory rating score on yacon slices blanched after 4.0 min as affected by temperature (°C) and citric acid (%).

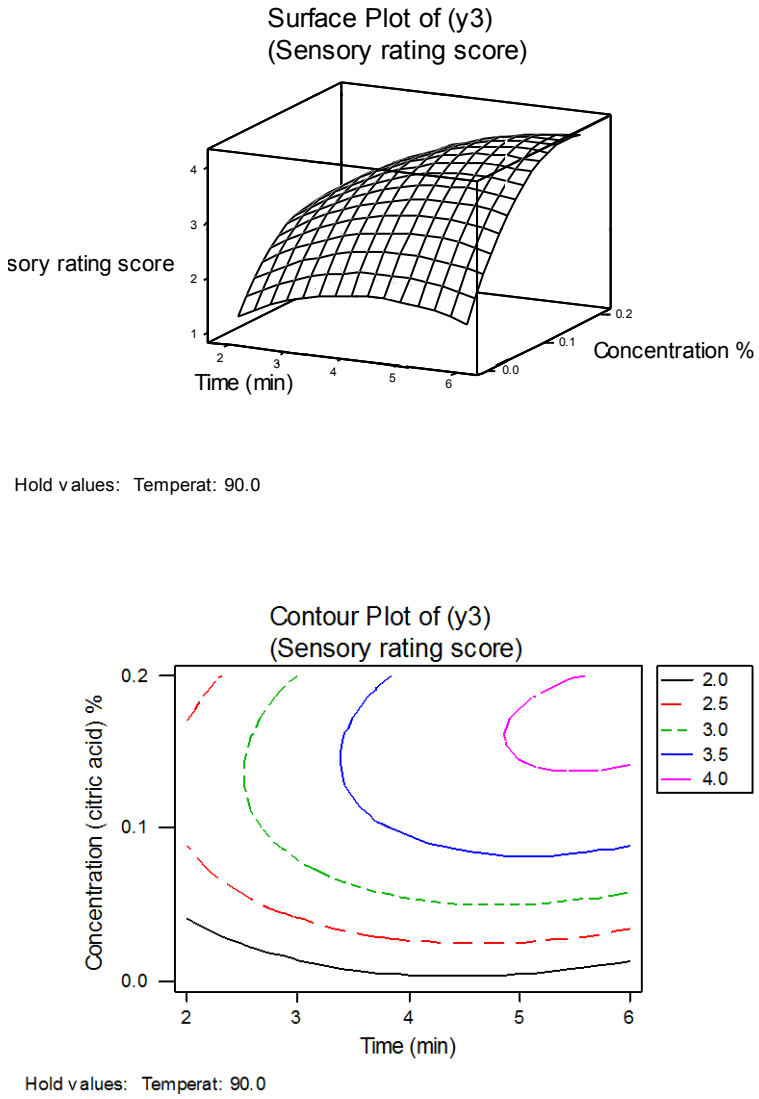
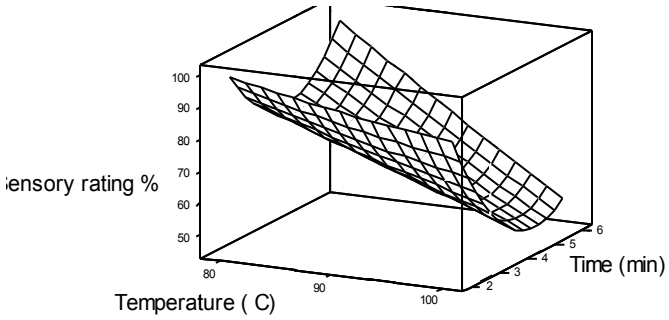


Figure 4.12 Surface and contour plots for sensory rating score on yacon slices blanched at 90˚C as affected by time (min) and citric acid (%).

Surface Plot of Sensory Browning Percentage



Contour Plot of Sensory Browning Percentage

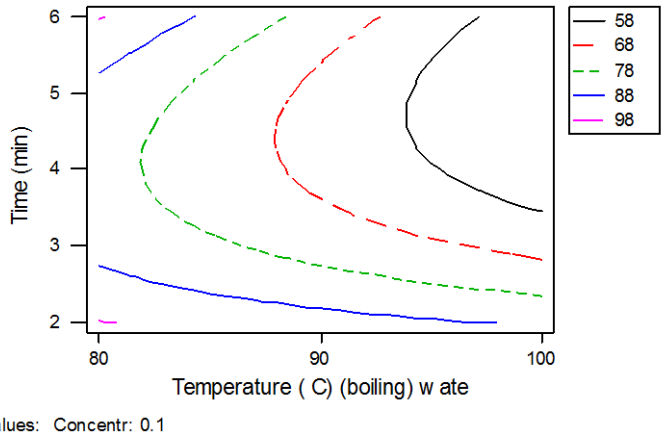
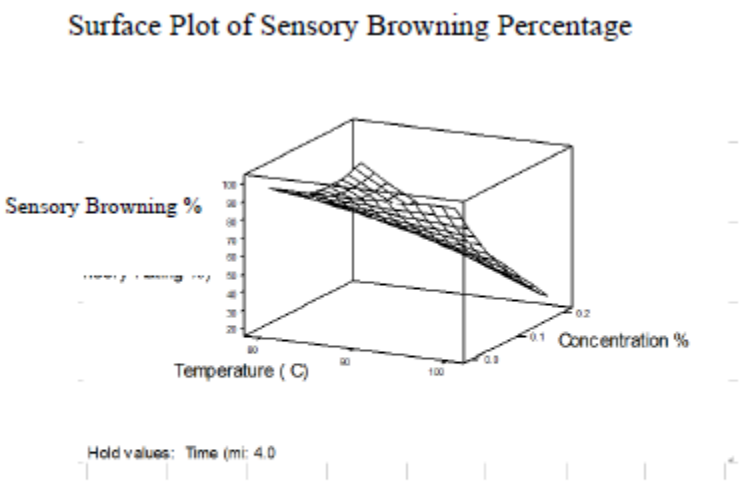


Figure 4.13 Surface and contour plots for sensory browning percentage on yacon slices blanched with 0.1% citric acid as affected by temperature (°C) and time (min)



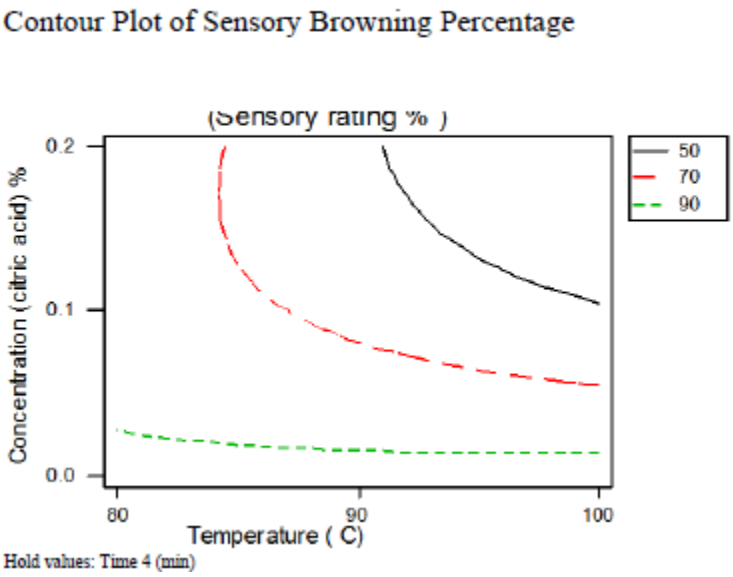


Figure 4.14 Surface and contour plots for sensory browning percentage on yacon slices blanched after 4.0 min as affected by temperature (°C) and citric acid (%)

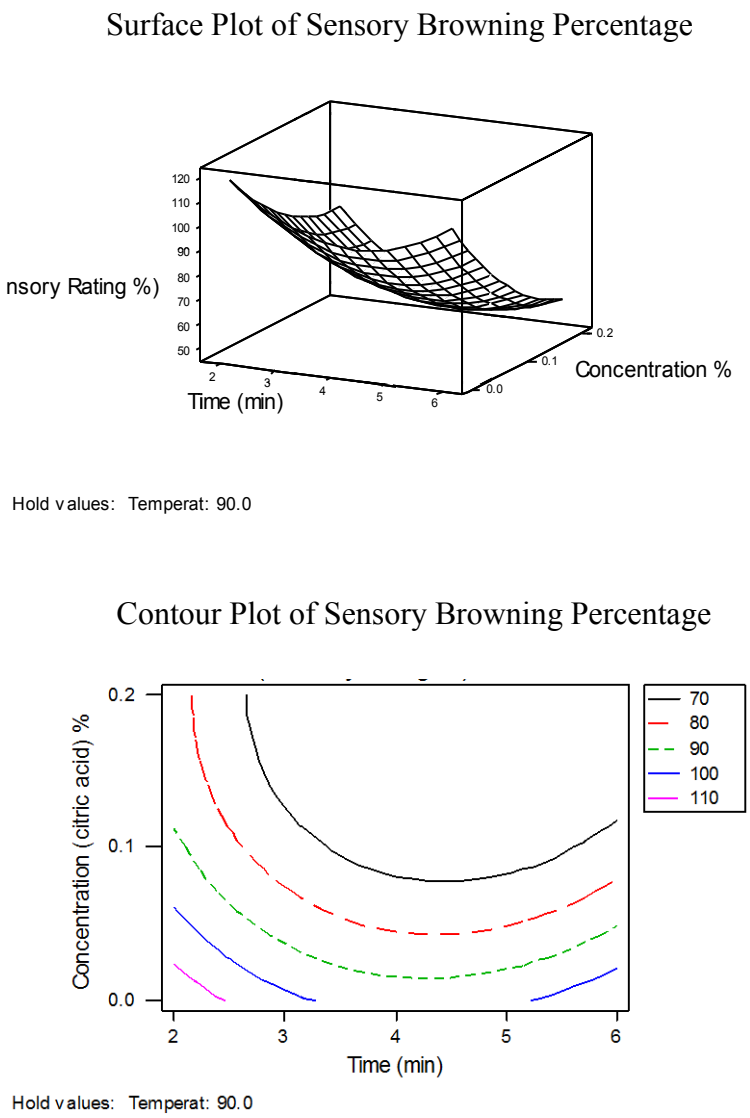


Figure 4.15 Surface and contour plots for sensory browning percentage on yacon slices blanched at 90°C as affected by time (min) and citric acid (%)

Optimization- overlapped contour plot analysis

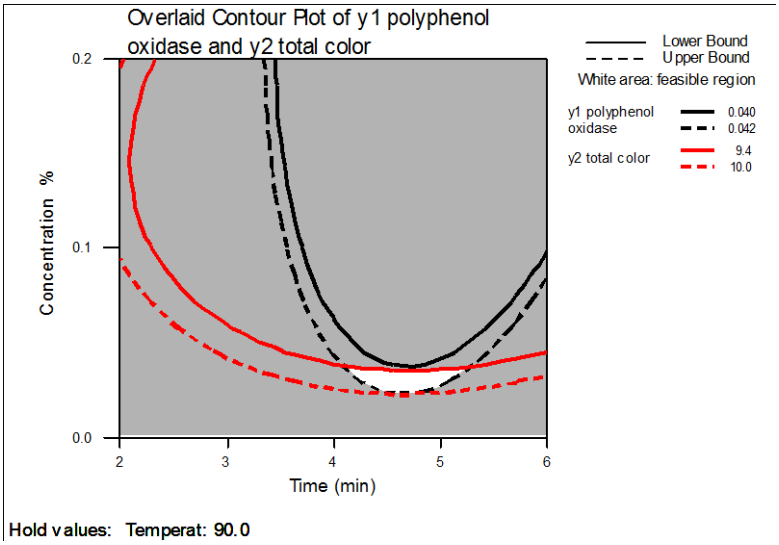


Figure 4.16 Overlapped contour plot for PPO activity and total color difference of blanched yacon slices at 90°C as affected by blanching time and citric acid concentration.

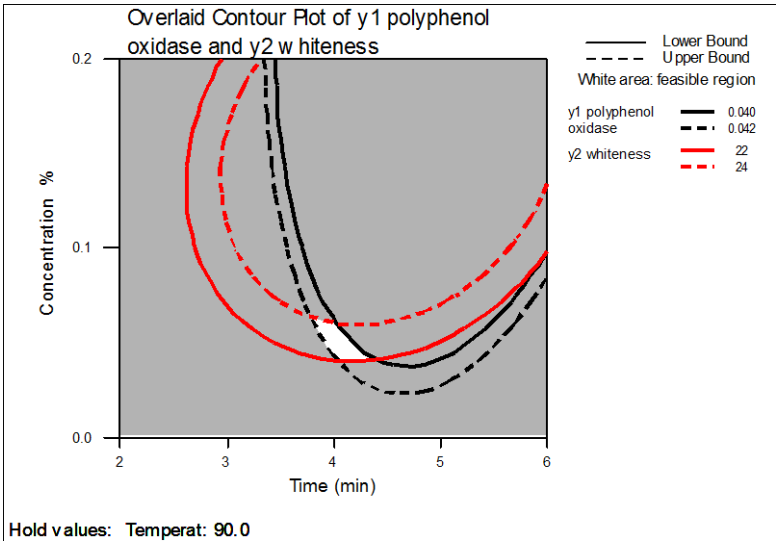


Figure 4.17 Overlapped contour plot for PPO activity and whiteness of blanched yacon slices at 90°C as affected by blanching time and citric acid concentration.

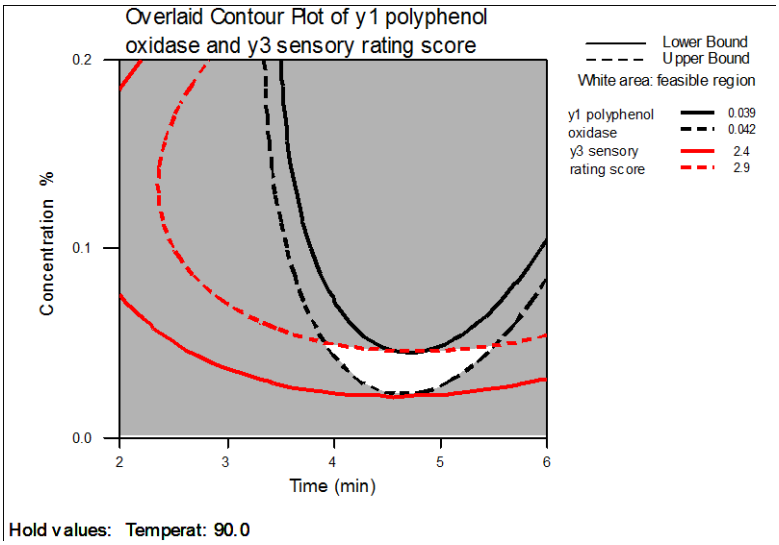


Figure 4.18 Overlapped contour plot for PPO activity and sensory rating scores of blanched yacon slices at 90°C as affected by blanching time and citric acid concentration.

