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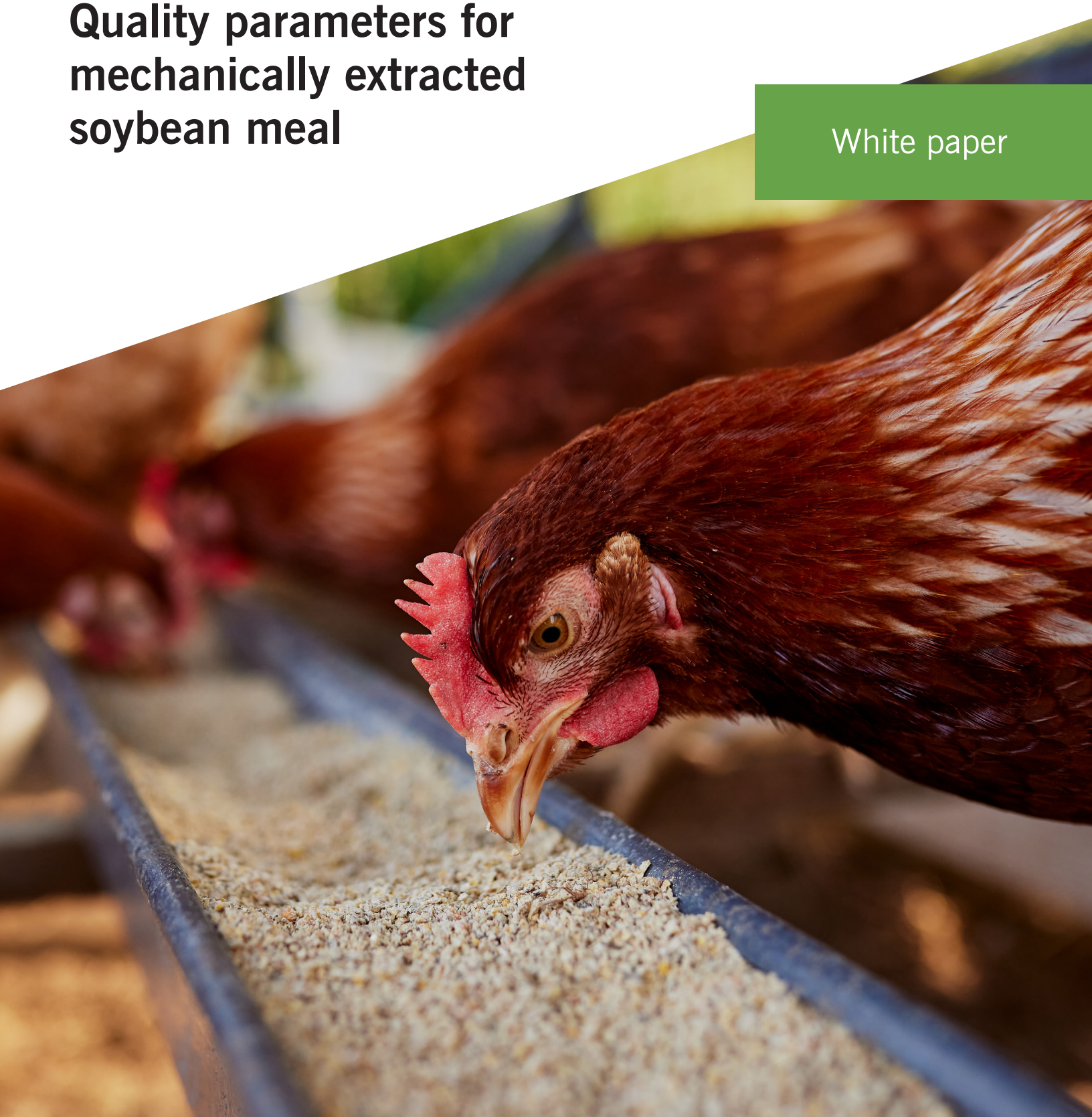
Quality parameters for mechanically extracted soybean meal

White paper



Executive Summary

First, the objective of this project was to define the quality parameters that correlate with poultry amino acid digestibility in a set of samples derived from commercial lots of mechanically extracted soybean meal.

