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Use of a Dry Fractionation Process To Manipulate the Chemical Profile and Nutrient Supply of a Coproduct from Bioethanol Processing

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ABSTRACT: With an available processing technology (fractionation), coproducts from bioethanol processing (wheat dried distillers grains with solubles, DDGS) could be fractionated to a desired/optimal chemical and nutrient profile. There is no study, to the author's knowledge, on manipulating nutrient profiles through fractionation processing in bioethanol coproducts in ruminants. The objectives of this study were to investigate the effect of fractionation processing of a coproduct from bioethanol processing (wheat DDGS) on the metabolic characteristics of the proteins and to study the effects of fractionation processing on the magnitude of changes in chemical and nutrient supply to ruminants by comparing chemical and nutrient characterization, in situ rumen degradation kinetics, truly absorbed protein supply, and protein degraded balance among different fractions of coproduct of wheat DDGS. In this study, wheat DDGS was dry fractionationed into A, B, C, and D fractions according to particle size, gravity, and protein and fiber contents. The results showed that the fractionation processing changed wheat DDGS chemical and nutrient profiles. NDF and ADF increased from fraction A to D (NDF, from 330 to 424; ADF, from 135 to 175 g/kg DM). Subsequently, CP decreased (CP, from 499 to 363 g/kg DM), whereas soluble CP, NPN, and carbohydrate increased (SCP, from 247 to 304 g/kg CP; NPN, from 476 to 943 g/kg SCP; CHO, from 409 to 538 g/kg DM) from fraction A to D. The CNCPS protein and carbohydrate subfractions were also changed by the fractionation processing. Effective degradability of DM and CP and total digestible protein decreased from fraction A to D (EDDM, from 734 to 649; EDCP, from 321 to 241; TDP, from 442 to 312 g/kg DM). Total truly absorbed protein in the small intestine decreased from fraction A to D (DVE value, from 186 to 124 g/kg DM; MP in NRC-2001, from 193 to 136 g/kg DM). Degraded protein balance decreased from wheat DDGS fractions A–D (DPB in the DVE/OEB system, from 245 to 161 g/kg DM; DPB in NRC-2001, from 242 to 158 g/kg DM). The fractionation processing had a great impact on the chemical and nutrition profiles. Total truly digested and absorbed protein supply and degraded protein balance were decreased. The processing relatively optimized the protein degraded balance of the coproducts to dairy cattle. Compared with the original wheat DDGS (without fractionation), fractionation processing decreased truly absorbed protein supply of DVE and MP values. In conclusion, fractionation processing can be used to manipulate the nutrient supply and N-to-energy degradation synchronization ratio of coproducts from bioethanol processing. Among the fractions, fraction A was the best in terms of its highest truly absorbed protein DVE and MP values. Fractionation processing has great potential to fractionate a coproduct into a desired and optimal chemical and nutrient profile. To the author's knowledge, this is the first paper to show that with fractionation processing, the coproducts from bioethanol processing (wheat DDGS) could be manipulated to provide a desired/optimized nutrient supply to ruminants.

KEYWORDS: *fractionation processing, degraded protein balance, truly absorbed protein supply, coproducts from bioethanol processing*

INTRODUCTION

Dried distillers' grains with solubles (DDGS) are important coproducts of fermentation during bioethanol production.[1,2] As a result of government policies for stimulating the expansion and consumption of bioethanol in North America, various DDGS products are currently manufactured.[1] Because the starch is removed and the other components are concentrated, the characteristics of high protein, high fiber, high fat, and low starch make DDGS an attractive ingredient in dairy and beef cattle diets,[2−4] but not desired for monogastric animals, poultry, and swine diets. In recent years, detailed research that evaluated the nutritive values of original DDGS has been done by Neuz-Ortin and Yu[5−9] and on corn DDGS,[4−13] wheat DDGS,[5−9,14−17] barley DDGS,[18] and blended DDGS.[5−9] However, so far, no research has been conducted on the effects of fractionation of DDGS on feed quality and nutrient profile in ruminants.

With an available processing technology (fractionation), food can be fractionated to a desired/optimal chemical and nutrient profile.[19−21] However, this fractionation processing has not been used in the bioethanol coproduct industry. There is no study, to our knowledge, on manipulating nutrient profiles of coproducts from bioethanol processing through fractionation

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Table 1. Particle Size, Yield, Chemical Composition of Various Wheat DDGS Fractions Produced by Particle Size, and Weight Separation Using Continuous-Flow and Vibratory Equipment

fraction no.	equipment	fraction name	particle size, μm	yield %, DM	CP %, DM	ADF %, DM	NDF %, DM
1	SWECO	Thrus 30M	232 ± 3.3	22.85 ± 1.09	52.67 ± 1.21	12.38 ± 1.56	22.16 ± 1.11
2	SWECO	Thrus 40M	302 ± 50.4	3.84 ± 1.09	52.12 ± 0.79	10.91 ± 1.29	22.00 ± 2.13
3	gravity table	GT1	471 ± 36.4	9.17 ± 0.75	49.97 ± 0.83	13.41 ± 1.89	23.92 ± 1.97
4	gravity table	GT2	543 ± 33.3	20.50 ± 1.22	45.07 ± 1.11	14.75 ± 1.39	29.10 ± 1.78
5	gravity table	GT3	711 ± 14.0	30.67 ± 1.97	40.52 ± 0.37	17.02 ± 1.60	33.54 ± 0.90
6	gravity table	GT4	766 ± 30.6	32.67 ± 1.75	38.17 ± 0.98	18.01 ± 1.52	38.18 ± 1.01
7	gravity table	GT5	619 ± 28.7	5.67 ± 1.37	32.09 ± 0.28	21.76 ± 0.97	42.96 ± 0.71

processing in ruminants. The objectives of this study were to investigate the effect of fractionation processing of coproducts from bioethanol processing (wheat DDGS) on changes of feed nutritive quality by comparing the wheat DDGS fractions in terms of (1) chemical profiles; (2) protein and carbohydrate fractions using the Cornell Net Carbohydrate and Protein System (CNCPS); (3) truly digestible nutrients and energy values in ruminants; (4) in situ rumen degradation kinetics; (5) intestinal digestibility of rumen undegraded protein; and (6) metabolizable protein and truly absorbable protein supply. The hypothesis of this study was that by fractionation processing, chemical and nutrient profiles from different fractions of wheat DDGS were changed, therefore resulting in a significant impact on chemical profiles and nutrient utilization and availability. To our knowledge, this is the first paper to show that with fractionation processing the coproducts from bioethanol processing (wheat DDGS) could be manipulated to provide a desired/optimized nutrient supply to ruminants.

