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Original Research Article

# Net energy, energy utilization, and nitrogen and energy balance affected by dietary pea supplementation in broilers



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#### ABSTRACT

Pea starch consists predominantly of C-type of amylopectin chain which is more resistant to digestive enzymes than A-type of starch thus slowly digested in poultry. It was hypothesized that the presence of slowly digested pea starch in broiler diets will increase net energy and the efficiency of energy utilization in broilers. Two experiments were performed to investigate starch digestibility of pea at different incubation times (in vitro study) and the effect of dietary pea on heat increment and net energy in broilers using an open-circuit respiratory calorimetry system (in vivo study). One-day-old Ross 308 male broilers were fed a common starter crumble from d 1 to 10 and standard grower diets thereafter. At d 21, birds were transferred to the chambers each housing 2 birds. Each treatment was replicated 6 times with 2 identical runs of 3 replicates per treatment. A wheat-soybean meal-based diet was used as a control and the treatment diet contained 500 g of pea/kg pea. In vitro study showed that pellet processing increased (P < 0.001) starch digestibility, particularly at shorter times for wheat and a much larger response for pea. Birds offered the pea-based diet had lower (P = 0.002) feed intake, lower (P = 0.020) body weight gain, but a similar (P > 0.05) FCR compared to those offered the wheat-based diet. Net energy (NE) and apparent metabolizable energy (AME) values were higher in the pea-based diet than in the wheat-based diet (P = 0.037 for NE and P = 0.018 for AME). Heat production, respiratory quotient, heat increment of feed, efficiency of utilization of gross energy for AME, and efficiency of utilization of AME for NE did not differ (P > 0.05) between the 2 treatments. There was no effect (P > 0.05) of pea on the total tract digestibilities of dry matter, crude protein and ash, but the total tract digestibility of starch was higher (P = 0.022) in the pea-based diet compared to the wheat-based diet. This study provides insight into the energy metabolism of broilers offered a pea-based diet and indicates that dietary pea supplementation increases dietary AME and NE but has no effect on heat increment of feed and the efficiency of energy utilization in broilers.

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#### 1. Introduction

Field pea (*Pisum sativum* L.) has a moderate level of energy and protein (Igbasan and Guenter, 1996) and can be used to partially replace wheat and soybean meal in broiler diets. Field pea contains up to 490 g/kg (DM) starch (Wang and Daun, 2004) which contributes largely to its apparent metabolizable energy (AME). Pea starch consists predominantly of C-type of amylopectin chain, with a high amylose-to-amylopectin ratio and granules ranging from 10 to 40  $\mu$ m (Daveby et al., 1998; Eliasson and Gudmundsson, 2006). In

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poultry, amylose is usually less digestible than amylopectin and C-type of starch is more resistant to digestive enzymes than A-type of starch. Thus, pea starch is slowly digested, as a consequence it often has a lower digestibility than starch from cereal grains (Weurding et al., 2001b; Meng and Slominski, 2005). These properties of pea starch have been shown to improve broiler feed efficiency (Weurding et al., 2003b; Gutierrez del Alamo et al., 2009; Herwig et al., 2019).

As starch is the major energy source in poultry diets, the extent of starch digestion affects AME values (Wiseman et al., 2000). Furthermore, starches with different rates of digestion may have the same extent of starch digestion, but the site of starch digestion and glucose absorption in the small intestine sections are different. These differences may affect the efficiency of starch utilization as well as other aspects of gastrointestinal function in broilers (Weurding et al., 2001a). The possible benefits of feeding a slowly digestible starch such as that of pea to poultry have been reported (Weurding et al., 2003a,b; Herwig et al., 2019). Rapidly digested starch, such as that in wheat, is digested more in the proximal parts of the small intestine, whereas slowly digested starch in pea is digested throughout the small intestine (Herwig et al., 2020). This results in increased flow and supply of glucose into the distal end of the small intestine, which provides the enterocytes with more glucose. Glucose can readily be metabolized as an energy source, sparing the use of amino acids for this purpose by the lower part of the small intestine (Enting et al., 2005). Consequently, this improves energy and protein utilization (Gary, 1992). Further, a moderate and prolonged glucose supply from slowly digested starch sources will lead to a lower but longer insulin curve resulting in efficient muscle protein deposition (Björck et al., 2000). These benefits of slowly digested starch have been linked to improved performance and amino acid digestibility in broilers when a mixture of slow and rapidly digested starch was fed compared to a rapidly digestible starch alone (Weurding et al., 2003a,b). It was hypothesized that dietary supplementation of slowly digested starch (e.g., pea) as opposed to rapidly digested starch (e.g., wheat) will increase dietary net energy and the efficiency of energy utilization in broilers. Two experiments were performed to investigate starch digestibility of pea at different incubation times (in vitro study) and the effect of pea on total tract nutrient digestibility, nitrogen and energy balance, and energy utilization efficiency of broilers using an open-circuit respiratory calorimetry system (in vivo study).

