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## Evaluation of an equation for predicting metabolisable energy concentration in compound feeds for pigs

Angelika Grümpel-Schlüter<sup>a</sup>, Andreas Berk<sup>a</sup>, Martin Schäffler<sup>b</sup>, Hubert Spiekers<sup>b</sup> and Sven Dänicke<sup>b</sup> 

<sup>a</sup>Institute of Animal Nutrition, Friedrich-Loeffler-Institut, Federal Research Institute for Animal Health, Braunschweig, Germany; <sup>b</sup>Bavarian State Research Center for Agriculture, Institute for Animal Nutrition and Feed Management, Poing-Grub, Germany

### ABSTRACT

It is useful to predict metabolisable energy (ME) concentration based on crude nutrients which can be determined on a laboratory scale to formulate compound feeds for pigs based on ME concentration and to control the declared concentration. In 2008 such an equation was derived premised on 290 balance experiments showing strong associations between ME predicted by digestible crude nutrients and by crude nutrients themselves. Since the suitability of a regression-based prediction equation might be strongly influenced by the number of observations, the current study aimed at 1) checking the suitability of the existing prediction equation by including more datasets and 2) deriving a revised prediction equation.

The equations were evaluated by correlation and regression analyses using the energy content calculated on the basis of crude nutrients according to the previously used ( $ME_S$ ) and the newly derived ( $ME_{S_{new}}$ ) equations as well as the energy content calculated on the basis of digestible nutrients ( $ME_D$ ).  $ME_D$  was correlated with  $ME_S$  ( $r_s = 0.784$ ;  $p < 0.001$ ) and  $ME_{S_{new}}$  ( $r_s = 0.802$ ;  $p < 0.001$ ). The root mean square error or the adjusted  $r^2$  was 0.332 MJ/kg DM or 0.830 for the regression of  $ME_S$  on  $ME_D$ , and 0.323 MJ/kg DM or 0.839 for the regression of  $ME_{S_{new}}$  on  $ME_D$ . Although the regressive evaluation for the prediction of ME revealed satisfying results, the remaining residual variation not explainable by the regression model should be considered. The minimum span of the prediction interval of the regression of  $ME_S$  or  $ME_{S_{new}}$  on  $ME_D$  covered a range of 0.65 and 0.64 MJ/kg DM, suggesting the variability of ME estimations to be expected when based on crude nutrients. The quality parameters for the newly derived equation were minimally better and the correlation coefficient between  $ME_D$  and both,  $ME_{S_{new}}$  and  $ME_S$ , was strong. Since there is also a non-negligible inaccuracy in the estimation of ME content using the newly derived equation and as the quality parameters were only slightly better, there is at this point no need to introduce the new equation. In future studies, alternative analytical methods for determining the concentration of ME in compound feeds should be considered to improve the accuracy of estimation equations.

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Metabolisable energy; compound feeds; pigs; estimation equation; energy concentration; feed controlling

## 1. Introduction

The formulation of compound feeds for pigs is premised on analysed and/or tabulated nutrient contents of the individual components. The respective energy concentration of the diets is usually calculated by estimation equations based on either crude or digestible crude nutrients and nutrients that cannot be designated as crude nutrients (e.g. starch [St] or sugar [Su]). For this purpose, the estimation of the concentration of metabolisable energy (ME) offers a possible approach. The calculation of the ME content in single feed components should be based on the analysed crude nutrient contents weighted with the tabulated digestibilities. Consequently, the ME content of feed mixtures can be calculated according to the mixture proportions (Spiekers et al. 2013). However, such equations are characterised by unavoidable errors leading to inherent prediction uncertainties.

Studies to determine the ME by balancing gross energy intake and the associated energy excretion via analyses of faeces, urine and methane are regarded to an increasing degree critically from an animal welfare point of view and they are additionally labour and cost expensive. Thus, estimation equations based on multiple correlations between the digestible nutrients or crude nutrients, respectively, and ME are increasingly applied. In 2006, an equation using the content of digestible nutrients was developed (ME<sub>D</sub>; Equation (1); GfE 2006).

$$\begin{aligned} \text{ME}_D [\text{MJ}/\text{kg DM}] = & 0.0205 \cdot \text{Digestible crude protein} [\text{g}/\text{kg DM}] \\ & + 0.0398 \cdot \text{Digestible ether extract} [\text{g}/\text{kg DM}] \\ & + 0.0173 \cdot \text{Starch} [\text{g}/\text{kg DM}] \\ & + 0.0160 \cdot \text{Sugar} [\text{g}/\text{kg DM}] \\ & + 0.0147 \cdot \text{Digestible organic residue} [\text{g}/\text{kg DM}] \end{aligned} \quad (1)$$

where digestible organic residue is calculated as the difference between digestible OM and the sum of digestible crude protein, digestible ether extract, St and Su.

