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The effects of extrusion on nutrient content of Canadian pulses with a focus on protein and amino acids

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Abstract

Alternative sources of protein will be required in both human and animal nutrition to support ingredient sustainability and nutrient demands of a growing world population. Extrusion is one technique utilized to process pulses and is reported to increase starch and protein digestibility but also has the potential to transform nutrients into non-nutritious compounds. This study sought to compare the effects of extrusion on nutrient composition in Amarillo peas, Dun peas, lentils, chickpeas, and faba beans, with soybean meal (control). Each pulse was extruded at 18% or 22% moisture and 110, 130, or 150 °C. Compared to whole samples, extrusion increased crude protein content of Amarillo and Dun peas, and lentils, and it decreased in soybean meal ($P < 0.05$). Compared with whole samples, extrusion increased methionine content in chickpeas and lentils ($P < 0.05$), with no effect in Amarillo or Dun peas, faba beans, and soybean meal. Cysteine content increased in extruded Amarillo peas compared with whole pulses, and decreased in soybean meal ($P < 0.05$). Results suggest that extrusion can positively affect protein and amino acid content of pulses, however, specific changes differ by pulse/legume type.

Key words: pulses, extrusion, nutrient composition, amino acids

1. Introduction

Pulses are a group of leguminous seeds grown as staple crops globally. The United Nations Food and Agriculture Organization defines pulses as "crops harvested solely for dry seed, excluding crops harvested green for food, oil extraction, or crops grown and harvested exclusively for sowing purposes"). As the global population continues to grow, there will be increased demand from agri-food industry to meet the nutritional needs of the population. The animal protein supply will reach maximum production; which will not meet the protein demands of the global population (Aiking 2011). Thus, the agri-food industry will need to focus on non-animal proteins to ensure adequate quality protein sources are available (Aiking 2011).

Pulses are a rich source of carbohydrates, protein, micronutrients, and bioactive compounds. However, like other plant proteins, their digestibility is generally lower in their natural (unprocessed) form when compared to animal protein sources (Young and Pellett 1994). Additionally, due to the limiting nature of certain indispensable amino acids (AA), pulses often have lower protein quality compared to animal protein sources (Young and Pellett 1994). For example, pulses have double the amount of protein compared to cereal grains, but are limiting in sulfur AA (methionine and cysteine) and (or) tryptophan (Trp) ; conversely cereal grains tend to be rich in sulfur AA and limiting in lysine (Lys; Evans and Boulter 1980; Young and Pellett 1994).

Dry pulses can undergo various household and industrial processes prior to consumption These processes include, but are not limited to, dehulling, boiling, fermentation, and extrusion (Malcolmson and Han 2019). Extrusion has gained a critical role in food production in both human and animal nutrition sectors. While soybean has been the main leguminous ingredient for development of extruded protein foods, there is growing interest in pulses as primary proteins in extruded foods. Briefly, extrusion is a high-temperature, short-time process, where starchy and (or) proteinaceous materials are forced through a heated barrel and die at high pressure to form the final product (Guy 2001; Berrios et al. 2012). In addition, extrusion offers many advantages over batch cooking methods with high throughput, versatility in ingredients used and products produced, lower cost, improved or maintained product quality, and nutrient retention (Guy 2001; Berrios et al. 2012). Extrusion cooking can also improve starch and protein digestibility of ingredients (Srihara and Alexander 1984; Alonso et al. 2000a, 2000b; Rathod and Annapure 2016) and, in some cases, the retention of AA, such as Lys (Jeunink and Cheftel 1979; Björck and Asp 1983), and reduce

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anti-nutritional factors (ANF) such as trypsin inhibitors and tannins (Alonso et al. 1998, 2000a; Abd El-Hady and Habiba 2003; Rathod and Annapure 2016). However, high temperature processing can also cause irreversible changes in nutrients which may negatively affect their bioavailability. For example, exposing proteins to heat can cause racemization of AA from the L to D form, which can impair bioavailability (Gilani et al. 2005). Heat-induced Maillard reactions, and the formation of cross-linked AA, such as lysinoalanine (LAL), can decrease levels and bioavailability of Lys (Björck and Asp 1983; Gilani et al. 2005). Other indispensable amino acids, such as arginine (Arg), cysteine (Cys), histidine (His), and Trp, may also be susceptible to heat damage (Iwe et al. 2001; Gilani et al. 2005).

As the need for alternative protein sources emerges, interest in the incorporation of other legumes, such as pulses, into extruded food products has developed. Information on the nutrient content and impact of common processing techniques will be critical to increasing the use of pulses as alternative proteins in both human and animal nutrition. Thus, the objectives of this study were to characterize the nutrient content, determine the effects of processing by extrusion on nutrient composition, and compare samples extruded at different moistures/temperatures in Canadian peas, chickpeas, lentils, and faba beans.

2. Materials and methods

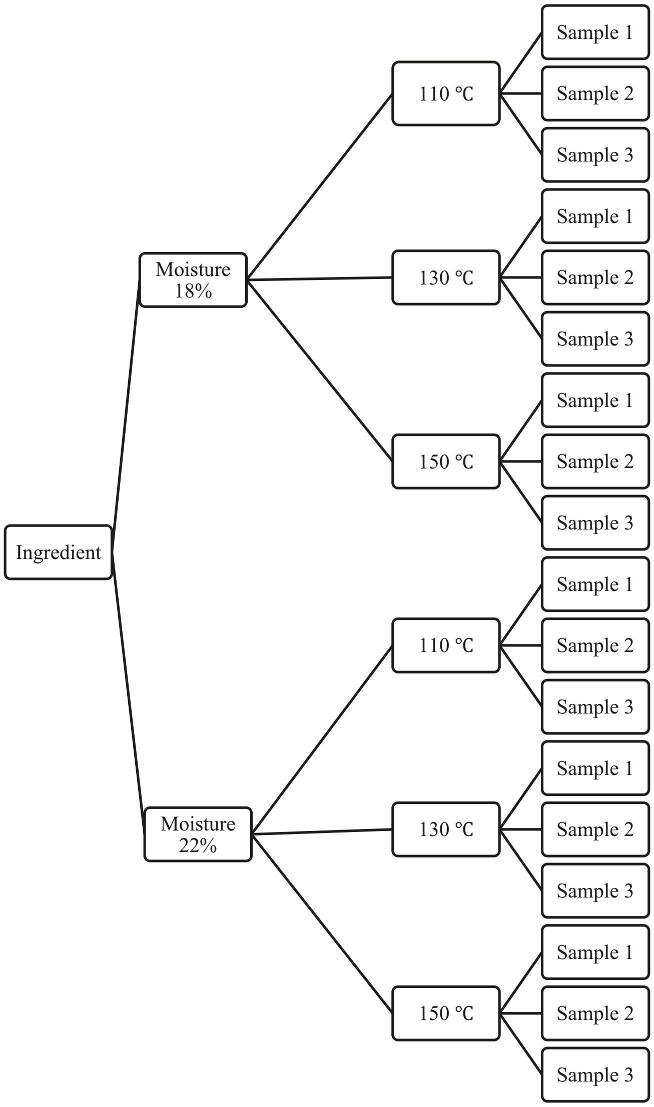
2.1. Ingredients

Five Canadian grown pulses as the ingredients of interest. Soybean meal (SBM) was used as a control as its nutrient content and the effects of processing have been well-characterized, making it ideal for comparison. Selected pulses included Amarillo and Dun field pea varieties (IFN 5–08-481; CDC Amarillo Variety; Oren and Marlene Robinson, Landis, SK, Canada and CDC Dakota; Faba Canada, Tisdale SK, Canada), chickpeas (Kabuli variety; AGT Foods, Regina, SK, Canada), faba beans (IFN 5–09-262; Snowbird variety; Faba Canada, Tisdale SK, Canada), lentils (Laird variety; AGT Foods, Regina, SK, Canada), and SBM (IFN 5–04-604; Cargill Animal Nutrition; North Battleford, SK, Canada).