#### 2. Materials and methods

This study was approved by the Animal Ethics Committee of the Jilin Academy of Agricultural Sciences and the experimental procedures were performed according to the guidelines for animal experiments set by the National Institute of Animal Health, China.

#### 2.1. Experimental treatments

The details of ingredients and nutrient composition of the experimental diets are presented in Table 1. A wheat—soybean meal-based diet was used as a control and the treatment diet contained 500 g of pea/kg. Pea was included in the diet by partially replacing wheat and soybean meal. The diets were formulated based on the Ross 308 nutrient specifications (Aviagen, 2014) and were iso-energetic and iso-nitrogenous with the same levels of added oil. Energy levels were adjusted and made similar in both the diets by adding Celite (an indigestible inert filler) in a wheat-soy diet so that only the effect of added dietary pea was determined. The treatment diets were cold-pelleted at 65 °C to produce 3-mm

 Table 1

 Ingredients and nutrient composition of the experimental diets (as-fed basis).

Item	Wheat-based diet	Pea-based diet
Ingredient, %		
Wheat	48.2	21.8
Pea	0	50
Soybean meal	36.8	19.2
Canola oil	5.2	5.2
Limestone	1.39	1.36
Dicalcium phosphate	0.44	0.54
Sodium chloride	0.3	0.35
Vitamin mineral premix <sup>1</sup>	1.0	1.0
Choline chloride	0.1	0.1
DL-Methionine	0.21	0.35
L-Threonine	0.00	0.05
Diluent <sup>2</sup>	6.33	0.00
Total	100	100
Calculated composition, %		
Dry matter	87.5	88.4
AME, MJ/kg	12.34	12.34
Crude protein	23.8	23.0
Crude fat	6.3	6.2
Starch	28.9	33.1
Calcium	0.87	0.87
Chloride	0.24	0.26
Available phosphorus	0.44	0.44
Total phosphorus	0.49	0.51
Sodium	0.18	0.18
Linoleic acid	1.66	1.75
Digestible Arg	1.43	1.54
Digestible Iso	0.88	0.79
Digestible Leu	1.53	1.36
Digestible Lys	1.10	1.21
Digestible Met	0.50	0.57
Digestible Met + Cys	0.83	0.83
Digestible Thr	0.73	0.73
Digestible Trp	0.28	0.20
Digestible Val	0.95	0.84

<sup>&</sup>lt;sup>1</sup> Provided per kilogram of diet: vitamin A, 12,500 IU; vitamin  $D_3$ , 3,500 IU; vitamin E ( $D_1$ -α-tocopheryl acetate), 20 IU; vitamin  $E_3$ , 3 mg; thiamine hydrochloride, 0.01 mg; riboflavin, 8.00 mg; pyridoxine hydrochloride, 4.5 mg; vitamin  $B_{12}$ , 0.02 mg; nicotinic acid, 34 mg; calcium pantothenate 12 mg; folic acid, 0.5 mg; biotin, 0.2 mg; Fe, 80 mg; Cu, 8 mg; Zn, 80 mg; Mn, 80 mg; I, 0.7 mg; Se 0.3 mg.

<sup>2</sup> Celite was used as a diluent.

pellets with a KJ200 pelleter (Huaxiang Machinery Co. Ltd., Zhangqiu, Shandong, China).

### 2.2. Bird housing and management

One-day-old Ross 308 male broiler chicks were purchased from the Yonghong Husbandry hatchery in Tieling, Liaoning, China and fed a commercial starter crumble (Hefeng Group, Gongzhuling, Jilin) from d 1 to 14 and then the grower treatment diets were applied at d 14 for the adaptation of the diets before the measurements. The birds were reared in a climate-controlled room with ad libitum access to feed and water. The lighting system followed the general breed management practice of 18 h of light and 6 h of darkness. At 21 d of age, the birds were transferred to the open circuit chambers for adaptation for chamber environment. Each chamber consisted of 2 birds, and each treatment was replicated 6 times with 2 identical runs of 3 replicates per treatment. The birds were subjected to the measurements of gaseous exchanges and energy and nitrogen balances in the open circuit respiratory chambers from d 25. During the measurement period, the amount of O2 consumption and CO2 production of broilers per chamber were determined to calculate heat production (HP) using the Brouwer equation (Brouwer, 1965). The respiration quotient (RQ) was determined as the volume of CO<sub>2</sub> produced divided by the volume of O2 consumed. The excreta were collected daily and

pooled for each chamber over 3 d and stored in a freezer at  $-20\,^{\circ}\text{C}$ . The initial and final body weight of each bird and daily feed intake per chamber were recorded.

