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Effect of high pressure treatment on thermal and rheological properties of lentil flour slurry

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ABSTRACT

Thermal characteristics of high pressure (HP) treated lentil flour dispersions were studied as function of pressure level (350–650 MPa) and moisture content (14–58 g per 100 g of flour). Differential scanning calorimeteric (DSC) measurement of pressure treated lentil dispersions indicated incomplete denaturation of lentil proteins. The protein dentauration temperature (T_d) shifted with applied pressure and moisture content non-systematically. No starch gelatinization peak was detected during thermal scanning of lentil slurries (untreated or treated) irrespective of moisture content or heating rate. High pressure treatment of lentil dispersion significantly reduced the retrogradation behavior compared with that obtained from the thermally gelatinized sample. Dynamic rheological measurement indicated pressure treated lentil slurries exhibited a true viscoelastic fluid. Slurries gradually transformed from solid-like behavior to liquid-like behavior as function of moisture content and pressure level. Fourier transform infrared (FTIR) spectroscopy confirmed insignificant change in amide band of pressure treated slurry. This study has provided complementary information of pressure-induced structural changes on both the molecular and the sub-molecular level of lentil protein.

1. Introduction

Lentils (*Lens culinaris*) are important crop in many developing countries. The Canadian legume industry has increased enormously in recent years and Canada becomes the largest exporter of lentils. Lentils contain about 25 g proteins, 56 g carbohydrate, and 1.0 g fat per 100 g seeds and these are considered as one of the best and cheapest sources of vegetable proteins (Adsule, Kadam, & Leung, 1989). Lentil flour has potential for traditional and newer product developments with health benefits since it contains higher amount of protein, gluten free in nature and low glycemic index. Lentil soup is consumed with cereal (rice or bread) to enrich protein intake in developing countries. Legume proteins are primarily storage proteins comprised of two principle globulins, legumin and vicilin (Swanson, 1990). Lentil protein isolate produced a milk of intermediate quality equivalent to milk prepared from soy protein isolates. Currently, lentil flours are incorporating to other flours for manufacturing extruded snacks and gluten free bakery products. It would be of practical interest to utilize legumes, for instance, in the formulation of manufactured foods for diabetics (Urbano et al., 1995). Both starch and protein fractions of lentils offer new source of novel ingredients. New sources of cheaper proteins provide new alternatives for the dairy industry, where cheaper proteins are required to replace existing proteins (Lee, Htoon, Uthayakumaran, & Paterson, 2007). Extensive research works have been carried out on cereal, potato, sweet potato and cassava starches due to their ready availability and wide usage in food and non-food applications. However, there is a little information available on structure–property relationship of lentil flour.

High pressure processing is considered one of the best emerging or non-thermal technologies for food. The technology has advantage over others in terms of functionality and consumers' acceptance for producing value-added fresh-like food products in addition to microbial and enzymatic inactivation. Numerous literatures are available on functionality of protein and carbohydrate foods under high hydrostatic pressure. High pressure treatment changes the conformation and coagulation of proteins by opening the native structures, resulting in denaturation and aggregation, affects the melting properties of starches; the rearrangement of the polymorphic forms in lipids; inactivates microorganisms, and induces chemical changes at low temperatures (Cheftel, 1992; Ledward, 1995; Lopez-Fandino, 2006). To determine the structural

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changes of proteins upon pressurization, Fourier transform infrared (FTIR) spectroscopy and differential scanning calorimetry (high sensitive micro-DSC) are commonly used tools. The information obtained by DSC is on a macroscopic level, it enables to assess the overall structure of the protein molecule. Fourier transform infrared (FTIR) spectroscopy – a vibrational spectroscopic technique has provided better understanding of secondary structure change of plant proteins after the influence of high pressure. FTIR measurement has considered a complementary approach to DSC because it provides direct structural information about the protein at a sub-molecular level (Brandes, Welzel, Werner, & Kroh, 2006). The technique has recently been used for conformational study of globular protein on the amide I region (Ahmed, Ramaswamy, Ayad, Alli, & Alvarez, 2007; Brandes et al., 2006; Carbonaro, Maselli, Dore, & Nucara, 2008; Ellepola, Choi, & Ma, 2005).

Starch is semi-crystalline biopolymer and contains both crystalline and amorphous components. Starch is known to absorb considerable amounts of water. During heating in presence of water, starch undergoes thermal transition known as gelatinization, causing disruption of the starch granule structure. The semi-crystalline structure gradually transforms to amorphous state which shows some physical change with time (Slade & Levine 1993). On cooling, an amorphous polymer undergoes glassy state from rubbery phase which is known as glass transition temperature (T_g). Because of their relevance in foods, starch–water systems have been extensively investigated (Slade & Levine, 1993; Tananuwong & Reid, 2004). Pressure could also delay starch gel retrogradation (Douzals, Marechal, Coquile, & Gervais, 1996). Due to increase demand of pressure treated food products it is relevant to study high pressure effect on completely gelatinized legume starch in excess water.