2.2. Processing

Prior to extrusion all samples were ground through a 2/64″ screen using a hammermill (G.J. Vis. Model: VISHM2014, Oakbluff, MN, Canada) at the Canadian Feed Research Centre (North Battleford, SK, Canada). Average particle size for ingredients are as follows: ground Amarillo peas (255 μm), ground chickpeas (216 μm), ground Dun peas (278 μm), ground faba beans (272 μm), ground lentils (296 μm), and SBM (370 μm). Extrusion of ingredients were completed at the Agri-Food Innovation Centre (Saskatoon, SK, Canada). Approximately 25 kg of each fine ground ingredient was extruded using a twin-screw extruder (Clextral EV 32 twin-screw extruder, Firminy, France) through a 2.7 mm die. The extruder was operated at 397 ± 2 rpm with a flow rate of 30.5 ± 0.2 kg/h. Ingredients were extruded according to a 2 × 3 factorial design with 2 moistures (18% and 22%) and 3 temperatures (110, 130, and 150 °C; Fig. 1). Extrusion temperatures and moistures were determined based on current/common extrusion parameters being utilized for pulse in both research and industry. Barrel zone temperatures (zones 3–6) were held constant at 110, 130, and 150 °C. Die temperature was variable at 110 ± 18, 130 ± 11, and 150 ± 13 °C. Samples were collected only after the ingredients had been extruded at a steady temperature for 1 min. Prior to processing, a test run was conducted to determine the length of time it took to extrude 25 kg of sample. Samples were collected for all runs at the beginning (sample 1), middle (sample 2), and end (sample 3) of the extrusion run for each ingredient processing parameter. Extrudates were dried using an electric dryer after passing through the die and allowed to cool to room temperature prior to storage.

Fig. 1. Overview of processing procedures for ingredients of interest.



2.3. Analytical methods

2.3.1. Proximate analysis

Prior to analysis, samples were ground through a 0.5 mm screen centrifugal mill (Ultra Centrifugal Mill Type ZM 200

Retsch Part #20.823.0003 Serial #1214030238P). Samples were analyzed for moisture (AOAC 930.15 1999), crude protein (CP) (AOAC 990.03 2002), crude fat (AOCS AM 5-04 2017), and ash (AOAC 942.05 1943) via SGS Agri-Foods Laboratories (Guelph, ON, Canada).

2.3.2. Amino acids analysis

Amino acid content was determined in ingredients via hydrolysis and ultra-performance liquid chromatography (UPLC; Waters Corporation, Milford, MA, United States) analysis. Amino acid content, except Cys, Met, and Trp, were determined using acid hydrolysis adapted from AOAC method 994.12 (2012). In brief, ~0.1 g of sample and 5 mL of 6 N hydrochloric acid (HCl) phenol solution were added to glass digestion tubes. The tubes were flushed with nitrogen gas, sealed, and digested at 110 °C for 24 h, after which samples were removed from the digestion block and cooled to room temperature. Samples were mixed with 1 mL of internal norvaline standard (5 mM) after which 1 mL aliquots of each sample were transferred to microcentrifuge tubes and stored at −20 °C until analysis. Prior to UPLC analysis, samples were thawed and neutralized by mixing 120 μL of sample with 100 μL of NaOH (6 N) and 400 μL of deionized water.

Sulfur AA (Cys and Met) and lysinoalanine (LAL) content of samples were determined using oxidative hydrolysis adapted from AOAC method 994.12 (2012). In brief ~0.1 g of sample and 2.5 mL of ice-cold oxidation solution (9:1 ratio of phenolic formic acid (88%) and 30% hydrogen peroxide, respectively) were added to glass digestion tubes, capped loosely, placed in an ice water bath and stored for 18–20 h in a fridge. Samples were removed from the fridge and 0.4 mg of sodium metabisulfite (Sigma, Oakville, ON, Canada) was added to each sample. Samples were rested for 2 h with occasional mixing to decompose excess performic acid. After 2 h, 2.5 mL of concentrated HCl (12 N) was added to each sample, tubes were flushed with N_2 gas, sealed, and digested at 110 °C for 24 h, after which samples were treated as previously stated. Prior to UPLC analysis, samples were thawed and neutralized by mixing 100 μL of sample with 160 μL of NaOH (6 N) and 400 μL of deionized water.

Tryptophan content of samples was determined using alkaline hydrolysis adapted from AOAC method 988.15 (2012). In brief, ~0.1 g of sample and 2.5 mL of 6 N NaOH were added to glass digestion tubes. Tubes were flushed with N_2 gas, sealed, and digested for 20 h at 110 °C, after which samples were removed from digestion block and cooled to room temperature. In an ice cold water bath 7.5 mL of 1% phenolic-HCl (2 N) and 1 mL of internal norvaline standard (5 mM) were added to each sample and gently mixed. After mixing, 500 μL of sample was mixed with 500 μL of deionized water in a microcentrifuge tube and stored at −20 °C until further analysis.

Amino acid standards and samples were derivatized using an AccQ-Tag Ultra derivatization kit (Waters Corporation, Milford, MA, United States). Derivatized AA (1 μL injection volume) were separated in a column (2.1 × 200 mm, 1.7 μL) maintained at 55 °C using UPLC with ultraviolet detection at a wavelength of 260 nm. Amino acid peak areas were analyzed using Waters Empower 2 Software (Waters Corporation, Milford, MA, United States).

2.3.3. Statistical analysis

Data were analyzed using a fixed models via PROC GLIMMIX in SAS (SAS v 9.4, SAS Institute Inc., Cary, NC, USA). A fixed model was used to compared whole vs extruded samples, where processing was the fixed effect. A second fixed model was used to compared extruded samples where moisture and temperature were fixed effects and moisture x temperature interaction was included.

The factorial model was set up as such:

$$y_{ijk} = \mu + A_i + B_j + (AB)_{ij} + \epsilon_{ijk}$$

where:

y_{ijk} = observation k in level i of factor A and level j of factor B for nutrient of interest, where $k = 1 \ldots n$ and n = number of observations for each A × B combination,
μ = the overall mean
A_i = the effect of level i of moisture, where $i = 1 \ldots a$ and $a = 2$,
B_j = the effect of level j of temperature, where $j = 1 \ldots b$ and $b = 3$,
$(AB)_{ij}$ = the effect of the interaction of level i of moisture with level j of temperature, and
ε_{ijk} = random error with mean 0 and variance σ^2.

A Tukey HSD test was used for means comparisons between samples. Differences were deemed significant when $P \leq 0.05$.

3. Results

3.1. Amarillo peas

3.1.1. Proximate analysis

Data are reported in Table 1 for proximate analysis. When compared with whole samples, extruded samples had a higher DM content ($P < 0.05$). There were no effects of moisture or temperature on DM content when extruded samples were compared with each other. When compared with whole samples, extruded samples had higher CP content ($P < 0.05$). There were no effects of moisture or temperature on CP content when extruded samples were compared with each other. When compared with whole samples, extruded samples had lower crude fat content ($P < 0.01$). There were no effects of moisture or temperature on crude fat content when extruded samples were compared with each other. There were no differences in ash content when whole and extruded samples were compared with one another. There were no effects of moisture or temperature on ash content when extruded samples were compared with each other.

