

## CHEMICAL AND BIOLOGICAL EVALUATION OF EGYPTIAN CANOLA MEAL FOR CHICKENS DIETS

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### ABSTRACT

Canola meal (CM) is used in all types of poultry feeds, however, because of its relatively low energy; its use in high energy broiler diets is restricted. No study has been conducted to evaluate the nutritional composition of CM derived from an Egyptian variety (Serw 4) grown in Egypt for poultry diets. The present study focused on the nutritional evaluation of Egyptian CM (ECM) compared with soybean meal (SBM). ECM contained lower crude protein (35.98 vs. 50.92%); more ether extract (EE) (10.35 vs. 1.69%), crude fiber (12.58 vs. 3.60), calcium (0.53 vs. 0.32%) and total phosphorus (1.16 vs. 0.73%) on dry matter basis. ECM contained more inhibitor contents like phytic acid (2.13 vs. 1.29%) as compared to SBM whereas as it had 22.47  $\mu\text{mol/gm}$  glucosinolates while it was not determined for SBM. Compared with SBM, as fed basis ECM contained total essential amino acids (20.44 vs. 19.66%); lower essential amino acid index (51.32 vs. 54.55%). The first, second and third limiting amino acids of ECM were methionine, phenyl alanine and isoleucine, while those of SBM were valine, methionine and cysteine, respectively. The biological evaluation of ECM and SBM was investigated with force-fed adult cockerels assay. Comparable with SBM, ECM had a lower apparent and true metabolizable energy than that of SBM (1.75 vs. 2.28 Kcal/g) and (2.39 vs. 3.12 Kcal/g), respectively. The calculated protein replacement value of ECM from SBM in poultry diets was 92.22%. Compared with SBM, ECM had nearly similar true amino acids availability mean (109.56 vs. 109.06) and similar TAAA of most AA; therefore, it could be a good alternative protein source for SBM in poultry diets. Generally, the presence of these essential nutrients implies that CM could be utilized as a safe protein in poultry diets with no detrimental effect on growth performance.

**Key words:** Egypt, canola meal, chickens, chemical, biological evaluation, metabolizable energy, amino acids availability, protein replacement value

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### INTRODUCTION

The scarcity and high prices of protein ingredients for animal feeding highlight the need for extensive studies of all possible sources of protein for efficient poultry production. Soybean meal is considered the main protein source in poultry feed formulations worldwide including Egypt. Low availability of locally produced soybean meal in Egypt caused a dependence mainly on importing large amounts of it and increasing its prices. It is important to know that, Egypt's soybean production prediction for the 2021-22 marketing year stays constant at 25,000 tonnes while Egypt's soybean consumption is predicted to rise by 2.2 percent from 4.54 to 4.64 million tonnes in the 2021-22 marketing year, attributed to rising demand from soy crush facilities. Thus, soybean imports are expected to rise by 0.2 million tonnes to 4.6 million tonnes in the 2021-22 marketing year, according to the USDA (2021).

At the same time, the declining profit of poultry industry due to increasing prices of the main raw feedstuffs forced them to search for cheaper alternative protein. In Egypt, canola has been recently more popular

as a promising new vegetable oil crop to overcome some of the local deficit of vegetable edible oil production as 98% of the consumption needs of edible oil are being met with imported oils (Aswaq Financial Co., 2018). Besides, providing canola meal as a byproduct rich in protein from oil extraction process which would be a good source of protein in both ruminants and poultry diets.

Several advantages are favoring canola to be grown in Egypt such as it is an annual winter oil crop with short duration period (4-5 months), limited water requirements, high seed yield in new reclaimed soils and relatively high oil content (El-agroudy *et al.*, 2018). The success of canola cultivation in a newly reclaimed land outside the old one of Nile valley contributed to avoiding the strong competition with other strategic winter season crops like wheat and Egyptian clover. There were several canola varieties either foreign origin like AD201, Pactol and Silvo or local origin like Serw 4 and Serw 6 grown under the environmental conditions of Egypt. All of these varieties including the local varieties containing low oil's Erucic acid (less than 2%) which ranges from 0.26-1.53% and low meal's glucosinolates which ranges from 4.30-

5.97  $\mu\text{mol/gm}$  (Mekki, 2013) and less than 30  $\mu\text{mol/gm}$  according to Canola Council of Canada (2019).

The efficient use of relatively low-cost non-conventional feed resources depends on their chemical and physical properties for sustainable animal production (Sebola *et al.*, 2017). Canola meal is a good source of most vitamins and minerals especially sulphur, selenium, phosphorus and B vitamins (Canola Council of Canada, 2019). Even though canola meal has lower mineral availability than that of soybean meal, it still contains higher available mineral content that attributed to its higher contents of total minerals compared with soybean meal. Therefore, it would be considered a better source of minerals (Bederska-Łojewska *et al.*, 2017). With all these advantages, still Egyptian feed companies are not using canola meal in animal and poultry diets formulations as a raw feed material that could be a good alternative protein source for soybean meal.

The term dietary fiber refers to edible components of plant cell walls, which are not digestible in the small intestine, but are fermented in the large intestine by gut microbiota (Williams *et al.*, 2019). Non-starch polysaccharides, lignin and polyphenols, and structural proteins and Maillard reaction products are all part of dietary fiber (Yvonne and Victoria, 2016). Fiber components of canola meal include non-starch polysaccharides, which mainly originate from the hull fraction of the seed and account for 18-20% of the meal (Khajali and Slominski, 2012). The major non-starch polysaccharides polymers of canola meal are Pectic polysaccharides which include rhamnogalacturonans and xylogalactouronan backbones with associated side chains of arabinose, galactose and xylose polymers. Other polysaccharides such as arabinans, arabinogalactans, galactans, xyloglucans, galactomannans and mannans are also exist (Pustjens *et al.*, 2013).

The non-starch polysaccharides are not degraded by poultry endogenous enzymes, and thus increase gut viscosity and the passage rate of digesta and consequently decreases nutrient utilization (Raza *et al.*, 2019). Canola generally contains biologically active chemical compounds such as phytates, non-starch polysaccharides and tannins (Khajali and Slominski, 2012), which can reduce the digestibility of nutrients. Although the protein quality of canola meal is comparable to that of soybean meal, certain important amino acids, notably lysine, is shown to have lesser bioavailability than that of soybean meal due to the occurrence of mallard reaction during canola cake/meal processing (Feng and Zuo, 2003).