#### 2.3. Calorimetry chambers

The design of the open-circuit respiration chamber has been previously described by Liu et al. (2017). Briefly, the respiratory chamber was air-conditioned to maintain a constant temperature (22 to 24 °C) and humidity (50% to 70%) using an air conditioner and a heater installed inside the chamber. Gas was extracted continuously from the respiration chamber by a vacuum pump. The concentrations of O<sub>2</sub> and CO<sub>2</sub> in and out of each chamber were measured at approximately 21-min intervals by an analyzer for a duration of 3 min with residual air flashed before each measurement. Oxygen was measured with a zirconium oxide sensor (Model 65-4-20; Advanced Micro Instruments, Huntington Beach, CA, USA), whereas CO<sub>2</sub> was measured with a non-dispersive infrared sensor (AGM 10; Sensors Europe GmbH, Erkrath, Germany) residing in the analyzer. The analyzer measured a range of 0 to 25% of O<sub>2</sub> and 0 to 2.5% of CO<sub>2</sub>.

#### 2.4. Chemical analyses

Prior to diet formulation, the ingredients were analysed for dry matter, crude protein, starch, calcium, phosphorus, and ash contents. Diets and excreta samples were analysed for starch, ether extract, moisture, nitrogen, ash, and gross energy (GE) according to AOAC (2006) procedures. Dry matter contents of diets and excreta samples were determined by placing the samples in a forced hot air oven at 105 °C to constant weight. All samples were ground through a mill equipped with a 1-mm screen to ensure a homogeneous mixture. Starch was determined using the Megazyme Total Starch Kit (Megazyme Inc., Chicago, IL, USA) following method 996.11 in AOAC (2006). Gross energy was measured in a bomb calorimeter (C2000, IKA, Guangzhou, China) using benzoic acid as a standard. The nitrogen contents of diet and excreta samples were determined using the Kjeldahl method (GB/T6432-2018, China) on the Auto Kjeldahl Analyzer (K9860, Hanon, Shandong, China) and derived values were subsequently multiplied by 6.25 to convert to crude protein (CP). Total ash was measured by placing duplicate samples in a muffle furnace at 580 °C for 13 h.

In vitro starch digestion of wheat and pea samples as well as the wheat and pea diets used in this study were performed using a method described by Karunaratne et al. (2018). Briefly, an incubation temperature of 41  $^{\circ}$ C was used, a small intestine buffer pH of 5.6 was used and small intestine enzyme levels were increased to increase the rate of starch digestion to more closely match in vivo digestion in chickens. The data were compared to the starch digestion of Canadian samples of wheat (Transcend Canadian Wheat — CTR1417) and pea (Striker Canadian Green Pea — CTR 0812).

#### 2.5. Calculation

The nitrogen balance data were expressed as gram per d for each bird and calculated as follows: RN =  $N_i - N_e$ , where RN is the retained nitrogen in the body,  $N_i$  is the nitrogen intake from the diet (g/d per bird) and  $N_e$  is the nitrogen excreted (g/d per bird); Nitrogen efficiency = RN/ $N_i \times 100$ .

The AME and AMEn values of the diets were determined using the following equations:

 $AME (kcal/kg \ DM) = (GE_i - GE_e)/FI,$ 

AMEn (kcal/kg DM) = AME  $- [8.22 \times RN]/FI$ ,

where GE<sub>i</sub> and GE<sub>e</sub> are the gross energy intake (kcal/d per bird) from the diet and the gross energy output from excreta (kcal/d per bird), respectively; the FI is the feed intake (g DM/d per bird); 8.22 is the nitrogen correction factor for each gram of nitrogen retained in the body (kcal/g; Hill and Anderson, 1958);

Daily ME intake (MEI, kcal/d per bird) = AME  $\times$  FI (g/d).