3.1.2. Amino acids

Data are reported in Table 1 for indispensable AA and LAL and Table S1 for dispensable amino acids. There were no differences in indispensable AA and LAL content when whole

Table 1. Effect of extrusion temperature and moisture on nutrient composition in whole and extruded Amarillo peas, % dry matter basis.

				Moisture 18%			Moisture 22%				*P* value		
	Whole[1]	Extruded[1]	*P* value	110 °C	130 °C	150 °C	110 °C	130 °C	150 °C	SEM	Moisture	Temp.	Moisture × temp.
Dry matter	86.83^{b} ± 0.66	95.27^{a} ± 0.22	<0.0001	95.00	96.21	95.81	94.43	95.32	94.84	0.49	0.06	0.14	0.91
Crude protein	21.27^{b} ± 0.13	21.56^{a} ± 0.04	0.04	21.55	21.51	21.68	21.46	21.47	21.72	0.09	0.70	0.06	0.76
Crude fat	1.62^{a} ± 0.1	1.20^{b} ± 0.03	0.0006	1.18	1.17	1.22	1.09	1.32	1.23	0.08	0.71	0.36	0.35
Ash	2.53 ± 0.07	2.56 ± 0.02	0.74	2.58	2.56	2.50	2.49	2.63	2.60	0.06	0.66	0.57	0.2
Indispensable amino acids													
Histidine	0.47 ± 0.03	0.44 ± 0.01	0.33	0.46	0.41	0.43	0.48	0.43	0.42	0.02	0.61	0.11	0.79
Isoleucine	0.67 ± 0.04	0.69 ± 0.01	0.63	0.69	0.68	0.76	0.65	0.68	0.69	0.02	0.09	0.11	0.40
Leucine	1.39 ± 0.05	1.32 ± 0.02	0.21	1.34	1.28	1.36	1.37	1.26	1.32	0.04	0.79	0.18	0.73
Lysine	1.25 ± 0.08	1.27 ± 0.03	0.77	1.26	1.30	1.30	1.27	1.25	1.25	0.07	0.59	0.99	0.85
Methionine	0.20 ± 0.03	0.23 ± 0.01	0.30	0.23	0.25	0.19	0.25	0.26	0.22	0.02	0.15	0.02	0.83
Phenylalanine	0.97 ± 0.06	0.91 ± 0.02	0.27	0.93	0.83	0.93	0.98	0.87	0.90	0.05	0.56	0.12	0.69
Threonine	0.74 ± 0.03	0.70 ± 0.01	0.20	0.71	0.69	0.75	0.72	0.69	0.66	0.02	0.19	0.49	0.15
Tryptophan	0.68 ± 0.07	0.61 ± 0.02	0.43	0.17	0.15	0.14	0.21	0.13	0.18	0.03	0.42	0.20	0.43
Valine	0.14 ± 0.03	0.16 ± 0.01	0.87	0.77	0.77	0.83	0.72	0.76	0.76	0.03	0.09	0.26	0.45
Cross-linked amino acid													
Lysinoalanine	0.02 ± 0.01	0.01 ± 0.00	0.27	0.01	0.01	0.01	0.01	0.01	0.01	0.01	0.43	0.94	0.30

[1] Data for whole compared to extruded samples are presented as mean ± standard error of the mean.

[a–b] Means with differing superscripts in the same row are different ($P \leq 0.05$). Means with two sets of superscripts indicate effects of moisture (first value) and temperature (second value). Means with a significant interaction effect (moisture × temperature) only report differences between means for interaction effect.

and extruded samples were compared. There was no effect of moisture or temperature on indispensable amino acid content when extruded samples were compared with each other. There was no effect of moisture or temperature on LAL content when extruded samples were compared with each other. When compared with whole samples, extruded samples had a higher Cys content ($P < 0.05$). When compared with whole samples, extruded samples had lower alanine (Ala; $P < 0.01$), aspartate (Asp; $P < 0.01$), and glutamate (Glu; $P < 0.01$) content. Samples extruded at 130 °C had higher Cys content when compared with samples extruded at 150 °C ($P < 0.05$). Samples extruded at 110 °C had higher glycine (Gly) and proline (Pro) content compared to samples extruded at 130 °C ($P < 0.05$). Samples extruded at 22% moisture/110 °C had higher serine (Ser) content compared to samples extruded at 22% moisture/150 °C ($P < 0.05$). There was no effect of extrusion moisture or temperature on LAL content between extruded samples.

3.2. Chickpeas

3.2.1. Proximate analysis

Data are reported in Table 2 for proximate analysis. When compared with whole samples, extruded samples had a higher DM content ($P < 0.01$). Dry matter content was highest in samples extruded at 18% moisture/all temperatures compared with samples extruded at 22%/all temperatures ($P < 0.01$). Samples extruded at 22% moisture/110 or 150 °C were higher compared with samples extruded at 22% moisture/130 °C ($P < 0.05$). There were no differences in CP content when whole and extruded samples were compared with each other. There were no effects of moisture or temperature on CP content when extruded samples were compared. When compared with whole samples, extruded samples had lower crude fat content ($P < 0.05$). Samples extruded at 22% moisture/110 °C had higher crude fat content compared with all other extruded samples ($P < 0.05$). Samples extruded at 18% moisture/110 °C had higher crude fat content compared with samples extruded at 18% and 22% moisture/150 °C ($P < 0.05$). Samples extruded at 18% moisture/130 °C had higher crude fat content compared with samples extruded at 18% moisture/150 °C ($P < 0.05$). When compared with whole samples, extruded samples had a higher ash content ($P < 0.05$). There were no effects of moisture or temperature on ash content when extruded samples were compared with each other.

3.2.2. Amino acids

Data are reported in Table 2 for indispensable amino acids and LAL and Table S2 for dispensable amino acids. With the exception of methionine, which was higher in extruded samples compared to whole ($P < 0.01$), there were no differences in indispensable AA content of extruded samples compared to whole. Samples extruded at 150 °C had higher His content compared to samples extruded at 130 °C ($P < 0.05$). Samples extruded at 150 °C had higher isoluecine (Ile) and leucine (Leu) content compared to samples extruded at 110 and 130 °C ($P < 0.05$). Samples extruded at 18% moisture had higher Leu and Trp content compared with samples extruded at 22% moisture ($P < 0.05$). Samples extruded at 18% moisture/150 °C had higher theonine (Thr) content when compared with all other extruded samples ($P < 0.05$). There were no differences in dispensable AA content of extruded samples compared to whole. Samples extruded at 18% moisture had higher Ala and Pro content when compared with samples extruded at 22% moisture ($P < 0.05$). Samples extruded at 150 °C had higher Ala content compared with samples extruded at 110 °C ($P < 0.05$). Samples extruded at 150 °C had higher Pro content compared to samples extruded at 130 °C ($P < 0.05$). Samples extruded at 18% moisture/150 °C had higher Asp content compared with all other extruded samples ($P \leq 0.05$), and samples extruded at 18% moisture/110 and 130 °C and 22% moisture/110 °C had higher Asp content compared with samples extruded at 22% moisture/130 and 150 °C ($P < 0.05$). Samples extruded at 18% moisture/150 °C had higher Glu content compared to all other extruded samples ($P < 0.05$), and samples extruded at 18% moisture/130 and 150 °C had higher Glu content compared with samples extruded at 22%/130 °C ($P < 0.05$). There was no effect of extrusion moisture or temperature on LAL content between extruded samples.