Metabolizable energy is the most commonly term used for describing the calorific value of diets formulated for poultry. It is particularly useful for poultry due to its ease of measurement since excreta represents both urinary and fecal energy output. In this respect, a rapid and simple technique in which the energy excreted is corrected for metabolic and endogenous energy losses

to obtain the true metabolizable energy values (Kim *et al.*, 2010). A major advantage of the true metabolizable energy system is that it removes the variations due to differences in feed intake. On the other hand, the determination of the nutritive value of a protein feed requires knowledge not only of the total but also the available amino acid content of the protein. The bioassay of true metabolizable energy has been extended to include determination of amino acid availability (Kim *et al.*, 2010).

Evaluating the potential of Egyptian canola meal, as a non-conventional feedstuff in diets of chickens, is important since the cost of feeding chickens for optimum growth performance has become high. The ability of chickens to digest and utilize Egyptian canola meal is largely unknown yet and is critical, if this feed resource is to be incorporated in chicken diets.

The aim of the present study was to determine the chemical composition and biological values of Egyptian canola meal produced from canola seed variety locally bred and cultivated in Egypt. Also, the objective of study was to highlight its potential as feed ingredient (protein source) in poultry diets through determining the energy values, the amino acids availability values and the protein biological value of Egyptian canola meal as compared to soybean meal.

## MATERIALS AND METHODS

Animal housing and handling procedures were approved by the Institutional Animal Care and Use Committee (CU-IACUC), Cairo University, Egypt, with approval number CU-II-F-17-20, on March 2020.

Local variety of canola seed (Serw 4) was obtained from Oil Crops Research Department, Field Crops Research Institute (FCRI), Agricultural Research Center, Egypt. Double mechanical cold pressing was done in Elmahala Alkobra- Algharbia Governorate-Egypt, to obtain Egyptian canola meal (ECM). Soybean meal (SBM) was obtained from Aldakahlia for poultry production company, Egypt.

For ECM and SBM, proximate composition including moisture, crude protein (CP), ether extract (EE), crude fibre (CF), ash, amino acids (AA), total calcium (Ca) and total phosphorus (P) were determined by the methods of Association of Official Analytical Chemists (AOAC, 2019) using triplicate samples for each determination. The amount of nitrogen free extract (NFE) was obtained by difference. Glucan and mannan non-starch polysaccharides (NSP) were determined by High Performance Liquid Chromatography (HPLC) as described by Casterlin *et al.* (1999).

Fractions of neutral detergent fiber (NDF), acid detergent fiber (ADF) and acid detergent lignin (ADL) were determined according to the methods of Van Soest *et al.* (1991) using an ANKOM 220 fiber analyzer

(ANKOM Technology Corporation, NY, USA). Hemicellulose was calculated as NDF – ADF, cellulose as ADF – ADL and lignin as ADL – insoluble ash.

Glucosinolates (GLS) were determined by High-performance liquid chromatographic (HPLC) using the procedure of Herzallah and Holley (2012). Total phenolic compounds were assayed according to the Folin–Ciocalteu method (Attard, 2013). Phytic acid was determined by using wade method according to AOAC (2019). ECM was subjected to phytochemical analysis for qualitative determination of tannins according to the methods described by Nyam *et al.* (2009).

On the basis of AA composition, the chemical score (CS) and essential AA index (EAAI) were calculated according to Block and Mitchel (1946) and Oser (1951), respectively. Cs represents the first limiting AA (the essential amino acid in the greatest deficit) is calculated by the following equation:

$$Cs = \frac{g \text{ amino acid per } 100g \text{ of test protein}}{g \text{ amino acid per } 100g \text{ of reference material}}$$

Essential amino acid index (EAAI) is calculated as follows:

$$EAAI = \sqrt[n]{\frac{100a}{a_e} \times \frac{100b}{b_e} \times \frac{100c}{c_e} \dots \times \frac{100j}{j_e}}$$

Where: a, b, c,.....j are percentages of the essential amino acids in the tested feed protein.

$a_e, b_e, c_e, \dots, j_e$  are percentages of the same essential amino acids in albumin protein.

n is the number of amino acids entering into the calculation.

The ECM biological evaluation was conducted on a number of 12 adult Single Comb Rhode Island (S. C. R. I.) cockerels obtained from El-Takamoly project, El-Fayoum Governorate. Cockerels of nearly similar body weight were individually allocated in metabolic cages present in digestion chamber, Regional Center for Food and Feed (RCFF) providing facilities to collect excreta free from feathers or feed. Feed and water were provided *ad libitum* till starting the assay.

The technique used for true metabolizable energy determination was carried out according to Kim *et al.* (2010). Four birds were used for each test material. Birds were fasted for 24 hrs to empty their digestive tract and then two groups of four cockerels were force-fed 30.1 g of SBM (45.32% CP) and 40 g of ECM (34.11% CP) using a long stem funnel. Besides, four birds of the third group were kept starving and used for determining the endogenous losses. Excreta were collected after 48 hrs, freed from any feathers, dried at 55° C, equilibrated with atmospheric moisture, weighed, ground and kept for gross energy (GE) and AA analysis. GE was determined on the test materials and the dried excreta using an

adiabatic oxygen bomb calorimeter "Parr 6400" depending on operation instruction manual for models produced after October 2010, NO. 587M Revision 05/16/2011 "Parr Instrument Company". The energetic values, either apparent or true metabolizable energy (AME or TME) were calculated as described by Kim *et al.*, (2010) using the following equations:

$$AME \left( \frac{Kcal}{g} \right) = \frac{EI - EE_f}{FI_f}$$

$$TME \left( \frac{Kcal}{g} \right) = \frac{EI - (EE_f - EE_s)}{FI_f}$$

Where: EI: Energy intake (feed intake in grams × gross energy in Kcal/g).

$EE_f$ : Energy voided in excreta of fed birds (grams of dried excreta of fed birds × gross energy in Kcal/g).

$EE_s$ : Energy voided in excreta of starved birds (grams of dried excreta of starved birds × gross energy in Kcal/g).

$FI_f$ : Feed intake of fed birds in g/bird.