Equation (1) is physiologically closely related to ME, since it considers that only those nutrients that are digested can be used in intermediate metabolism for generation of energy. However, since the determination of digestible nutrients is associated with a complex methodology, in 2008 an alternative estimation equation for the determination of energy content in compound feeds was derived using Equation (1) as reference (GfE 2008; Bulang and Rodehutschord 2009). The resulting equation estimates the ME concentration based on the crude nutrients (ME<sub>S</sub>; Equation (2); Bulang and Rodehutschord 2009; GfE 2008).

$$\begin{aligned} \text{ME}_S [\text{MJ}/\text{kg DM}] = & 0.021503 \cdot \text{Crude protein} [\text{g}/\text{kg DM}] \\ & + 0.032497 \cdot \text{Ether extract} [\text{g}/\text{kg DM}] \\ & - 0.021071 \cdot \text{Crude fibre} [\text{g}/\text{kg DM}] \\ & + 0.016309 \cdot \text{Starch} [\text{g}/\text{kg DM}] \\ & + 0.014701 \cdot \text{Organic residue} [\text{g}/\text{kg DM}] \end{aligned} \quad (2)$$

where organic residue (OR) is calculated as the difference between organic matter (OM) and the sum of crude protein (CP), ether extract (EE), crude fibre (CF) and St.

The rationale behind Equation (2) is the assumption that the relationship between crude nutrients and ME is still strong enough for a reliable ME estimation in compound feeds even if the level of digestion of nutrients is excluded. Nevertheless, in practice for the formulation and evaluation of the ME content this equation is used as a standard, since the required values to estimate ME are usually available.

The derivation and evaluation of Equation (2) is based on a dataset including 290 observations from different digestibility studies performed between 1988 and 2004 in six different facilities (Bulang and Rodehutscord 2009). One aim of the present study was to check the suitability of Equation (2), including more recent datasets since both compound feed and pig genetics as well as plant genetics have developed considerably over the years which presumably influences the digestibility of crude nutrients (Fabian et al. 2003; Da Silva et al. 2012). Another aim of the present study was to derive a revised estimation equation and to check its suitability and its benefit compared to Equation (2).

## 2. Material and methods

### 2.1. Data description

Five hundred and twenty-four datasets from digestibility studies with compound feeds were used for the analysis. The dataset included 290 data lines used by Bulang and Rodehutscord (2009) for the derivation of Equation (2). This dataset was supplemented with additional datasets provided by three institutions: Institute for Animal Nutrition and Feed Management (Bavarian State Research Center for Agriculture) in Grub, Institute of Animal Nutrition, Livestock Products and Nutrition Physiology (University of Natural Resources and Life Sciences) in Wien and Institute of Animal Nutrition (Friedrich-Loeffler-Institut) in Braunschweig. The requirement for the use of the datasets was that the digestibility studies were carried out in accordance with the guidelines for trials to determine energetic value of feedstuffs (GfE 2005).

In 379 out of 524 digestibility studies compound feed for fattening pigs covering a body weight range of 30 to 110 kg were examined. In the remaining 145 studies pigs with a body weight between 5 and 25 kg, corresponding to weaner/rearing pigs, were used to determine the digestibility of compound feed for those pig categories. As Bulang and Rodehutscord (2009) had shown that subdividing the pigs into different age groups does not have a significant effect on the resulting equation, all studies were used jointly as one data basis in the following procedure.

Most of the compound feeds were cereal-based, with wheat and barley being the main components. However, in 134 feeds no information was available about the main components. Based on the fact that information on diet components is not essential for a regressive evaluation of chemically determined nutrients, all mentioned available studies were used in the following evaluations in order to obtain the largest possible data basis covering the widest possible range of crude nutrients.

Datasets contained information about the dry matter (DM) contents and the concentrations of crude nutrients: crude ash (CA), OM, CP, EE, CF, N-free extractives (NfE), St and Su. The concentrations of neutral detergent fibre (NDF) and acid detergent fibre were requested from the institutions but not included in the further analysis because of the low number of datasets containing this information. Digestibility of crude nutrients

**Table 1.** Chemical composition and digestibility of compound feeds for piglets and growing finishing pigs ( $n = 524$ ).