The objectives of this work were to study thermal and rheological characteristics of lentil flour slurry (at various moisture level) after high pressure treatment and to understand the pressure susceptibility of major lentil components (starch and protein) with the results obtained by the differential scanning calorimetry (DSC) measurements, and the change of the protein secondary structure by FTIR measurements.

2. Materials and methods

2.1. Materials

Canadian grown lentil flour was obtained locally and used for high pressure treatments. Mean values for proximate analysis (g/100 g) of lentil flour samples (as per analyzed by AOAC method, 1996) are: moisture, 6.47 ± 0.21, ash, 2.31 ± 0.03, protein (N × 6.25), 26.25 ± 0.23, and crude fat 1.01 ± 0.10. The starch content was estimated as per method described by Dubois, Gilles, Hamilton, Rebers, and Smith (1956) and found to be about 52 g per 100 g lentil flour. The lentil samples were ground with a coffee grinder (Model KSM 2, type 4041, Braun Canada), to obtain flour and 80% of the particles went through #100 mesh screen aperture (0.15 mm sieve opening as per ASTME II).

2.2. Preparation of sample

A calculated amount of distilled water was added to lentil flour to achieve desired levels of moisture content (14.9, 24.6, 29.1, 37.2, 46.7 and 57.6 g water per 100 g ground flour). The dispersions were kept for an hour at room temperature (20 ± 1 °C) for hydration. All measurements were conducted with the same lot of the product obtained at one time. The moisture content has been varied to evaluate changes in thermal and rheological properties as influenced by high pressure treatment. Measurements were made in triplicate.

2.3. High-pressure treatment

Lentil dispersions (approx. 20–25 g) were packed in low-density polyethylene bags (Whirl-Pak®, USA) and heat sealed. The headspace in the sealed pack was kept to a minimum. Samples were then transferred to a 5 L pressure reactor unit (ACIP 6500/5/12VB; ACB Pressure Systems, Nantes, France) equipped with temperature and pressure regulator device. Water was used as the pressure transmitting medium. Samples were pressure treated at specified pressure level (350, 450, 550 and 650 MPa) for a holding time of 15 min. The pressurization rate was about 4.4 MPa/s and released at 26 MPa/s. The initial temperature of pressure medium was 18 °C which was sharply increased to 24.8 and 28.5 °C due to the adiabatic effect during pressurization of 350 and 650 MPa however, temperature became steady at 22 and 26 °C during holding period at those pressure levels. At 450 and 550 MPa, the increases in temperatures were 26 and 27.5 °C and equilibrated at 23.5 and 25 °C respectively at similar condition. All experiments were carried out in duplicate. Pressure treated samples were stored in refrigerator for further use.

In a separate experiment, lentil flour dispersion (58 g water/100 g sample) was isothermally heated in a temperature controlled bath at 85 °C for 30 min, cooled and stored in refrigerator.

2.4. Differential scanning calorimetric (DSC) measurement

A differential scanning calorimeter (DSC) (TA Q100, TA Instruments, New Castle, DE, USA) was employed to measure the thermal analysis for control and pressure treated lentil slurries. The DSC was calibrated with indium and sapphire for temperature and heat capacity calibration (TA Instruments, 2002). The samples (10–12 mg) were run at a 10 °C/min heating/cooling ramp in heating–cooling cycles in a nitrogen atmosphere (flow rate 50 mL/min). The samples were heated from 0 to 150 °C to detect thermal gelatinization of starch and protein denaturation in flour. An empty pan was used as a reference. The DSC measurements were done more than 5 times. Thermal transitions of lentil slurries were measured for the peak gelatinization (T_m) and the denaturation temperature (T_d). The enthalpy (δH) of the transition (associated with starch gelatinization and protein denaturation) was calculated from the area of the peak endotherm using the Universal Analysis Software (version 3.6C, TA Instruments, New Castle, DE, USA). One set of DSC run was carried out for both thermally treated control and pressure treated lentil slurry after one week to locate recrystallization of starch during retrogradation.

2.5. Rheological measurements

Rheological measurements of control and pressure treated lentil flour dispersions were performed on a controlled-stress rheometer (AR 2000, TA Instruments, New Castle, DE) with parallel plate geometry (60-mm diameter). The width of the gap between two plates was 1000 μm. The sample temperature was internally controlled by peltier system (−20 to 200 °C with an accuracy of ±0.1 °C) attached with a water circulation unit. A platinum resistance thermometer sensor positioned at the centre of the plate ensured temperature control and measurement. For each test, a measured volume of thoroughly mixed sample (approximately 2 mL) was placed at the centre of the rheometer plate for 5 min for stress relaxation and temperature equilibration before the actual measurements. It is worth to mention that sample with moisture