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Effect of extrusion conditions on the physico-chemical properties and *in vitro* protein digestibility of canola meal

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ABSTRACT

Canola meal has potential as a high protein food ingredient. The extrusion-induced changes in color, pH, extractable protein and *in vitro* protein digestibility of canola meal under different extrusion conditions was assessed. The extrusion barrel moisture (24%, 30% or 36%) and screw kneading block length (0, 30 or 60 mm) were used as independent process parameters. Extrusion at high barrel moisture (36%) favored protein aggregation resulting in lower extractable protein compared to extrusion at the lowest barrel moisture (24%). At lower barrel moisture contents (24% and 30%), a longer kneading block length increased extractable protein but this was not the case at 36% barrel moisture. Canola protein digestibility was improved upon extrusion at 30% barrel moisture but there was no significant change at lower (24%) or higher (36%) barrel moisture. The kneading block length of the screw had no significant effect on the canola protein digestibility within the same barrel moisture level. The relationship between the physico-chemical parameters and *in vitro* digestibility was examined. This study highlighted the complex interplay of extrusion processing variables that affect protein degradation and the interaction of components, with consequent effects on protein digestibility.

1. Introduction

Canola is a rapeseed variety characterized by low erucic acid (< 2%) in the oil portion and low glucosinolate content (< 30 μmol/g) in the meal portion. The canola meal, a by-product of oil extraction, contains up to 50% protein on a dry basis (Aachary, Thiyam-Hollander, & Eskin, 2014). The canola proteins have largely been used for animal feed but there is interest in adding value to canola meal (Wanasundara, 2011). The amino acid profile of canola proteins is well balanced for human nutrition in addition to desirable functional properties (Aider & Barbana, 2011). The nutritional profile of canola proteins made them good candidates as food ingredients (Wu & Muir, 2008).

Plant proteins such as those from legumes and cereals had low *in vitro* digestibility, which has been related to a lower nutritional value (Carbonaro, Maselli, & Nucara, 2012). Low digestibility of plant proteins has been related to the coexistence of protease inhibitors, like trypsin inhibitor as well as to the inherent structural properties of plant proteins (Carbonaro et al., 2008; Deshpande & Damodaran, 1989). There are differences between the nutritional values of different plant proteins. For example, the bioavailability of the canola meal protein (80.1%) in poultry is significantly lower than that of soy bean meal protein (88.9%) (Zuprizal, Chagneau, & Lessire, 1991).

Extrusion is used for the production of a wide range of food ingredients and products. The high temperature and high shear of the process results in the changes to the physical, chemical and nutritional quality of the extruded food product (Alam et al., 2016). The *in vitro* protein and starch digestibility of lentil splits were improved on extrusion and attributed to the inactivation of anti-nutritional factors such as trypsin inhibitors, phytic acid and tannins (Rathod & Annapure, 2016). The denaturation of proteins, which exposes new sites for enzymes to attack, is also a contributory factor for improved protein digestibility upon extrusion (Singh, Gamlath, & Wakeling, 2007). The *in vitro* protein digestibility of sorghum protein was increased on extrusion, with extrusion temperature being the key influencer of digestibility (Fapojuwo, Maga, & Jansen, 1987). In contrast, post-heat treatment of extruded sorghum (mixed with boiling water and heated for 5 min) reduced *in vitro* protein digestibility.

Extrusion variables, such as the temperature, screw speed, screw configuration, are determinants of the mechanical and thermal energy inputs and the residence time, which influence the degradation and interaction of components and hence the product attributes (Chen et al.,

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2010; Owolabi et al., 2008; Zhang et al., 2015). There is a fine balance between the effects of processing conditions on protein digestibility and the interplay between the input processing variables (*i.e.* barrel moisture content and extruder screw profile). In this work the effect of input processing variables on the physico-chemical properties and *in vitro* protein digestibility of canola meal was examined, as a function of barrel moisture and kneading block length of the screw, as they affect the shear-induced changes to the material. The changes in color, pH, alkali-extractable protein and the molecular weight profile of the extractable protein were measured. The relationship of these physico-chemical characteristics to the *in vitro* protein digestibility of canola meal was assessed. The understanding from this study may provide a more informed basis for applying the extrusion process to improve protein digestibility of canola meal.

2. Materials and methods

2.1. Materials

Canola meal was provided by ROBE (Riverina Oil & BioEnergy Pty Ltd., Wagga Wagga, Australia). The oil extraction process, outlined by the manufacturer, indicated that canola seeds were cooked at 80–105 °C for 15–20 min after flaking. Physical pressing of oil was conducted at 105 °C, followed by a solvent extraction of residue oil and desolvenization at around 105 °C for ~20 min. Moisture, protein, fat, ash and carbohydrates content of canola meal were measured by National Measurement Institute (Melbourne, Australia).

Trypsin and chymotrypsin were purchased from Sigma-Aldrich, Inc. (St Louise, MO, USA). Peptidase was provided by Amano Enzyme Inc. All other chemicals and reagents were purchased from Merck Pty Ltd. (Kilsyth, VIC, Australia).