	Mean	SD*	CV <sup>#</sup> [%]	Minimum	Maximum	Q25 <sup>¶</sup>	Q75 <sup>‡</sup>
Chemical composition [g/kg DM]							
Crude ash	54.4	13.4	25	17.3	181.0	48.3	58.5
Organic matter	945.7	13.5	1	819.0	983.0	941.6	951.7
Crude protein	194.8	25.6	13	93.0	259.3	181.7	212.3
Ether extract	35.9	13.6	38	9.0	145.2	26.6	41.8
Crude fibre	43.0	14.2	33	16.0	131.0	37.0	45.8
N-free extractives	684.7	43.5	6	523.0	856.1	657.0	713.0
Starch	478.9	79.0	17	95.4	763.0	442.3	507.6
Sugar	42.9	21.8	51	5.0	239.7	31.9	46.8
Digestibility [%]							
Organic matter	87.3	3.5	4	66.1	98.7	86.3	89.0
Crude protein	87.2	6.1	7	62.0	99.9	84.2	90.9
Ether extract	74.9	12.3	17	-5.7	99.9	69.7	82.5
Crude fibre	40.5	10.7	26	8.2	99.5	33.9	46.6
N-free extractives	90.6	3.4	4	74.5	98.1	88.4	92.9
ME <sub>D</sub> <sup>§</sup>	15.1	0.8	5	10.4	18.4	14.9	15.5
ME <sub>S</sub> <sup>§</sup>	15.1	0.7	5	10.7	17.8	15.0	15.4

\*SD, standard deviation; <sup>#</sup>CV, coefficient of variation; <sup>¶</sup>Q25, lower 25% quantile; <sup>‡</sup>Q75, upper 25% quantile; <sup>§</sup>ME<sub>D</sub>, metabolisable energy calculated based on digestible nutrients; <sup>§</sup>ME<sub>S</sub>, metabolisable energy content calculated based on crude nutrients.

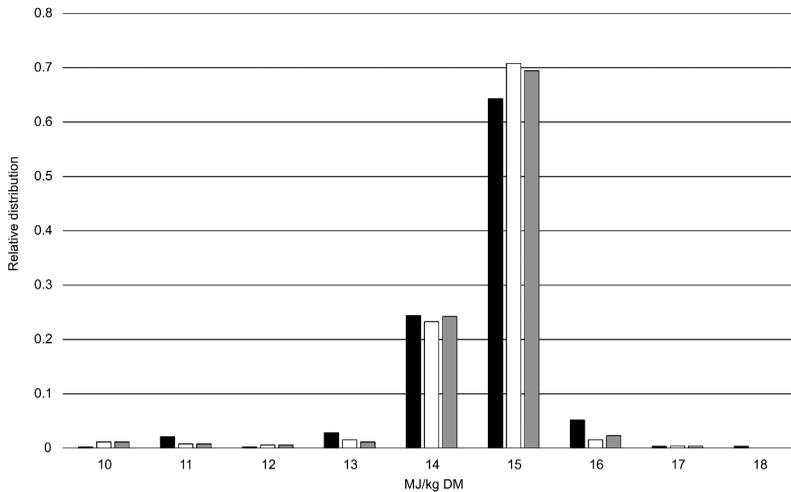
was also included in the datasets. The analysed contents of crude nutrients and the digestibility showed sufficient variation, since this dataset covered wider ranges than that used for the derivation of Equation (2) (Table 1).

The energy concentration was calculated using Equation (1) based on digestible nutrients (ME<sub>D</sub>) as well as using Equation (2) based on crude nutrients (ME<sub>S</sub>). Both energy concentrations were used in the further statistical procedure. The calculated energy concentration for the combined dataset varied greatly between the compound feeds. However, the concentrations were not evenly distributed; most of the feeds had an energy concentration between 14 and 15 MJ/kg DM for ME<sub>D</sub> as well as for ME<sub>S</sub>. The number of compound feeds in the ME<sub>D</sub> classes 10, 11, 12, 13, 14, 15, 16, 17, 18 MJ/kg DM were 1, 11, 1, 15, 128, 337, 27, 2 and 2, respectively. The number of compound feeds in the ME<sub>S</sub> classes 10, 11, 12, 13, 14, 15, 16, 17 MJ/kg DM were 6, 4, 3, 8, 122, 371, 8 and 2, respectively (Figure 1).