An experiment was also undertaken to determine the protein replacement value (PRV) of ECM as replacer of SBM protein throughout the same technique (Kim *et al.*, 2010) by using the following equation:

$$PRV = \frac{A - B}{N \text{ intake}}$$

Where: A = N balance for standard protein in mg per basal Kcal.

B = N balance for protein under investigation in mg per basal Kcal.

N intake: the nitrogen intake for the two proteins where they had the same value of nitrogen.

The true AA availability (TAAA) value was calculated as described by Kim *et al.* (2010).

The recorded/calculated data for different parameters were subjected to statistical analysis through T test procedure of SAS program (SAS, 2010).

## RESULTS AND DISCUSSION

**Chemical Analysis:** The proximate composition of the tested ECM as fed and on dry matter (DM) basis is shown in Table 1. Egyptian Canola meal (ECM) is the residue remaining after the removal of the greater part of oil from Egyptian cultivar origin canola seed. ECM had significantly lower moisture, lower CP, higher EE, higher CF, similar ash and similar NFE than the corresponding values for SBM.

The moisture content of ECM (5.2%) was less than 10% indicating the possibility of storing such material for a long time without deleterious effects. The high CF content of ECM (12.58%) would reduce its nutritive

value for poultry. This would indicate that ECM could not be used as a sole feed in chicken diets, but could only partially replace SBM in poultry feeds based on corn-SBM. The obtained results of proximate analysis of ECM were in general agreement with the proximate analysis of CM reported by Nega and Woldes (2018). The higher CF content of ECM (12.58% on DM basis) than CF content of SBM (3.60% on DM basis) agreed with the difference

between CF level of CM ranged from 8.0-9.91% and the range (3.38-4.53%) of SBM content (Chen *et al.* 2015) and the CF content of CM ranged from 9.54-12.20% and CF content of SBM (7.0%), reported by Nega and Woldes (2018). The variation between CF content of both ECM and SBM in the present study and that reported in previous studies may be due to de-hulling and extraction processes of CM.

**Table 1: Proximate analysis (%) of Egyptian canola meal (ECM) and soybean meal (SBM).**

Nutrient	ECM	SBM	Difference	Pr >  t
<b>As fed (%)</b>				
Moisture	5.20±0.64	11.00±0.58	-5.81	0.0006
CP	34.11±2.34	45.32±0.86	-11.21	<.0001
EE	9.81±0.89	1.50±0.24	8.31	<.0001
CF	11.93±0.97	3.20±0.04	8.73	<.0001
Ash	6.40±0.52	6.30±0.30	0.10	0.8930
NFE	32.55±1.75	32.68±1.21	-0.13	0.9277
<b>DM basis (%)</b>				
Moisture	0	0	-	-
CP	35.98±1.61	50.92±2.77	-14.94	<.0001
EE	10.35±0.64	1.69±0.31	8.66	<.0001
CF	12.58±0.69	3.60±0.19	8.98	<.0001
Ash	6.75±0.58	7.08±0.50	-0.33	0.7021
NFE	34.34±1.32	36.72±1.28	-2.38	0.1484

**DM basis: dry matter basis.**

The CP content of the tested ECM (34.11%) was significantly lower than that of SBM (45.32%) as fed basis. The lower CP content of the tested ECM than that of SBM is at the same line of previous studies that reported a significantly lower CP level of CM than that of SBM (Gorski *et al.*, 2017; Nega and Woldes, 2018). Regard to the CP content of ECM, some studies (Kasprzak *et al.*, 2017; Wang *et al.*, 2017) reported a higher CP content of CM (37.8-43.9% and 36.2-41.9%, respectively). In contrast, there are other studies reported a lower CP content of CM compared with CP content of the tested ECM, In this connection, Jahan *et al.* (2013) and Bryan *et al.* (2017) indicated that CM contained 28.5% and (29.4-31.3%) CP, respectively. These analysis of CM variations attributed to the variation of cultivar, growing conditions especially weather and soil characteristics, extraction method, processing effect, Ether extract (EE) content of the meal (Canola Council of Canada, 2019) and the amount of hulls which are removed. Although the CP content of tested ECM (34.11%) is lower than that of tested SBM (45.32%), it compared favorably with some oilseed meals which are used in poultry diets such as cottonseed meal, sunflower meal, linseed meal, flaxseed meal and peanut meal (Ravindran and Blair, 1992).

The EE values of CM varied with variation of extraction method and the processes used. As known, generally the expeller pressed (double pressed) CM has

higher EE value than the pre-press solvent extracted CM but lower than that resulted from the cold pressing method. The present EE value of 9.81% for the double mechanical pressed tested ECM was markedly higher than the 1.50 % of the tested SBM. This indicated EE value is in the range of EE values for the expeller pressed CM (8-12%) reported by Canola Council of Canada (2019). The higher of EE content of ECM than that of SBM is in concurrent with the difference between EE content of CM and that of SBM that determined by Chen *et al.* (2015). It is well known that the solvent extraction method is more efficient in terms of oil removal and less than 2% of the oil could remain in the meal. Expeller processed meals on the other hand, usually contain higher residual oil (8-12%). Pre-press solvent extraction is another method usually used to remove the oil from canola seed and yields meals of the lowest oil content. In this regard, Canola Council of Canada (2019) reported that expeller processed meal contained more EE than pre-press solvent meals because a higher level of residual oil is intentionally left in expeller processed meal to prevent damage to its nutritional quality caused by higher processing temperature. Therefore, using different methods for oil extraction could explain the wide range of EE content of CM reported in the literature.

It was also found that de-hulling the seeds before oil extraction could affect the nutrient content of CM by improving the nutritional value of the obtained

meal (Khajali and Slominski, 2012). Therefore, a meal containing the lowest values for both CF and EE and the highest value for CP could be produced from removing the hulls and using solvent extraction method during processing.

The ash content (6.75% on DM basis) of tested ECM was similar to that of SBM (6.3%) and within the range of published data for expeller pressed CM ash of several investigators (Leming and Lember, 2005; Thacker and Petri, 2009). Whereas ash level of the tested SBM in the present study is in the same line of the ash content of SBM (6.47% as average) reported by Ravindran *et al.* (2014).

In general, the wide variation in the chemical composition of CM among the reported values in the literature and those of the tested sample (ECM) could be mainly attributed to the different varieties or cultivars used. Moreover, the values may vary widely depending on the degree of de-hulling, processing method, oil extraction procedure and even the size of the seed.