3.3. Dun peas

3.3.1. Proximate analysis

Data are reported in Table 3 for proximate analysis. When compared with whole samples, extruded samples had a higher DM content ($P < 0.0001$). Samples extruded at 18% moisture had higher DM content compared with 22% moisture ($P < 0.0001$). Samples extruded at 130 and 150 °C had higher DM content compared with samples extruded 110 °C ($P < 0.0001$). While the interaction for type III tests of fixed effects indicated no difference in DM content, differences between means were indicated (Table 3; $P \leq 0.05$). When compared with whole samples, extruded samples had higher CP content ($P \leq 0.01$). There were no effects of moisture or temperature on CP content when extruded samples were compared with each other. When compared with whole samples, extruded samples had lower crude fat content ($P \leq 0.05$). There were no effects of moisture on crude fat content when extruded samples are compared to each other, however, samples extruded at 130 and 150 °C had higher fat content when compared with samples extruded at 110 °C ($P < 0.01$). While the interaction for type III tests of fixed effects indicated no difference in crude fat content across extruded samples, differences between means were indicated (Table 3; $P \leq 0.05$). There were no differences in ash content when whole and extruded samples were compared with one another. Samples extruded at 18% moisture/150 °C had higher ash content compared to all other extruded samples ($P < 0.05$).

3.3.2. Amino acids

Data are reported in Table 3 for indispensable amino acids and LAL and Table S3 for dispensable amino acids. There were no differences in indispensable amino acid and LAL content when whole and extruded samples were compared. Samples

Table 2. Effect of extrusion temperature and moisture on nutrient composition in whole and extruded chickpeas, % dry matter basis.

				Moisture 18%			Moisture 22%				*P* value		
	Whole[1]	Extruded[1]	*P* value	110 °C	130 °C	150 °C	110 °C	130 °C	150 °C	SEM	Moisture	Temp.	Moisture × temp.
Dry matter	88.52^{b} ± 0.74	95.45^{a} ± 0.25	<0.0001	96.37^{a}	96.58^{a}	96.09^{a}	95.23^{b}	93.80^{c}	94.60^{b}	0.16	<0.0001	<0.0001	<0.0001
Crude protein	22.85 ± 0.36	22.82 ± 0.08	0.95	22.90	22.63	23.03	22.56	22.97	22.86	0.21	0.74	0.60	0.30
Crude fat	6.04^{a} ± 0.59	4.42^{b} 0.20	0.02	5.02^{b}	4.57^{bc}	3.54^{d}	5.79^{a}	3.69^{bc}	3.88^{cd}	0.15	0.55	<0.0001	0.0005
Ash	2.69^{b} ± 0.07	2.87^{a} ± 0.02	0.03	2.81	2.84	2.87	2.82	3.00	2.88	0.05	0.16	0.17	0.30
Indispensable amino acids													
Histidine	0.53 ± 0.09	0.51 ± 0.02	0.88	0.48^{ab}	0.51^{b}	0.67^{a}	0.51^{ab}	0.44^{b}	0.53^{a}	0.04	0.13	0.04	0.19
Isoleucine	0.72 ± 0.09	0.84 ± 0.03	0.25	0.77^{b}	0.80^{b}	1.10^{a}	0.83^{b}	0.75^{b}	0.85^{a}	0.06	0.14	0.02	0.07
Leucine	1.49 ± 0.14	1.58 ± 0.05	0.56	$1.67^{a,xy}$	$1.60^{a,y}$	$1.97^{a,x}$	$1.55^{b,xy}$	$1.37^{b,y}$	$1.45^{b,x}$	0.07	0.0008	0.03	0.07
Lysine	1.10 ± 0.12	1.39 ± 0.05	0.05	1.37	1.51	1.22	1.52	1.28	1.36	0.12	0.84	0.53	0.21
Methionine	0.25^{b} ± 0.01	0.35^{a} ± 0.00	<0.0001	0.35	0.35	0.34	0.34	0.37	0.35	0.01	0.25	0.12	0.15
Phenylalanine	1.23 ± 0.16	1.20 ± 0.05	0.87	1.15	1.24	1.32	1.35	1.01	1.13	0.14	0.51	0.61	0.27
Threonine	0.71 ± 0.07	0.78 ± 0.03	0.37	0.80^{b}	0.78^{b}	0.98^{a}	0.78^{b}	0.66^{b}	0.74^{b}	0.03	0.0004	0.0018	0.02
Tryptophan	0.17 ± 0.05	0.20 ± 0.02	0.58	0.21^{a}	0.20^{a}	0.29^{a}	0.18^{b}	0.12^{b}	0.19^{b}	0.03	0.02	0.10	0.53
Valine	0.75 ± 0.07	0.84 ± 0.02	0.21	0.80	0.84	0.91	0.85	0.78	0.88	0.06	0.84	0.44	0.61
Cross-lnked amino acid													
Lysinoalanine	0.02 ± 0.01	0.01 ± 0.00	0.25	0.01	0.01	0.01	0.01	0.02	0.01	0.01	0.34	0.37	0.41

[1] Data for whole compared to extruded samples are presented as mean ± standard error of the mean.

[a–d, x–y] Means with differing superscripts in the same row are different ($P \leq 0.05$). Means with two sets of superscripts indicate effects of moisture (first value) and temperature (second value). Means with a significant interaction effect (moisture × temperature) only report differences between means for interaction effect.

Table 3. Effect of extrusion temperature and moisture on nutrient composition in whole and extruded Dun peas, % dry matter basis.

				Moisture 18%			Moisture 22%				*P* value		
	Whole[1]	Extruded[1]	*P* value	110 °C	130 °C	150 °C	110 °C	130 °C	150 °C	SEM	Moisture	Temp.	Moisture × temp.
Dry matter	87.44^{a} ± 0.73	94.8^{b} ± 0.24	<0.0001	95.03^{bc}	95.66^{ab}	96.05^{a}	93.04^{d}	94.33^{c}	94.71^{bc}	0.21	<0.0001	<0.0001	0.23
Crude protein	22.82^{b} ± 0.19	23.82^{a} ± 0.06	0.0001	23.94	23.87	23.86	23.72	23.63	23.90	0.12	0.19	0.58	0.48
Crude fat	1.39^{a} ± 0.17	0.52^{b} ± 0.06	0.0001	0.34^{b}	0.50^{a}	0.77^{a}	0.26^{b}	0.71^{a}	0.54^{a}	0.09	0.65	0.0038	0.08
Ash	2.5 ± 0.06	2.64 ± 0.02	0.05	2.60^{b}	2.62^{ab}	2.78^{a}	2.60^{b}	2.67^{ab}	2.59^{b}	0.04	0.10	0.15	0.01
					Indispensable amino acids								
Histidine	0.52 ± 0.06	0.52 ± 0.02	0.95	0.56^{b}	0.44^{b}	0.46^{b}	0.54^{a}	0.56^{a}	0.61^{a}	0.04	0.02	0.40	0.12
Isoleucine	0.85 ± 0.12	0.85 ± 0.03	0.96	0.82^{b}	0.72^{b}	0.77^{b}	0.85^{a}	0.96^{a}	0.99^{a}	0.05	0.0020	0.66	0.12
Leucine	1.51 ± 0.14	1.59 ± 0.05	0.59	1.66^{b}	1.36^{b}	1.41^{b}	1.67^{a}	1.68^{a}	1.77^{a}	0.10	0.01	0.36	0.18
Lysine	1.43 ± 0.21	1.69 ± 0.08	0.25	1.82^{ab}	1.26^{c}	1.40^{bc}	1.85^{a}	1.96^{a}	1.85^{a}	0.08	0.0002	0.04	0.0065
Methionine	0.22 ± 0.01	0.23 ± 0.00	0.56	0.24^{a}	0.24^{a}	0.23^{a}	0.23^{b}	0.22^{b}	0.22^{b}	0.01	0.03	0.40	0.93
Phenylalanine	1.02 ± 0.09	1.03 ± 0.03	0.95	1.03^{b}	0.94^{b}	0.92^{b}	1.06^{a}	1.11^{a}	1.11^{a}	0.07	0.05	0.91	0.51
Threonine	0.80 ± 0.08	0.80 ± 0.03	0.96	0.79^{b}	0.67^{b}	0.76^{b}	0.91^{a}	0.89^{a}	0.76^{a}	0.05	0.02	0.24	0.14
Tryptophan	0.15 ± 0.04	0.20 ± 0.01	0.28	0.17	0.19	0.17	0.30	0.19	0.20	0.03	0.06	0.21	0.11
Valine	0.87 ± 0.10	0.95 ± 0.03	0.42	0.93^{b}	0.80^{b}	0.88^{b}	0.98^{a}	1.09^{a}	1.12^{a}	0.06	0.0033	0.67	0.18
					Cross-linked amino acid								
Lysinoalanine	0.03 ± 0.01	0.01 ± 0.00	0.11	0.01	0.01	0.02	0.01	0.01	0.01	<0.01	0.24	0.67	0.59