## 2.2. Statistics

### 2.2.1. Equation derivation

A new equation was derived using the entire dataset. The derivation was aligned with the procedure of Bulang and Rodehutsord (2009). First, the Spearman correlation coefficient between the energy concentration calculated based on digestible nutrients and crude nutrients CA, CP, EE, CF, St, Su and OR was determined (proc corr). Organic residue was calculated as the difference between organic matter and the sum of CP, EE, CF and St. In a second step, the crude nutrients which were significantly correlated with the energy concentration were modelled in a linear regression model with the energy concentration as dependent variable (proc reg). The crude nutrients exhibiting a significant relation to the energy concentration were used to define the new estimation equation.



**Figure 1.** Relative distribution of energy concentration calculated using Equation (1) ( $ME_D$ ; based on digestible nutrients; black bars), Equation (2) ( $ME_S$ ; based on crude nutrients; white bars) or Equation (3) ( $ME_{S_{new}}$ ; based on crude nutrients; grey bars) for 524 compound feeds.

The linear regression was calculated throughout without intercept, since the variation in the energy concentration should only be explained by the crude nutrients and a residual error. In order to validate the derived regression coefficients, a validation dataset was generated using a bootstrapping method. For this purpose, 5,000 additional datasets of the same size as the initial dataset ( $n = 524$ ) were created in 5,000 iterations (proc surveyselct). Using these 5,000 datasets, the regression analysis was performed to derive regression coefficients.

Application ranges for the usage of the estimation equation were defined based on the given data of the crude nutrients which were included in the newly derived equation. The application ranges were premised on twice the standard deviation which was added to or subtracted from the mean so that at least 95% of the observations were included in these ranges.

Results with a  $p$ -value  $\leq 0.05$  were regarded to be significant. All statistical calculations were made using appropriate procedures in SAS 9.3 (SAS Institute Inc. 2016).

### 2.2.2. Validation of prediction equations

Both the newly derived and the already defined Equation (2) based on crude nutrients were checked for their suitability. For this purpose, on the one hand, the Spearman correlation between  $ME_S$  and  $ME_D$  was calculated (proc corr). On the other hand, the linear regression was performed (proc reg) with  $ME_D$  as dependent variable and  $ME_S$ , each calculated with Equation (2) and the newly derived equation as independent variable. In order to evaluate the suitability of the equations the adjusted r-square as well as the root mean square error (RMSE) was used.

All statistical calculations were made using appropriate procedures in SAS 9.3 (SAS Institute Inc. 2016).

### 3. Results

#### 3.1. Equation derivation

A significant correlation with  $ME_D$  was found for six out of the eight used crude nutrients. CA ( $r_s = -0.272$ ,  $p < 0.001$ ), CP ( $r_s = 0.115$ ,  $p = 0.009$ ), EE ( $r_s = 0.288$ ,  $p < 0.001$ ), CF ( $r_s = -0.588$ ,  $p < 0.001$ ), St ( $r_s = 0.282$ ,  $p < 0.001$ ) and OR ( $r_s = -0.335$ ,  $p < 0.001$ ) were significantly correlated to  $ME_D$ . Between  $ME_D$  and NfE and Su no significant correlation was found. Consequently, only those nutrient fractions with a significant correlation to  $ME_D$  were used to construct the new prediction equation as they contribute to the ME concentration.

The linear regression analysis showed a significant relation between  $ME_D$  and OR (partial regression coefficient  $b_1 = 0.01412$ ,  $p < 0.001$ ), CP ( $b_2 = 0.02039$ ,  $p < 0.001$ ), EE ( $b_3 = 0.03702$ ,  $p < 0.001$ ), CF ( $b_4 = -0.01775$ ,  $p < 0.001$ ) and St ( $b_5 = 0.01648$ ,  $p < 0.001$ ). The partial regression coefficient between  $ME_D$  and CA was not significant ( $b_1 = -0.000699$ ,  $p = 0.547$ ). The RMSE was 0.321 MJ/kg DM and adjusted  $r^2$  was 0.999. Using the estimate values of the significant crude nutrients, a new equation to predict the energy content of compound feeds was derived ( $ME_{S_{new}}$ ; Equation (3)).

$$\begin{aligned} ME_{S_{new}}[\text{MJ/kg DM}] = & 0.02039 \cdot \text{Crude protein [g/kg DM]} \\ & + 0.03702 \cdot \text{Ether extract [g/kg DM]} \\ & - 0.01775 \cdot \text{Crude fibre [g/kg DM]} \\ & + 0.01648 \cdot \text{Starch [g/kg DM]} \\ & + 0.01412 \cdot \text{Organic residue [g/kg DM]} \end{aligned} \quad (3)$$

where OR is calculated as the difference between OM and the sum of CP, EE, CF and St.