Canola meal is considered as a quality source of essential minerals (Table 2). The current study showed that ECM had a significantly higher Ca and P as compared to SBM (Table 2). Gorski *et al.* (2017) and Wang *et al.* (2017) determined that CM contains higher Ca and P contents than SBM. Although 65 % of the total P in CM is in the phytate form which is not available for monogastric animals, it is still a better source of Ca and P compared to SBM where CM is containing 0.38% nonphytate (available) P compared to other ingredients 0.28, 0.23, 0.09, 0.26, 0.07, and 0.13% for SBM, cottonseed meal, wheat, wheat bran, corn, and barley, respectively (Chen *et al.*, 2015; Gorski *et al.*, 2017).

The results obtained from this study revealed that ECM contains 0.53% and 1.16% for total Ca and total P, respectively, on DM basis. These results are in close agreement with those provided for Australian canola meal and Canadian canola meal (Spragg and Mailer, 2007; Canola Council of Canada, 2019).

**Table 2: Minerals and anti-nutritional factors of Egyptian canola meal (ECM) and soybean meal (SBM)**

Analysis	ECM	SBM	Difference	Pr >  t
<b>As fed</b>				
Ca (%)	0.50±0.05	0.29±0.01	0.21	<.0001
P (%)	1.10±0.12	0.65±0.01	0.45	<.0001
<b>Total phenolic compounds (gallic acid) (ppm)</b>	7868	NA	-	-
Phytic acid (%)	2.02±0.42	1.15±0.15	0.87	<.0001
Tannins (%)	1.10±0.21	0.14±0.01	0.96	<.0001
Mannan (%)	5.63±0.85	1.24±0.05	4.39	<.0001
Glucan (%)	10.52±0.65	4.63±0.06	5.89	<.0001
NDF (%)	20.98±0.86	11.90±0.48	9.08	<.0001
ADF (%)	18.24±0.47	8.20±0.14	10.04	<.0001
ADL (%)	9.01±0.66	0.58±0.01	8.43	<.0001
Hemi cellulose (%)	2.73±0.45	3.70±0.01	-0.97	<.0001
Cellulose (%)	9.23±0.75	7.62±0.49	1.61	<.0001
Lignin (%)	8.16±0.02	1.85±0.01	6.31	<.0001
Glucosinolates ( $\mu\text{mol. g}^{-1}$ )	21.30	NA	-	-
<b>DM basis</b>				
Ca (%)	0.53±0.04	0.32±0.01	0.21	<.0001
P (%)	1.16±0.01	0.73±0.01	0.43	<.0001
<b>Total phenolic compounds (gallic acid) (ppm)</b>	8300	NA	-	-
Phytic acid (%)	2.13±0.18	1.29±0.03	0.84	<.0001
Tannins (%)	1.16±0.08	0.15±0.01	1.01	<.0001
Mannan (%)	5.64±0.78	1.39±0.58	4.25	<.0001
Glucan (%)	11.09±1.10	5.20±0.22	5.89	<.0001
NDF (%)	22.13±1.78	13.37±0.58	8.76	<.0001
ADF (%)	19.24±0.96	9.21±0.37	10.03	<.0001
ADL (%)	9.50±0.01	0.65±0.01	8.85	<.0001
Hemi cellulose (%)	2.88±0.28	4.16±0.02	-1.28	<.0001
Cellulose (%)	9.74±0.88	8.56±0.18	1.18	<.0001
Lignin (%)	8.61±0.76	2.08±0.01	6.53	<.0001
Glucosinolates ( $\mu\text{mol. g}^{-1}$ )	22.47	NA	-	-

Ca: calcium, P: phosphorus, NDF: neutral detergent fiber, ADF: acid detergent fiber, ADL: acid detergent lignin, NA: not applicable.

Regard to the inhibitors content of ECM, the total GLS content of 22.47  $\mu\text{mol. g}^{-1}$  meal (on DM basis) as shown in Table- 2 agreed with (Chen *et al.*, 2015; Aljuobori *et al.*, 2016). Whereas was at least twice higher than the GLS values previously determined in expeller extracted as well as solvent extracted CM samples (Toghyani *et al.*, 2014; Canola Council of Canada, 2019), although was still lower than 30.0  $\mu\text{mol.g}^{-1}$ , which is the defined value for CM (00-rapeseed) (Canola Council of Canada, 2019). Processing temperature during oil extraction affects the content and composition of GLS in CM. Spragg and Mailer (2007) reported that increasing level of heat during processing, with both expeller and solvent extracted meals showing a decline of GLS levels compared to cold press meals. It seems that both seed source and lack of added heat during double cold press oil extraction may have contributed to the higher GLS content of the ECM used in the present study.

Data illustrated in Table-2 reveal that ECM also contains 2.13% phytic acid (on DM basis), whereas SBM contains lower phytic acid compared with ECM (1.29% on DM basis) (Table 2). Phytate has the ability to chelate cations such as iron, sodium, sulfur, calcium, zinc, copper as well as proteins (Khajali and Slominski, 2012).

Tannins are complex polyphenolic structures with the molecular weight of 500 to 3000 Da. CM contains 1.5 to 3% tannins with higher levels observed in brown-seeded canola in comparison with its yellow-seeded counterpart (Khajali and Slominski, 2012; Canola Council of Canada, 2019). The tannins in CM are primarily insoluble and associated with the hull, and do not appear to have the same negative effects on

palatability and protein digestibility that they do in other plants (Khajali and Slominski, 2012). On dry matter basis, the percent tannins obtained from ECM was 1.16 significantly higher than the tannins content 0.15% of SBM (Table 2). The ECM tannins content in the present study was in agreement with previous studies (Khattab *et al.*, 2010; Mansoori *et al.* 2015). Results of current study showed that NSP-glucan and NSP-mannan for ECM were 11.09% and 5.94% on DM basis, respectively. Whereas tested SBM had a significantly lower NSP-glucan and NSP-mannan (5.20% and 1.39% on DM basis) as compared with ECM contents.