MATERIALS AND METHODS

Fractionation Processing of Coproduct. The fractionation processing of coproduct (wheat DDGS) was carried out in Dr. E. Beltranena' laboratory at Agri-Food Discovery Place (Edmonton, AB, Canada). Dry fractions of wheat DDGS were produced by particle size and weight separation using continuous-flow and vibratory equipment. Separation by particle size was conducted using a SWECO ZS30 vibro-separator (SWECO Inc., Florence, KY, USA) equipped with three circular sieves, 30 M (600 μm), 40 M (425 μm), at a rate of 24 kg/h. The material that remained and suspended over the 30 M sieve was separated by differential weight using a Westrup LA-K gravity separator with feed vibration of 6.5 (0−10), air supply of 1 (0−10), long side inclination of 2.0, and short side inclination of 2.5 at a rate of 23 kg/h into fractions (Table 1). The fractions of similar CP and ADF content were pooled. The two SWECO fractions were combined as fraction A. The gravity table 1 (GT1) and gravity table 2 (GT2) fractions were combined as fraction B. The GT3 fraction was renamed fraction C, and the GT4 and GT5 fractions were combined as fraction D. In total we obtained four fractions through this fractionation processing. Table 1 provides detailed information.

Animals and Diets. The in situ experiments were carried out at the Livestock Research Station, University of Saskatchewan, Canada. Three lactating Holstein cows with flexible rumen cannulae (10 cm internal diameter, Bar Diamond Inc., Parma, ID, USA) were used in this study. Cows were housed in pens of approximately 1.5 × 3 m in the research barn. The cows were fed twice daily at 8:00 a.m.and 4:00 p.m. with equal allotments of a diet consisting of 51% barley silage, 15% chopped alfalfa hay, and 34% concentrate (56% barley, 5% wheat, 5% oats, 33% dairy supplement pellets, and 1% molasses) according to the dairy nutrient requirements defined by NRC.[22] Water was supplied ad libitum. All animal care and handling used in this study was in accordance with the guidelines approved by the Canadian Council on Animal Care.[23]

In Situ Rumen Degradation Kinetics. Rumen degradation parameters were determined using the in situ method.[24] Before rumen incubation, the wheat DDGS fraction samples A−D were processed using a Sven roller mill (Apollo Machine and Products Ltd., Saskatoon, Canada). The roller gap was adjusted to a size of 0.203 mm to equalize the particle size of all samples according to published suggestion.[24] Approximately 7 g of wheat DDGS fraction sample was placed into each numbered nylon bag (10 × 20 cm) with the pore size of 41 μm (Nitex 03-41/31, Screentec Crop., Mississagua, ON, Canada). The ratio of sample size to bag surface area was equal to 17.5 mg cm^{-2}, which is within the range recommended by a published report.[25] A polyester mesh bag was used to hold the sample bags in the rumen, which was 45 × 45 cm with a 90 cm length of rope to be anchored to the cannula. Sample bags were added to the polyester mesh bag according to the gradual addition−all out schedule and incubated for 60, 48, 36, 24, 12, 8, 4, and 0 h. The number of bags for each treatment at each incubation time in each experiment run were 2, 2, 2, 3, 3, 4, 5, and 5 bags for incubation times of 0, 4, 8, 12, 24, 36, 48, and 60 h, respectively. The maximum number of bags in the rumen at any one time was around 32.[26] Treatments were randomly assigned to the three lactating cows in two in situ experimental runs. After incubation, the bags were removed from the rumen and, including those samples for 0 h, and rinsed under cold water to remove excess ruminal contents. The bags were washed with cool water without detergent six times. Then samples in washed bags were dried in a 55 °C forced-air oven for 48 h and then stored at 4 °C until analysis.

Rumen degradation characteristics of dry matter (DM) and crude protein (CP) were calculated using the NLIN procedure of the statistical package of SAS.[27] The first-order kinetics equation by Tamminga et al.[28] is

$$R(t) = U + D \times \exp^{(-K_d \times (t - T_0))}$$

where $R(t)$ stands for residue of the incubated material after t h of rumen incubation (g/kg); U and D (in g/kg) stand for undegradable and potentially degradable fractions, respectively; lag time (T_0) is in h; and the degradation rate (K_d) is in %/h.