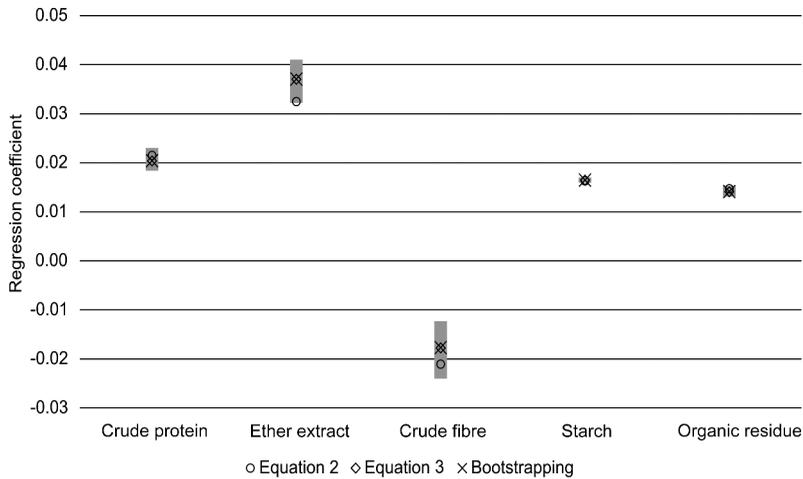
The regression analysis using the bootstrapping data sets resulted in a mean regression coefficient of 0.02044 (minimum: 0.01839 | maximum: 0.02303) for CP, 0.03707 (0.03224 | 0.04104) for EE,  $-0.01769$  ( $-0.02402$  |  $-0.01230$ ) for CF, 0.01649 (0.01594 | 0.01697) for St and 0.01413 (0.01311 | 0.01526) for OR. Both the regression coefficients from Equations (2) and (3) fall within the ranges of coefficients determined using the bootstrapping data set, with the coefficients from Equation (3) closer to those from Equation (2) (Figure 2).

In order to define application ranges twice the standard deviation of CP, EE and CF were used (Table 1). For the newly derived estimation equation the application range for CP content was 140 to 250 g/kg DM, for EE the range was  $\leq 60$  g/kg DM and for CF a range of 10 to 70 g/kg DM could be defined.

#### 3.2. Validation of prediction equations

The concentration of energy, calculated using Equation (3), led to the following distribution of compound feeds on the  $ME_{S_{new}}$  classes 10, 11, 12, 13, 14, 15, 16 and 17 of 6, 4, 3, 6, 127, 364, 12 and 2, respectively (Figure 1).

In order to assess the suitability of the equation the Spearman correlation and the linear regression were calculated.  $ME_D$  was significantly positively correlated with  $ME_S$



**Figure 2.** Regression coefficients defined in Equation (2) (circle), Equation (3) (diamond) or bootstrapping data set (cross). Grey bars show the range from minimal to maximal regression coefficient determined using bootstrapping data sets ( $n = 5,000$ ).

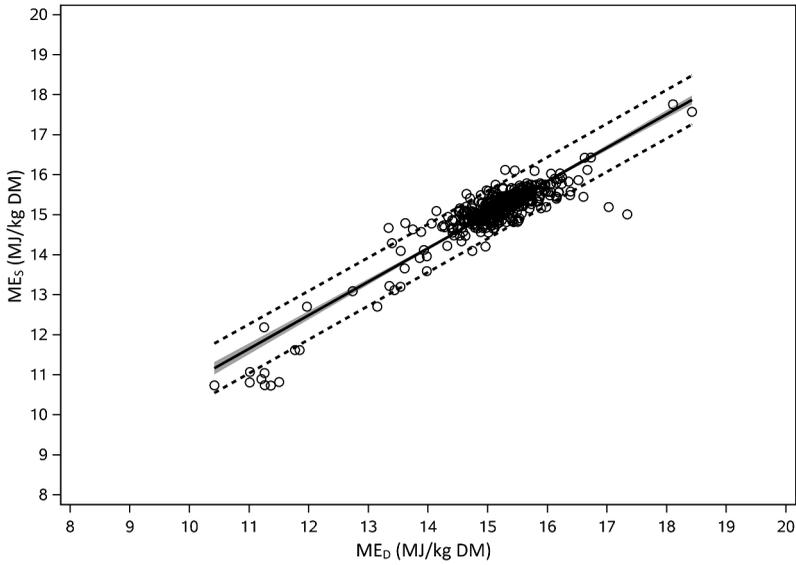
and  $ME_{S_{new}}$  (Table 2). The correlation coefficient was higher for  $ME_D$  and  $ME_{S_{new}}$  than for  $ME_D$  and  $ME_S$ , even if the differences were small. The correlation coefficients between  $ME_D$  and  $ME_S$  or  $ME_{S_{new}}$ , respectively, correspond to a strong correlation.