Heat production (HP) values were estimated from the volumes of  $CO_2$  expired and  $O_2$  consumed by birds using the modified Brouwer equation (excluding methane and nitrogen in exhaled gas from the equation due to the negligible amount of these produced in avian). The equation (Brouwer, 1965) is as follows:

$$HP (kcal) = 3.866 \times VO_2 (L) + 1.200 \times VCO_2 (L),$$

where  $VO_2$  and  $VCO_2$  are volumes of  $O_2$  consumed and  $CO_2$  exhaled, respectively. Heat increment (HI) of feed was calculated as follows: HI = HP - FHP, where FHP is the fasting heat production. A 450 kJ/ (kg BW<sup>70</sup>· d) per bird for broilers reported by Noblet et al. (2015) was used in the calculation. This FHP value corresponds to the asymptotic HP (at zero activity) during a 24-h fasting period. The NE values of feeds were calculated using the following equation:

NE (kcal/kg DM) = (MEI - HI)/FI.

Retained energy (RE) was calculated as follows:

RE = MEI - HP,

RE as protein (kcal/d per bird) = RN  $\times$  6.25  $\times$  5.7,

where 6.25 is the protein equivalent of 1-g nitrogen, and 5.7 is the energy equivalent of 1-g protein (kcal);

RE as fat (kcal/d per bird) = RE - RE as protein.

The results for the MEI, HP, HI, and RE were expressed as (kcal/kg  $BW^{0.70}$  per d). FCR was calculated as feed intake (g DM/d per bird) divided by weight gain (g/d per bird).

#### 2.6. Statistical analysis

In vitro results were analysed in triplicates using 1-way ANOVA in JMP statistical software v. 14 (SAS Institute Inc, Cary, NC) and Tukey's HSD test was used to separate means between the treatments when there was a significant difference. The means between the 2 treatments for all parameters of the in vivo study were analysed and compared using the independent-samples t-test in SPSS 19.0 (2010, SPSS Inc., Chicago, IL, USA). Differences between the treatments were considered significant at P < 0.05.

#### 3. Results and discussion

The performance of birds in the present study met or exceeded the Ross 308 performance objectives (Aviagen, 2014) for daily feed intake, daily weight gain and FCR (Table 2). This was an essential starting point for the study as it aimed to detect subtle but important differences elicited by the 2 diets.

During the experiment, birds offered the pea-based diet had lower (P = 0.002) feed intake, lower (P = 0.020) body weight gain, but a similar (P > 0.05) FCR compared to those offered the wheat-based diet (Table 2). Others reported similar findings when high

**Table 2**Growth performance of broilers offered the experimental diets during d 25 to 28.

Item	Wheat-based diet	Pea-based diet	SEM	<i>P</i> -value
Mean BW, kg	1,525	1,505	18.8	0.460
Daily feed intake, g DM	147.2	132.5	2.3	0.002
Daily BW gain, g	119.7	110.6	5.4	0.020
FCR, g/g DM	1.230	1.198	0.017	0.133
Daily ME intake	2,279	2,187	48	0.226
Daily NE intake	1,756	1,678	43	0.296
ME cost of BW gain, kJ/g	19.04	19.77	0.27	0.097
NE cost of BW gain, kJ/g	14.66	15.17	0.29	0.273

levels of pea were included in broiler diets (Cowieson et al., 2003; McNeill et al., 2004; Herwig et al., 2019). Inclusion levels of 200 to 300 g/kg have been recommended for broilers (Farrell et al., 1999; Igbasan and Guenter, 1996). Although low levels of slow digested starch improved protein and energy availability and are considered beneficial for broiler performance (Weurding et al., 2003b; Gutierrez del Alamo et al., 2009), excessive amounts of pea starch in broiler diets have been reported to have adverse effects on performance (Herwig et al., 2019).

In vitro results (Table 3) showed that starch digestion differed (P < 0.001) between the treatments at all incubation times. A period of 15, 60, and 120 min is generally representative of in vivo starch digestibility in the distal duodenum, jejunum and ileum, respectively (Karunaratne et al., 2018). In the present study, in vitro starch digestibility of Chinese wheat sample was similar to that of Canadian wheat sample at incubation temperatures longer than 60 min. Pea starch was slowly digested compared to wheat and the digestibility was lower than wheat at all the incubation times similar to Canadian pea (P < 0.001). The starch digestibility of pea-based diet was higher (P < 0.001) than that of pea sample at all incubation times. For example, at 90 and 120 min of incubation, starch digestibility values for pea-based diet were 83.0% and 85.2% respectively compared to 55.0% and 63.5% for pea sample. However, the starch digestibility of wheat-based diet was higher than that of wheat sample only at incubation times below 60 min but at or after 60 min of incubation there was no difference in starch digestibility of wheat sample and the wheat-based diet. This demonstrates the effect of pellet processing on improved starch digestibility, particularly at shorter times for wheat and a much larger response for pea. Thus, in vitro results demonstrated that the samples of wheat and pea used in this study were similar to the Canadian varieties.