Chemical Analysis. The samples of wheat DDGS fractions and in situ residues for chemical analysis were ground through a 1 mm screen (Retsch ZM-1, Brinkmann Instruments Ltd., Mississauga, ON, Canada). All samples were then analyzed for DM (AOAC method 930.15), ash (AOAC method 942.05), ether extract (EE; AOAC method 920.39), and CP (AOAC method 984.13; Kjeltec 2400).[29] Neutral detergent insoluble protein (NDICP) and acid detergent insoluble protein (ADICP) were determined according to the procedures of Lacitra et al.[30] Soluble CP (SCP) was determined by incubating the sample with bicarbonate−phosphate buffer and filtering through Whatman no. 54 filter paper.[31] Nonprotein nitrogen (NPN) was analyzed by the precipitation of true protein in the filtrate with tungstic acid and determined as the difference between total nitrogen and the nitrogen content of the residue after filtration. The starch was analyzed by using the Megazyme Total Starch Assay Kit (AOAC method 996.11).[32] The neutral detergent fiber (NDF), acid detergent fiber (ADF), and acid detergent lignin (ADL) were analyzed by Ankom filter bag method (Ankom A200 filter bag technique, Ankom Technology, Fairport, NY, USA) according to the procedures of Van Soest et al.[33] A heat-stable α-amylase was used in NDF determination. The contents of total carbohydrate (CHO), nonfiber CHO (NFC), hemicellulose, and cellulose were calculated according to the NRC.[22]

Table 2. Effect of Fractionation Processing of Coproducts from Bioethanol Processing (Wheat DDGS) on Chemical Profiles

item	fractionation of coproduct (wheat DDGS)			
	A	B	C	D
basic chemical composition (g/kg DM)				
dry matter (DM, g/kg)	923 ± 0.2	926 ± 0.3	920 ± 0.1	930 ± 0.7
ash	54 ± 0.2	54 ± 0.0	54 ± 0.1	54 ± 0.2
ether extract (EE)	48 ± 0.1	50 ± 0.3	47 ± 0.5	46 ± 0.1
structural and non-structural CHO profiles (g/kg DM)				
total carbohydrate (CHO)	409 ± 1.1	474 ± 3.9	515 ± 1.4	538 ± 2.2
starch	21 ± 0.4	16 ± 0.6	14 ± 0.3	14 ± 1.2
neutral detergent fiber (NDF)	330 ± 6.1	372 ± 4.3	416 ± 0.9	424 ± 3.9
acid detergent fiber (ADF)	135 ± 0.2	162 ± 0.6	173 ± 4.6	175 ± 1.5
acid detergent lignin (ADL)	65 ± 0.7	65 ± 0.2	58 ± 15.5	57 ± 2.7
hemicellulose	195 ± 8.3	211 ± 4.9	243 ± 5.5	249 ± 5.4
cellulose	70 ± 1.5	97 ± 0.4	115 ± 10.9	118 ± 4.2
protein profile				
crude protein (CP, g/kg DM)	499 ± 0.9	422 ± 3.6	384 ± 1.0	363 ± 2.3
soluble CP (SCP, g/kg CP)	247 ± 3.7	269 ± 6.9	271 ± 3.6	304 ± 0.5
neutral detergent insoluble CP (NDICP, g/kg CP)	387 ± 10.8	402 ± 5.3	416 ± 0.5	404 ± 1.0
acid detergent insoluble CP (ADICP, g/kg CP)	144 ± 10.3	150 ± 5.3	145 ± 0.6	124 ± 0.5
nonprotein nitrogen (NPN, g/kg CP)	118 ± 10.4	156 ± 14.2	244 ± 26.4	287 ± 16.0
nonprotein nitrogen (NPN, g/kg SCP)	476 ± 35.2	579 ± 38.1	899 ± 85.6	943 ± 53.9

Protein and Carbohydrate Fractions Partitioning. Crude protein and CHO of each wheat DDGS fraction were partitioned into five and four subfractions, respectively, using the CNCPS.[34] These fractions are associated with degradation rate and behaviors.[34] In the CNCPS system,[34] CP is divided into three subfractions (PA, PB, and PC) according to their rates of degradation and availability in the rumen. Due to different K_d in the rumen, subfraction PB is further divided into three fractions (PB_1, PB_2, and PB_3). Similarly, the subfractions of CHO include subfraction CA, which is rapidly degradable with a K_d of 300%/h, subfraction CB_1, which is intermediately degradable with an intermediate K_d of 20−50%/h, subfraction CB_2, which is slowly degraded in the rumen with a low degradation rate of 2−10%/h, and subfraction CC, which is undegradable CHO and calculated on the basis of ADL.

Truly Digestible Nutrients and Energy Values. The values of truly digestible CP (tdCP), truly digestible fatty acid (tdFA), truly digestible NDF (tdNDF), and truly digestible NFC (tdNFC) were determined on the basis of the formulas of Nutrient Requirements of Dairy Cattle.[22,34] The values of total digestible nutrients at a maintenance level (TDN_{1x}), digestible energy at a maintenance level (DE_{1x}), metabolizable energy at a production level when intake is 3 times the maintenance intake (ME_{3x}), and net energy for lactation when intake is 3 times the maintenance intake (NE_{L3x}) were estimated on the basis of the chemical summative approach,[22] whereas net energy for maintenance for growing animal (NE_m) and net energy for retention or gain (NE_g) were calculated using the formulas given in *Nutrient Requirements of Beef Cattle*.[36]

Prediction of Potential Nutrient Supply Using the DVE/OEB System and NRC Model. The DVE/OEB system[28] and NRC-2001 model[22] were used in this study for estimating the truly absorbed protein supply in the small intestine and degraded protein balance. The detailed concept and principle have been reported by Tamminga et al.[28] and NRC-2001.[22] The comparison between two systems (DVE/OEB vs NRC-2001) has been reported by Yu[37] in concentrate and by Yu et al.[38] in forage.

Briefly, in the DVE/OEB system, DVE is truly absorbed protein in the small intestine, mainly contributed by (1) truly absorbed rumen synthesized microbial protein in the small intestine ($AMCP^{DVE}$), (2) truly absorbed rumen undegraded protein in the small intestine ($ARUP^{DVE}$), and (3) a correction for endogenous protein losses in the digestive tract (ENDP) and calculated as $DVE = AMCP^{DVE} + ARUP^{DVE} - ENDP$. Degraded protein balance (DPB^{OEB}) reflects the (im)balance between microbial protein synthesis potentially possible from available rumen degraded CP ($MCP_{RDP}{}^{DVE}$) and that potentially possible from the energy extracted during rumen anaerobic fermentation (MCP_{FOM}, based on fermented organic matter (FOM) in the rumen) and calculated as $DPB^{OEB} = MCP_{RDP}{}^{DVE} - MCP_{FOM}$. A positive DPB^{OEB} value indicates potential loss of nitrogen from the rumen. A negative DPB^{OEB} value indicates that microbial protein synthesis may be impaired due to a shortage of N in the rumen. The optimum DPB^{OEB} value in a ration should be therefore zero or slightly above.