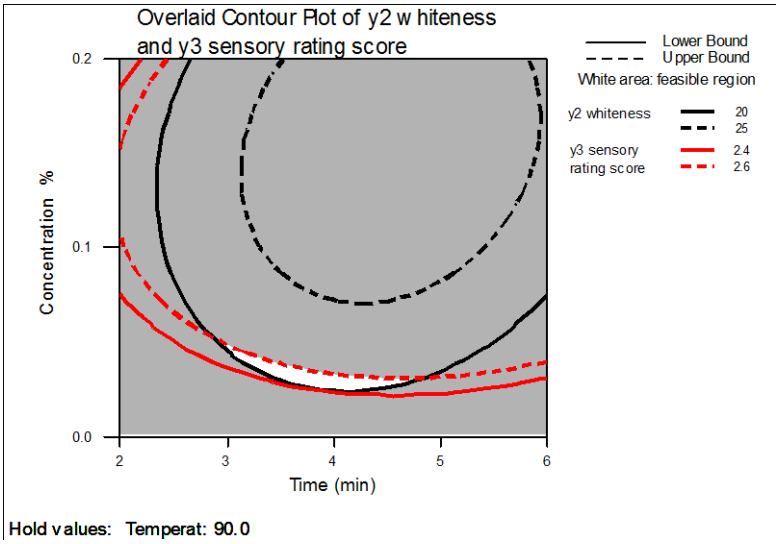


Figure 4.19 Overlapped contour plot for whiteness and sensory rating scores of blanched yacon slices at 90°C process as affected by blanching time and citric acid concentration

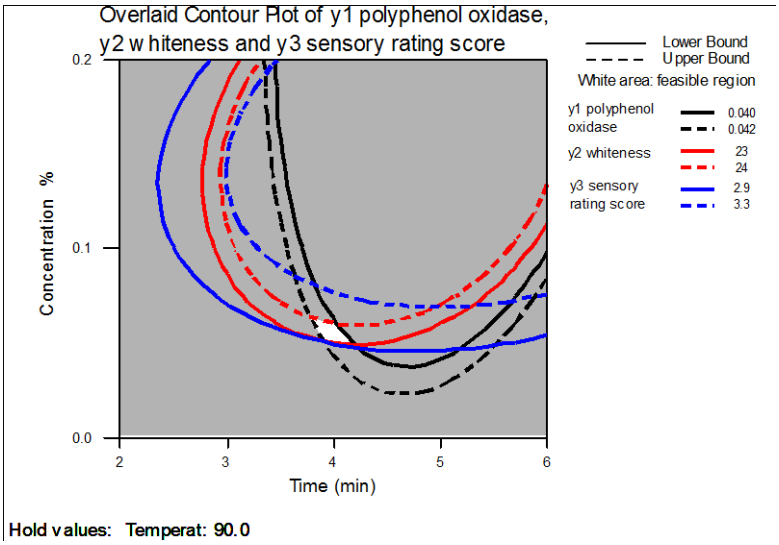


Figure 4.20 Overlapped contour plot for PPO activity, whiteness and sensory rating scores of blanched yacon slices at 90°C as affected by blanching time and citric acid concentration.

Experiment II: Optimization of Blanching Process for Yacon (*Smallanthus Sonchifolius*) Root Slices

Based on results from experiment I and further experiments (data not shown), yacon slices were blanched in boiling water with and without citric acid for 6 min. There was no interaction ($P>0.05$) between time and treatment with regard to polyphenol oxidase (PPO) activity (Table A.2); but blanching time had an effect ($P<0.05$) on PPO activity (Table A.2). PPO activity in yacon slices blanched for 1.5 min decreased ($P<0.05$) to 0.004 (ΔABS/min); when the blanching time was increased to 3.0 min or over, the PPO activity further decreased ($P<0.05$) to a negligible level. As described in the discussion of experiment I, PPO plays an important role in most fruits and vegetables. It is directly responsible for the enzymatic browning reaction in fruits and vegetables that are damaged during post harvest processing due the oxidization of the phenolic substrates to quinones (Lee and others 1990; Ayaz and others 2008). The difference in the degree of

browning is caused by the difference in phenolic content and PPO activity among a variety of fruits and vegetables (Lee and others 1990). Whole Russet Burbank potatoes were blanched for up to 60 min at 50 °C without the appearance of browning; and the PPO activity ranged from1.58 ΔABS/min (control) to 0.7ΔABS/min (Yemenicioglu 2002). Further, blanching banana slices in boiling water for 11 min followed by freezing at -24°C inhibited color deterioration in the frozen product (Cano and others 1990). Heat treatment on freshly-cut Chinese water chestnut in boiling water for 30 s effectively prevented browning associated with PPO activity (Peng and Jiang 2004). The discoloration of canned beet root slices radically moved toward the center of the slice when the steam time was increased; one of the reasons for this is the thermal stability of PPO (Im and others 1990). In potato slices, the PPO activity decreased over 50°C with adequate treatment time and was destroyed at 80°C (Severini and others 2003; Vamos-Vigyazo 1981). Moreover, PPO in eggplant was completely inactivated at 75°C for 30 min or 80°C for 5 min (Fujita and Tono 1988), half of the PPO activity in mango was lost at 85°C for 2.1 min or 80°C for 4.0 min (Park and others 1980), and the PPO activity in both Bosc and Red pears was completely inactivated at 75°C for 30 min (Siddiq and others 1993). Thus, PPO heat resistance depends on the species and cultivars of fruits and vegetables.

Addition of citric acid to water for blanching had no effect ($P>0.05$) on PPO activity (Table A.5). Citric acid has been reported for its inhibitory effect on PPO through the chelation of the copper at the active site of the PPO; thus it is called an anti-browning agent in minimally processed fruits and vegetables (Altunkaya and others 2009; Ahvenaien 1996). The inhibition of PPO activity prevents the activation of quinine, which causes the yacon slices to become brown when in storage (Yoruk and Marshell

2003). In certain studies, PPO activity in potato cubes (2 × 2 × 2 cm) was completely inactivated at 20°C when the cubes were immersed in a citric acid solution (0.5 or 1.0%) (Eshtiaghi and Knorr 1993). Further, PPO activity in the banana pulp was strongly inhibited by a solution of citric acid and acetic acid (Yang and others 2000). The PPO in avocado was controlled by immersing it in a combination of ascorbic acid, citric acid, and subjecting it to heat treatment (Almeida and Nogueira 1995). The PPO of litchi fruits was controlled to prevent browning under a combination of 10 mmol litre−1 glutathione and 100 mmol litre−1 citric acid (Jiang and Fu 1998). However, in another study, citric acid solutions (0.2-10 g/L range) showed little effect in terms of inhibiting PPO activity in apple cubes (Pizzocaro and others1993). Thus, the effect of citric acid on PPO activity not only depends on the combination with other chemicals but also on the type of fruits and vegetables.

There was no interaction ($P>0.05$) between blanching treatment and time with regard to whiteness of yacon slices (Figure 4.22). Whiteness was not affected ($P>0.05$) by treatment, but it was affected ($P<0.05$) by blanching time. Whiteness of blanched samples was higher ($P<0.05$) than the whiteness of unblanched samples, regardless of the inclusion of citric acid. Whiteness was highest ($P<0.05$) after blanching for 1.5 to 4.5 min, but decreased ($P<0.05$) after blanching for 6.0 min (Figure 4.22). There was no interaction ($P>0.05$) between time and treatment with regard to total color difference (TCD) (Figure 4.23). Moreover, TCD was not affected ($P>0.05$) by treatment but was affected ($P<0.05$) by blanching time. The TCD increased for the first 1.5 min and then had no effect ($P>0.05$) for time. This is consistent with Fante and others (2012) who observed an increase in ΔE values during the first 2 min, but after this time these values no longer changed significantly ($P > 0.05$). There was an interaction ($P<0.05$) between

time and treatment with regard to the hue angle of blanched yacon slices (Figure 4.24). The hue angle of blanched yacon slices in water with citric acid was higher ($P<0.05$) than the hue angle of blanched yacon slices in water without citric acid for each blanching time, except 3.0 min (Figure 4.24). In water treatment , the hue angle increased for 1.5 min and had no effect ($P>0.05$) after this time. Fante and others (2012) showed the hue angle changes were more significant after 2 min ($P > 0.05$). The increase in hue angle means a decrease in the redness and an increase in the intensity of yellowness (Fante and others 2012). There was no interaction ($P>0.05$) between time and treatment with regard to chroma of blanched yacon slices (Figure 4.25). Chroma was the same ($P>0.05$) regardless of treatment but decreased ($P<0.05$) after blanching for 6 min.

As discussed in experiment I, the heat treatment causes the color of fruits and vegetables to change. Blanching yacon slices causes them to change from chalky to translucent in visual experiments. The chroma value changed from 9.4 to 5.9 (Table A.3. to Figure 4.25). However, in the steam blanched yacon, the chroma value increased with time (Fante and others 2012). The regular or total transmission measurement could measure a transparent product because the transparent product allowed some light to pass through, but specularly and diffusely reflect light as well (Anonymous 2008). Translucent objects are not easy to measure because any variations in their thickness and background will affect their color (Anonymous 2008). During the blanching, the factors that caused the color change were the dilution of pigment or intercellular air and others (Mark 1949).

Heat treatment helps produce high–quality, minimally processed fruits (Kim and others 1993). Numerous researchers have shown that blanching potato slices before frying can improve the color of the potato chips because blanching could cause reducing sugars to be released from the potato tissues; potato slices that are blanched before frying

have a light color and do not brown after frying (Pedreschi and others 2005; Andersson 1994). Pre-storage heating also increases the rate of color changes in fruits and vegetables (Mark and Lurie 1992 1992). Heat treatment of apple slices at 45°C causes less browning and firmer texture compared to untreated apple slices (Kim and others 1993). The hue angle of blanched yacon slices in water with citric acid was higher ($P<0.05$) than the hue angle of blanched yacon slices in water without citric acid in each blanching time except at 3.0 min (Figure 4.24). Citric acid functions as an antioxidant in processed fruits and vegetables. Perez-Gago and others (2005) used an antioxidant to coat freshly cut apples and this reduced browning. Yabuta and others (2001) illustrate that the green pigment in food materials like sweet potato is caused by quinone or semiquinone-type products of trihydroxy benzacridine derivatives or charge-transfer complexes of the quinhydrone-type between them. Apart from citric acid, ascorbic acid or $NaBH_4$ can reduce the green pigment (Yabuta and others 2001).

There was an interaction ($P<0.05$) between time and treatment with regard to the total phenolics in yacon slices (Figure 4.26). There was a higher amount of total phenolics ($P<0.05$) in yacon slices blanched in water without citric acid than yacon slices blanched in water with citric acid. The total phenolics in the blanched samples was lower ($P<0.05$) than the total phenolics in the unblanched sample, regardless of the addition of citric acid. In the water-blanched sample, the total phenolics decreased ($P<0.05$) to the lowest level after 4.5 min. Blanching in water with citric acid led to a decrease in total phenolics ($P<0.05$) to the lowest level after blanching for 6.0 min. There was an interaction ($P<0.05$) between time and treatment with regard to chlorogenic acid in yacon slices, with the same result as in total phenolics (Figure 4.26 and 4.27).

Silva and others (1991) illustrated that after-cooking, darkening was highly correlated with phenolic acid content ($r = 0.85$, $P < 0.01$) in Spartan Pearl potatoes. Chlorogenic acid and its derivatives are antioxidants in fruits and vegetables such as yacon, potato, apple, tomato, papaya, banana, and blueberry (Ojansivu and others 2011; Ohnishi and others 1994; Cano and others 1996; Murata and others 1993; Kader and others 1997). During food processing and cooking, the condensation reaction of two molecules of chlorogenic acid or caffeic acid ester with one molecule of a primary amino compound under aeration in alkaline solution causes the formation of a green pigment (Yabuta and others 2001). Further, greening is observed during the alkali extraction of protein from sunflower meal due to its chlorogenic acid (Yabuta and others 2001; Sabir and others 1974). The chlorogenic acid content in sunflower kernels is found to decrease when the temperature increases (5°C, 15°C, and 40°C) after storage for 120 days (Pomenta and others 1971). Kader and others (1997) showed that chlorogenic acid decreases drastically in the first 10 min, reaches a minimum in 30 min, and then maintains a stable low content in homogenized fresh blueberries. However, the optimal chlorogenic acid oxidase in head lettuce (Lactuca Sativa) was stable in the pH range 6–8 at 5°C for 20 h (Fujita and others 1991). Chlorogenic acid plays an important role in the defense system of plant tissue (Namiki and others 2001). Namiki and others (2001) reported that caffeic acid derivatives such as chlorogenic acid yield a semiquinone-type free radical. The caffeate radicals may polymerize with each other to form brown products; moreover, there is a trihydroxy benzacridine derivative, which is reactive with oxygen that is also formatted with an amino compound. It could be hypothesized that chlorogenic acid leaches in boiling water, thus not available in yacon slices for browning. As described previously, citric acid serves as an antioxidant in food products (Perez-

Gago and others 2006; Guilbert 1986; Baldwin and others 1995). Fresh yacon contains approximately 12.9 to 13.8 mg per 100g dry weight of chlorogenic acid and is about 80% of total phenolics in yacon slices (Table A.3). In potato, chlorogenic acid constitutes approximately 90% of the total phenolic compounds and its concentration ranges from 10 to 20 mg per 100 g of fresh weight (Singh and others 1998; Mondy and others 1988).

There was an interaction ($P<0.05$) between time and treatment with regard to the total sugars in blanched yacon slices (Figure 4.28). Total sugars decreased ($P<0.05$) as blanching time was increased in yacon slices blanched in water without citric acid. However, total sugars increased in yacon slices blanched in water with citric acid after 4.5 min blanching time; thus, there was a higher amount of total sugars in this sample as compared to those blanched in water without citric acid for 4.5min. Further, there was an interaction ($P<0.05$) between time and treatment with regard to the inulin in blanched yacon slices (Figure 4.29). There was lower inulin ($P<0.05$) in yacon slices blanched in water without citric acid than in yacon slices blanched in water with citric acid at each time of blanching. The inulin in yacon slices is lost during blanching because the inulin types of FOS hydrolyze to reducing sugars over 70°C (Scher and others 2009). There was an interaction ($P<0.05$) between time and treatment with regard to the sucrose in blanched yacon slices (Figure 4.30). Sucrose decreased ($P<0.05$) when yacon slices were blanched in water without citric acid. Further, there was an interaction ($P<0.05$) between time and treatment with regard to the glucose in blanched yacon slices (Figure 31). The highest glucose amount was present in yacon slices when blanched in water without citric acid after 1.5 min. The glucose in this sample was more stable than that in yacon slices blanched in water with citric acid. Further, there was an interaction ($P<0.05$) between time and treatment with regard to fructose in blanched yacon slices (Figure 32). The

amount of fructose decreased ($P<0.05$) with an increase in blanching time, except in yacon slices blanched in water with citric acid after 4.5min. The amount of fructose was higher in the yacon slices blanched in water without citric acid than in yacon slices blanched in water with citric acid after 4.5 min. Furthermore, the water soluble chemical compounds measured in yacon slices blanched in water with citric acid—such as total phenolic compounds (4.6 mg/g more), chlorogenic acid (3.52 mg/g more), inulin (5.41% more), sucrose (5.94% more), glucose (3.27% more), fructose (20.29% more), and total sugars (34.91% more)—were all higher than slices blanched in water without citric acid after 4.5 min.