**Table 2.** Spearman correlation coefficients ( $r_s$ ) and respective  $p$ -values as well as regression equation and respective F-value and  $p$ -value of analysis of variance, root mean square error (RMSE), adjusted r-square and results of t-test for regression coefficients (Intercept and ME) between  $ME_D$  and  $ME_S$ ,  $ME_D$  and  $ME_{S_{new}}$  and  $ME_S$  and  $ME_{S_{new}}$  calculated for all compound feeds ( $n = 524$ ).

	$ME_D$ * $ME_S$	$ME_D$ * $ME_{S_{new}}$	$ME_S$ * $ME_{S_{new}}$
Spearman correlation			
$r_s$	0.783	0.802	0.979
$p$	< 0.001	< 0.001	< 0.001
Regression analysis			
Regression model	$y = 0.14 + 0.99 \cdot x$	$y = -0.02 + 1.00 \cdot x$	$y = 0.00 + 1.00 \cdot x$
F-value ( $p$ )	2559.73 (< 0.001)	2726.56 (< 0.001)	49,785.1 (< 0.001)
RMSE [MJ ME/kg DM]	0.332	0.323	0.075
Adjusted r-square	0.830	0.839	0.990
Coefficient intercept t-value ( $p$ )	0.47 (0.636)	- 0.07 (0.946)	- 0.04 (0.965)
Coefficient $ME_S/ME_{S_{new}}$ t-value ( $p$ )	50.59 (< 0.001)	52.22 (< 0.001)	223.13 (< 0.001)

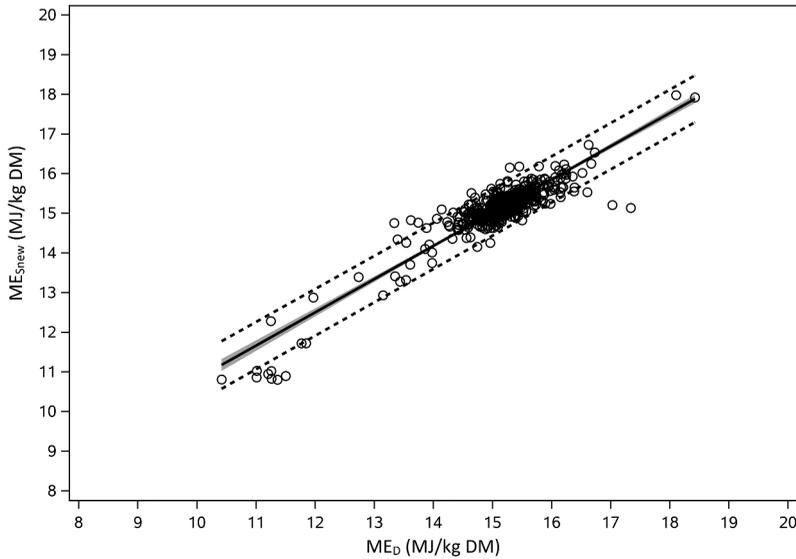
The linear regression between  $ME_D$  and  $ME_S$  was significant, the predicted values varied on average by 0.33 MJ/kg DM around the observed values (Table 2). The linear regression model enabled to explain 83.03% of the variance in  $ME_S$  by  $ME_D$  (Table 2). The lower limit of the confidence intervals was 0.03 MJ/kg DM, the upper limit was 0.17 MJ/kg DM, and the prediction interval was 0.65 MJ/kg DM (Figure 3).