Similarly, in the NRC-2001 model, metabolizable protein (MP) is defined as the true protein that is digested postruminally and the component amino acids absorbed by the intestine, contributed by (1) truly absorbed rumen synthesized microbial protein in the small intestine (AMP^{NRC}), (2) truly absorbed rumen undegraded protein in the small intestine ($ARUP^{NRC}$), and (3) endogenous CP (AECP) and calculated as $MP = AMP^{NRC} + ARUP^{NRC} + AECP$. The degraded protein balance (DPB^{NRC}) reflects the difference between the potential microbial protein synthesis based on rumen degraded protein and that based on energy (TDN) available for microbial fermentation in the rumen, calculated as $DPB^{NRC} = RDP^{NRC} - 1.18MCP_{TDN}$[37,38] The optimum DPB^{NRC} value in a ration should be zero or slightly above.[28,37,38]

Statistical Analysis. Statistical analysis was performed with SAS 9.2.[27] The data of chemical and nutrient profiles from fractions A−D were analyzed as mean and standard deviation (laboratory replications). The data of in situ rumen degradation and estimated intestinal digestion were analyzed using the Mixed Procedure with the model $Y_{ij} = \mu + a_i + b_j + \varepsilon_{ij}$, where Y_{ij} was the measured variable; μ was the overall mean; a_i was the effect of DDGS fractions of A−D, as a fixed effect; b_j was the random effect of in situ run; and ε_{ij} was the random error associated with observation ij. For all statistical analyses, significance was declared at $P < 0.05$. Treatment means were compared using the Tukey method.

RESULTS AND DISCUSSION

Effects of Fractionation Processing on Chemical Characteristics and Profiles. The chemical and nutrient profiles of wheat DDGS affected by fractionation processing are presented in Table 2. By comparison among the four fractions (A−D), the fractionation processing did not change DM and ash contents, but changed other chemical compositions. For structural and nonstructural CHO profiles (Table 2), the

Table 3. Effect of Fractionation Processing of Coproducts from Bioethanol Processing (Wheat DDGS) on CNCPS[a] Protein and Carbohydrates Subfractions

item	fractionation of coproduct (wheat DDGS)			
	A	B	C	D
protein subfractions (g/kg CP)				
rapidly degradable protein subfraction (PA)	118 ± 10.4	156 ± 14.2	244 ± 26.4	287 ± 16.0
rapidly degradable protein subfraction (PB_1)	129 ± 6.8	113 ± 7.4	27 ± 22.9	17 ± 16.4
intermediately degradable protein subfraction (PB_2)	366 ± 14.5	329 ± 13.0	313 ± 4.1	292 ± 0.6
slowly degradable protein subfraction (PB_3)	243 ± 21.1	252 ± 0.8	271 ± 0.1	280 ± 0.5
undegradable protein subfraction (PC)	144 ± 10.3	150 ± 5.3	145 ± 0.6	124 ± 0.5
carbohydrate subfractions (g/kg CHO)				
rapidly fermented carbohydrate subfraction (CA)	575 ± 5.7	538 ± 3.4	475 ± 1.5	457 ± 10.9
intermediately degraded carbohydrate subfraction (CB_1)	47 ± 0.7	34 ± 0.8	28 ± 0.4	27 ± 2.5
slowly degraded carbohydrate subfraction (CB_2)	0 ± 0.0	97 ± 2.4	227 ± 73.5	263 ± 4.8
unavailable cell wall (CC)	378 ± 5.1	330 ± 1.8	270 ± 71.6	253 ± 13.2

[a]CNCPS, Cornell Net Carbohydrate and Protein System.

results showed that total CHO was numerically increased by 32% in fraction D when compared with fraction A. The structural carbohydrates of NDF, ADF, hemicellulose, and cellulose contents were also numerically increased by 28, 30, 28, and 69%, respectively, in fraction D when compared with fraction A. The highest increase was cellulose. As to nonstructural carbohydrates, starch and NFC were reduced by 33 and 5%, respectively, in fraction D when compared with fraction A.

With regard to protein profile, by fractionation processing, CP and ADICP from fractionation A–D decreased, but SCP, NDICP, and NPN increased (Table 2). The CP was numerically decreased by 27% from fraction A to fraction D. However, SCP and NDICP were numerically increased by 23 and 4%, respectively. The highest increase from fraction A to D was NPN (2.4 times). The results indicate that the fractionation processing manipulates the chemical profiles of wheat DDGS.

In the literature, the detailed chemical and nutrient profiles of original DDGS (without fractionation) have been reported by Neuz-Ortin and Yu.[5] However, there is no report on the chemical and nutrient profiles of DDGS after fractionation, so there is no comparison that could be made with the current study in DDGS fractions. However, the use of fractionation processing to fractionate plant seeds (canola, flax, oats, barley, wheat, lentils, and peas) has been reported by Zijlstra et al.[21] They concluded that the plant seeds can be fractionated successfully for animal nutrition.

Compared with original DDGS reported by Neuz-Ortin and Yu,[5] the values of chemical profiles in wheat DDGS after fractionation processing in the present study were completely different. The changes through fractionation had a great effect on the nutrient profiles and subsequently the quality of wheat DDGS as a feed in ruminants.