In the results, it shows 25.8 g /100g dry matter in yacon roots (Table A.4). There are different amounts of FOS in the dry root of yacon, and it ranges between 22–77 g/100g (Hermann and others 1999); in a fully ripe banana, ranging between 297 to 1600 µg/g of dry matter (Agopian and others 2008). During storage, the inulin content decreases in tubers. Inulin is decomposed by an internal enzyme, inulase (Takeuchi and others 2011; Cabezas and others; Modler and others 1993). To maintain the inulin content, heat treatment processing above 60°C has been reported to inactivate inulase (Takeuchi and others 2011). In some studies, dry heating of inulin from chicory at between 135°C to 195°C for 60 min caused significant decomposition of the fructan ranging from 20% to 100% (Bohm and others 2005). Bohm and others (2005) illustrated that heat treatment of inulin causes the long fructose chains to be decomposed into other products, such as fructose. In the tubers of Cichorium intybus, the decrease in inulin was associated with an increase in glucose and sucrose; in the tube of Helianthus tuberosus (Jerusalem artichoke), there was an increase in the proportion of fructan when sucrose reached its maximum content (Cabezas and others 2002). However, inulin, sucrose,

glucose, and fructose in the yacon slices could have leached in the boiling water. Raul and others (1984) demonstrated that the reducing sugars in the water after blanching potato slices totaled 0.09 % in 1000mL. In a 4 mm potato slice, the losses of glucose amounted to 12.50% at 25°C, 14.55% at 45°C, 34.90% at 55°C, 50.39% at 65°C and 63% at 85°C, and the estimated apparent diffusivity of glucose in potato were 8.20 × 10−10 m2/sec at 65°C and 12.50 × 10−10 m2/sec at 85°C.

There was an interaction ($P<0.05$) between time and treatment with regard to inulin in the blanched yacon slices (Figure 4.29).There was a larger amount of sucrose, glucose, and fructose in yacon slices blanched in water without citric acid than in yacon slices blanched in water with citric acid for 1.5 and 3 min. This is because the inulin in yacon slices breaks down into simple sugars. Scher and others (2009) showed that the FOS hydrolyzed to reducing sugars at 70 °C, and they indicated the reducing sugars increased from 36.65 ± 0.54–44.10 ± 0.96% and the non-reducing sugars decrease from 31.62 ± 0.55–26.18 ± 0.29%.

Overall, there was no interaction ($P>0.05$) between time and treatment with regard to the whiteness, total color difference, chroma, and PPO activity of blanched yacon slices, but they were affected ($P<0.05$) by blanching time. However, there was an interaction between time and treatment with regard to hue angle, total phenolics, chlorogenic acid, total sugars, inulin, sucrose, glucose, and fructose in blanched yacon slices. The blanching treatment with citric acid can maintain a higher proportion ($P<0.05$) of important chemical compounds, total phenolics, chlorogenic acid, total sugars, inulin, and fructose than the blanching treatment with only water. Further, heat treatment can inhibit enzymes, such as PPO and pectinesterase. Steinbuch (2006) showed that pectinesterase is inactivated at temperatures of 80°C and higher, while decreasing the

hardening of the bean texture. Invertase activities and reducing sugar concentration increased in roots stored at low temperatures; for example, in sweet potatoes, acid invertase is the most effective in determining and reducing sugar content (Huang and others 1999). Further, reducing the sugar level was correlated to acid and total invertase activity, regardless of the cultivar in sweet potatoes (Huang and others 1999). Heat treatment at higher temperatures and for a shorter time can avoid heat damage. Peng and Jiang (2004) showed that in Chinese water chestnuts, heat treatment prevents browning associated with PPO and total phenolic content and prevents the decay in quality that reduces the amount of total soluble solids (Peng and Jiang 2004). Garlic products that are blanched in hot water at 90°C for 15 min before packing are more stable, have better quality, and retain the best color as compared with those that are not blanched (Rejano and others 1997).

Reduction of enzymatic browning focuses on the chemical inhibitors of enzymatic activity, removing oxygen and phenolics or rendering a substrate to avoid the formation of brown pigments (Perez-Gago and others 2006). Citric acid is a weak organic acid that is a natural preservative and anti-browning compound; no change in flavor was found after the reduction in pH levels as a result of food processing (Altunkaya and others 2009; Boun and others 1991). Citric acid has been reported for its inhibitory effect on PPO activity through the chelation of copper at the active site of PPO; thus, it is used as an anti-browning agent in minimally processed fruits and vegetables (Altunkaya and others 2009; Ahvenaien 1996). Similar to the effect of some inhibitory chemicals of metallic ions such as Ca2+, Mn2+, Co2+, and Ni2+ on chlorogenic acid oxidase activity, potassium cyanide and sodium diethyldithiocarbamate markedly inhibit chlorogenic acid oxidase activity in lettuce (Fujita and others 1991). Further, citric acid has the function of

stabilizing the phenolic compound—chlorogenic acid—in yacon. It chelates iron and a high ratio of citric acid to chlorogenic acid can decrease the tendency of after-cooking darkening in potatoes (Kermasha and others 1993; Hughes and others 1967). Further, the acidification of lettuce through citric acid also appeared to stabilize phenolic compounds (Altunkaya 2009). Thus, citric acid has been considered as both an anti-browning and antioxidant agent (Wang and others 2003). Both the inhibitor and method used affect the type and degree of the inhibitory effect of aromatic carboxylic acid (Kermasha and others 1993). Chlorogenic acid is often used as a substrate for studies on PPO inhibition (Kermasha and others 1993; Oszmianski and others 1990). The after-cooking darkening in potato tubers is due to the reaction between chlorogenic acid and iron (Kermasha and others 1993; Hughes and others 1962). Singh and others (1998) illustrate that the incorporation of 1.2% of citric acid prevents the darkening of the extract and neutralizes the inhibitory effect of chlorogenic acid during the extraction of the nucleic acid from potato.

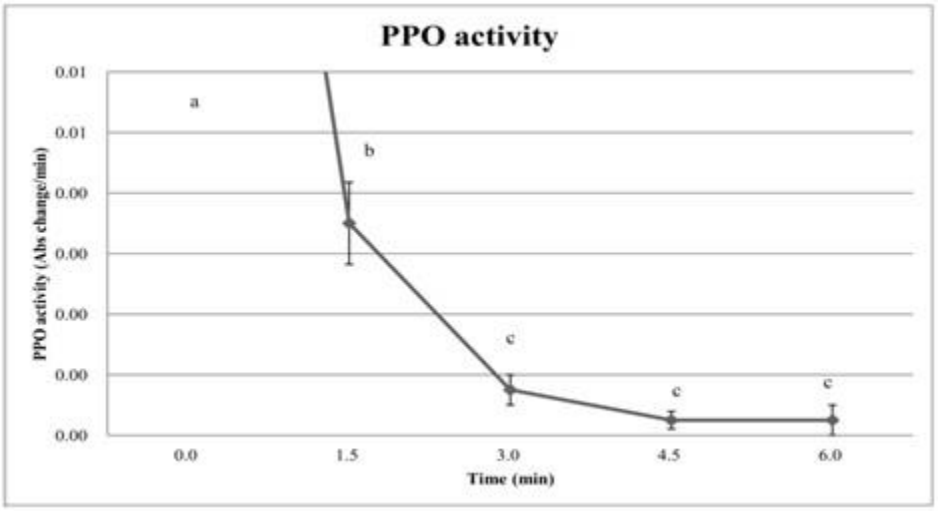


Figure 4.21 Polyphenol oxidase (PPO) activity of yacon slices blanched in water with and without citric acid as affected by blanching time.

Means denoted by the same letters are not significantly different (P<0.05).

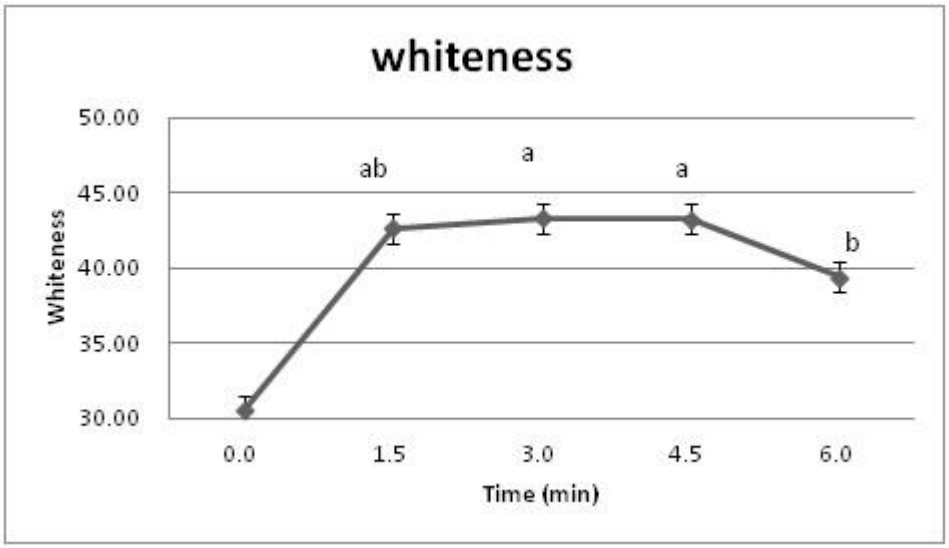


Figure 4.22 Whiteness of yacon slices blanched in water with and without citric acid as affected by blanching time.

Means denoted by the same letters are not significantly different (P<0.05).

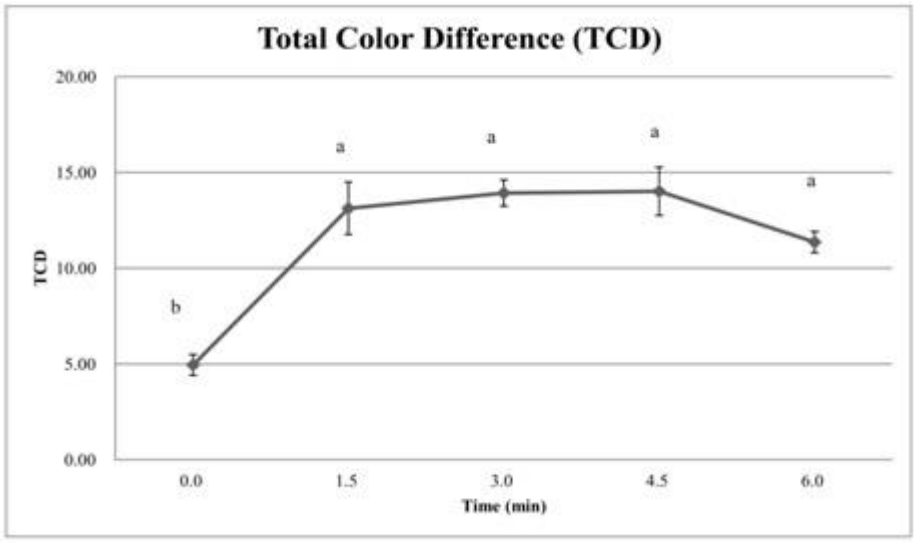


Figure 4.23 Total color difference (TCD) of yacon slices blanched in water with and without citric acid as affected by blanching time.

Means denoted by the same letters are not significantly different (P<0.05).

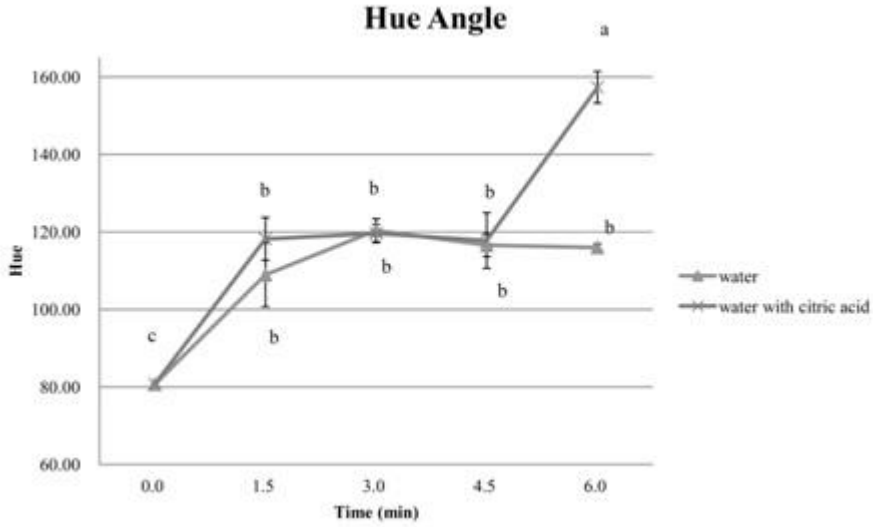


Figure 4.24 Hue angle of yacon slices blanched in water with or without citric acid as affected by blanching time.

Means denoted by the same letters are not significantly different (P<0.05).

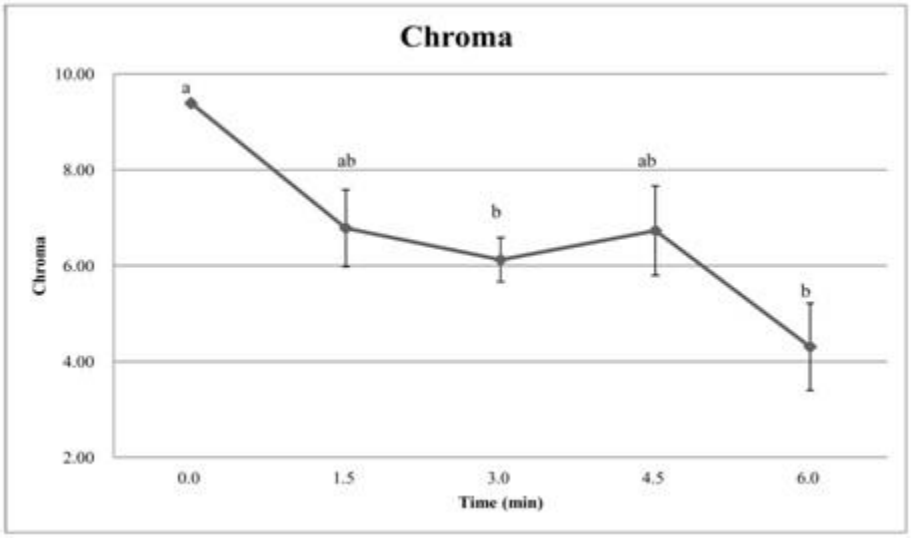


Figure 4.25 Chroma of yacon slices blanched in water with and without citric acid as affected by blanching time.

Means denoted by the same letters are not significantly different (P<0.05).