Energy values of the experimental diets are presented in Table 4. The measured energy values (AME, AMEn, and NE) of the pea-based diet were higher than the wheat-based diet (P=0.018 for AME, P=0.015 for AMEn, and P=0.037 for NE). Processing of peas by heat treatment, pelleting and micronization has been shown to improve protein and starch digestibilities and AME values of pea in previous studies (Grosjean et al., 1999; Igbasan and Guenter, 1996; Longstaff and McNab, 1987). Grosjean et al. (1999) reported

**Table 4**Diet energy and the energy utilization efficiency of broilers offered the experimental diets.

Item	Wheat-based diet	Pea-based diet	SEM	P-value
Energy values, MJ/kg DM				
AME	15.48	16.52	0.26	0.018
AMEn	14.48	15.53	0.25	0.015
NE	11.92	12.67	0.20	0.037
Heat increment of feed	3.56	3.85	0.19	0.316
Energy utilization				
AME/GE	0.798	0.779	0.013	0.322
AMEn/GE	0.746	0.733	0.012	0.446
NE/AME	0.770	0.768	0.010	0.886
NE/AMEn	0.823	0.817	0.011	0.700

AME = apparent metabolizable energy; AMEn = AME corrected to zero N retention; NE = net energy; GE = gross energy.

improvements in AME values of field peas and coloured peas by 1.25 MJ/kg DM and 1.48 MJ/kg DM respectively when feed was offered in pellet form compared to mash form. Improved AME value of field pea after pelleting was also observed in other studies (Carré et al., 1987; Carré et al., 1991; Barrier-Guillot et al., 1995) which is thought to be due to improvements in both starch and protein digestibilities (Grosjean et al., 1999). Grosjean et al. (1999) reported an improvement in starch digestibility of field pea from 89.5% to 98.5% when mash diet was pelleted and fed to broilers. Pelleting breaks down the cell walls of pea cotyledons and increases the accessibility of nutrients to digestive enzymes, thus increasing the digestibility of nutrients (Carré et al., 1991). In the present study, the calculated AME values of the diets were the same for both the treatments (i.e., 12.34 MJ/kg) suggesting that AME values were underestimated during feed formulations by using the table values and that the effect of pelleting may have to be considered in future to determine the precise AME values of peas.

Reduced feed intake in birds offered the pea-based diet may have diminished potential differences (P > 0.05) in AME and NE intakes between the 2 groups (Table 2). Although it may be speculated that a higher-than-expected energy content in the diet may have resulted in reduced feed intake in birds offered the pea-based diet, recent research suggests a reduced ability of broilers to regulate feed intake in response to dietary energy content (Classen, 2017). Slower starch digestibility may also reduce the passage rate of feed and affect feed intake. Also, the possible anti-nutritional effects of tannins and fibre in pea on feed intake cannot be neglected when pea is included at higher level in diets. In addition, there was no difference in heat increment of feed between the 2 treatments (P > 0.05, Table 4), but the NE value was higher for the pea-based diet, which was likely related to the higher AME content of the diet.

Table 5 shows the nitrogen and energy balance of broilers offered the experimental diets. Birds offered the wheat-based diet had higher intake (P = 0.004) and higher retention of nitrogen (P < 0.001) than those offered the pea-based diet, but there was no