[1] Data for whole compared to extruded samples are presented as mean ± standard error of the mean.

[a–b] Means with differing superscripts in the same row are different ($P \leq 0.05$). Means with two sets of superscripts indicate effects of moisture (first value) and temperature (second value). Means with a significant interaction effect (Moisture x Temperature) only report differences between means for interaction effect.

extruded at 22% moisture had higher His, Ile, Leu, phenylalanine (Phe), Thr, and valine (Val) content when compared with samples extruded at 18% moisture ($P < 0.05$). Samples extruded at 18% moisture had higher Met content compared with samples extruded at 22% moisture ($P < 0.05$). Samples extruded at 22% moisture/110, 130, and 150 °C had higher Lys content compared with samples extruded at 18% moisture/130 and 150 °C, and samples extruded at 18% moisture/110 °C had higher Lys content than samples extruded at 18% moisture/130 °C ($P < 0.05$). There was no effect of extrusion moisture or temperature on LAL content between extruded samples.

3.4. Faba bean

3.4.1. Proximate analysis

Data are reported in Table 4 for proximate analysis. When compared with whole samples, extruded samples had a higher DM content ($P < 0.01$). There were no effects of moisture or temperature on DM content when extruded samples were compared with each other. There were no differences in CP content when whole and extruded samples were compared. There was no effect of temperature on CP content when extruded samples were compared with each other, however, samples extruded at 18% moisture had higher CP content when compared to samples extruded at 22% moisture ($P < 0.05$). When compared with whole samples, extruded samples had lower crude fat content ($P < 0.01$). Samples extruded at 130 and 150 °C had higher crude fat content when compared with samples extruded at 110 °C ($P < 0.01$). There were no differences in ash content when whole and extruded samples were compared with one another. There were no effects of moisture or temperature on ash content when extruded samples were compared with each other.

3.4.2. Amino acids

Data are reported in Table 4 for indispensable amino acids and LAL and Table S4 for dispensable amino acids. There were no differences in indispensable amino acid content when whole and extruded samples were compared. When compared with whole samples, extruded samples had lower LAL content ($P < 0.01$). Samples extruded at 18% moisture had higher Val content when compared with samples extruded at 22% moisture ($P < 0.01$). Samples extruded at 130 °C had higher Leu ($P < 0.05$) and Val ($P < 0.01$) content when compared with samples extruded at 110 and 150 °C. Samples extruded at 130 °C had higher Lys content compared to samples extruded at 150 °C ($P \leq 0.05$). Samples extruded at 22% moisture/130 °C had higher His content compared with samples extruded at 22% moisture/110 °C ($P \leq 0.05$). Samples extruded at 22% moisture/130 °C had higher Phe content compared to samples extruded at 22% moisture/150 °C ($P \leq 0.05$). With the exception of Gly, where extruded samples had lower content compared to whole samples ($P < 0.01$), there were no differences between extruded and whole samples for dispensable AA. Samples extruded at 110 and 130 °C had higher Ala and Asp content when compared with samples extruded at 150 °C ($P < 0.05$). Samples extruded at 130 °C had higher Arg, Gly, and Pro content compared with samples extruded at 150 °C ($P < 0.05$). There was a moisture x temperature effect ($P < 0.05$) for Trp, but when means were compared there were no differences. There was no effect of extrusion moisture or temperature on LAL content between extruded samples.

3.5. Lentil

3.5.1. Proximate analysis

Data are reported in Table 5 for proximate analysis. When compared with whole samples, extruded samples had a higher DM content ($P < 0.01$). Samples extruded at 18% moisture\110 °C had higher DM content compared to samples extruded at 22% moisture/110 and 150 °C ($P \leq 0.05$). Samples extruded at 18% moisture/130 °C had higher DM content compared to samples compared to samples extruded at 22% moisture/110 °C. When compared with whole samples, extruded samples had higher CP content ($P < 0.01$). Samples extruded at 150 °C had higher CP content compared to samples extruded at 130 °C. Samples extruded at 18% moisture/150 °C had higher CP content compared to all extruded samples except samples extruded at 22% moisture/110 °C. When compared with whole samples, extruded samples had lower crude fat content ($P < 0.05$). There were no effects of moisture or temperature on crude fat content when extruded samples were compared with each other. When compared with whole samples, extruded samples had higher ash content. There were no effects of moisture or temperature on ash content when extruded samples were compared with each other.

3.5.2. Amino acids

Data are reported in Table 5 for indispensable amino acids and LAL and Table S5 for dispensable amino acids. When compared with whole samples, extruded samples had higher Ile ($P = 0.01$), Leu ($P < 0.05$), Met ($P < 0.01$) and Lys ($P < 0.05$), Trp ($P < 0.05$), and Val ($P < 0.01$) content. There were no differences in LAL content when whole and extruded samples were compared. Samples extruded at 130 °C had higher Met content compared to samples extruded at 110 and 150 °C ($P < 0.01$). With the exception of Gly, where extruded samples had lower content compared to whole samples ($P < 0.05$), there were no differences between extruded and whole samples for dispensable AA. Samples extruded at 18% moisture had higher tyrosine (Tyr) content compared with samples extruded at 22% moisture ($P < 0.01$). Samples extruded at 110 °C had higher Tyr content compared with samples extruded at 130 °C ($P < 0.05$). Samples extruded at 22% moisture/130 °C had higher Cys content compared with samples extruded at 18% moisture/110 and 150 °C and 22% moisture/110 and 150 °C ($P < 0.05$). Samples extruded at 18% moisture/130 °C had higher Cys content compared with samples extruded at 18% moisture/150 °C and 22% moisture/110 and 150 °C ($P < 0.05$). Samples extruded at 22% moisture/150 °C had higher Cys content compared to samples extruded at 22% moisture/110 °C ($P < 0.05$). There was no effect of extrusion

Table 4. Effect of extrusion temperature and moisture on nutrient composition in whole and extruded faba beans, % dry matter basis.