**Amino Acids (AA):** The essential AA (EAA), the calculated values for chemical score (CS) and essential amino acid index (EAAI), as well as the limiting AA (LAA) in the tested ECM compared to those of SBM are shown in Table 3. Compared with SBM, ECM contained significantly similar percentage of threonine, isoleucine, histidine, lysine, methionine and tryptophan, whereas it contained significantly higher percentage of valine, (methionine + cysteine) and total EAA. On the other hand, it contained significantly lower percentage of (glycine + serine), leucine, (phenylalanine + tyrosine), phenylalanine, arginine and EAAI. With exception of valine percentage which was significantly lower in CM compared with SBM, the profile of AA in ECM compared with that of SBM in current study was in the same line of the profile AA of CM compared with that of SBM reported by Gorski *et al.* (2017); Wang *et al.* (2017) and Nega and Woldes (2018).

**Table 3: Essential amino acid (EAA) composition (%), chemical score (CS), limiting amino acid (LAA) and essential amino acid index (EAAI) of Egyptian canola meal (ECM) and soybean meal (SBM), based upon albumin as standard**

Amino acid	ECM (%)	SBM (%)	Difference	Pr >  t	Alb. Std (%)
Threonine (THR)	1.21±0.14	1.49±0.11	-0.28	0.2817	2.69
Glycine (GLY)+ Serine (SER)	2.68±0.16	3.43±0.08	-0.75	0.0194	6.62
Valine (VAL)	1.71±0.17	1.22±0.13	0.49	0.0540	4.31
Isoleucine (ILE)	1.28±0.11	1.71±0.01	-0.43	0.1193	3.32
Leucine (LEU)	2.19±0.17	3.18±0.16	-0.99	0.0058	5.17
Phenylalanine (PHE)+Tyrosine (TYR)	2.51±0.21	3.48±0.32	-0.97	0.0064	6.26
Phenylalanine (PHE)	1.54±0.11	2.13±0.14	-0.59	0.0471	3.86
Histidine (HIS)	1.00±0.10	1.16±0.01	-0.16	0.5244	1.19
Lysine (LYS)	2.00±0.23	2.28±0.64	-0.28	0.2817	3.59
Arginine (ARG)	2.20±0.22	2.85±0.11	-0.65	0.0335	3.24
Methionine (MET)+Cysteine (CYS)	2.44±0.11	1.40±0.03	1.04	0.0046	4.21
Methionine (MET)	0.82±0.19	0.71±0.01	0.11	0.6586	2.36
Tryptophan (TRP)	0.48±0.01	0.65±0.01	-0.17	0.4998	1.21
Total EAA	20.44±1.14	19.66±0.88	0.78	0.0165	46.18
EAAI	51.32±1.13	54.55±1.74	-3.23	<.0001	100
CS	34.75±0.96	28.31±0.63	6.44	<.0001	
First LAA	MET	VAL	-	-	
Second LAA	PHE	MET	-	-	
Third LAA	ILE	CYS	-	-	

Most of the AA values in ECM (Table 3) were in general agreement with the published data of AA profile of CM by several investigators (Radfar *et al.*, 2017; Bryan *et al.*, (2019); Canola Council of Canada, 2019). Variability in AA composition of CM in literature could not only be due to the cultivar or variety used, but might be also due to the processing conditions specially the method of fat extraction (Canola Council of Canada, 2019). In addition, this variation is largely due to the method of AA determination.

Based on the AA content of albumin (Alb.) protein as standard (Std), the CS values were calculated. The CS values calculated in the present study clearly showed that the methionine was the first LAA in ECM with value of 34.75% which was significantly higher than 28.31% that represents the value of the first LAA (valine) in SBM for chickens. The second LAA of ECM was phenyl alanine, while, isoleucine was the third LAA. The corresponding ones of SBM were methionine and cysteine, respectively. Conflicting results were reported

about the LAA in CM by Newkirk (2011) and Ivanova *et al.* (2016) who reported that lysine is the first limiting amino acid of CM.

The present results of AA composition showed that ECM contained a reasonable amount of EAA which compared favorably with some other oilseed meals such as SBM and cotton seed meal (Khajali and Slominski, 2012). Wickramasuriya *et al.*, (2015) reported that AA content of CM compares well with that of SBM, the former being richer in methionine while the poorer in lysine.

**Biological Evaluation:** The values of AME and TME of the tested ECM (1.75 and 2.39 Kcal/g, respectively) were significantly lower than that of SBM which were 2.28 and 3.12 Kcal/g, respectively, although, the GE of ECM (4.66 Kcal/g) showed higher value than that of SBM (4.23 Kcal/g). While, the determined protein replacement value of ECM protein was 92.22% from SBM protein (Table 4).

**Table 4: Gross energy (GE), apparent and true metabolizable energy (AME and TME) values (Kcal/g) of Egyptian canola meal (ECM) and soybean meal (SBM) and the protein replacement value (PRV) (%) of ECM from SBM**

Test material	ECM	SBM	Difference	Pr >  t
GE	4.66±0.03	4.23±0.01	0.43	<.0001
AME	1.75±0.01	2.28±0.02	-0.53	<.0001
TME	2.39±0.01	3.12±0.04	-0.73	<.0001
PRV	92.22	-	-	-

The GE value of ECM (4.66 Kcal/g) was within the range of GE of CM determined by Olukosi *et al.* (2017) and Veluri and Olukosi (2020). Generally, variations in GE contents of CM that reported in literature could be attributed to the nutrient composition of such meals, especially the EE content. In this regard, the expeller processed CM contained more EE than prepress solvent meals which is directly reflected on the energetic value of such meal (Canola Council of Canada, 2019). The GE content of a feedstuff is not necessarily all available to the bird. Therefore, the bioavailability of energy was measured as metabolizable energy (ME), which is considered the most available value in evaluating poultry feeds.

The present AME value of ECM is in general agreement with that reported by Askbrant (1988) who recorded a range of 1.737 to 1.934 Kcal/g DM for three different cultivars of CM and lower than that reported by Lee *et al.* (1995) wherein nitrogen corrected AME (AMEn) was 1.980 Kcal/g.

The correction of AME values for metabolic fecal plus endogenous urinary energy (endogenous energy losses "EEL") causes the TME values to be greater than the corresponding AME values for the tested ECM as well as for SBM. The resulting TME of ECM

increased by 36.57% than AME while its value of SBM increased by 36.84% than AME. In this connection, Yamazaki (1987) obtained a wider range of variation among the increased values of TME when he calculated the difference between AMEn and TME values being from 9.5% for sesame seed meal to 30.6% for CM.