Effects of Fractionation Processing on CNCPS Protein and Carbohydrate Subfractions. Fractionation processing of wheat DDGS into A, B, C, and D fractions changed CNCPS CP and CHO subfraction profiles (Table 3). According to Sniffen et al.,[34] protein can be partitioned into PA, PB_1, PB_2, PB_3, and PC fractions. These fraction profiles have a significant impact on protein degradation kinetics and bypassed protein. Because CHO fractions have different degradation rates, carbohydrate can be partitioned into CA, CB_1, CB_2, and CC fractions.[34] Again, these CHO fractions are associated with different CHO degradation kinetics and affect rumen fermentable energy for microorganism synthesis. In this study, subfraction PA (infinity degradable protein fraction) was increased by 2.4 times from fraction A to fraction D. Subfractions PB_1 (rapidly degradable protein fraction) and PB_2 (intermediately degradable fraction) were decreased by 7.6 and 1.3 times, respectively, whereas fraction PB_3 (slowly degradable protein fraction) was increased by 15% from fraction A to fraction D. There was not much change in unavailable protein fraction of PC (Table 3). These results indicated that the fractionation processing changed protein subfractions of PA and PB without much affect on the PC fraction. This is a favorite change.

In subfractions of CHO, CB_2 (slowly degradable CHO fraction) was increased dramatically (0 vs 263 g/kg CHO), whereas subfractions CA (rapidly degradable), CB_1 (intermediately degradable CHO fraction), and CC (unavailable carbohydrate associated with lignin and cell wall) were decreased by 21, 42, and 7% from fraction A to fraction D. These changes exactly coincided with their chemical changes in Table 1.

Again, there is no report on the protein and CHO profiles of DDGS after fractionation. No comparison could be made with the current study. Nuez-Ortin and Yu[5,6] reported the CNCPS protein and carbohydrate subfractions for both wheat and wheat DDGS (wDDGS) with PA = 218 (in wheat) and 163 (in wDDGS) versus 118–287 (in wDDGS fractions A–D); PB_1 = 28 (wheat) and 0 (wDDGS) versus 129–17 (wDDGS fractions); PB_2 = 619 (wheat) and 277 (wDDGS) versus 366–292 (wDDGS fractions); PB_3 = 135 (wheat) and 512 (wDDGS) versus 243–280 (wDDGS fractions); PC = 0 (wheat) and 49 (wDDGS) versus 144–124 g/kg CP (wDDGS fractions); CA = 74 (wheat) and 359 (wDDGS) versus 575–475 (in wDDGS fractions A–D); CB_1 = 739 and 124 versus 47–27 (fractions); CB_2 = 158 and 313 versus 0–263 (fractions); and CC = 29 and 204 vs 378–253 (fractions) g/kg CHO. Compared to their results, fractions A–D of wheat DDGS in the present study had different profiles from original wheat and wheat DDGS reported by Neuz-Ortin and Yu[5] and Azarfar et al.[39] There is no report in DDGS fractions in the literature. Our results indicate that fractionation processing could be used to manipulate CNCPS protein and carbohydrate subfractions of wheat DDGS. These changes will result in changes in hourly effective nitrogen to energy synchronization ratio described by Tamiminga et al.,[40] Sinclair,[41] Yu et al.,[42] and

Table 4. Effect of Fractionation Processing of Coproducts from Bioethanol Processing (Wheat DDGS) on Truly Digestible Nutrients and Energy Values Using a Chemical Summary Approach (NRC, 2001)

item	fractionation of coproduct (wheat DDGS)			
	A	B	C	D
truly digestible nutrients (g/kg DM)				
truly digestible nonfiber carbohydrate (tdNFC)	267 ± 0.4	266 ± 5.6	254 ± 2.0	255 ± 6.4
truly digestible crude protein (tdCP)	471 ± 1.2	397 ± 2.7	362 ± 0.9	345 ± 2.4
truly digestible neutral detergent fiber (tdNDF)	21 ± 0.8	55 ± 1.1	94 ± 17.6	108 ± 0.5
truly digestible fatty acids (tdFA)	28 ± 0.1	40 ± 0.3	37 ± 0.5	36 ± 0.1
total digestible nutrient at a maintenance level				
total digestible nutrients ($TDN_{1\times}$, g/kg DM)	751 ± 2.2	738 ± 1.1	723 ± 17.6	719 ± 4.2
predicted energy values (Mcal/kg DM) (NRC-2001 Dairy; NRC-1996 Beef)				
digestible energy at maintenance level $DE_{1\times}$	3.81 ± 0.01	3.65 ± 0.00	3.54 ± 0.08	3.49 ± 0.01
metabolizable energy at 3× maintenance intake ($ME_{p3\times}$)	3.08 ± 0.01	2.94 ± 0.00	2.84 ± 0.07	2.80 ± 0.01
net energy for lactation at 3× maintenance intake ($NE_{L3\times}$)	1.98 ± 0.01	1.89 ± 0.00	1.81 ± 0.05	1.78 ± 0.01
net energy for maintenance in growing animal (NE_m)	2.36 ± 0.01	2.23 ± 0.00	2.14 ± 0.06	2.10 ± 0.01
net energy for retention or gain (NE_g)	1.64 ± 0.01	1.54 ± 0.00	1.46 ± 0.05	1.43 ± 0.01