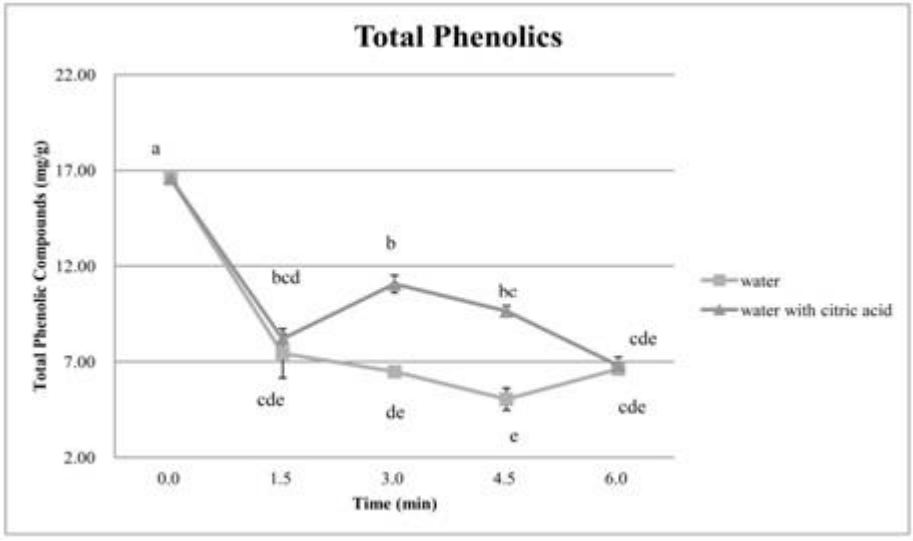


Figure 4.26 Total phenolics in yacon slices blanched in water with or without citric acid as affected by blanching time.

Means denoted by the same letters are not significantly different (P<0.05).

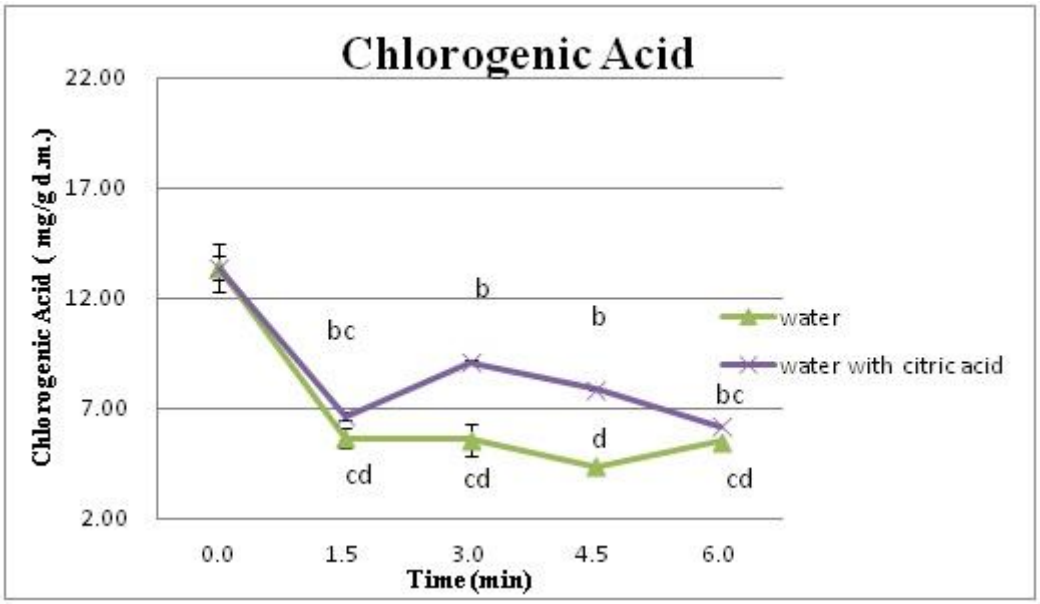


Figure 4.27 Chlorogenic acid in yacon slices blanched in water with or without citric acid as affected by blanching time.

Means denoted by the same letters are not significantly different (P<0.05).

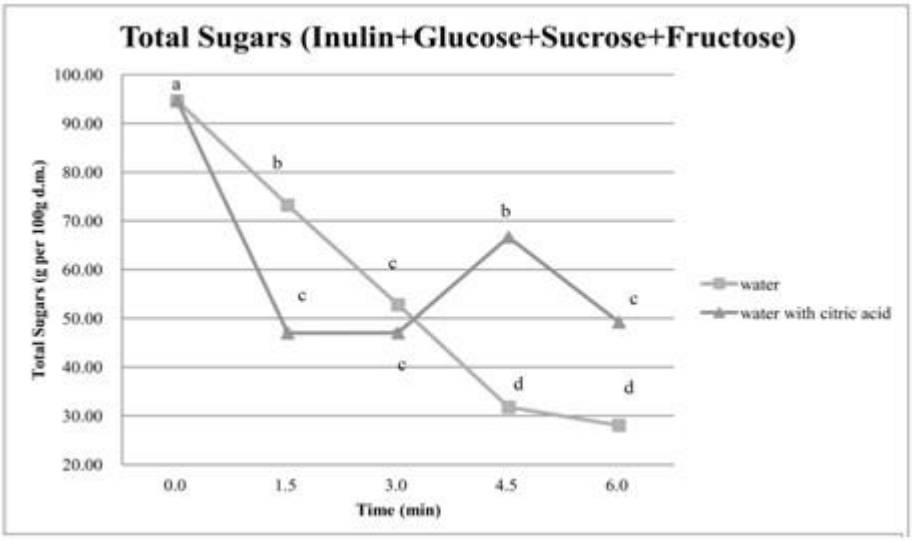


Figure 4.28 Total sugars (Inulin + Glucose + Sucrose + Fructose) in yacon slices blanched in water with or without citric acid as affected by blanching time.

Means denoted by the same letters are not significantly different (P<0.05).

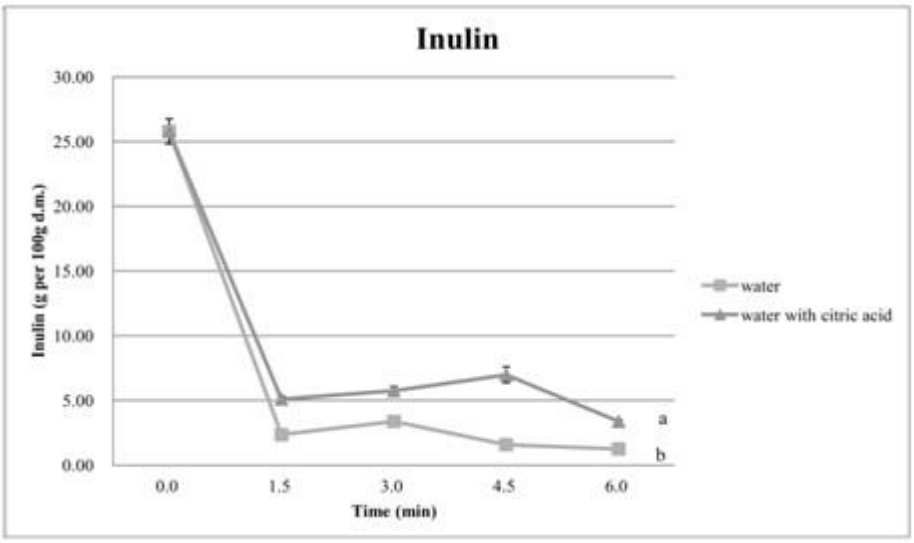


Figure 4.29 Inulin in blanched yacon slices in water with or without citric acid as affected by blanching time.

Means denoted by the same letters are not significantly different (P<0.05).

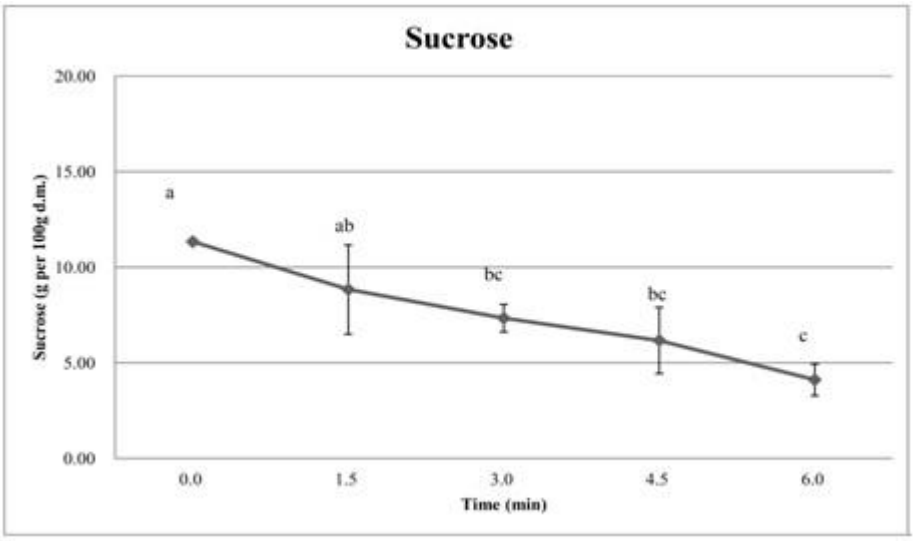


Figure 4.30 Sucrose in yacon slices blanched in water with and without citric acid as affected by blanching time.

Means denoted by the same letters are not significantly different (P<0.05).

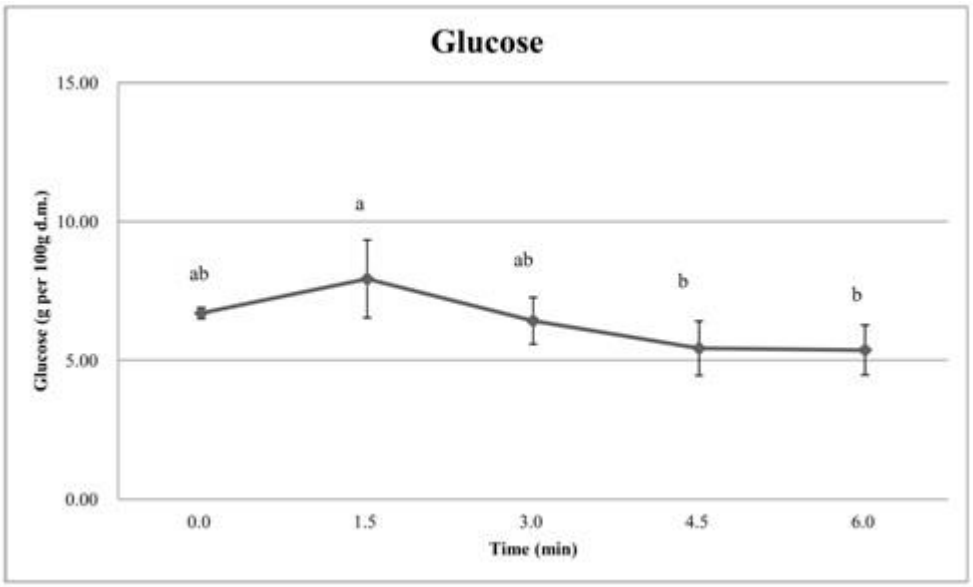


Figure 4.31 Glucose in yacon slices blanched in water with and without citric acid as affected by blanching time.

Means denoted by the same letters are not significantly different (P<0.05).

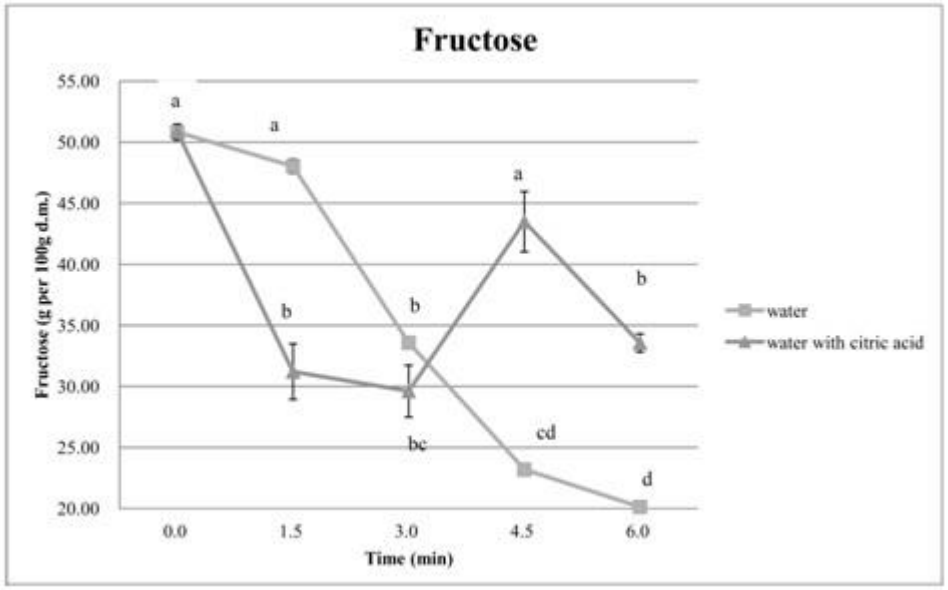


Figure 4.32 Fructose in yacon slices blanched in water with or without citric acid as affected by blanching time.

Means denoted by the same letters are not significantly different (P<0.05).

CHAPTER V

SUMMARY AND CONCLUSIONS

The present study demonstrated that polyphenol oxidase (PPO) activity was completely inactivated through blanching with 0.07% citric acid after 4 min at 90°C. The optimization condition on multiple quality characteristics indicates the lowest PPO activity (0.040–0.042 Abs), 23–24 whiteness value, and a sensory rating score of 3 at 90°C, with 0.05%–0.07% citric acid concentration, for 3.9–4.1 min through an overlapped contour plot. Moreover, yacon slices blanched at 90°C, for 5 min, with 0.1% citric acid concentration showed the lowest PPO activity content; yacon slices blanched at 90°C, for 5 min, and with 0.18% citric acid concentration showed the highest whiteness value; yacon slices blanched at 90°C, for 5.5 min, and with 0.18% citric acid concentration showed the highest image sensory appearance score and lowest browning percentage by panelists.

In the second experiment, a comparison was conducted with the whiteness, total color difference, hue angle, chroma, PPO activity, total phenolic compounds, chlorogenic acid, total sugars, inulin, sucrose, glucose, and fructose between yacon slices blanched in water without citric acid and yacon slices blanched in water with 0.07% citric acid. There was no interaction ($P>0.05$) between time and treatment with regard to whiteness, total color difference, chroma, and PPO activity of blanched yacon slices, but they were affected ($P<0.05$) by blanching time. Blanching after 3 min decreased ($P<0.05$) PPO

activity to a negligible level. Moreover, there was an interaction between time and treatment with regard to hue angle, total phenolics, chlorogenic acid, total sugars, inulin, sucrose, glucose, and fructose in blanched yacon slices. The blanching treatment with citric acid generally maintained higher ($P<0.05$) total phenolics, chlorogenic acid, total sugars, inulin, and fructose than the blanching treatment with only water. Therefore, blanching treatment with citric acid can lower the PPO activity to prevent the yacon from browning and stabilize the important chemical compounds, such as chlorogenic acid and inulin, and increase the percentage of total sugars in yacon slices blanched in water with citric acid as compared with yacon slices blanched in water only.