The linear regression between  $ME_D$  and  $ME_{S_{new}}$  was also significant, the predicted values differed on average by 0.32 MJ/kg DM from the observed values (Table 2). Based on the model, 93.90% of the variance in  $ME_{S_{new}}$  could be explained by  $ME_D$ . The lower



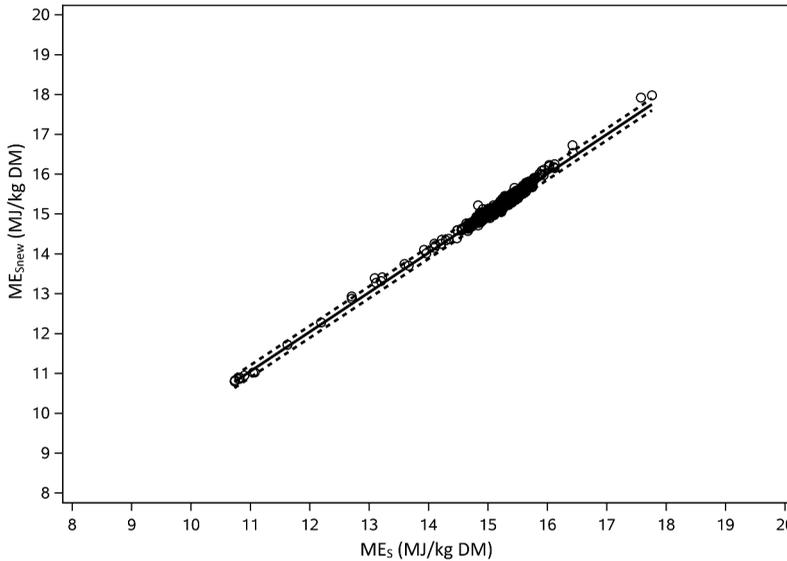
**Figure 3.** Regression between ME<sub>D</sub> and ME<sub>s</sub>. The black line shows the regression line, circles show the observations ( $n = 524$ ), grey shaded area shows the 95% confidence interval, the dashed lines the 95% prediction interval. The regression model is  $y = 0.14 + 0.99 \cdot x$  with RMSE = 0.332 MJ/kg and adjusted  $r^2 = 0.830$ .

limit of the confidence interval was 0.03 MJ/kg DM, the upper limit was 0.17 MJ/kg DM, and the prediction interval was 0.64 MJ/kg DM (Figure 4).



**Figure 4.** Regression between ME<sub>D</sub> and ME<sub>Snew</sub>. The black line shows the regression line, circles show the observations ( $n = 524$ ), grey shaded area shows the 95% confidence interval, the dashed lines the 95% prediction limits. The regression model is  $y = -0.02 + 1.00 \cdot x$  with RMSE = 0.323 MJ/kg and adjusted  $r^2 = 0.839$ .

Comparing  $ME_S$  and  $ME_{S_{new}}$  by calculating the Spearman correlation and the linear regression, a strong relation between both was found.  $ME_{S_{new}}$  was significantly strongly correlated with  $ME_S$  ( $r_s = 0.981$ ;  $p < 0.001$ ). The linear regression between  $ME_S$  and  $ME_{S_{new}}$  was significant, too, the predicted values differed on average by 0.08 MJ/kg DM from the observed values (Table 2). Based on the model, 98.96% of the variance in  $ME_{S_{new}}$  could be explained by  $ME_S$ . The lower limit of the confidence interval was 0.01 MJ/kg DM, the upper limit was 0.04 MJ/kg DM, and the prediction intervals was 0.15 MJ/kg DM (Figure 5).



**Figure 5.** Regression between  $ME_S$  and  $ME_{S_{new}}$ . The black line shows the regression line, circles show the observations ( $n = 524$ ), grey shaded area shows the 95% confidence interval, the dashed lines the 95% prediction limits. The regression model is  $y = 0.00 + 1.00 \cdot x$  with  $RMSE = 0.075$  MJ/kg and adjusted  $r^2 = 0.990$ .

#### 4. Discussion

Deriving the equation based on data from 524 digestibility studies, the same crude nutrients that appear in the prediction equation deduced by Bulang and Rodehutscord (2009) were found to be relevant. The derivation of partial regression coefficients using 5,000 datasets generated using bootstrapping led to very similar coefficients for individual crude nutrients. Moreover, there is a very close connection between  $ME_S$  and  $ME_{S_{new}}$  which is mainly based on sharing partially similar datasets. Additionally, the inclusion of additional datasets in estimation of  $ME_{S_{new}}$  did not alter the partial regression coefficients markedly.