**Table 3**Starch in vitro digestion (%) of ingredients and diets.

Item	15 min	30 min	45 min	60 min	90 min	120 min
Wheat	34.0 <sup>b</sup>	51.8°	74.8 <sup>b</sup>	86.3ª	96.6ª	94.6ª
Pea	15.5 <sup>c</sup>	20.7 <sup>e</sup>	38.9 <sup>d</sup>	41.1 <sup>c</sup>	55.0 <sup>c</sup>	63.5 <sup>c</sup>
Wheat-based diet	43.7 <sup>a</sup>	59.7 <sup>b</sup>	85.1 <sup>a</sup>	88.9 <sup>a</sup>	96.4 <sup>a</sup>	94.9 <sup>a</sup>
Pea-based diet	30.6 <sup>b</sup>	44.1 <sup>d</sup>	65.4 <sup>c</sup>	69.3 <sup>b</sup>	83.0 <sup>b</sup>	85.2 <sup>b</sup>
Transcend Canadian Wheat – CTR1417 <sup>1</sup>	44.1 <sup>a</sup>	67.1 <sup>a</sup>	88.0 <sup>a</sup>	92.5 <sup>a</sup>	95.7 <sup>a</sup>	94.3 <sup>ab</sup>
Striker Canadian Green Pea – CTR 0812 <sup>1</sup>	14.1 <sup>c</sup>	19.3 <sup>e</sup>	35.0 <sup>d</sup>	35.1 <sup>d</sup>	45.8 <sup>d</sup>	53.8 <sup>d</sup>
SEM	1.059	1.083	0.933	1.087	1.520	1.492
P-value	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001

 $<sup>\</sup>overline{a^{-e}}$  Values in the same column with different superscripts are significantly different (P < 0.05).

<sup>&</sup>lt;sup>1</sup> Canadian samples of wheat and pea.

**Table 5**Nitrogen and energy balance of broilers offered the experimental diets.

Item	Wheat-based diet	Pea-based diet	SEM	<i>P</i> -value
Nitrogen balance				
Intake, g/d	6.20	5.64	0.10	0.004
Retained, g/d	4.28	3.81	0.06	0.0001
Efficiency, %	69.0	67.6	0.8	0.259
Energy balance, kJ/kg BW <sup>0.70</sup>				
ME intake	1,697	1,645	42.5	0.409
Heat production	390	383	21.4	0.826
Retained energy				
Total	857	811	34.0	0.402
As protein	474	427	7.8	0.002
As fat	383	385	28.6	0.971
NE intake	1307	1261	34.0	0.296
Respiratory quotient	1.012	1.029	0.010	0.282

difference (P>0.05) in the efficiency of nitrogen utilization between the 2 treatments. Total energy gain and energy gain as fat were similar (P>0.05) between the 2 treatments, but the energy gain as protein was higher (P=0.002) in birds offered the wheatbased diet compared to those offered the pea-based diet. This is in line with the lower daily bodyweight gain in birds fed pea-based diet than wheat-based diet. Heat production, respiratory quotient, heat increment of feed, efficiency of utilization of GE for AME (AME/GE and AMEn/GE), and efficiency of utilization of AME for NE (NE/AME and NE/AMEn) did not differ (P>0.05) between the 2 treatments. This implies that pea as a main ingredient up to the level of 50% shows comparable metabolism and energy efficiency to the wheat-based diet.

The effect of dietary treatments on the total tract digestibility of nutrients in broilers are presented in Table 6. There was no effect (P > 0.05) of pea on the total tract digestibilities of dry matter, crude protein and ash, but the total tract digestibility of starch was higher (P = 0.022) in the birds offered the pea-based diet compared to those offered the wheat-based diet. In vitro results showed that only 85.2% of starch in pea-based diet was digested at 120 min of incubation (corresponding to digestion at distal ileum) compared to 94.9% in wheat-based diet. These results are supported by a few other in vitro and in vivo studies on wheat and pea starch digestibilities showing higher proportion of undigested pea starch compared to wheat starch in the small intestine (Weurding et al., 2001a; Herwig, 2018; Herwig et al., 2019). However, a time of 120 min of incubation which represents a digestion at distal ileum in broilers (Herwig et al., 2019) is possibly based on the assumption that passage rate of all the ingredients/diets are similar in the in vitro system. As pea starch is slowly digested, the passage rate of pea-based diet must have been reduced thus the corresponding digestion time at distal ileum should be higher than 120 min. Therefore, the lower in vitro starch digestibility of pea or pea diet at 120 min may not necessarily suggest lower ileal digestibility of pea starch in this study. Instead, reduced feed passage rate may lower feed intake and make starch more available to the birds leading to a higher total tract digestibility of starch in birds fed the pea-based diet as shown in the present study. It may also be possible that a greater proportion of starch from pea entered the caeca and was fermented resulting in higher total tract digestibility of starch in the