				Moisture 18%			Moisture 22%				*P* value		
	Whole[1]	Extruded[1]	*P* value	110 °C	130 °C	150 °C	110 °C	130 °C	150 °C	SEM	Moisture	Temp.	Moisture × temp.
Dry matter	87.96 ± 0.44	95.36 ± 0.15	<0.0001	94.87	95.14	95.75	95.49	95.22	95.72	0.36	0.45	0.25	0.63
Crude protein	27.31 ± 0.40	27.75 ± 0.10	0.29	28.07^{a}	27.82^{a}	28.05^{a}	27.88^{b}	27.27^{b}	27.51^{b}	0.19	0.02	0.10	0.57
Crude fat	1.53^{a} ± 0.16	0.81^{b} ± 0.05	0.0004	0.67^{b}	0.97^{a}	0.97^{a}	0.50^{b}	0.76^{a}	0.98^{a}	0.09	0.13	0.0040	0.50
Ash	3.31 ± 0.06	3.41 ± 0.02	0.11	3.39	3.42	3.45	3.41	3.35	3.42	0.04	0.47	0.55	0.58
Indispensable amino acids													
Histidine	0.64 ± 0.04	0.59 ± 0.01	0.24	0.60^{ab}	0.61^{ab}	0.55^{ab}	0.51^{b}	0.66^{a}	0.58^{ab}	0.02	0.83	0.01	0.05
Isoleucine	0.85 ± 0.07	0.94 ± 0.02	0.25	0.91	1.01	0.92	0.93	1.02	0.85	0.06	0.78	0.10	0.68
Leucine	1.81 ± 0.10	1.82 ± 0.03	0.93	1.86^{b}	1.93^{a}	1.77^{b}	1.60^{b}	1.95^{a}	1.75^{b}	0.07	0.14	0.01	0.13
Lysine	1.47 ± 0.14	1.59 ± 0.05	0.41	1.64^{ab}	1.74^{a}	1.37^{b}	1.57^{ab}	1.71^{a}	1.49^{b}	0.10	0.94	0.03	0.65
Methionine	0.19 ± 0.02	0.21 ± 0.01	0.25	0.23^{a}	0.21^{b}	0.18^{b}	0.24^{a}	0.19^{b}	0.20^{b}	0.01	0.52	0.0003	0.15
Phenylalanine	1.10 ± 0.05	1.08 ± 0.02	0.61	1.07^{ab}	1.07^{ab}	1.11^{ab}	1.06^{ab}	1.17^{a}	1.00^{b}	0.04	0.81	0.17	0.05
Threonine	0.84 ± 0.07	0.79 ± 0.02	0.73	0.91	0.96	0.85	0.86	0.96	0.85	0.04	0.62	0.06	0.83
Tryptophan	0.18 ± 0.05	0.23 ± 0.02	0.33	0.30	0.25	0.18	0.16	0.27	0.21	0.04	0.31	0.16	0.03
Valine	0.94 ± 0.25	1.02 ± 0.07	0.36	1.00^{b,x}	1.11^{a,x}	1.05^{b,x}	0.85^{b,y}	1.11^{a,y}	0.93^{b,y}	0.03	0.0085	0.0008	0.11
Cross-linked amino acid													
Lysinoalanine	0.04^{a} ± 0.00	0.01^{b} ± 0.00	<0.0001	0.01	0.01	0.01	0.01	0.01	0.01	<0.01	0.96	0.62	0.06

[1] Data for whole compared to extruded samples are presented as mean ± standard error of the mean.

[a–b, x–y] Means with differing superscripts in the same row are different ($P \leq 0.05$). Means with two sets of superscripts indicate effects of moisture (first value) and temperature (second value). Means with a significant interaction effect (moisture × temperature) only report differences between means for interaction effect.

Table 5. Effect of extrusion temperature and moisture on nutrient composition in whole and extruded lentils, % dry matter basis.

				Moisture 18%			Moisture 22%				*P* value		
	Whole[1]	Extruded[1]	*P* value	110 °C	130 °C	150 °C	110 °C	130 °C	150 °C	SEM	Moisture	Temp.	Moisture × temp.
Dry matter	90.45^{b} ± 0.64	95.47^{a} ± 0.21	<0.0001	96.48^{a}	96.08^{ab}	95.43^{abc}	94.34^{c}	95.50^{abc}	95.01^{bc}	0.27	0.0005	0.14	0.01
Crude protein	23.72^{b} ± 0.25	26.78^{a} ± 0.08	<0.0001	26.63^{b}	26.66^{b}	27.18^{a}	26.84^{ab}	26.70^{b}	26.67^{b}	0.08	0.20	0.02	0.0014
Crude fat	0.99^{a} ± 0.16	0.64^{b} ± 0.05	0.05	0.76	0.67	0.71	0.83	0.51	0.38	0.11	0.16	0.09	0.23
Ash	2.49^{b} ± 0.07	2.89^{a} ± 0.02	<0.0001	2.78	2.90	2.90	2.90	2.97	2.93	0.06	0.16	0.25	0.70
Indispensable amino acids													
Histidine	0.53 ± 0.08	0.62 ± 0.03	0.32	0.68	0.58	0.62	0.59	0.67	0.54	0.08	0.73	0.69	0.44
Isoleucine	0.67^{b} ± 0.09	1.01^{a} ± 0.03	0.0024	1.08	1.02	0.95	0.99	0.98	1.05	0.09	0.87	0.87	0.53
Leucine	1.49^{b} ± 0.16	1.87^{a} ± 0.05	0.03	2.08	1.85	1.87	1.84	1.74	1.85	0.15	0.33	0.51	0.76
Lysine	1.20^{b} ± 0.17	1.69^{a} ± 0.06	0.02	1.85	1.61	1.63	1.74	1.52	1.81	0.15	0.94	0.33	0.59
Methionine	0.16^{b} ± 0.01	0.21^{a} ± 0.00	0.0002	0.20^{b}	0.23^{a}	0.20^{b}	0.21^{b}	0.23^{a}	0.20^{b}	0.01	0.30	0.0007	0.18
Phenylalanine	1.03 ± 0.14	1.22 ± 0.05	0.25	1.40	1.17	1.27	1.21	1.22	1.04	0.13	0.26	0.50	0.53
Threonine	0.63 ± 0.13	0.68 ± 0.03	0.07	1.02	0.87	0.94	0.90	0.85	0.95	0.07	0.51	0.35	0.68
Tryptophan	0.14^{b} ± 0.04	0.23^{a} ± 0.01	0.04	0.21	0.21	0.30	0.24	0.18	0.24	0.03	0.43	0.11	0.38
Valine	0.79^{b} ± 0.10	1.13^{a} ± 0.03	0.0039	1.23	1.13	1.07	1.12	1.07	1.19	0.09	0.82	0.75	0.46
Cross-linked amino acid													
Lysinoalanine	0.02 ± 0.01	0.02 ± 0.00	0.62	0.02	0.01	0.01	0.01	0.02	0.01	0.01	0.82	0.61	0.43

[1] Data for whole compared to extruded samples are presented as mean ± standard error of the mean.

[a–c] Means with differing superscripts in the same row are different ($P \leq 0.05$). Means with two sets of superscripts indicate effects of moisture (first value) and temperature (second value). Means with a significant interaction effect (moisture × temperature) only report differences between means for interaction effect.

moisture or temperature on LAL content between extruded samples.

3.6. SBM

3.6.1. Proximate analysis

Data are reported in Table 6 for proximate analysis. When compared with whole samples, extruded samples had a higher DM content ($P < 0.01$). Samples extruded at 18% moisture had higher DM content compared with samples extruded at 22% moisture ($P < 0.01$). When compared with whole samples, extruded samples had lower CP content ($P < 0.01$). Samples extruded at 18% moisture had higher CP content compared with samples extruded at 22% moisture ($P < 0.01$). Samples extruded at 110 and 130 °C had higher CP content compared to 150 °C ($P < 0.01$). When compared with whole samples, extruded samples had higher crude fat content ($P < 0.05$). There were no effects of moisture or temperature on crude fat in extruded samples. When compared with whole samples, extruded samples had lower ash content ($P < 0.01$). Samples extruded at 18% moisture had higher ash content compared to samples extruded at 22% moisture ($P < 0.01$).