Yacon is a highly economical and valuable product owing to its nutraceutical properties, high prebiotic fiber, and phenolic content; but, yacon is easily deteriorated (browned) after cutting in a short time. The optimal blanching process to inhibit browning of yacon slices is at 90°C, for 3.9–4.1min with a 0.05%–0.07% citric acid concentration. Although blanching with citric acid can help retain the important functional components in yacon slices more than blanching without citric acid, most of the chemical compounds in yacon are lost in the water. The use of other blanching methods may be useful to retain important functional components and nutrients in yacon slices, such as steam, microwave, high-pressure blanching, or by chemical inactivation by the additive in yacon slices of ascorbic acid, acetic acid, and others in modified atmosphere packaging (removal of oxygen).

REFERENCES CITED

Agopian RGD, Soares CA, Purgatto E, Cordenunsi BR, Lajolo, FM. 2008. Identification of fructooligosaccharides in different banana cultivars. J Agr Food Chem 56:3305-3310.

Ahvenaien R. 1996. New approaches in improving the shelf life of minimally processed fruits and vegetables. Trends Food Sci Tech 7:179–187.

Almeida MEM, Nogueira JN. 1995. The control of polyphenol oxidase activity in fruits and vegetables. Plant Food Hum Nutr 47:245-256.

Altunkaya A, Gokmen V. 2009. Effect of various anti-browning agents on phenolic compounds profile of fresh lettuce (*L. sativa*). Food Chem 117:122-126.

Andersson, A. 1994. Modeling of potato blanching. Ph.D. Thesis, Lund University: Sweden

Anonymous. 2008. What is color and how is it measured? Hunter Associates Laboratory, Inc. 12:1-8. Avilable from http://www.hunterlab.com/appnotes/an05_00.pdf Accessed 2013 Jun 16.

Antolovich, M., Prenzler, P., Robards, K., & Ryan, D. 2000. Sample preparation in the determination of phenolic compounds in fruits. Analyst 125:989-1009.

Arogba SS, Ajiboye OL, Ugboko, S.Y. Essienette, Afolabi PO.1998. Properties of polyphenol oxidase in Mango (*Mangifera indica*) kernel. J Sci Food Agr 77:459-462.

Ayaz FA, Demir O, Torun H, Kolcuoglu Y, Colak A. 2008. Characterization of polyphenoloxidase (PPO) and total phenolic contents in medlar (*Mespilus germanica* L.) fruit during ripening and over ripening. Food Chemistry 106:291-298.

Babu US, Sommers K, Harrison LM, Balan KV. 2012. Effects of fructooligosaccharide-inulin on Salmonella-killing and inflammatory gene expression in chicken macrophages. Vet Immunol Immunop 149:92-96.

Baldwin EA, Nisperos-Carriedo MO, Baker RA. 1995. Use of edible coatings to preserve quality of lightly (and slightly) processed products. Crit Rev Food Sci Nutr 35:509–524.

Barnes HM, Feldman JR, White WV. 1950. Isochlorogenic acid. Isolation from coffee and structure studies. J Am Chem Soc 72:4178-4182

Bevilacqua MD, Amore A, and Polonara F. 2004. A multi-criteria decision approach to choosing the optimal blanching–freezing system. J Food Eng 63: 253-263.

Bizzarri G, Andreotti R, Massini R. 1981. Enzyme inactivation in spinach and potato: Catalase, peroxidase and phenolase. Ind Conserve 56:97-102.

Boun HR, Huxsoll CC. 1991. Control of Minimally Processed Carrot (Daucus carota) Surface Discoloration Caused by Abrasion Peeling. J Food Sci 56:416–418.

Brako L, Zarucchi JL. 1993. Catalogue of the flowering plants and gymnosperms of Peru. Missouri Botanical Garden, St. Louis, USA.

Böhm A, Kaiser I, Trebstein A, Henle T. 2005. Heat-induced degradation of inulin. Eur Food Res Technol 220:466-471.

Butler G, Rivera D. 2004. Innovations in Peeling Technology for Yacon. International Potato Center. Available from: http://cipotato.org/artc/cip_crops/1919-Yacon_Syrup.pdf. Accessed 2012 Aug 7

Cabezas MJ, Rabert C, Bravo S, Shene C. 2002. Inulin and Sugar Contents in *Helianthus tuberosus* and *Cichorium intybus* Tubers: Effect of Postharvest Storage Temperature. J Food Sci 67: 2860–2865.

Campos D, Betalleluz-Pallardel I, Chirinos R, Aguilar-Galvez A, Noratto G, Pedreschi R. 2012. Prebiotic effects of yacon (*Smallanthus sonchifolius* Poepp. & Endl), a source of fructooligosaccharides and phenolic compounds with antioxidant activity. Food Chem 135:1592–1599.

Cano MP, Lobo MG, de Ancos B, Galeazzi, MAM. 1996. Polyphenol oxidase from Spanish hermaphrodite and female papaya fruits (Carica papaya Cv. Sunrise, Solo Group). J Agr Food Chem 44:3075-3079.

Cano MP, Hernandez A, Ancos BD. 1997. High pressure and temperature effects on enzyme inactivation in strawberry and orange products. J Food Sci 62:85-88.

Cano P, Marin MA, Fuster C. 1990. Freezing of banana slices. Influence of maturity level and thermal treatment prior to freezing. J Food Sci 55:1070-1072

Centers for Disease Control and Prevention. 2011. Obesity – at a glance. Available from: http://www.cdc.gov/chronicdisease/resources/publications/aag/pdf/2011/Obesity_AAG_WEB_508.pdf. Accessed 2011 July.

Chan HT, Linse E. 1989. Conditioning cucumbers for quarantine heat treatments. HortScience 24, 985 – 989.

Cho YK, Ahn HK, 1999. Purification and characterization of polyphenol oxidase from potato. I. Purification and properties. J Biochem-Tokyo 23:577-592.

CIE. 1986. Colorimetry (2nd ed.). Wien, Austria: CIE Central Bureau Kegelgasse. CIE Publication 15.2.

Cortes H, Pardio V, Garcia M. 1999. Color parameter changes in banana slices during osmotic dehydration. Dry Technol 17:955-960.

Dykes L, Rooney LW. 2007. Phenolic compounds in cereal grains and their health benefits. Cereal Food World 52:105-111.

Embs RJ, Markakis P. 1965. The Mechanism of Sulfite Inhibition of Browning Caused by Polyphenol Oxidase. J Food Sci 30: 753–758.

Eshtiaghi, MN, Knorr D. 1993. Potato Cubes Response to Water Blanching and High Hydrostatic Pressure. J Food Sci 58:1371–1374.

FAO. 2007. *Polymnia sonchifolia.* Available from: http://ecocrop.fao.org/ecocrop/srv/en/cropView?id=8852. Accessed 2012 Jun.

Fante L, Scher C, Norena C, Rios A. 2012. Study of enzyme inactivation using steam in yacon (*Smallanthus sonchifolius*) roots. J Food Process Pres 37:16-24.

Foca G, Masino F, Antonelli A, Ulrici A. 2011. Prediction of compositional and sensory characteristics using RGB digital images and multivariate calibration techniques. Anal Chim Acta 706:238-245.

Fujita S, Tono T. 1988. Purification and some properties of polyphenoloxidase in eggplant (Solanum melongena). J Sci Food Agr 46:115-123.

Fujita S, Tono T, Kawahara H. 1991. Purification and properties of polyphenol oxidase in head lettuce (Lactuca sativa). J Sci Food Agr 55:643-651.

Galeazzi MAM, Sgarbieri VC, Constantinides, SM. 1981. Isolation, Purification and Physicochemical Characterization of Polyphenoloxidases (PPO) from a Dwarf Variety of Banana (*Musa cavendishii*, L). J Food Sci 46:150–155.

Garcia EL, Barrett DM, Lamikanra O. 2002. Preservative treatments for fresh-cut fruits and vegetables. Sci Technol Market 267–303

Gardiner WP, Gettinby G. 1998. Experimental design techniques in statistical practice: A practical software-based approach. Horwood, England: Horwood Publishing Limited 370p.

Genta SCW. 2009. Yacon syrup: Beneficial effects on obesity and insulin resistance in humans. Clin Nutr 28:182–187.

Genta SB, Cabrera WM, Grau A, Sanchez SS. 2005. Subchronic 4-monthoral toxicity study of dried Smallanthus sonchifolius (yacon) roots as a diet supplement in rats. Food Chem Toxicol 43:1657–1665.

Gonazlez-Martinez G, Ahrne L, Gekas V, Sjoholm I. 2004. Analysis of temperature distribution in potato tissue during blanching and its effect on the absolute residual pectin methylesterase activity. J Food Eng 65:433–441.

Granato D, Castro IA, Piekarski FVB, Beninca C, Masson ML. 2011. Influence of passion fruit juice on colour stability and sensory acceptability of non-sugar Yacon-based pastes. Braz Arch Biol Techn 54:147-159.

Guilbert S. 1986. Technology and application of edible protective films. In: Matholouthi, M. (Ed.), Food Packaging and Preservation: Theory and Practice. Elsevier Applied Science Publishing Co., London 371–393.

Hallfrisch J. 1990. Metabolic effects of dietary fructose. Faser J 4:2652-60

Hermann M, Freire I, Pazos C. 1999. Compositional diversity of the yacon storage root, in: Impact on a changing world. International Potato Centre (CIP), Lima (Peru) 425–432. Avilable from http://www.cipotato.org/market/PgmRprts/pr97-98. Accessed 2013 Jun 12.

Hill B, Roger T, Vorragen F. 1997. Comparative Analysis of the Quantization of Color Spaces on the Basis of the CIELAB Color-Difference Formula. ACM T GRAPHIC 16:109-154.

Hsu CL, Chen W, Weng YM, Tseng CY. 2003. Chemical composition, physical properties, and antioxidant activities of yam flours as affected by different drying methods. Food Chem 83:85-92.

Huang YH, Picha DH, Kilili AW, Johnson CE. 1999. Changes in Invertase Activities and Reducing Sugar Content in Sweetpotato Stored at Different Temperatures. J Agr Food Chem 47:4927-4931.

Hughes JC, Evans JL. 1967. Studies on after-cooking blackening in potatoes. IV. Field experiments. Eur Potato J 10:16–36.

Im JS, Parkin KL, Elbe JH. 1990. Endogenous polyphenoloxidase activity associated with the “black ring” defect in canned beet (*Beta vulgaris* L.) root slices. J Food Sci 55: 1042-1045

Jackson JC, Bourne MC, Barnard J. 1996. Optimization of blanching for crispness of banana chips using response surface methodology. J Food Sci 61:165-166.

Jacobi KK, Wong LS, Giles JE, 1996. Postharvest quality of zucchini (*Cucurbita pepo* L.) following high humidity hot air disinfestation treatments and cool storage. Postharv Biol Technol 7:309 – 316

Jenkins DJ, Kendall CW, Faulkner D, Vidgen E, Trautwein EA. 2002. A dietary portfolio approach to cholesterol reduction: combined effects of plant sterols, vegetable proteins, and viscous fibers in hypercholesterolemia. Metabolism 51: 1596-1604.

Jeong HL, Jin WJ, Kwang DM, Kee JP. 2008. Effects of anti-browning agents on Polyphenoloxidase Activity and Total Phenolics as Related to Browning of Fresh-Cut 'Fuji' Apple. ASEAN Food J 15:79-87.

Jiang Y, Fu J. 1998. Inhibition of polyphenol oxidase and the browning control of litchi fruit by glutathione and citric acid. Food Chem 62:49-52.

Jiang, Y. 2000. Role of anthocyanins, polyphenol oxidase and phenols in lychee pericarp browning. J Sci Food Agr 80:305-310.

Ju Z, Bramlage WJ. 1999. Phenolics and lipid-soluble antioxidants in fruit cuticle of apples and their antioxidant activities in model systems. Postharvest Biol Tec 16:107-118.

Kader F, Rovel B, Girardin M, Metche M. 1997. Mechanism of Browning in Fresh Highbush Blueberry Fruit (*Vaccinium corymbosum* L). Role of Blueberry Polyphenol Oxidase, Chlorogenic Acid and Anthocyanins. J Sci Food Agric 74: 31–34.

Kermasha S, Goetghebeur M, Monfette A, Metche M, Rovel B. 1993. Studies on inhibition of mushroom polyphenol oxidase using chlorogenic acid as substrate. J Sci Food Agr 41:526-531.

Klein JD, Lurie S. 1990. Prestorage heat treatment as a means of improving poststorage quality of apples. J Amer Soc Hort Sci 115:265-269.

Klein JD, Lurie S. 1992. Heat treatments for improved postharvest quality of horticultural crops. Hortechnology 2:316-320.

Kim DM, Smith NL, Lee CY. 1993. Apple cultivar variations in response to heat treatment and minimal processing. J Food Sci 58:1111-1114.

Kim DO, Padilla-Zakour OI. 2004. Jam Processing Effect on Phenolics and Antioxidant Capacity in Anthocyanin-rich Fruits: Cherry, Plum, and Raspberry. J Food Sci 69:395-S400.

Kim T, Silva J, Weng W, Chen W, Corbitt M, Jung Y., & Chen, Y. 2009. Inactivation of *Enterobacter sakazakii* by water-soluble muscadine seed extracts. Int J Food Microbiol 129:295-299

Klein JD, Lurie S. 1992. Heat treatments for improved postharvest quality of horticultural crops. HortTechnology 2:316-320.

Koukounaras A, Diamantidis G, Sfakiotakis E. 2008. The effect of heat treatment on quality retention of fresh-cut peach. Postharvest Biol Tec 48:30-36

Kumar VBA, Mohan TCK, Murugan K. 2008. Purification and kinetic characterization of polyphenol oxidase from Barbados cherry (*Malpighia glabra L.*). Food Chem 110:328-333.

Lachman J, Fernandez EC, Orsak M. 2003. Yacon (Smallanthus sonchifolia [Poepp. Et Endl.] H. Robinson) chemical composition and use – a review. Plant Soil Environ 49: 283–290.

Lee CY, Kagan V, Jaworski AW, Brown SK. 1990. Enzymic browning in relation to phenolic compounds and polyphenoloxidase activity among various peach cultivars. J Agr Food Chem 38:99-101.

Lee, H. 2000. HPLC analysis of phenolic compounds. In: Nollet, LML, editor. Food Analysis by HPLC. New York: Marcel Dekker, Inc. 775-824.

Lee JY, Park HJ, Lee CY, Choi WY. 2003. Extending shelf-life of minimally processed apples with edible coatings and antibrowning agents. Lebensm Wiss Technol 36:323–329

Lomax AR, Calder PC. 2009. Prebiotics, immune function, infection and inflammation: a review of the evidence. Brit J Nutr 101:633-658.

Lhomme C, Puigserver A, Biagini A. 2003. Effect of food-processing on the degradation of fructooligosaccharides in fruit. Food Chem 82:533-537.

Lichter A, Dvir O, Rot I, Akerman M, Regev R, Wiseblum A, Fallik E., Zauberman G, Fuchs Y. 2000. Hot water brushing: an alternative method to SO2 fumigation for color retention of litchi fruit. Postharvest Biol. Tec 18, 235–244

Liu, FW. 1978. Modification of apple quality by high temperature. J Amer Soc Hort Sci. 103:730-732.