Table 5. Effect of Fractionation Processing of Coproducts from Bioethanol Processing (Wheat DDGS) on in Situ Rumen Degradation Kinetics of Dry Matter and Crude Protein

item	fractionation of coproduct (wheat DDGS)[a]				SEM[b]	*P* value
	A	B	C	D		
effect of fractionation of wheat DDGS on in situ kinetics of DM						
degradation rate (K_d, %/h)	6.20	6.00	4.90	4.30	0.634	0.2517
lag time (T_0, h)	0.5	0.6	0.2	0.0	0.28	0.4704
soluble fraction (*S*, g/kg)	526	485	470	450	15.3	0.0612
insoluble, but potentially degradable fraction (*D*, g/kg)	410 b	425 ab	443 ab	461 a	9.6	0.0430
undegradable fraction (*U*, g/kg)	65	91	70	107	10.4	0.1308
ruminal bypass dry matter (%BDM)	26.6 c	30.4 b	33.8 a	35.1 a	0.44	0.0006
effective degradability of dry matter (%EDDM)	73.4 a	69.6 b	66.2 c	64.9 c	0.43	0.0005
effect of fractionation of wheat DDGS on in situ kinetics of CP						
degradation rate (K_d, %/h)	6.75	8.30	6.55	8.00	0.915	0.3417
lag time (T_0, h)	0.8	1.6	1.5	0.6	0.44	0.2167
soluble fraction (*S*, g/kg)	403	412	419	435	21.2	0.6302
insoluble, but potentially degradable fraction (*D*, g/kg)	555	525	535	492	15.1	0.1360
undegradable fraction (*U*, g/kg)	42 b	63 ab	46 b	73 a	6.2	0.0306
ruminal bypass protein (%BCP)	30.5	28.4	30.2	28.5	0.71	0.2193
effective degradability of protein (%EDCP)	69.6	71.6	69.9	71.5	0.70	0.2182

[a]Means with different letters in the same row are significantly different ($P < 0.05$). Multitreatment comparison method: Tukey. [b]SEM, standard error of mean.

Tas et al.[43] as well as nutrient flow from rumen to the small intestine.

Effects of Fractionation Processing on Truly Digestible Nutrients and Energy Values. The results of truly digestible nutrients and energy values of wheat DDGS affected by fractionation processing are presented in Table 4. The results show that fractionation processing reduced tdNFC by 4% and tdCP by 27% and highly increased tdNDF by 414% and tdFA by 29% from fraction A to fraction D. However, the total $TDN_{1\times}$ from fractions A–D was only slightly reduced from 751 to 719 g/kg DM. For the energy content, the $DE_{1\times}$, $ME_{p3\times}$, $NE_{L3\times}$, NE_m, and NE_g values were all reduced by 8–13% from the fraction A to fraction D (Table 4). These results indicate that fractionation processing changes the energy density of wheat DDGS.

Neuz-Ortin and Yu[5,9] reported the energy value and truly digestible nutrient for original wheat DDGS (without fractionation) using both NRC chemical summary[5,35] and biological approaches.[9] By comparison of these two studies with the present results, the fractionation processing changed the energy density of wheat DDGS (A–D fractions), for example, $TDN_{1\times}$ from 751 to 719 g/kg DM in wheat DDGS fractions versus 834 g/kg DM in wheat and 760 g/kg DM in wheat DDGS and $NE_{L3\times}$ from 1.98 to 1.78 Mcal/kg DM in wheat DDGS fractions versus 1.89 Mcal/kg DM in wheat and 1.94 Mcal/kg DM in wheat DDGS.

Effects of Fractionation Processing of Wheat DDGS on in Situ Rumen Degradation Kinetics. Rumen degradation kinetics are important parameters for rumen microbial protein synthesis and energy supply as well as bypassed nutrients to the small intestine.[28,44] The results of in situ rumen degradation kinetics and degradability of DM and CP of wheat DDGS after fractionation are presented in Table 5. The results show that fractionation processing changed original degradation kinetics of wheat DDGS in comparison with the results from original wheat DDGS reported by Nuez-Ortin and Yu.[6,7] For DM degradation kinetics, fractionation processing (fractions A–D) did not significantly change K_d (average, 5.35%/h), *S* fraction (483 g/kg), and *U* fraction (83 g/kg), but increased *D* fraction by 12%, which was a major reason for the

Table 6. Effect of Fractionation Processing of Wheat DDGS on Estimated Intestinal Digestion of Protein

item	fractionation of coproduct (wheat DDGS)[a]				SEM[b]	*P* value
	A	B	C	D		
rumen phase (g/kg DM)						
ruminally undegraded protein in DVE/OEB system (RUP^{DVE})	169 a	133 b	129 b	115 b	3.5	0.0017
ruminally undegraded protein in NRC-2001 model (RUP^{NRC})	152 a	120 b	116 b	103 b	3.2	0.0017
effective degradation of protein (EDCP)	321 a	280 b	247 c	241 c	3.0	0.0001
intestinal phase						
intestinal digestibility of rumen undegraded protein (%dRUP)	86.0	78.0	84.5	74.5	2.42	0.0769
intestinally absorbable rumen undegraded protein (IADP, %CP)	26.2	22.1	25.5	21.2	1.01	0.0576
intestinally absorbable protein (IADP, g/kg DM)	121 a	86 b	90 b	71 b	4.1	0.0046
total digestible protein (TDP, %CP)	95.8	93.7	95.4	92.7	0.62	0.0656
total digestible protein (TDP = EDCP + IADP, g/kg DM)	442 a	366 b	337 c	312 d	2.3	<0.0001

[a]Means with different letters in the same row are significantly different ($P < 0.05$). Multitreatment comparison method: Tukey. [b]SEM, standard error of mean.

Table 7. Effect of Fractionation Processing of Coproducts from Bioethanol Processing (Wheat DDGS) on Predicted Nutrient Supply to Dairy Cattle Using the DVE/OEB Modeling Approach