3.6.2. Amino acids

Data are reported in Table 6 for indispensable amino acids and LAL and Table S6 for dispensable amino acids. With the exception of His, where extruded samples had higher content when compared with whole samples ($P < 0.01$), there were no differences between whole and extruded samples for all indispensable AA and LAL. Samples extruded at 22% moisture/110 °C had higher His content compared with samples extruded at 22% moisture/150 °C ($P < 0.01$). Samples extruded at 130 °C had higher Phe content compared with samples extruded at 150 °C ($P < 0.05$). There was a moisture x temperature effect ($P < 0.01$) for Val, but when means were compared there were no differences. There was no effect of extrusion moisture or temperature on LAL content between extruded samples. Extruded samples had higher Ala, Asp, Glu, and Ser ($P \leq 0.05$) content and lower Cys ($P < 0.01$) content when compared with whole samples. There were no effects of moisture or temperature on dispensable AA content of extruded samples.

4. Discussion

While pulses are regarded as nutrient dense and sustainable ingredients for animals and humans, further understanding of how different extrusion parameters can affect nutrient composition is desirable. The objective of this study was to document variation in chemical composition of Canadian pulses and SBM when subjected to the same extrusion conditions. This study demonstrated that extrusion conditions can differentially affect the nutrient profile of types of Canadian peas, lentils, chickpeas, and faba beans, and SBM.

Crude protein content of whole pulses and SBM ranged from 21%-51%. Other studies have reported similar CP content in pulses and SBM with ranges of 19%–27% and 45%–50%, respectively (Alonso et al. 1998,2000a; Abd El-Hady and Habiba 2003; Lagos and Stein 2017; Nosworthy et al. 2017; 2018*b*, 2018*a*, 2020). In a variety of peas and in faba beans, previous analyses reported no differences in CP content of raw samples compared to extruded when peas and faba beans were extruded at 25% moisture/148 °C and 25% moisture/152 °C, respectively (Alonso et al. 1998, 2000a). Conversely, extrusion of beans, chickpeas, yellow split peas, and red and green lentils increased protein content, however, these changes are relatively small, thus authors concluded that extrusion did not drastically change crude protein content of pulse ingredients (Nosworthy et al. 2017, 2018*a*; 2018*b*, 2020). These results are similar to the current study, where the effect of extrusion on CP differs by pulse type, but overall changes are relatively small. Abd El-Hady and Habiba 2003 reported that extrusion temperature, but not moisture (18% and 22%), significantly changed CP content of faba beans, chickpeas, kidney beans, and peas; samples extruded at 180 °C had significantly lower CP content compared to samples extruded at 140 °C. However, similarly to the data reported by Nosworthy et al. 2017, 2018*a*; 2018*b*, 2020, changes were small (less than 1%). Interestingly, faba bean samples processed at 18% moisture had higher CP content compared to those processed at 22% moisture. These results differ from Abd El-Hady and Habiba (2003), who reported no differences in CP content due to moisture content. In the current study, and differing from the pulses, CP content of SBM decreased when samples were extruded. In extruded soy flour and soy/acha blends, this decrease was not observed (Dust et al. 2004; Anuonye et al. 2010), however, consideration of product type (soy flour vs SBM) may be the reason there are differences. One other consideration is changes in protein content could be due to grinding prior to extrusion and related to pulse type. Crude protein is reported to increase in raw dehulled peas compared to whole peas (Alonso et al. 1998; Wang et al. 2008). In the current study, pulses were not dehulled prior to additional processing, but a portion of the hull may have been lost during grinding, resulting in an apparent increase in CP content of ground pulses due to losses of other nutrients (e.g., fibre) present in the hull. It appears that changes in protein content are not only affected by extrusion parameters, but exhibit differential effects between pulse types. Thus, depending on the on nutritional targets for the final extruded product, ingredient type and prior processing should be considered.

Indispensable and dispensable AA content of whole pulses were similar to those reported in literature (Alonso et al. 2000b; Nosworthy et al. 2017, 2018*a*; 2018*b*, 2020; Mayer Labba et al. 2021). Amino acid content of SBM was similar to those reported by Lagos and Stein (2017), but lower than those reported by Grieshop et al. (2003). Indispensable AA analysis of pulses and SBM in this study were consistent with previous analysis with sulfur AA or Trp being the limiting AA.

For the purpose of this discussion, the focus will be primarily on the limiting AA of legumes/pulses and potential loss of AA. In this study, changes in Met content were ingredient dependent. There were no significant changes in Met content of Amarillo and Dun peas, and SBM due to processing. This lack

Table 6. Effect of extrusion temperature and moisture on nutrient composition in whole and extruded soybean meal, % dry matter basis.

				Moisture 18%			Moisture 22%				*P* value		
	Whole[1]	Extruded[1]	*P* value	110 °C	130 °C	150 °C	110 °C	130 °C	150 °C	SEM	Moisture	Temp.	Moisture × temp.
Dry matter	87.71^{b} ± 0.26	96.64^{a} ± 0.09	<0.0001	96.96^{a}	96.87^{a}	96.87^{a}	96.62^{b}	96.19^{b}	96.32^{b}	0.15	0.0012	0.24	0.55
Crude protein	51.66^{a} ± 0.67	49.2^{b} ± 0.22	0.0025	$50.05^{a, x}$	$50.10^{a, x}$	$48.51^{a, y}$	$49.34^{b, x}$	$49.62^{b, x}$	$47.56^{b, y}$	0.15	<0.0001	<0.0001	0.30
Crude fat	1.11^{b} ± 0.14	1.51^{a} ± 0.05	0.01	1.32	1.55	1.63	1.49	1.64	1.46	0.08	0.63	0.07	0.10
Ash	6.69^{a} ± 0.16	6.11^{b} ± 0.06	0.0050	6.26^{a}	6.33^{a}	6.21^{a}	6.13^{b}	5.81^{b}	5.80^{b}	0.09	0.0017	0.17	0.17
Indispensable amino acids													
Histidine	1.02^{b} ± 0.06	1.23^{a} ± 0.02	0.0076	1.16^{ab}	1.24^{ab}	1.20^{ab}	1.35^{a}	1.26^{ab}	1.13^{b}	0.04	0.20	0.09	0.03
Isoleucine	1.75 ± 0.14	1.98 ± 0.05	0.14	1.75	2.04	2.04	2.13	2.09	1.73	0.09	0.59	0.13	0.0086
Leucine	3.56 ± 0.14	3.75 ± 0.05	0.23	3.58	3.81	3.73	3.86	3.89	3.54	0.10	0.48	0.11	0.09
Lysine	2.80 ± 0.12	2.86 ± 0.04	0.64	2.70	2.85	2.86	2.78	3.15	2.86	0.09	0.12	0.06	0.24
Methionine	0.66 ± 0.03	0.62 ± 0.01	0.24	0.65	0.64	0.60	0.61	0.59	0.61	0.03	0.27	0.67	0.55
Phenylalanine	2.38 ± 0.12	2.37 ± 0.04	0.92	2.27^{ab}	2.47^{a}	2.31^{b}	2.51^{ab}	2.52^{a}	2.15^{b}	0.08	0.52	0.01	0.09
Threonine	1.67 ± 0.12	1.63 ± 0.03	0.11	1.78	1.87	1.91	1.76	1.92	1.79	0.05	0.51	0.12	0.31
Tryptophan	0.62 ± 0.07	0.66 ± 0.02	0.56	0.67	0.69	0.74	0.53	0.69	0.66	0.05	0.09	0.11	0.41
Valine	1.61 ± 0.20	2.02 ± 0.05	0.06	1.81	2.09	2.12	2.14	2.19	1.80	0.09	0.63	0.14	0.01
Cross-linked amino acid													
Lysinoalanine	0.02 ± 0.00	0.01 ± 0.00	0.53	0.02	0.02	0.01	0.01	0.02	0.01	0.02	0.82	0.21	0.90

[1] Data for whole compared to extruded samples are presented as mean ± standard error of the mean.