Loaiza-Velarde JG, Mangrich ME, Campos-Vargas R. Saltveit ME. 2003. Heat shock reduces browning of fresh-cut celery petioles. Postharvest Biol Tec 27:305–311

Lobo, A. R. 2007. Effects of fructans-containing yacon (Smallanthus sonchifolius Poepp & Endl.) flour on caecum mucosal morphometry, calcium and magnesium balance, and bone 435 calcium retention in growing. Brit J Nutr 97:776-785.

Loiselle F, Tai GCC, Christie, BR. 1990. Genetic components of chip color evaluated after harvest, cold storage and reconditioning. Am Potato J 67:633–646.

Lurie, S. 1998. Postharvest heat treatments. Postharvest Biot Tec14:257-269.

Lurie S, Klein JD, Ben-Arie R. 1990. Postharvest heat treatment as a possible means of reducing superficial scald of apples. J Hort Sci 65:503–509.

Manrique I, Parraga A, Hermann M. 2004. Yacon syrup principles and processing. International Potato Center (CIP), Lima Peru. Avilable from http://cipotato.org/publications/pdf/002250.pdf Accessed 2012 Nov 21.

Manzocco L, Rumignani A, Lagazio C. 2012. Use of images in shelf life assessment of fruit salad. J Food Sci 77:258–262

Marinova D, Ribarova F, Atanassova M. 2005. Total phenolics and total flavonoids in Bulgarian fruits and vegetables. J U Chem Tech Metall, 40(3), 255-260.

Mayer-Miebach E, Spieß W. 2003. Influence of cold storage and blanching on the carotenoid content of Kintoki carrots. J Food Eng 56:211-213.

Modler HW, Jones JD, Mazza G. 1993. Observations on long-term storage and processing of Jerusalem artichoke tubers (*Helianthus tuberosus*). Food Chem 48:279–284

Mondy NI, Gosselin B. 1988. Effect of peeling on total phenols, total glycoalkaloids, discoloration and flavor of cooked potatoes. J Food Sci 53:756–759

Mark EM. 1949. Advances in food research. Academic Press 2:330-331.

Müftügil N. 1985. The peroxidase enzyme activity of some vegetables and its resistance to heat. J Sci Food Agric 36: 877–880

Murata M, Kurokami C, Homma S, Matsuhashi, C. 1993. Immunochemical and immunohistochemical study of apple chlorogenic acid oxidase. J Agr Food Chem 41:1385-1390.

Muthukumar M, Sargunamani D, Selvakumar N, Venkata Rao J. 2004. Optimisation of ozone treatment for colour and COD removal of acid dye effluent using central composite design experiment. Dyes Pigments 63:127-134.

Namiki M, Yabuta G, Koizumi Y, Yano, M. 2001. Development of free radical products during the greening reaction of caffeic acid esters (or chlorogenic acid) and a primary amino compound. Biosci Biotech Bioch 65:2131-2136.

Nasirizadeh N, Dehghanizadeh H, Yazdanshenas M, Karimi A, Moghadam M. 2012. Optimization of wool dyeing with rutin as natural dye by central composite design method. Ind Crop Prod 40:361-366.

Ndiaye C, Xu SY, Wang Z. 2013. Steam blanching effect on polyphenoloxidase, peroxidase and colour of mango (*Mangifera indica L.*) slices. Food Chem 11:392-95

Neves VA, Da Silva MA. 2007. Polyphenol oxidase from yacon roots (*Smallanthus sonchifolius*). J Agr Food Chem 55:2424-2430.

Niness KR. 1999. Inulin and oligofructose: what are they? J Nutr, 129 :1402–1406

Ohnishi M, Morishita H, Iwahashi H, Toda S, Shirataki Y, Kimura M, Kido R. 1994. Inhibitory effects of chlorogenic acids on linoleic acid peroxidation and haemolysis. Phytochemistry 36:579-583.

Ohyama T, Ito O, Yasuyoshi S, Ikarashi T, Minamisawa K, Kubota M, Asami T. 1990. Composition of storage carbohydrate in tuber roots of Yacon (*Polymnia sonchifolia*). Soil Sci Plant Nutr 36:67–171.

Ojansivu I, Ferreira CL, Salminen S. 2011. Yacon, a new source of prebioticoligosaccharides with a history of safe use. Trends Food Sci Tech 22:40–46.

Oszmianski J, Lee CY. 1990. Enzymic oxidative reaction of catechin and chlorogenic acid in a model system. J Agr Food Chem 38:1202-1204.

Ozoglu H, Bayind A. 2002. Inhibition of enzymic browning in cloudy apple juice with selected antibrowning agents. Food Control 13:213-221.

Park JS, Yang JS, Hwang, BY, Yoo BK. 2009. Hypoglycemic effect of yacon tuber extract and its constituent, chlorogenic acid, in streptozotocininduced diabetic rats. Biomolecules & Therapeutics 17:256–262.

Park YK, Sato HH, Almeida TD, Moretti RH. 1980. Polyphenol oxidase of mango (Mangifera indica var. Haden). J Food Sci 45:1619-1621

Paull RE. Chen NJ. 1990. Heat shock response in field grown ripening papaya fruit. J. Am Soc Hort Sci 115,623–631

Pedreschi F, Moyano P, Kaack K, Granby K. 2005. Color changes and acrylamide formation in fried potato slices. *Food Research International*, *38*(1), 1-9.

Pedreschi R, Campos D, Noratto G, Chirinos R, Cisneros Zevallos L. 2003. Andean yacon root (*Smallanthus sonchifolius Poepp.Endl*) fructooligosaccharides as a potential novel source of prebiotics. J Agric Food Chem 51:5278-5284.

Rejano L., Sanchez, A. H., Castro, A. D., & Montano, A. (1997). Chemical characteristics and storage stability of pickled garlic prepared using different processes. J Food Sci 62:1120-1123.

Peng L, Jiang Y. 2004. Effects of heat treatment on the quality of fresh-cut Chinese water chestnut. Int J Food Sci Tech 39:143-148.

Perez-Gago MB, Serra M, Río MD. 2006. Color change of fresh-cut apples coated with whey protein concentrate-based edible coatings. Postharvest Biol Tec 39:84-92

Pizzocaro F, Torreggiani D, Gilardi G. 1993. Inhibition of apple polyphenoloxidase (PPO) by ascorbic acid, citric acid and sodium chloride. J Food Process Pres 17:21-30.

Polreich S. 2003. Establishment of a Classification Scheme to structure the Post-Harvest Diversity of Yacon Storage Roots (*Smallanthus sonchifolius* (Poepp. & Endl.) H. Robinson) University of Kassel. Thesis

Pomenta JV, Burns EE. 1971. Factors affecting chlorogenic, quinic and caffeic acid levels in sunflower kernels. J Food Sci 36:490-492.

Prusky D, Fuchs Y, Kobiler I, Roth I, Weksler A, Shalom Y, Fallik E, Zaurberman G, Pesis, E, Akerman M, Yekutieli O, Wiseblum A, Regev R, Artes L. 1999. Effect of hot water brushing, prochloraz treatment and waxing on the incidence of black spot decay caused by Alternaria alternate in mango fruit. Postharvest Biol Tec 15:165–174

Quinteros, ET. 2000. Produção com tratamento enzimático e avaliação do suco de Yacon. PhD Thesis, Universidade Estadual de Campinas, Campinas, Brasil.

Raul L, Ricardo AB, Enrique RS. 1984. Effect of soaking-blanching conditions on glucose losses in potato slices. Can I Food Sc Tech J. 17:111-113.

Reis F, Masson M, Waszczynskyj N. 2008. Influence of a blanching pretreatment on color oil uptake and water activity of potato sticks, and its optimization. J Food Process Eng 31:833-852.

Rejano L, Sanchez AH, Castro AD., Montano A. 1997. Chemical characteristics and storage stability of pickled garlic prepared using different processes. J Food Sci 62:1120-1123.

Renuka B, Kulkarni SG, Vijayanand P, Prapulla SG. 2009. Fructooligosaccharide fortification of selected fruit juice beverages: Effect on the quality characteristics. LWT-Food Sci Tech 42:1031-1033.

Ridder H, Blommaert FJ, Fedorovskaya EA. 1995. Naturalness and image quality: chroma and hue variation in color images of natural scenes. In Proc. SPIE 2411: 51-61.

Rish B, Herrmann K. 1988. Die Gehalte Hydroxyzimtsare-verbindungen und Catechinen in Kern und Steinobst. Zeitsch-rift fur Lebensmittal – Untersuchung Und – Forschung 186:225–230.

Roberfroid M. 1993. Dietary fiber, inulin, and oligofructose: a review comparing their physiological effects. Critical Reviews in Food Science & Nutrition, 33: 103-148.

Roberfroid M, Slavin L. 2000. Nondigestible oligosaccharides. Crit Rev Food Sci Nutr 40:461–480

Rojas-Graü MA, Soliva-Fortuny R, Martín-Belloso O. 2009. Edible coatings to incorporate active ingredients to fresh-cut fruits: a review. Trends Food Sci Tech 20:438-447.

Rossi M, Nani R, Torreggiani D, Scalzo R, Giussani E, Morelli R. 2003. Effect of fruit blanching on phenolics and radical scavenging activity of highbush blueberry juice. Food Res Int 36:999-1005.

Roussos PA, Pontikis CA. 2001. Phenolic compounds in olive explants and their contribution to browning during the establishment stage in vitro. Gartenbauwissenschaft 66: 298-303.

Sabir, MA, Sosulski FW, Finlayson A. 1974. Chlorogenic acid-protein interactions in sunflower. J Agric Food Chem 22: 575-578.

Saltveit ME. 2000. Wound induced changes in phenolic metabolism and tissue browning are altered by heat shock. Postharvest Biol Tec 21:61–69

Scher CF, Rios AO, Norena CPZ. 2009. Hot air drying of yacon (*Smallanthus sonchifolius*) and its effect on sugar concentrations. Int J Food Sci Technol 44: 2169–2175.

Severini C, Baiano A, Pilli T, Romaniello R, Derossi A. 2003. Prevention of enzymatic browning in sliced potatoes by blanching in boiling saline solutions. *LWT-Food Science and Technology*, *36*(7), 657-665.

Seymour GB, John P, Thompson AK. 1987. Inhibition of degreening in the peel of bananas ripened at tropical temperatures. II. Role of ethylene, oxygen and carbon dioxide. Ann Appl Biol 110:153 – 161

Siddiq M, Cash JN, Sinha, NK, Akhter P. 1993. Characterization and inhibition of polyphenol oxidase from pears (Pyrus communis L. cv. Bosc and Red). J Food Biochem 17:327-337

Silva GH, Chase RW, Hammerschmidt R, Cash JN. 1991. After-cooking darkening of Spartan Pearl potatoes as influenced by location, phenolic acids, and citric acid. J Agr Food Chem 39:871-873.

Simonovska B, Vovk I, Andrensek S, Valentová K, Ulrichová J. 2003. Investigation of phenolic acids in yacon (*Smallanthus sonchifolius*) leaves and tubers. J Chromatogr A 1016:89-98.

Singh RP, Singh M, King RR. 1998. Use of citric acid for neutralizing polymerase chain reaction inhibition by chlorogenic acid in potato extracts. J Virol Methods 74:231-235.

Singleton VL. 1965. Colorimetry of total phenolics with phosphomolybdic-phosphotungstic acid reagents. Am J Enol Viticult 16:144-158.

Steinbuch E. 1976. Technical Note: Improvement of texture of frozen vegetables by stepwise blanching treatments. Int J Food Sci Tech. 11: 313–316.

Stewart ML, Timm DA, Slavin JL. 2008. Fructooligosaccharides exhibit more rapid fermentation than long-chain inulin in an in vitro fermentation system. Nutrition Research, 28:329-334.

Sun J, Chu YF, Wu X, Liu RH. 2002. Antioxidant and antiproliferative activities of common fruits. J Agr Food Chem: 7449-7454.

Takeuchi J, Nagashima T. 2011. Preparation of dried chips from Jerusalem artichoke (*Helianthus tuberosus*) tubers and analysis of their functional properties. Food Chem 126:922-926

Toivonen P, Brummell DA. 2008. Biochemical bases of appearance and texture changes in fresh-cut fruit and vegetables. Postharvest Biol Tec 48:1-14.

Unal MU. 2007. Properties of polyphenol oxidase from Anamur banana (*Musa cavendishii*). Food Chem 100:909-913.

Valentová K, Lebeda A, Dolezalová I, Jirovský D, Simonovska B, Vovk I, ... & Ulrichová J. 2006. The biological and chemical variability of yacon. J Agr Food Chem 54: 1347-1352.

Vámos-Vigyázó L. 1981. Polyphenol oxidase and peroxidase in fruits and vegetables Critical Reviews in Food Science and Nutrition, 15:49–127

Van Loo J, Coussement P, Leenheer L, Hoebregs H, Smits G. 1995. On the presence of inulin and oligofructose as natural ingredients in the western diet. Crit Rev Food Sci 35: 525-552

Van Loo J, Coussement P, Leenheer L, Hoebregs H, Smits G. 1995. On the presence of inulin and oligofructose as natural ingredients in the western diet. Crit Rev Food Sci 35: 525-552.

Vaughn KC, Duke SO. 1984. Function of polyphenol oxidase in higher plants.Physiol Plant 60:106-112.

Walter WM, Giesbrecht FG. 1982. Effect of lye peeling conditions on phenolic destruction, starch hydrolysis, and carotene loss in sweet potatoes. J Food Sci 47:810-812.

Wang L, Kim D, Park J. 2003. Various antibrowning agents and green tea extract during processing and storage. J Food Process Pres 27:213–225

Waterhouse AL. 2001. Determination of total phenolics. In: Wrolstad, RE, editor. Current Protocols in Food Analytical Chemistry. NY: John Wiley & Sons, Inc I1-I8.

Whitaker JR, Lee CY. 1995. Recent advances in chemistry of enzymatic browning. In Enzymatic browning and its prevention. Washington DC: American Chemistry Society, Symposium Series 600:2-7

Weaver WW. 2006. Yummy Yacon. Mother Earth News. Avilable from http://www.motherearthnews.com/organic-gardening/yummy-yacon.aspx#axzz2XiP8edtC Accessed 2012 Nov 21.

Wissemann KM, Lee CY. 1981. Characterization of polyphenoloxidase from Ravat 51 and Niagara grapes. J Food Sci 46:506–508.

Wuyts N, De Waele D, Swennen R. 2006. Extraction and partial characterization of polyphenol oxidase from banana (*Musa acuminata Grande naine*) roots. Plant Physiol Bioch 44:308-314.

Xin Y, Yong T. 2010. Blanching Color Protection of Yacon by Orthogonal Experimental Design. Academic Periodical of Farm Products Processing 11:010.

Yabuta G, Koizumi Y, Namiki K., Hida M, Namiki, M. 2001. Structure of Green Pigment Formed by the Reaction of Caffeic Acid Esters (or Chlorogenic acid) with a Primary Amino Compound. Biosci Biotech Bioch 65:2121-2130

Yan X, Suzuki M, Ohnishi-Kameyama M, Sada Y, Nakanish T, Nagat, T. 1999. Extraction and identification of antioxidants in the roots of yacon (*Smallanthus sonchifolius*). J Agric Food Chem 47:4711-4713.