**Table 6**Total tract digestibility of nutrients in broilers offered the experimental diets (%).

Item	Wheat-based diet	Pea-based diet	SEM	P-value
Protein	69.0	67.6	0.81	0.259
Starch	95.8	97.6	0.29	0.022
Ash	45.4	42.3	2.29	0.371
Dry matter	74.3	73.1	1.34	0.544

pea-based diet compared to the wheat-based diet. A fairly high fermentation of semi-purified pea starch in the caeca has been reported in a recent broiler study (Herwig et al., 2020). Similarly, in pigs fed a diet with high amylose content, starch digestibility was lower at the ileal level but almost complete at the faecal level (Fouhse et al., 2015) suggesting that considerable amount of fermentation may have occurred in the caeca. Increased fermentation may impact heat increment but a lack of dietary pea starch effect on heat increment and comparatively lower total tract starch digestibility in this study indicate that chickens may not have the same ability to ferment starch like pigs.

#### 4. Conclusion

This study provides insight into the energy metabolism of broilers offered a pea-based diet and indicates that dietary supplementation of pea increases dietary AME and NE but does not affect heat increment of feed and the efficiency of utilization of AME to NE in broilers. However, it should be noted that varietal differences and processing conditions of pea may affect the results and thus these potential effects should be further explored.

#### **Author contributions**

**Nishchal K. Sharma**: data curation, writing - original draft preparation; **Zhibin Ban**: data curation, formal analysis, methodology, investigation, reviewing and editing; **Hank L. Classen**: conceptualization; methodology, data curation, reviewing and editing; **Huaming Yang**: investigation, methodology, data curation, reviewing and editing; **Xiaogang Yan**: data curation, reviewing and editing; **Mingan Choct**: conceptualization, reviewing and editing; **Shu-Biao Wu**: conceptualization, data curation, investigation, project administration, reviewing and editing.

#### **Conflict of interest**

We declare that we have no financial and personal relationships with other people or organizations that might inappropriately influence our work, and there is no professional or other personal interest of any nature or kind in any product, service and/or company that could be construed as influencing the content of this paper.

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#### References

AOAC. Official methods of analysis. 18th ed. Arlington, VA, USA: Association of Official Analytical Chemists; 2006.

Aviagen. Aviagen Ross broiler management manual. 2014. http://pt.aviagen.com/assets/Tech\_Center/Ross\_Broiler/Ross\_Broiler\_Manual.

Barrier-Guillot B, Metayer JP, Grosjean F, Peyronnet C. Feeding value of pea presented in mash or pellets in adult cockerels, laying hens, broilers and turkey poults. In: 10th European symposium on poultry nutrition, Antalya, Turkey; 1995. p. 286–7.

Björck I, Liljeberg H, Ostman E. Low glycaemic—index foods. Br J Nutr 2000;83: 149–55.

Brouwer E. Report of sub-committee on constants and factors. In: Blaxter KL, editor. Energy metabolism. London, UK: Academic Press; 1965. p. 441–3.

Carré B, Escartin R, Melcion JP, Champ M, Roux G, Leclercq B. Effect of pelleting and associations with maize or wheat on the nutritive value of smooth pea (Pisum sativum) seeds in adult cockerels. Br Poult Sci 1987;28:219–29.

Carré B, Beaufils E, Melcion JP. Evaluation of protein and starch digestibility and energy value of pelleted or unpelleted pea seeds from winter or spring cultivars in adult and young chickens. J Agric Food Chem 1991;39:468–72.