The lower AME of ECM compared with SBM in the current study agreed with the results reported by Gorski *et al.* (2017); they reported a lower range of AME of CM (2.0-2.2 Kcal/g) than the AME of SBM which recorded 2.44 Kcal/g. The TME value of the tested ECM (2.39 Kcal/g) was in general agreement with those ranged from 2.20 to 2.53 Kcal/g as reported by several investigators (Sibbald *et al.*, 1990; Rostagno *et al.*, 2017).

Such discrepancies among the present results of ME values of ECM and those reported previously by some investigators could be due to the cultivar of the tested sample as well as the method of processing, especially the method of fat extraction. Moreover, the presence of toxic materials in CM (Conaway *et al.*, 2002) might adversely affect the ME value of CM. Moreover, the available carbohydrate content (Perween *et al.*, 2016) and the age of the birds (Stefanello *et al.*, 2016) could be other reasons for such discrepancies. In general, although the tested ECM contained lower ME value than those

recorded for SBM, it is still compares favorably with those of other oilseed meals such as cotton seed meal and sun flower meal (Khajali and Slominski, 2012).

The results of True AA availability (TAAA) for ECM and SBM are shown in Table-5. The damage of AA availability may occur as a result of the reaction within the protein or with other components like reducing sugars

or with lipids (Toda *et al.*, 2014). This observation was supported by the low moisture content of such test material (5.2%). It seemed that an overheating process was used to dry the material for the purpose of prolonged storage or used during the processing procedure to alleviate the effects of the toxic substances present in seeds.

**Table 5: True amino acids availability (TAAA) of Egyptian canola meal (ECM) compared to soybean meal (SBM) (%)**

Amino acid	TAAA			
	ECM	SBM	Difference	Pr >  t
Aspartic (ASP)	104.46±2.21	99.85±1.22	4.61	0.0367
Threonine (THR)	96.56±2.18	105.82±1.03	-9.26	0.0017
Serine (SER)	107.26±1.33	108.88±0.99	-1.62	0.3833
Glutamic (GLU)	99.48±1.15	100.85±1.11	-1.37	0.4568
Glycine (GLY)	181.88±3.44	165.62±1.26	16.26	<.0001
Alanine (ALA)	101.46±1.02	96.32±1.13	5.14	0.0245
Valine (VAL)	95.48±0.047	102.46±0.84	-6.98	0.0067
Isoleucine (ILE)	100.42±1.02	100.19±1.05	0.23	0.8982
Leucine (LEU)	104.17±1.18	100.52±1.08	3.65	0.0584
Tyrosine (TYR)	121.51±0.21	117.04±0.18	4.47	0.0410
Phenylalanine (PHE)	117.19±0.01	108.61±0.24	8.58	0.0025
Histidine (HIS)	129.12±1.58	121.84±1.44	7.28	0.0055
Lysine (LYS)	92.39±0.87	95.91±0.47	-3.52	0.0571
Arginine (ARG)	109.31±1.12	104.80±0.96	4.51	0.0397
Proline (PRO)	108.70±1.22	115.21±0.87	-6.51	0.0092
Cysteine (CYS)	94.50±1.23	106.28±1.73	-11.78	0.0005
Methionine (MET)	98.70±2.11	103.76±1.54	-5.06	0.0260
Tryptophan (TRP)	ND	ND	-	-

ND: not detected

The storage condition might be another factor for such damage of lysine. The canola seeds might have been stored for a long time without controlled conditions under the relatively high environmental temperature at summer weather in Egypt. Such kind of feeds must be stored under controlled conditions to keep the fat content in good quality i.e. prevent the rancidity and avoid any deterioration to the protein quality of canola seeds or CM. Therefore, the nutritionists must be aware of this complex situation as they are responsible for the precise assessment of the AA requirements to ensure efficient use of protein resources. For meeting the protein requirements of a bird, it is necessary to match the AA composition of the feed with the needs of the bird. The best way of doing this is to use appropriate available AA values rather than total content.

However, Canola Council of Canada (2019) reported possibility of available lysine reduction because of heat treatment that used through CM production. It is obvious that the true lysine availability mean value of ECM (92.39%) is lower than that of SBM (95.91%). The true lysine availability value of 92.39 % in the tested ECM was markedly similar to the published value that being 92% (Ravindran and Bryden, 1999), whereas it was higher than 82.8% that reported by Lee *et al.* (1995). The

low availability of lysine in ECM could be due to some factors such as light, heat, alkali and reducing sugars (Canola Council of Canada, 2019), as a result of which some lysine becomes nutritionally unavailable.

The TAAA of ECM ranged between 92.39% for lysine and 129.12% for histidine which is higher than the reported TAAA range of 82.40% for tyrosine to 92.6% for histidine that obtained by Lee *et al.* (1995), whereas that of SBM ranged between 95.91% and 121.84% for the same amino acids of ECM (Table 2), respectively, indicating the high protein value of ECM. Therefore, the PRV of ECM was biologically determined based on SBM protein as the standard. The results showed that ECM is a rich source of protein that could successfully replace SBM protein up to 92.22% in poultry feeds (Table 4).

Compared with the SBM, the mean TAAA of the tested ECM (109.56%) was nearly similar to that calculated for the tested SBM (109.06%); therefore, it could be a good alternative protein source for SBM in poultry diets. The average TAAA value of ECM (109.56%) was higher than the reported range of 87 to 90% that obtained by Lee *et al.* (1995) for CM.

**Conclusion:** Based on the data observed, ECM could be a good source of nutritional and chemical components

that can be exploited in poultry feeding. The presence of these important components means that ECM could be a valuable dietary ingredient to improve poultry performance. However, the anti-nutritional factors present in ECM could be reduced through the use of specific enzymes.