Yang CP, Fujita S, Ashrafuzzaman MD, Nakamura N, Hayashi N. 2000. Purification and characterization of polyphenol oxidase from banana (Musa sapientum L.) pulp. JAgr Food Chem48:2732-2735.

Yang M, Liu X, Huang J, Zhan H. 2012. Vacuum sugar process system optimization yacon. Academic Periodical of Farm Products Processing 2012:01

Yemenicioglu A. 2002. Control of polyphenol oxidase in whole potatoes by low temperature blanching. European Food Research and Technology. Eur Food Res Technol 214:313-319.

Yemenicioglu A, Ozkan M, Cemeroglu B. 1997. Heat inactivation kinetics of apple polyphenoloxidase and activation of its latent form. J Food Sci 62:508–510.

Yoruk R, Marshall MR. 2003. Physicochemical properties and function of plant polyphenoloxidase. J Food Bioch 27:361-422.

Yoshida M, Ono H, Mori Y, Chuda Y, Mori M. 2002. Oxygenation of Bisphenol A to Quinones by Polyphenol Oxidase in Vegetables. J Agr Food Chem 50:4377-4381.

APPENDIX A

RAW DATA, STATISTIC ANALYSIS RESULTS, A SENSORY APPEARANCE RATING SAMPLE, AND STANDARD CURVES

Table A.1 Result from different roots on optimization of blanching treatment for yacon (raw data).

Central Composite Design for yacon blanching (three factors)																							
Run	Natural variables			Coded variables			Responses (Polyphenol oxidase results of yacon)				Responses (y2-1) (Whiteness)				Responses (y2) (TCD)				Responses (y3) (Sensory Rating Score)			Responses (y3) (Sensory Rating %)		
	Temperature (C) (boiling) water	Time (min)	Concentration (citric acid) %	x1	x2	x3	y1 (5min)	y1 (6min)	y1 (7min)	y1 (10min)	Whiteness (3 set data)	2 days	6 days	10 days	Total color (3 set data)	2 days	6 days	10 days	2 days	6 days	10 days	2 days	6 days	10 days
1	84	2.8	0.04	-1	-1	-1	0.06	0.06	0.05	0.05	18.4	18.4	13.7	20.8	9.09	9.09	13.9	7.13	2.8	2.5	2.2	94	96	96
	84	2.8	0.04	-1	-1	-1	0.06	0.06	0.05	0.05	13.7				13.9									

Table A.1 continued

	84	2.8	0.04	-1	-1	-1	0.06	0.06	0.06	0.06	20.8				7.13								
2	84	2.8	0.16	-1	-1	1	0.06	0.06	0.06	0.06	22.0	22.0	16.0	21.8	5.60	5.60	12.0	6.51	2.8	2.7	2.3	92	94	94
	84	2.8	0.16	-1	-1	1	0.06	0.06	0.05	0.05	16.0				12.0								
	84	2.8	0.16	-1	-1	1	0.06	0.06	0.06	0.05	21.8				6.51								
3	84	5.2	0.04	-1	1	-1	0.04	0.04	0.04	0.04	17.6	17.6	15.3	21.6	9.85	9.86	12.4	6.70	2.8	2.7	2.7	92	94	94
	84	5.2	0.04	-1	1	-1	0.07	0.06	0.06	0.05	15.3				12.4								
	84	5.2	0.04	-1	1	-1	0.04	0.04	0.04	0.04	21.6				6.70								

Table A.1 continued

4	84	5.2	0.16	-1	1	1	0.04	0.03	0.03	0.03	27.1	27.1	21.0	28.3	2.48	2.48	8.17	6.55	4.0	3.6	3.2	92	96	96
	84	5.2	0.16	-1	1	1	0.04	0.03	0.03	0.03	21.0				8.17									
	84	5.2	0.16	n-1	1	1	0.03	0.03	0.03	0.02	28.3				6.55									
5	96	2.8	0.04	1	-1	-1	0.06	0.05	0.06	0.06	18.6	18.6	12.1	24.3	8.90	8.90	16.2	7.44	2.1	2.1	2.3	98	94	96
	96	2.8	0.04	1	-1	-1	0.06	0.05	0.05	0.05	12.1				16.2									
	96	2.8	0.04	1	-1	-1	0.06	0.06	0.05	0.05	24.3				7.44									

Table A.1 continued

6	96	2.8	0.16	1	-1	1	0.06	0.05	0.05	0.05	23.2	23.2	17.5	28.1	5.86	5.86	12.2	8.32	3.6	3.2	2.9	69	81	94
	96	2.8	0.16	1	-1	1	0.05	0.05	0.05	0.05	17.5				12.2									
	96	2.8	0.16	1	-1	1	0.06	0.05	0.05	0.05	28.1				8.32									
7	96	5.2	0.04	1	1	-1	0.04	0.04	0.04	0.03	22.7	22.7	15.9	25.5	5.39	5.39	13.8	7.91	2.5	2.7	2.6	89	92	92
	96	5.2	0.04	1	1	-1	0.05	0.04	0.04	0.04	15.9				13.8									
	96	5.2	0.04	1	1	-1	0.04	0.04	0.04	0.04	25.5				7.91									
8	96	5.2	0.16	1	1	1	0.04	0.04	0.04	0.04	28.9	28.9	22.4	31.3	3.35	3.35	8.23	8.73	4.0	4.0	3.8	58	58	65

Table A.1 continued

	96	5.2	0.16	1	1	1	0.04	0.04	0.04	0.04	22.4				8.23								
	96	5.2	0.16	1	1	1	0.04	0.04	0.04	0.03	31.3				8.73								
9	80	4	0.1	-1.682	0	0	0.06	0.06	0.05	0.05	30.9	30.9	25.3	31.7	3.96	3.96	5.55	7.30	4.6	3.8	3.5	3.8	69	71
	80	4	0.1	-1.682	0	0	0.06	0.05	0.05	0.05	25.3				5.55								
	80	4	0.1	-1.682	0	0	0.06	0.06	0.06	0.05	31.7				7.30								

Table A.1 continued

10	100	4	0.1	1.682	0	0	0.04	0.04	0.04	0.04	29.3	29.3	36.5	29.4	4.30	4.30	12.0	6.11	4.1	4.1	4.1	27	27	27
	100	4	0.1	1.682	0	0	0.04	0.03	0.03	0.04	36.5				12.0									
	100	4	0.1	1.682	0	0	0.04	0.04	0.04	0.04	29.4				6.11									
11	90	2	0.1	0	-1.682	0	0.08	0.07	0.07	0.07	21.3	21.3	14.5	21.8	6.72	6.72	14.2	7.25	2.7	2.9	2.9	83	75	77
	90	2	0.1	0	-1.682	0	0.09	0.07	0.07	0.07	14.5				14.2									

Table A.1 continued

	90	2	0.1	0	-1.682	0	0.09	0.07	0.06	0.07	21.8				7.25								
12	90	6	0.1	0	1.682	0	0.02	0.02	0.02	0.02	27.2	27.2	19.4	25.4	4.34	4.34	10.4	5.81	4.0	4.0	4.1	40	56	58
	90	6	0.1	0	1.682	0	0.03	0.03	0.03	0.03	19.4				10.4								
	90	6	0.1	0	1.682	0	0.04	0.04	0.03	0.03	25.4				5.81								
13	90	4	0	0	0	-1.682	0.05	0.04	0.04	0.04	19.2	19.2	10.3	24.4	8.09	8.09	17.4	6.97	2.1	2.1	1.8	92	94	98
	90	4	0	0	0	-1.682	0.06	0.05	0.04	0.04	10.3				17.4								

Table A.1 continued

	90	4	0	0	0	-1.682	0.04	0.04	0.04	0.04	24.4				6.97								
14	90	4	0.2	0	0	1.682	0.03	0.03	0.03	0.03	28.5	28.5	32.2	28.2	2.18	2.18	12.2	4.62	4.1	4.1	4.1	15	15	21
	90	4	0.2	0	0	1.682	0.03	0.03	0.03	0.02	32.2				12.2								
	90	4	0.2	0	0	1.682	0.03	0.03	0.03	0.03	28.2				4.62								
15	90	4	0.1	0	0	0	0.04	0.03	0.03	0.03	31.4	31.4	26.9	30.1	5.38	5.38	11.5	7.52	4.1	4.1	4.0	27	27	40
	90	4	0.1	0	0	0	0.03	0.04	0.04	0.04	26.9				11.5								
	90	4	0.1	0	0	0	0.04	0.04	0.04	0.04	30.1				7.52								

Table A.1 continued

16	90	4	0.1	0	0	0	0.04	0.04	0.04	0.04	34.7	34.7	28.6	21.8	9.12	9.12	5.24	6.17	3.4	3.1	2.5	81	83	96
	90	4	0.1	0	0	0	0.04	0.04	0.04	0.04	28.59				5.24									
	90	4	0.1	0	0	0	0.03	0.03	0.04	0.03	21.8				6.17									
17	90	4	0.1	0	0	0	0.04	0.04	0.04	0.04	33.8	33.8	31.5	28.4	8.09	8.09	9.17	8.65	3.8	3.8	3.6	52	52	70
	90	4	0.1	0	0	0	0.03	0.03	0.03	0.03	31.5				9.17									
	90	4	0.1	0	0	0	0.03	0.03	0.03	0.03	28.3				8.66									
18	90	4	0.1	0	0	0	0.04	0.04	0.04	0.04	33.6	33.6	27.9	24.4	9.04	9.04	5.83	7.41	4.0	3.4	2.5	50	75	96

Table A.1 continued

	90	4	0.1	0	0	0	0.04	0.04	0.04	0.04	27.9				5.83								
	90	4	0.1	0	0	0	0.04	0.04	0.04	0.04	24.4				7.42								
19	90	4	0.1	0	0	0	0.04	0.04	0.04	0.04	21.5	21.5	22.1	23.6	6.06	6.06	5.78	5.10	4.0	3.4	2.5	50	75	96
	90	4	0.1	0	0	0	0.04	0.04	0.04	0.04	22.1				5.78								
	90	4	0.1	0	0	0	0.03	0.03	0.03	0.03	23.6				5.10								
20	90	4	0.1	0	0	0	0.04	0.04	0.04	0.04	16.29	16.3	20.7	22.0	11.2	11.2	6.97	6.06	4.0	3.4	2.5	50	75	96
	90	4	0.1	0	0	0	0.04	0.03	0.03	0.03	20.7				6.98								

Table A.1 continued

	90	4	0.1	0	0	0	0.03	0.03	0.03	0.03	22.0				6.06								
Control							0.18	0.17	0.17	0.17													
							0.18	0.18	0.17	0.17													
							0.18	0.18	0.18	0.18													

Table A.2 P values for blanching variables effect on PPO activity, whiteness, total color difference, sensory rating, sensory browning %.

Source	PPO activity	Whiteness	Total Color Difference	Sensory Rating	Sensory Browning %
	P value	P value	P value	P value	P value
Constant	0.000	0.052	0.052	0.65	0.929
Temp	0.000	0.033	0.033	0.737	0.977
Time	0.001	0.237	0.237	0.944	0.962
Citric acid	0.372	0.325	0.325	0.367	0.326

Table A.3 Results of PPO activity, Color (L, a, b, whiteness, TCD, hue angle, chroma), Total phenolic compounds, and moisture from different roots on optimization of blanching treatment for yacon (raw data).

treatment	time	Rept	PPO	Color							Total phenolic compounds	Moisture	
			ΔABS/min	L	a	b	Whiteness	TCD	hue angle	chroma	mg/g in d.m.	Moisture%	D.M. (g)
Control 1	0	1	0.0215	31.0	1.5	9.4	30.3	5.9	80.8	9.5	16.37	90.86	0.46
Control 2	0	2	0.0195	31.5	1.5	9.2	30.8	4.0	80.7	9.3	16.76	91.31	0.43
water	1.5	1	0.0045	46.9	-3.3	6.6	46.3	16.7	117.2	7.4	8.73	91.21	0.44
water	1.5	2	0.0040	43.9	-1.4	7.2	43.4	13.2	100.7	7.3	6.15	90.73	0.46
water	3	1	0.0010	45.6	-2.8	5.4	45.2	15.5	117.4	6.0	6.33	90.89	0.46
water	3	2	0.0010	41.7	-3.0	4.6	41.3	12.3	123.4	5.5	6.64	89.77	0.51
water	4.5	1	0.0005	43.6	-2.6	4.8	43.3	14.0	119.5	5.5	4.46	88.36	0.58
water	4.5	2	0.0005	40.7	-2.6	6.0	40.3	11.0	113.6	6.6	5.62	88.51	0.57
water	6	1	0.0000	39.9	-2.4	5.1	39.6	10.5	117.0	5.7	6.81	92.01	0.40
water	6	2	0.0010	39.6	-2.5	5.5	39.3	10.4	115.0	6.1	6.41	91.08	0.45
water with citric acid	1.5	1	0.0040	40.0	-3.1	7.4	39.4	10.1	112.7	8.0	8.47	92.00	0.40
water with citric acid	1.5	2	0.0015	41.7	-2.5	3.7	41.5	12.5	123.7	4.4	7.92	87.18	0.64
water with citric acid	3	1	0.0010	43.5	-3.4	6.6	43.0	13.5	117.2	7.5	11.52	93.55	0.32
water with citric acid	3	2	0.0000	44.1	-2.7	4.8	43.8	14.4	121.9	5.5	10.61	92.44	0.38
water with citric acid	4.5	1	0.0000	47.4	-3.8	10.2	46.3	17.2	113.4	9.4	9.94	92.78	0.36
water with citric acid	4.5	2	0.0000	43.4	-3.1	4.5	43.2	13.9	125.0	5.5	9.35	92.92	0.35
water with citric acid	6	1	0.0000	39.8	-2.6	0.9	39.7	12.7	161.4	2.8	7.24	93.76	0.31
water with citric acid	6	2	0.0000	39.0	-2.4	1.2	38.9	11.9	153.3	2.7	6.36	93.26	0.34

Table A.4 Results of Chlorogenic acid, total sugars, inulin, sucrose, glucose, fructose from different roots on optimization of blanching treatment for yacon (raw data).

treatment	time	Rept	Chlorogenic Acid	Total Sugars (Inulin+Sucrose+Glucose+Fructose)	Inulin	Sucrose	Glucose	Fructose
			ppm in D.M.	% in D.M.	% in D.M.	% in D.M.	% in D.M.	% in D.M.
Control 1	0	1	13811.75	94.44	26.76	11.18	6.34	50.16
Control 2	0	2	12923.71	94.84	24.83	11.49	7.05	51.47
water	1.5	1	6212.49	70.79	2.26	10.40	10.69	47.44
water	1.5	2	5081.94	75.66	2.46	14.64	9.94	48.61
water	3	1	6053.38	54.47	3.80	9.34	8.11	33.22
water	3	2	5157.48	51.12	2.96	6.63	7.59	33.93
water	4.5	1	3627.21	32.32	1.56	3.72	3.71	23.33
water	4.5	2	5066.22	31.22	1.59	2.69	3.89	23.06
water	6	1	5548.03	27.39	1.14	2.76	3.72	19.77
water	6	2	5489.56	28.70	1.37	2.83	4.02	20.48
water with citric acid	1.5	1	7730.41	48.47	4.89	4.12	5.98	33.48
water with citric acid	1.5	2	5571.48	45.52	5.31	6.18	5.11	28.93
water with citric acid	3	1	8882.72	50.43	6.07	7.34	5.27	31.74
water with citric acid	3	2	9298.59	43.65	5.41	6.03	4.72	27.49
water with citric acid	4.5	1	7945.71	64.52	7.59	9.48	6.42	41.02
water with citric acid	4.5	2	7787.54	68.83	6.38	8.79	7.71	45.95
water with citric acid	6	1	6243.16	49.73	3.24	6.24	7.44	32.81
water with citric acid	6	2	6116.60	48.72	3.51	4.61	6.30	34.31

Table A.5 ANOVA Results for PPO activity, whitness, TCD, Hue angle, chroma, and total phenolics in experiment II.