- Classen HL. Diet energy and feed intake in chickens. Anim Feed Sci Technol 2017;233:13–21.
- Cowieson AJ, Acamovic T, Bedford MR. Supplementation of diets containing pea meal with exogenous enzymes: effects on weight gain, feed conversion, nutrient digestibility and gross morphology of the gastrointestinal tract of growing broiler chicks. Br Poult Sci 2003;44:427—37.
- Daveby YD, Razdan A, Aman P. Effect of particle size and enzyme supplementation of diets based on dehulled peas on the nutritive value for broiler chickens. Anim Feed Sci Technol 1998;74:229–39.
- Eliasson AC, Gudmundsson M. Starch: physicochemical and functional aspects. In: Eliasson AC, editor. Carbohydrates in food. Boca Raton: Taylor and Francis Group: 2006.
- Enting H, Pos J, Weurding RE, Veldman A. Starch digestion rate affects broiler performance. In: Proc Aust Poult Sci Symp; 2005. p. 17–20.
- Farrell DJ, Perez-Maldonado RA, Mannion PF. Optimum inclusion of field peas, faba beans, chick peas and sweet lupins in poultry diets. II. Broiler experiments. Br Poult Sci 1999;40:674—80.
- Fouhse JM, Ganzle MG, Regmi PR, van Kempen T, Zijlstra RT. High amylose starch with low in vitro digestibility stimulates hindgut fermentation and has a bifidogenic effect in weaned pigs. J Nutr 2015;145:2464–70.
- Gary GM. Starch digestion and absorption in nonruminants. J Nutr 1992;122:172–7. Grosjean F, Barrier-Guillot B, Bastanelli D, Rudeaux F, Bourdillon A, Peyronnet C. Feeding value of three categories of pea (Pisum sativum, L.) for poultry. J Anim Sci 1999:69:591–9
- Gutierrez del Alamo A, Verstegen MWA, Den Hartog LA, Perez de Ayala P, Villamide MJ. Wheat starch digestion rate affects broiler performance. Poult Sci 2009:88:1666–75.
- Herwig E, Schwean-Lardner K, van Kessel A, Savary R, Classen HL. Assessing the effect of starch digestion characteristics on ileal brake activation in broiler chickens. PLoS One 2020;15:e0228647. https://doi.org/10.1371/journal.pone.0228647.
- Herwig E, Abbott D, Schwean-Lardner KV, Classen HL. Effect of rate and extent of starch digestion on broiler chicken performance. Poult Sci 2019;1–9.
- Herwig E. Effect of rate and extent of starch digestion on performance, physiology and behaviour of broilers and laying hens. Ph.D. Diss. Saskatoon, Canada: Univ. Saskatchewan; 2018.

- Hill F, Anderson DL. Comparison of metabolizable energy and productive energy determinations with growing chicks. J Nutr 1958;64:587–603.
- Igbasan FA, Guenter W. The evaluation and enhancement of the nutritive value of yellow—, green— and brown—seeded pea cultivars for unpelleted diets given to broiler chickens. Anim Feed Sci Technol 1996;63:9—24.
- Karunaratne ND, Abott DA, Chibbar RN, Hucl PJ, Pozniak CJ, Classen HL. In vitro assessment of the starch digestibility of western Canadian wheat market classes and cultivars. Can J Anim Sci 2018;98:463—76.
- Liu W, Liu G, Liao R, Chang Y, Huang X, Wu Y, Yang H, Yan H, Cai H. Apparent metabolizable and net energy values of corn and soybean meal for broiler breeding cocks. Poult Sci 2017;96:135–43.
- Longstaff M, McNab JM. Digestion of starch and fibre carbohydrates in peas by adult cockerels. Br Poult Sci 1987;28:261—85.
- McNeill L, Bernard K, Macleod MG. Food intake, growth rate, food conversion and food choice in broilers fed on diets high in rapeseed meal and pea meal, with observations on sensory evaluation of the resulting poultry meat. Br Poult Sci 2004;45:519–23.
- Meng X, Slominski BA. Nutritive values of corn, soybean meal, canola meal, and peas for broiler chickens as affected by a multicarbohydrase preparation of cell wall degrading enzymes. Poult Sci 2005;84:1242—51.
- Noblet J, Dubois S, Lasnier J, Warpechowski M, Dimon P, Carré B, Milgen JV, Labussiére E. Fasting heat production and metabolic BW in group-housed broilers. Animal 2015;9:1138–44.
- Wang N, Daun JK. The chemical composition and nutritive value of Canadian pulses. Quality of western Canadian peas. Field pea (Pisum sativum) Internal Report. Winnipeg, MB: Grain Research Laboratory, Canadian Grain Commission; 2004.
- Weurding RE, Veldman A, Veen WAG, Van der Aar PJ, Verstegen MWA. In vitro starch digestion correlates well with rate and extent of starch digestion in broiler chickens. J Nutr 2001a;131:2336–42.
- Weurding RE, Veldman A, Veen WAG, Van der Aar PJ, Verstegen MWA. Starch digestion rate in the small intestine of broiler chickens differs among feedstuffs. I Nutr 2001b:131:2329–35.
- Weurding RE, Enting H, Verstegen MWA. The relation between starch digestion rate and amino acid level for broiler chickens. Poult Sci 2003a;82:279–84.
- Weurding RE, Enting H, Verstegen MWA. The effect of site of starch digestion on performance of broiler chickens. Anim Feed Sci Technol 2003b;110:175–84.
- Wiseman J, Nicol NT, Norton G. Relationship between apparent metabolisable (AME) values and in vivo/in vitro starch digestibility of wheat for broilers. Worlds Poult Sci J 2000;56:306–18.