2.2. Extrusion

All of the extrusion experiments were conducted using a lab-scale, co-rotating and intermeshed twin-screw lab extruder (KDT30-II, Jinan Kredit Machinery Co. Ltd., China). The extruder had 30 mm screw diameter (D), 20:1 screw length/diameter ratio, and a rectangular die with 2 × 5 mm^2 opening. The barrel was segmented into a feeding zone and 4 temperature-controlled zones, which were heated by an electric cartridge heating system and cooled with running water. The temperature and screw speed were controlled with a control panel, where the extruder responses, including the motor electric current and die pressure, were displayed.

The extruder barrel temperatures from the powder feed port to the die were set at 60, 90, 110 and 110 °C, and kept constant during experiment. Feed rate and screw speed were fixed at 30 kg/h and 288 rpm, respectively. Barrel moisture content and screw kneading block length were chosen as variables. It is expected that barrel moisture content will have significant effect on the physico-chemical changes of the canola meal during extrusion; and the kneading block length of the screw will affect the mechanical shearing and specific mechanical energy input to the canola meal. The barrel moisture content was selected as 24%, 30% and 36% based on preliminary experiments, which showed that barrel moisture (< 24%) led to blockage during extrusion while barrel moisture (> 36%) led to inability to form a structured product. The barrel moisture content was controlled by adjusting the water feed rate by a peristatic pump, which was calibrated before the extrusion run. The water port was located at around 2/3 of the screw length from the die end. The basic screw configuration with 0 mm kneading block was built from feed to the die with CE/37.5/37.5/8 and CE/30/45/10, which represented 8 conveying elements (CE) with 37.5 mm length and 37.5° helix angle and 10 conveying elements with 30 mm length and 45° helix angle, as shown in Fig. 1. Based on basic screw configurations, other screw configurations were designed as with 30 mm and 60 mm kneading block, to replace the conveying elements and located at end of the screw. The experiment design had 9 treatments with three barrel moisture contents (24%, 30% and 36%) and three screw profiles (with kneading block lengths of 0, 30 and 60 mm).

Extruded samples were collected for physicochemical analysis, when the process conditions reached steady state, as indicated by constant values for extruder electric current and die pressure. Two sets of samples, each about 5 kg, were collected and immediately put into plastic bags and stored at − 18 °C until taken for analysis. To assess the repeatability of the extrusion process, three extrusion treatments with three barrel moisture contents (24%, 30% and 36%) and one screw profile (with kneading block lengths of 30 mm) were repeated in a separate runs. The electric current, die pressure and yield were repeatable with the variance less than 1%. So for all other process conditions, one extrusion run was carried out.

The specific mechanical energy (SME, kJ/kg) was calculated from the mechanical energy output from the motor (E_m) and materials flow rate (M, kg/h, determined by the output of the extrudate), according to the following formula.

$$SME = 3.6 * E_m / M \quad (1)$$

where, 3.6 is the conversion from W·h/kg to kJ/kg; M is the material flow rate, [kg/h]; E_m is the mechanical energy output [W] from the motor:

$$E_m = 3^{1/2} \eta_e \eta_m UI \quad (2)$$

where, η_e is the power factor, 0.877; η_m is the efficiency, 0.857; U is the voltage of power supply, 420 [V]; I is the net electric current [A], which is the difference of the electric current that was recorded during extrusion and that with no feed materials.

2.3. Freeze drying of meals and extrudates

For the purpose of characterization and analysis, the raw canola meal and extruded samples were freeze dried in Dynavac Freeze Dryer (FD-5, Australia). The freeze dried canola meals were ground into powders with particle size < 125 μm using Retsch mill (MM400, Germany) and stored in sealed containers at ~22 °C until use.

2.4. Color and pH measurement

The color measurement was carried out using a Minolta CR-300 Chroma meter (Konica Minolta Sensing Americas, Ramsey, NJ). The system provides three color components; L* (black-white component, luminosity), a* (+ red to − green component) and b* (+ yellow to − blue component). Powder samples were measured in a 5 cm diameter plastic petri dish with at least 5 mm thickness of the canola meal to cover the bottom of the petri dish for each measurement. The color change, ΔE index, was calculated using Eq. (3):

$$\Delta E = \sqrt{(L_S - L_R)^2 + (a_S - a_R)^2 + (b_S - b_R)^2} \quad (3)$$

where, S denotes values of the extruded canola meals and R denotes values of the raw canola meal before extrusion.

The pH of canola meal suspensions (0.016 g/mL) was measured using a pH meter (LAQUA, F-71, Horiba Scientific). For preparation of the suspensions, 781 mg canola meal was dispersed in 50 mL de-ionized water with a magnetic stirrer at 37 °C for 2 min or for the time until pH value was stable. The amount of canola meal was calculated based on total protein content of the canola meal so a protein concentration in the suspension was 1 mg N/mL.

2.5. Characterization of protein components

2.5.1. Protein content determination

The protein content was determined using a Leco TruMac Nitrogen