Sorce	DF	PPO activity		Whiteness		Total Color Difference		Hue Angle		Chroma		Total Phenolics	
		F	P	F	P	F	P	F	P	F	P	F	P
Treatment	1	0.21	0.6608	0.21	0.6608	0.16	0.7022	14.28	0.0044	0.61	0.454	18.85	0.0019
Time	4	55.75	<.0001	55.75	<.0001	28.59	<.0001	43.61	<.0001	9.27	0.003	302.3	<.0001
Treatment * Time	4	3.2	0.0677	3.2	0.0677	3.15	0.0704	8.17	0.0046	2.24	0.1445	92.16	<.0001

R^2 values: 0.9908 (PPO activity), 0.9832 (whiteness), 0.9363 (total color difference), 0.9609 (Hue angle), 0.8462 (chroma), and 0.9848 (totlal phenolics)

Table A.6 ANOVA Results of PPO activity, whitness, TCD, Hue angle, chroma, and total phenolics in experiment II.

Sorce	DF	Chlorogenic acid		Total Sugars		Inulin		Sucrose		Glucrose		Fructose	
		F	P	F	P	F	P	F	P	F	P	F	P
Treatment	1	31.37	0.0003	18.85	0.0019	76.97	<.0001	0	0.9729	0.29	0.6046	8.55	0.0169
Time	4	84.7	<.0001	302.3	<.0001	947.69	<.0001	15.02	0.0005	13.15	0.0008	87.25	<.0001
Treatment * Time	4	5.71	0.0143	92.16	<.0001	9.01	0.0033	12.38	0.0011	34.13	<.0001	54.69	<.0001

R^2 values: 0.9777 (chlorogenic acid), 0.9944 (total sugars), 0.9977 (inulin), 0.9241 (sucrose), 0.9547 (glucose), and 0.9846 (fructose)

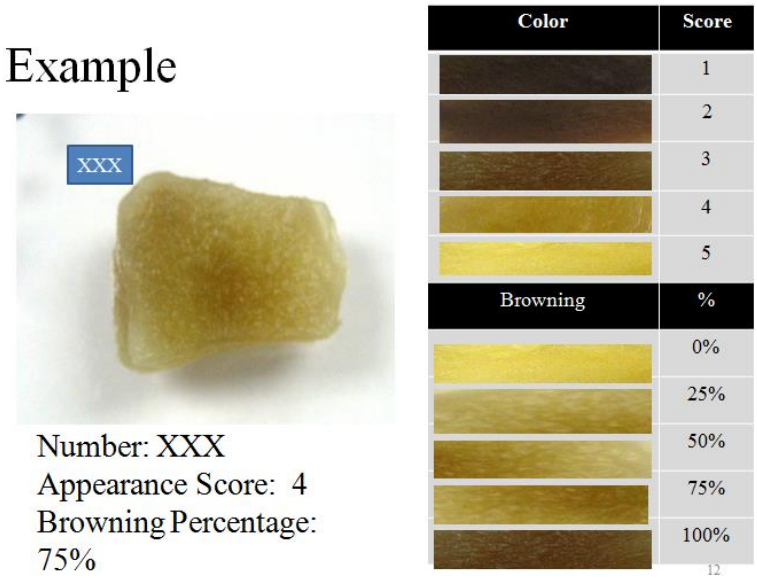


Figure A.1 Sensory rating score

Score where 1-dark brown, 2-red brown, 3-medium brown, 4-brown, 5-yellowish (original color), and sensory browning percentage were 0 %, 25 %, 50 %, 75 %, and 100 %.

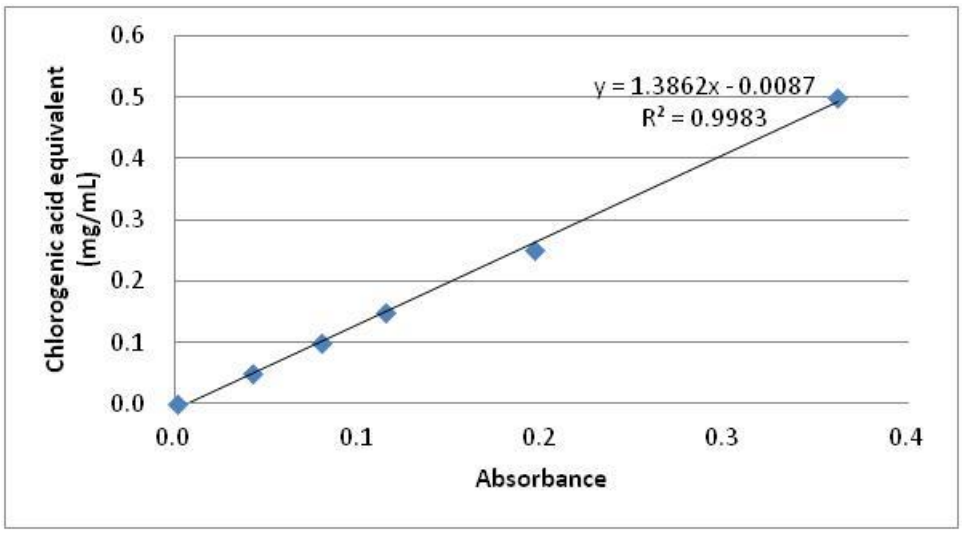


Figure A.2 Standard curve used for total phenolics calculation in Folin-Ciocalteu procedure.

Total phenolics were expressed as milligrams of chlorogenic acid equivalents per milliliter.

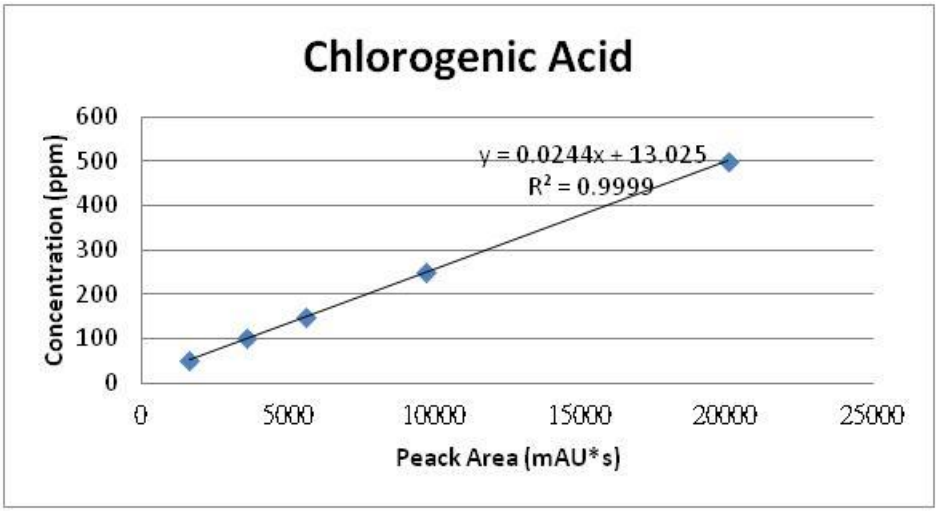


Figure A.3 Standard curve used to quantify chlorogenic acid in HPLC chromatograms

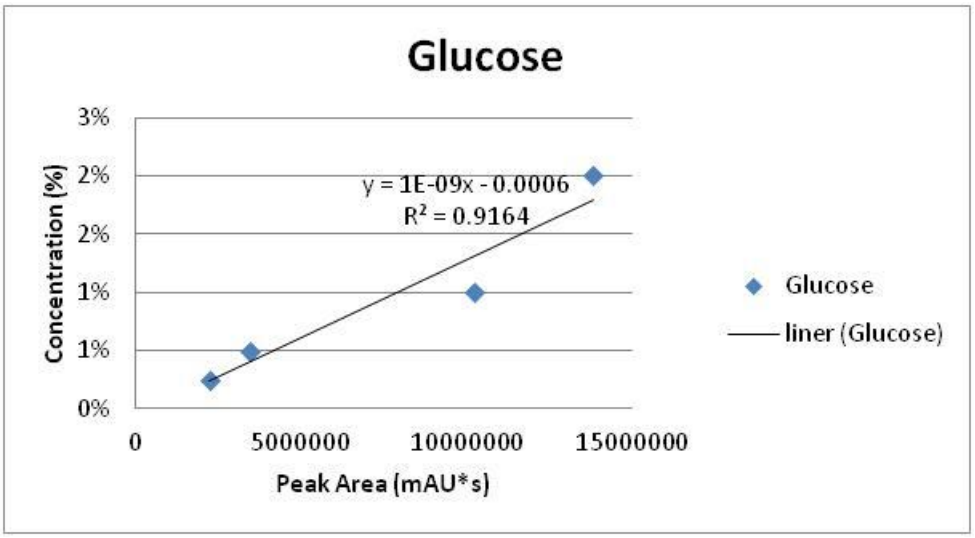


Figure A.4 Standard curve used to quantify chlorogenic acid in HPLC chromatograms

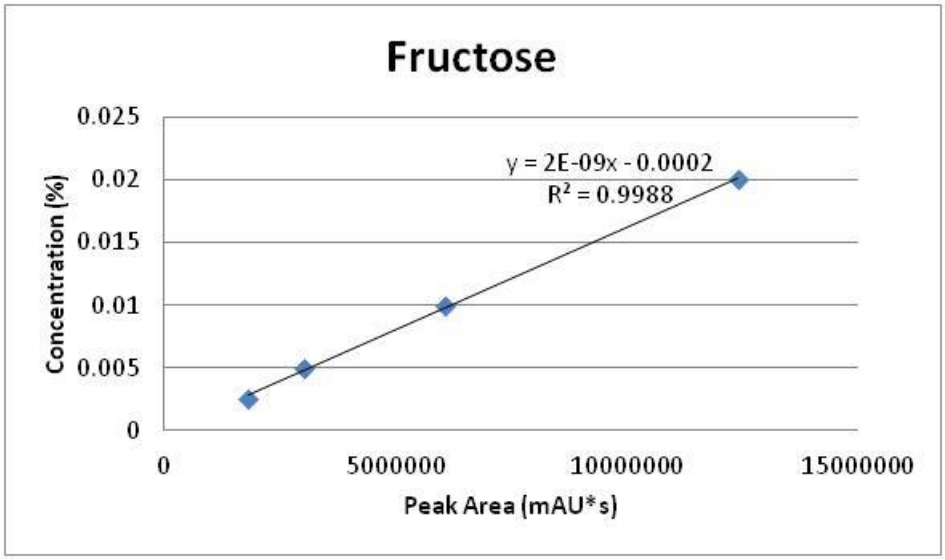


Figure A.5 Standard curve used to quantify chlorogenic acid in HPLC chromatograms

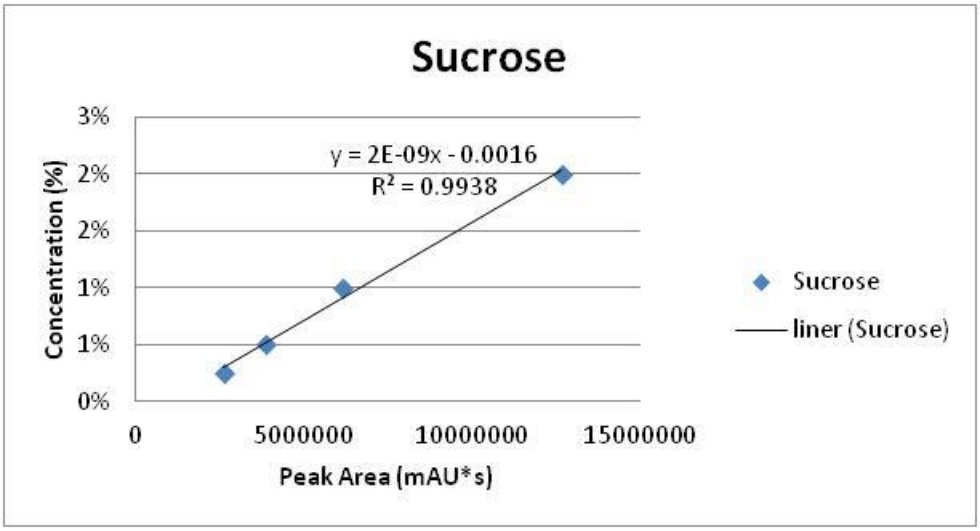


Figure A.6 Standard curve used to quantify chlorogenic acid in HPLC chromatograms

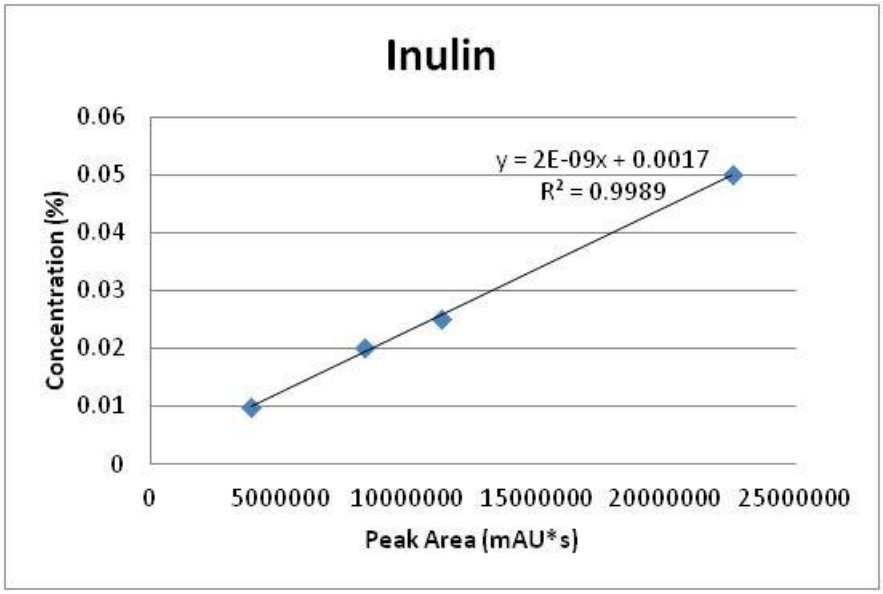


Figure A.7 Standard curve used to quantify chlorogenic acid in HPLC chromatograms