[a–b, x–y] Means with differing superscripts in the same row are different ($P \leq 0.05$). Means with two sets of superscripts indicate effects of moisture (first value) and temperature (second value). Means with a significant interaction effect (moisture × temperature) only report differences between means for interaction effect.

of change in peas is opposite to data reported by Alonso et al. (2000*b*), where extrusion (25% moisture, 145 °C) significantly decreased Met content of extruded peas from 1.02% to 0.67%. In extruded SBM protein concentrate there were no reported changes in Met content (Jeunink and Cheftel 1979). Increases in Met content of chickpeas, faba beans, and lentils between whole and extruded pulse samples could be due to inaccurate measurement of sulfur-containing amino acids (SAA) during analysis. Performic acid-oxidation and subsequent hydrolysis of whole pulses samples may not be sufficient in releasing all SAA during digestion. In contrast, exposing pulse samples to moisture, heat, and pressure through extrusion may have cause more SAA to become available in the sample, which were captured in the AA digestion and analysis process. This could suggest that thermal treatment of pulses may increases the available amounts of SAA in pulse ingredients compared to raw, but may also highlight an error with how SAA are analyzed in raw ingredients not exposed to thermal processing if total SAA are not being released during the digestion process.

Changes in Lys are also an important indication of how processing affects protein/AA. For the majority of samples analyzed in this study there were no significant changes in Lys content, suggesting there are little to no chemical alterations/losses of Lys in extruded pulses and SBM samples. This is further supported by the lack of significant differences in LAL content of pulses, with the exception of faba bean. Similarly, previous analysis of LAL reported no additional formation of LAL occurred when samples in extruded faba bean and soy protein concentrates (Jeunink and Cheftel 1979). In the present study, whole samples of faba had higher levels of LAL compared to extruded samples. It is hypothesized that significant formation of LAL during preparation (grinding) and (or) during analysis of the whole samples which could account for the increased LAL content; this occurrence was also hypothesized by Jeunink and Cheftel (1979). Alternatively, there may have been some analytical inaccuracies, such as the temperature of the sample increasing above the digestion temperature, which could account for the higher LAL content of the raw faba bean samples. Since there were no differences in Trp content of extruded pulses and SBM samples compared to whole/ground, extrusion may not improve protein quality if Trp is the dietary limiting AA.

Although there were changes in some dispensable AA, many of these changes do not directly impact protein quality measurements of pulses, with the exception of Cys and Tyr, which are incorporated in protein quality calculations for sulfur AA and aromatic AA, respectively. There were no significant differences in Phe or Tyr content in extruded pulse and SBM samples compared to whole/ground. Thus, changes in protein quality of extruded pulses are most likely linked to the changes in limiting AA Met, Cys, and Trp. Similarly to Met, changes in Cys due to processing were also ingredient dependent. In the current study, changes (or no difference) in Cys content were opposite to results reported by Jeunink and Cheftel (1979) who reported a decrease in cysteine + cystine in extruded faba bean protein concentrate and no change in extruded SBM protein concentrate. These differences in Cys content may be due to the form in which the pulses and SBM were processed (whole vs meal vs protein concentrate).

Overall, there were differences between means for some AA, depending on pulse type; yet, there was no ideal moisture or temperature which consistently increased AA content. This suggests that, similarly to CP, extrusion processing may alter AA content of pulses and SBM, but is not dependent on a specific moisture or temperature to do so. Ultimately, it appears AA generally remain stable during extrusion processing.

Dry matter content of whole pulses and SBM ranged from 86% to 90%. Dry matter content of whole pulses were slightly lower, or of similar content, compared to values reported in other studies, which range from 89–93% DM (de Almeida Costa et al. 2006; Rathod and Annapure 2016; Nosworthy et al. 2017, 2018*a*; 2018*b*, 2020). Dry matter content of SBM was similar to values reported by (Lagos and Stein 2017) which ranged from 88–89% and was similar to DM content of whole pulses (87%). Significant changes in DM content of extruded samples compared to whole are attributed to the drying process of the extrudate post-extrusion.

Crude fat content of whole pulses and SBM ranged from 0.99% to 6.03%, and is similar to values reported in literature which ranges from 1% to 6% for pulses (de Almeida Costa et al. 2006; Rathod and Annapure 2016; Nosworthy et al. 2017, 2018*a*; 2018*b*, 2020) and 1% to 3% for SBM/soy flours (Iwe et al. 2001; Anuonye et al. 2010; Lagos and Stein 2017). Crude fat content was significantly decreased due to extrusion in pulse samples. Extreme processing conditions, such as extrusion, can potentially affect lipid stability and decrease lipid content of extruded samples (Björck and Asp 1983). Another potential explanation for a decrease in fat content, is that fat may complex with amylose during extrusion, with maximum complexing occurring at temperatures from 110 to 140 °C (Bhatnagar and Hanna 1994).

Ash content of whole pulses ranged from 2% to 3%, with lentil and faba beans having the lowest and highest ash content of pulses, respectively, and is consistent with data reported by de Almeida Costa et al. (2006). Ash content of SBM was twice the amount of pulses, at ~6%, which is consistent with data reported by Lagos and Stein (2017), where a variety of SBM were reported to have ~6% ash content. Ash content of pulses either remained the same or increased due to extrusion, whereas ash content of extruded samples in SBM decreased. These changes could have implications on total mineral supply in extruded diets (e.g., dog kibble).

There were some limitations to this study, the primary limitation being that processing only focused on a singular ingredient rather than a food matrix comprised of multiple ingredients. During mixing and extrusion of ingredients there may be increased nutrient interactions that are not accounted for by the processing of a single ingredient. Additionally, the limiting AA of the processed pulses, may not be the same limiting AA in a diet or food matrix since ingredients can be complementary such as pulses (low in SAA, rich in Lys) and cereal grains (low in Lys, rich in SAA) when mixed in a food or diet matrix. Anti-nutritional factors were not measured in this study, but future research should consider running ANF analyses to help account for potential nutrient changes.

5. Conclusion

Overall, there is no ideal extrusion moisture or temperature for all ingredients investigated. Rather, it appears nutrient changes are ingredient dependent in addition to potential processing effects. This study assessed specific extrusion conditions, that could be modified and refined depending on the applications. There were limited losses in regards to AA, with no loss of sulfur AA. There was no loss of Lys content and LAL formation was minimal suggesting that processing of pulses at 18 and 22% moisture between 110 and 150 °C did not cause major chemical alterations of AA. Results further suggest that ingredient specific heating regimes, rather than standardized heating regime, should be utilized. Further investigation is warranted to determine optimal extrusion parameters for pulses and to develop a deeper set of data to truly understand the effects of extrusion.

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Supplementary material

Supplementary data are available with the article at https://doi.org/10.1139/cjas-2022-0088.

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