item	fractionation of coproduct (wheat DDGS)[a]				SEM[b]	*P* value
	A	B	C	D		
truly absorbed rumen synthesized microbial protein in small intestine (g/kg DM)						
fermented organic matter in the rumen (FOM)	572	568	572	578	5.6	0.6434
rumen microbial protein synthesized based on available energy-FOM (MCP_{FOM})	86	85	86	87	0.8	0.6459
ruminally degraded protein (RDP^{DVE})	331 a	289 b	256 c	248 c	3.5	0.0003
rumen microbial protein synthesized based on rumen degraded protein ($MCP_{RDP}{}^{DVE}$)	331 a	289 b	256 c	248 c	3.5	0.0003
truly absorbed rumen synthesized microbial protein in small intestine ($AMCP^{DVE}$)	55	54	55	55	0.5	0.6444
truly absorbed rumen undegraded protein in the small intestine (g/kg DM)						
ruminally undegraded protein (RUP^{DVE})	169 a	133 b	129 b	115 b	3.5	0.0017
truly absorbed rumen synthesized microbial protein in small intestine ($ARUP^{DVE}$)	145 a	104 b	109 b	85 b	4.9	0.0044
endogenous protein losses in the digestive tract (g/kg DM)						
digestible organic matter (DOM)	779 a	751 b	747 b	739 b	3.6	0.0058
undigested dry matter (UDM)	168 b	195 a	199 a	207 a	3.6	0.0058
endogenous protein losses in the digestive tract (ENDP)	14 b	16 a	16 a	17 a	0.3	0.0057
total truly absorbed protein in the small intestine (g/kg DM)						
DVE (=$AMCP^{DVE}$ + $ARUP^{DVE}$ − ENDP)	186 a	142 b	147 b	124 b	4.8	0.0033
degraded protein balance (DPB^{OEB}, g/kg DM)						
DPB^{OEB}	245 a	204 b	170 c	161 c	2.9	0.0001

[a]Means with different letters in the same row are significantly different ($P < 0.05$). Multitreatment comparison method: Tukey. [b]SEM, standard error of mean.

resulting increase in EDCP by 12% and decrease in %BDM by 32%. Nuez-Ortin and Yu[6,7] reported that original wheat DDGS had effective degradability (EDDM) of 577 g/kg DM versus 321 to 241 g/kg DM in wheat DDGS fractions A−D, the soluble fraction (*S*) of 291 versus 526 to 450 g/kg in wheat DDGS fractions A−D, potentially degradable fraction (*D*) of 601 versus 410 to 461 g/kg in wheat DDGS fractions A−D, and degradation rate of 6.0 versus 6.2 to 4.3%/h in wheat DDGS fractions A−D. From these results, it is clear that fractionation of wheat DDGS influenced rumen degradation kinetics of DM and thus had a great impact on nutrient degradability and availability in the rumen.

For CP degradation kinetics, fractionation processing (fractions A−D) did not change protein degradation kinetic parameters except the *U* fraction, which was increased from 42 to 73 g/kg of CP. The results indicate that the degradation kinetics of different nutrients affected fractionation processing in different patterns. Increasing NDF and ADF (Table 2) decreased EDDM of wheat DDGS in the rumen (Table 5) and increased BDM to the small intestine. However, increasing NDF and ADF (Table 2) did not affect fractions of EDCP and BCP of wheat DDGS in the rumen (Table 5). However, compared with original wheat DDGS,[6,7] the wheat DDGS fractions A−D showed dramatically different protein degradation rates (K_d = 4.5% /h in original wheat DDGS vs 8.3 to 6.5%/h in wheat DDGS fractions), protein soluble fraction (*S* = 81 vs 403 to 435 g/kg in fractions), and potentially degradable protein fraction (*D* = 895 vs 492 to 555 g/kg in the fractions). These results indicated that fractionation processing could manipulate the protein nutrient supply to rumen and small intestine.

Effect of Fractionation Processing on Rumen and Intestinal Digestion of Protein. Truly absorbed protein in the small intestine is an important parameter in judging a true protein value. If rumen undegraded or bypassed protein cannot be digested by enzymes and absorbed in the small intestine, it is useless. Therefore, intestinal digestion and absorption of rumen bypass protein is a key parameter for ruminant nutrition. Rumen and intestinal protein absorption of wheat DDGS affected by the fractionation processing are presented in Table 6. The results show that fractionation processing decreased both ruminally undegraded and degraded protein when

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Table 8. Effect of Fractionation Processing of Coproducts from Bioethanol Processing (Wheat DDGS) on Predicted Nutrient Supply to Dairy Cattle Using the NRC Modeling Approach

item	fractionation of coproduct (wheat DDGS)[a]				SEM[b]	*P* value
	A	B	C	D		
truly absorbed rumen synthesized microbial protein in the small intestine (g/kg DM)						
rumen microbial protein synthesized based on available RDP (MCP_{RDP}^{NRC})	295 a	257 b	228c	220 c	2.7	0.0001
rumen microbial protein synthesized based on available energy-TDN (MCP_{TDN})	90	88	86	86	0.8	0.0718
truly absorbed rumen synthesized microbial protein in the small intestine ($AMCP^{NRC}$)	57	56	55	55	0.5	0.0728
truly absorbed rumen undegraded protein in the small intestine (g/kg DM)						
ruminally undegraded protein (RUP^{NRC})	152 a	120 b	116 b	103 b	3.2	0.0017
truly absorbed rumen undegraded protein in the small intestine ($ARUP^{NRC}$)	131 a	93 b	98 b	77 b	4.5	0.0044
total truly absorbed protein in the small intestine (MP, g/kg DM)						
MP	193 a	154 b	158 b	136 b	4.6	0.0042
degraded protein balance (DPB^{NRC}, g/kg DM)						
DPB^{NRC}	242 a	198 b	167 c	158 c	3.6	0.0003

[a]Means with different letters in the same row are significantly different ($P < 0.05$). Multitreatment comparison method: Tukey. [b]SEM, standard error of mean.

expressed in grams per kilogram of DM. When the DVE/OEB system was applied, RUP^{DVE} decreased by 33% from 169 (fraction A) to 115 g/kg DM (fraction D). When the NRC-2001 model was applied, RUP^{NRC} decreased by 32% from 152 (fraction A) to 103 g/kg DM (fraction D). EDCP was decreased by 25% from 321 to 241 g/kg DM. Nuez-Ortin and Yu[6,7] reported that the original wheat DDGS had EDCP = 185 g/kg DM and RUP = 222 g/kg DM. Our results of wheat DDGS fractions are significantly different from those in wheat DDGS reported by Nuez-Ortin and Yu.[6]