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## REFERENCES

- Aljuobori, A., I. Zulkifli, A.F. Soleimani, N. Abdullah and J.B. Liang (2016). Growth performance, carcass characteristics and blood parameters of broiler chickens fed on canola meal under high environmental temperature. *Europ. Poultry Science* 80(127):1-11.
- AOAC (2019). Official Methods of Analysis. 21<sup>st</sup> ed. Association of Official Analytical Chemists, Washington, D.C (USA).
- Askbrant, S.U.S. (1988). Metabolisable energy content of rapeseed meal, soyabean meal and white flowered peas determined with laying hens and adult cockerels. *Br. Poult. Sci.* 29:445–455.
- Aswaq Financial Co. Analysis Report of, 2018. On the Strategic Commodities Market in Egypt; (24/1/2019). Provided by Engineer Atteia Shaaban, Consultant for Extraction & Refining of Edible Oils; Vice-President of Oils & Oils by-Products Division; Chamber of Food Industry; Federation of Egyptian Industry.
- Attard, E. (2013). A rapid microtitre plate Folin-Ciocalteu method for the assessment of polyphenols. *Open Life Sci.* 8:48–53.
- Bederska-Łojewska, D, S. Świątkiewicz, A. Arczewska-Włosek and T. Schwarz (2017). Rye non-starch polysaccharides: their impact on poultry intestinal physiology, nutrients digestibility and performance indices – a review, *Annals of Animal Science* 17(2):351–369.
- Block, R.J. and H.H. Mitchel (1946). The correlation of the amino acid composition of proteins with their nutritive value. *Nutr. Abstr. A Rev.* 16:248–278.
- Bryan, D.D.S.L., J.L. MacIsaac, B. Rathgeber, N. McLean and D.M. Anderson (2017). Meal residual oil level and heat treatment after oil extraction affects the nutritive value of expeller pressed canola meal for broiler chickens. *Canadian J. Anim. Sci.* 97(4): 658-667.
- Bryan, D.D.L.S., J.L. MacIsaac, N.L. McLean, B.M. Rathgeber and D.M. Anderson (2019). Nutritive Value of Expeller-Pressed Yellow Canola Meal for Broiler Chickens Following Enzyme Supplementation. *J. Applied Poultry Research* 28(4):1156–1167.
- Canola Council of Canada. (2019). Canola meal feed industry guide. Canola Council of Canada, Winnipeg, MB.
- Casterline, J.L., C.J. Oles and Y. Ku (1999). Measurement of Sugars and Starches in Foods by a Modification of the AOAC Total Dietary Fiber Method. *J. AOAC INTERNATIONAL* 82(3):759–765.
- Chen, X., C. Parr, P. Utterback and C.M. Parsons (2015). Nutritional evaluation of canola meals produced from new varieties of canola seeds for poultry. *Poultry Science* 94(5):1-8.
- Conaway, C., Y. Yang and F. Chung (2002). Isothiocyanates as Cancer Chemopreventive Agents: Their Biological Activities and Metabolism in Rodents and Humans. *Current Drug Metabolism* 3(3):233–255.
- El-agroudy, Nagwa M., Nadia Gad and Fatima A. Shafiq (2018). Economic Study of Oil Crops in Egypt. *Curr. Sci. Int.* 7(4):565-569.
- Feng, D. and J. Zuo (2003). Nutritional and anti-nutritional composition of rapeseed meal and its utilization as a feed ingredient for animal. Guangzhou: College of Animal science, South China Agricultural University; 265 p.
- Gorski, M., C. Foran, P. Utterback and C.M. Parsons (2017). Nutritional evaluation of conventional and increased-protein, reduced-fiber canola meal fed to broiler chickens. *Poultry Science* 96(7):2159–2167.
- Herzallah, S. and R. Holley (2012). Determination of sinigrin, sinalbin, allyl- and benzyl isothiocyanates by RP-HPLC in mustard powder extracts. *LWT Food Sci. Technol.* 47(2):293–299.
- Ivanova, P., V. Chalova, G. Uzunova, L. Koleva and I. Manolov (2016). Biochemical Characterization of Industrially Produced Rapeseed Meal as a Protein Source in Food Industry. *Agriculture and Agricultural Science Procedia* 10:55–62.
- Jahan, D., L. Hussain, M. Islam and M. Khan (2013). Comparative Study of Mustard Oil Cake and Soybean Meal Based Artificial Diet for Rohu, *Labeo rohita* (Ham.) Fingerlings. *The Agriculturists* 11(1):61–66.
- Kasprzak, M.M., J.G.M. Houdijk, O.A. Olukosi, H. Appleyard, S.P.J. Kightley, P. Carre and J. Wiseman (2017). The influence of oil extraction process of different rapeseed varieties on the ileal digestibility of crude protein and amino acids in broiler chickens. *Animal Feed Science and Technology* 227:68–74.