For the evaluation of Equations (2) and (3), both the correlation and the regression between  $ME_D$  and  $ME_S$  or  $ME_{S_{new}}$ , respectively, were calculated. It was found that there is a closer correlation between  $ME_D$  and  $ME_{S_{new}}$ , than between  $ME_D$  and  $ME_S$  but both correlations were strong. Nevertheless, the difference in the quality parameters  $RMSE$  and adjusted  $r^2$  for  $ME_D$  and  $ME_S$  or  $ME_{S_{new}}$ , respectively, is likewise small. Additionally,

inaccuracies in the estimation of energy concentration persist, even using the newly derived equation. The regression analysis between  $ME_D$  and  $ME_S$  or  $ME_{S_{new}}$ , respectively, showed that the prediction interval was almost equal for both equations. The prediction interval of 0.65 MJ/kg DM in the regression between  $ME_D$  and  $ME_S$  or 0.64 MJ/kg DM in the regression between  $ME_D$  and  $ME_{S_{new}}$ , respectively, indicates that, for example, a  $ME_S$  or  $ME_{S_{new}}$  content of 13.4 MJ/kg DM corresponds to a  $ME_D$  content in the range between 12.8 and 14.0, or 12.8 and 14.1 MJ/kg DM, respectively. This prediction uncertainty strongly hints at the limitations of ME prediction equations based on crude nutrients in general. Furthermore, the estimation equation shows a sufficient estimation accuracy only for the defined crude nutrient contents, while the estimations using contents outside these ranges have a significantly higher estimation error.

To improve the preciseness of the energy estimation, the availability of the crude nutrients, especially that of the fractions of dietary fibre, should be considered (Boisen and Eggum 1991; Choi et al. 2020; Sung and Kim 2021). According to Noblet and Perez (1993), the best estimate is made when CA, CP, EE and NDF are included in the equation. Whether the use of NDF leads to a better estimate cannot be deduced from the given data, because in most data sets this information is not available. Other statistical methods could possibly also be used to improve the prediction of energy content. Both artificial neural network and support vector machines including analysed contents of CP, EE, CF and St provide better predictions for energy content than conventional multiple regression (Ahmadi and Rodehutsord 2017). Estimating the content of ME by using the mentioned artificial neural networks and support vector machines and then comparing the results with the estimates using Equations (1), (2) and (3) could provide further information on how accurate the estimate is when using different statistical methods.

Another alternative to improve the estimation of the energy content is the use of near infrared reflectance spectroscopy (NIRS). NIRS determines the chemical feed composition and nutrient digestibility to calculate the energy content using estimation equations. The method is slightly better than mathematical models, but requires a large number of samples for calibration (Aufrère et al. 1996; Bastianelli et al. 2015). Further research is needed to make this technique standard. The same applies to the use of modelling animals. Satisfactory results were obtained when estimating the energy content using rats as models, at least when fat and fibre content was moderate (Jørgensen and Lindberg 2006). The same is true for weaner pigs as model animals for adult pigs, even if differences in the feeding strategies, i.e. *ad libitum* versus restricted feeding can be problematic (Świąch 2017). However, animal experiments are complex and cannot be performed for practical purposes. Additionally, in animal studies the animal individual effects can lead to a strong variation in the results (Boisen and Eggum 1991). This animal individual influence is completely excluded when *in vitro* methods are used to determine the energy content. In addition to two-step methods, which are applied to identify the ileal digestibility (Boisen and Fernández 1995), three-step methods can be used to determine the digestibility in the entire digestive tract (Boisen and Fernández 1997). Even if antinutritional effects, correlations between dry matter and fibre or complex bacterial interactions are only limited or not representable (Moughan 1999), the two- or three-step method is suitable to ascertain the precaecal or overall digestibility (Boisen and Eggum 1991; Boisen and Fernández 1997). In particular, the three-step method for determining digestibility throughout the entire digestive tract could be used largely to

figure out the energy content. However, it is necessary to check the procedure again in detail to optimise the process and get reliable, accurate energy values.

In future research results of analytical studies for determination of energy concentration should be considered in combination with data collected in animal-based studies in order to derive an estimation equation that is based on analytically determinable quantities and, at the same time, has a wide application range and a high estimation accuracy.

## 5. Conclusion

The newly derived estimation equation shows the statistical quality parameters of estimations minimally improved compared to the previously used one. However, due to the assumption of a close relationship between crude nutrients and ME the newly derived estimation equation does not overcome the inherent prediction uncertainty either. Hence, the replacement of the original equation to predict ME from crude nutrients ( $ME_S$ ) by the new estimation equation ( $ME_{S_{new}}$ ) is not recommended. Alternative analytical methods for determining the concentration of ME in compound feeds should be considered in future studies in order to improve the accuracy of the estimation equation. Additionally, the results from these methods could be correlated with animal experimental data and crude nutrient contents to derive an estimation equation on the basis of analytically determinable quantities.

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## Disclosure statement

No potential conflict of interest was reported by the author(s).

## ORCID

Sven Dänicke  <http://orcid.org/0000-0002-4913-4726>

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