Compared with RUP in wheat grain, which was 47 g/kg DM,[14] all wheat DDGS fractions from A to D were a superior source of RUP to wheat grain (169−115 vs 47 g/kg DM). Although there are no significant differences in intestinal digestibility of RUP (%dRUP) among the DDGS fractions with 81% of average %dRUP, the intestinally absorbable protein and total digestible protein (TDP, g/kg DM) were significantly decreased from the DDGS fractions A−D (Table 6). The results indicate that the fractionation processing did not affect the total (average 94% of total CP), rumen (71% of total CP), and intestinal protein digestion (24% of total CP). However, because of significant decreases of CP content in the fractions due to the fractionation processing, the total digestible protein was decreased from 442 to 212 g/kg DM. In comparison with TDP of 895 g/kg DM in original wheat DDGS reported by Nuez-Ortin and Yu,[6,7] the TDP value was decreased. No study has been found in the literature on the TDP value of DDGS after fractionation processing.

Effect of Fractionation of Wheat DDGS on Protein-Degraded Balance and Predicted Nutrient Supply. Using the DVE/OEB system published by Tamminga et al.[28,44] and the NRC-2001 model,[22] the prediction of nutrient supply in the small intestine and protein-degraded balance to dairy cattle are presented in Tables 7 and 8. In the DVE/OEB system,[28,34] there was no significant difference in fermentable organic matter (FOM) among the DDGS fractions, which resulted in no significant difference in potential microbial protein synthesis and absorption in terms of MCP_{FOM} and $AMCP^{DVE}$ (Table 7). However, the rumen undegraded protein value of RUP^{DVE} was decreased from wheat DDGS fractions A−D (RUP^{DVE} = 169 vs 115 g/kg DM). Therefore, absorbed RUP value of $ARUP^{DVE}$ decreased from 145 (fraction A) to 85 g/kg DM (fraction D), which led to a total truly absorbed protein DVE value decrease from wheat DDGS fractions A−D (186 vs 124 g/kg DM). Meanwhile, DPB^{OEB} decreased from wheat DDGS fractions A−D (245 vs 161 g/kg DM) (Table 7). The results showed that although fractionation processing changed the chemical profile (e.g., increases of NDF and ADF content and decrease of CP), the values of FOM (energy supply) was not affected, but the total truly absorbed protein value of DVE was dramatically changed. Fractionation processing also decreased the protein-degraded protein balance, which indicated it reduced potential nitrogen loss.[28] Again, there is no study found in the literature on DVE and OEB values of DDGS after fractionation processing. Nuez-Ortin and Yu[6,7] reported that original wheat DDGS had a total truly absorbed protein DVE value of 248 g/kg DM and a degraded protein balance OEB value of 42 g/kg DM. Compared to the values of original DDGS reported by Nuez-Ortin and Yu,[6,7] fractionation processing greatly affected the truly absorbed protein supply (186 to 124 in wheat DDGS fractions vs 248 g/kg DM in original wheat DDGS).

In the NRC-2001 model,[22] microbial protein synthesis was based on TDN as an energy source. There was no significant difference in total microbial protein synthesis of MCP_{TDN} (based on TDN) and absorbed microbial protein $AMCP^{NRC}$ value among the wheat DDGS fractions A−D. The value in absorbed rumen undegraded protein $ARUP^{NRC}$ of the fractions decreased, which led to total metabolizable protein MP value decrease from wheat DDGS fractions A−D (193 vs 136 g/kg DM) (Table 8). Similarly, the degraded protein balance DPB^{NRC} value decreased significantly from wheat DDGS fractions A−D (242 vs 158 g/kg DM). The results also support the summary above. Again, there is no study found in the literature that could be used in comparison with the results in the current study. There is no study on fractionation effect on metabolic characteristics of protein (MP, DPB values) estimated using the NRC-2001 model.[22] However, Nuez-Ortin and Yu[7,8] reported an MP value of 242 g/kg DM and a degraded protein balance value of 78 g/kg DM in original wheat DDGS. Compared to the original DDGS reported by Nuez-Ortin and Yu,[7,8,45] fractionation processing greatly affected the total MP supply (from 186 to 124 in wheat DDGS fractions vs 242 g/kg DM in original wheat DDGS).

The wheat DDGS fractions have been developed mainly as an excellent protein source for monogastric animals (such as broiler chickens and pigs). However, this study with dairy cattle illustrates, although the DDGS fractions have higher truly absorbed protein supply in the small intestine, and microbial

protein synthesis potentially possible from available rumen degradable CP (based on RDP) is still far more than potentially possible from the energy extracted during rumen anaerobic fermentation (based on FOM or TDN in the rumen). This suggests the need to improve the energy-nitrogen synchronization by combining other feeds in TMR of ruminant animals.

In summary, fractionation processing had a significant impact on the chemical profiles and nutrient quality of wheat DDGS fractions. With the contents of NDF and ADF of wheat DDGS fractions increasing, the predicted total truly digested and absorbed protein supply to dairy cattle and degraded protein balance were decreased. This optimized the protein degraded balance of wheat DDGS fractions to dairy cattle. However, there is a need to improve the energy-nitrogen synchronization further by combining other feeds in TMR of ruminant animals to prevent nitrogen loss. The best fraction was DDGS fraction A in terms of highest truly absorbed protein values of DVE and MP. Compared with original wheat DDGS, fractionation decreased the truly absorbed protein supply of DVE and MP values. In conclusion, fractionation processing can be used to manipulate nutrient supply of coproduct from bioethanol processing to dairy cattle. There is great potential to fractionate a feed into a desired and optimal chemical and nutrient profile in dairy cattle. Future study is needed to investigate the impact of fractionation processing on the molecular structure spectral profiles in fractions of wheat DDGS in relation to nutrient availability and utilization.

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