- Khajali, F. and B.A. Slominski (2012). Factors that affect the nutritive value of canola meal for poultry. *Poult. Sci.* 91:2564-2575.
- Khattab, R., E. Goldberg, L. Lin and U. Thiyam (2010). Quantitative analysis and free-radical-scavenging activity of chlorophyll, phytic acid, and condensed tannins in canola. *Food Chemistry* 122:1266-1272.
- Kim, E.J., C.M. Parsons, R. Srinivasan and V. Singh (2010). Nutritional composition, nitrogen-corrected true metabolizable energy, and amino acid digestibilities of new corn distillers dried grains with solubles produced by new fractionation processes. *Poult. Sci.* 89:44-51.
- LEE, K.H., G.H. QI and J.S. SIM (1995). Metabolizable Energy and Amino Acid Availability of Full-Fat Seeds, Meals, and Oils of Flax and Canola. *Poultry Science* 74(8):1341-1348.
- Leming, R. and A. Lember (2005). Chemical composition of expeller-extracted and cold-pressed canola meal. *Agraarteadus* 16:103-109.
- Mansoori, B., A. Rogiewicz and B.A. Slominski (2015). The effect of canola meal tannins on the intestinal absorption capacity of broilers using a D-xylose test. *J.Anim. Phys. Anim. Nutr.* 99 (6):1084-1093.
- Mekki, B. (2013). Yield and quality traits of some canola varieties grown in newly reclaimed sandy soils. *World appl.Sci.J.* 25(2):258-263.
- Nega, T. and Y. Woldes (2018). Review on Nutritional Limitations and Opportunities of using Rapeseed Meal and other Rape Seed by-Products in Animal Feeding. *J. Nutritional Health & Food Engineering* 8(1):43-48.
- Newkirk, R.W. (2011). Meal nutrient composition in Canola: Chemistry, Production, Processing, and Utilization. J. K. Daun, N. A. M. Eskin, D. Hickling, eds. AOCS Press, Urbana, IL. Pages 229-244
- Nyam, M.A., D.L. Wonang and C.O. Akueshi (2009). Phytochemical screening and antimicrobial studies on *Canarium schiveinfurthii* Linn ("Atili") fruits and oil. *Nigerian J. Botany* 22(2):247-253.
- Olukosi, O.A., M. Kasprzak, S. Kightely, P. Carre, J. Wiseman and J.G.M. Houdijk (2017). Investigations of the nutritive value of meals of double-low rapeseed and its influence on growth performance of broiler chickens. *Poult. Sci.* 96:3338-3350.
- Oser, B.L. (1951). Method for integrating essential amino acid content in the nutritional evaluation of protein. *J. Am. Dietetic Acc.* 27:396-402.
- Perween, S., K. Kumar, Chandramoni, S. Kumar, P.K. Singh, M. Kumar and A. Dey (2016). Effect of feeding different dietary levels of energy and protein on growth performance and immune status of Vanaraja chicken in the tropic. *Veterinary World* 9(8):893-899.
- Pustjens, A.M., H.A. Schols, M.A. Kabel and H. Gruppen (2013). Characterisation of cell wall polysaccharides from rapeseed (*Brassica napus*) meal. *Carb. Polymer* 98:1650-1656.
- Radfar, M., A. Rogiewicz and B.A. Slominski (2017). Chemical composition and nutritive value of canola-quality *Brassica juncea* meal for poultry and the effect of enzyme supplementation. *Animal Feed Science and Technology* 225:97-108.
- Ravindran, V. and R. Blair (1992). Feed resources for poultry production in Asia and the Pacific. II. Plant protein sources. *World's Poultry Science J.* 48(3):205-231.
- Ravindran, V. and W.L. Bryden (1999). Amino acid availability in poultry: invitro and invivo measurements. *Australian J. Agricultural Research* 50:889-908.
- Ravindran, V., M.R. Abdollahi and S.M. Bootwalla (2014). Nutrient analysis, metabolizable energy and digestible amino acids of soybean meals of different origins for broilers. *Poult. Sci.* 93:2567-2577.
- Raza, A., S. Bashir and R. Tabassum (2019). An update on carbohydrases: growth performance and intestinal health of poultry. *Heliyon* 5(4):e01437.
- Rostagno, H.S., L.F.T. Albino, M.I. Hannas, J.L. Donzele, N.K. Sakomura, F.G. Perazzo, A. Saraiva, M.L. Teixeira, P.B. Rodrigues, R.F. Oliveira, S.L. Barreto and C. de Oliveira Brito (2017). Tabelas Brasileiras para Aves e Suínos. in *Composicao de Alimentos e Exigencias Nutricionais*, 4<sup>th</sup> ed. Ed. H. S. Rostagno. Universidade Federal de Vicosa, Vicosa, Brazil.
- SAS (2010). SAS User's Guide: Statistics. Version 9.1 SAS Inst. Inc., Cary, NC., USA.
- Sebola, N.A., V. Mlambo and H. K. Mokoboki (2017). Chemical characterisation of *Moringa oleifera* (MO) leaves and the apparent digestibility of MO leaf meal-based diets offered to three chicken strains. *Agroforestry Systems* 93(1):149-160.
- Sibbald, I.R., D.D. Hall, M.S. Wolynetz, J.A. Fernández and H. Jørgensen (1990). Relationships between bioavailable energy estimates made with pigs and cockerels. *Animal Feed Science and Technology* 30(1-2), 131-142.
- Spragg, J. and R. Mailer (2007). Canola meal value chain quality improvement. A final report prepared for AOF and Pork CRC on the canola meal value

- chain quality improvement project Code: 1B-103-0506 31-31.
- Stefanello, C., S.L. Vieira, H.V. Rios, C.T. Simões and J.O.B. Sorbara (2016). Energy and nutrient utilisation of broilers fed soybean meal from two different Brazilian production areas with an exogenous protease. *Animal Feed Science and Technology* 221:267–273.
- Thacker, P.A. and D. Petri (2009). The effect of canola or mustard biodiesel Press cake on nutrient digestibility and performance of broiler chickens. *Asia– Aust J Anim Sci.* 22(11):1531–1539.
- Toda, M., M. Heilmann, A. Ilchmann and S. Vieths (2014). The Maillard reaction and food allergies: is there a link? *Clin. Chem. Lab. Med.* 52 (1):61–67.
- Toghyani, M., N. Rodgers, M.R. Barekatin, P.A. Iji and R.A. Swick (2014). Apparent metabolizable energy value of expeller-extracted canola meal subjected to different processing conditions for growing broiler chickens. *Poult. Sci.* 93:2227–2236.
- USDA (2021). Oil seeds and products annual: U.S. soybean exports to Egypt continue to increase. Foreign Agricultural Service, USDA.
- Van Soest, P. J., J.B. Robertson and B.A. Lewis (1991). Methods for dietary fiber, neutral detergent fiber, and nonstarch polysaccharides in relation to animal nutrition. *J. Dairy Sci.* 74:3583–3597.
- Veluri, S. and O.A. Olukosi (2020). Metabolizable energy of soybean meal and canola meal as influenced by the reference diet used and assay method. *Animals* 10:2132.
- Wang, L.F., E. Beltranena and R.T. Zijlstra (2017). Diet nutrient digestibility and growth performance of weaned pigs fed Brassica napus canola meal varying in nutritive quality. *Animal Feed Science and Technology* 223:90–98.
- Wickramasuriya, S., Y.J. Yi, J.C. Kim, J. Yoo, N.K. Kang and J.M. Heo (2015). A review of canola meal as an alternative feed ingredient for ducks. *J. Anim. Sci. and Technology* 57:29-38.
- Williams, B.A., D. Mikkelsen, B.M. Flanagan and M.J. Gidley (2019). "Dietary fibre": moving beyond the "soluble/insoluble" classification for monogastric nutrition, with an emphasis on humans and pigs. *J. Anim. Sci. and Biotechnology* 10:45.
- Yamazaki, M. (1987). The apparent and true metabolizable energy values of feed ingredients for poultry. *Japanese Poultry Science* 24(2):114–119.
- Yvonne, Maphosa and A. Jideani Victoria (2016). Dietary fiber extraction for human nutrition-A review, *Food Reviews International* 32(1):98-115.