Second, a total of 9 samples corresponding to an equal number of lots of commercial mechanically extracted soybean meal were used. Table 1 (page 5) shows their proximate composition.

Third, each of the 9 samples was chemically analyzed for proximate composition, KOH protein solubility, and trypsin inhibitor activity.

Fourth, an amino acid digestibility bioassay was conducted with single-comb white-leghorn roosters at the University of Illinois to determine the in vivo digestible amino acid coefficients in each of the 9 mechanically extracted soybean meal samples.

Fifth, statistical correlations were conducted between the KOH protein solubility values and the in vivo digestible amino acid coefficients, as well as between the trypsin inhibitor activity values and the in vivo digestible amino acid coefficients.

Sixth, the correlation between the KOH protein solubility values and lysine digestibility is strong, as indicated by the R^2 value of 0.66, which means that 66% of the variation of the KOH protein solubility value is explained by digestibility. In other words, the lower the value of KOH protein solubility, the lower the digestibility of lysine is.

Seventh, no correlation was observed between the trypsin inhibitor activity values and the in vivo digestible lysine (dLYS) coefficients, as the correlation between KOH protein solubility and dLYS coefficients was predominant.

Eighth, given the fact that the KOH protein solubility optimum and the trypsin inhibitor activity optimum move in opposite directions, the practical recommendation for mechanically extracted soybean meal manufacturers by dry extrusion is to have a target KOH protein solubility of about 77-78% and **simultaneously** trypsin inhibitor activity of no more than 10 TUI/mg (6.67 mg TId/g).

Objective

The objective of this project was to define the quality parameters that correlate with poultry amino acid digestibility in a set of samples derived from commercial lots of mechanically extracted soybean meal.

Justification

Ever since the landmark work of Osborne and Mendel[1] at the beginning of the last century, which concluded that for soybeans to be consumed by humans and animals, they need to be heat-treated, concerns about insufficient heat treatment, as well as its excess, have been on the minds of nutritionists and researchers. During the last 100 years that soybean meal has been available as an ingredient for animal nutrition,[2] several tests have been developed to correlate processing conditions to animal performance. The solubility of soybean meal protein in a dilute alkaline solution was devised as a laboratory method to test for overheating[3,4] while the measurement in the lab of the anti-nutritional factors called trypsin inhibitors is utilized to determine if insufficient heat treatment (i.e., underprocessing) during processing has occurred.[5,6]

These two tests, the solubility of the protein in potassium hydroxide (KOH) to detect overprocessing and the presence of trypsin inhibitors to detect underprocessing, were developed using solvent-extracted soybean meal. Today, a relatively accurate assessment of the quality of commercial solvent-extracted soybean meal is possible utilizing these two parameters, as correlations have been established with animal performance.[7,8] Mechanically extracted soybean meal is a different ingredient from solvent-extracted soybean meal because both processes are quite different. However, for mechanically extracted soybean meal, there is not enough information on the application and definition of these two parameters. Considering that both parameters are actually chemical analyses, a correlation with a biological measurement is necessary for the interpretation of the data.

In other words, looking at what happens to in vivo amino acid digestibility for a range of protein solubility values in samples of mechanically extracted soybean meal will allow producers to fine-tune their process and avoid overprocessing. Conversely, looking at what happens to amino acid digestibility for a range of trypsin inhibitor values in samples of mechanically extracted soybean meal will allow producers to fine-tune their process and avoid insufficient processing. This article presents the results of the correlations between amino acid digestibility and KOH protein solubility and the correlations between amino acid digestibility and trypsin inhibitors in representative samples of mechanically extracted soybean meal. These are the basis for defining the quality parameters for mechanically extracted soybean meal.

How were the correlations established?

To establish sound correlations between the laboratory parameters (chemical methods) and in vivo (that is, a biological measurement) performance — in this case, amino acid digestibility measured with roosters — it is necessary to collect mechanically extracted soybean meal samples that exhibit a wide range of values for both chemical parameters. The following paragraphs describe what was done to soundly establish the above-mentioned correlations.

Mechanically extracted soybean meal samples utilized in this study: A total of nine samples corresponding to an equal number of lots were used: 4 of the 9 samples were processed at the same plant, while the remaining 5 came from different sources. However, all 9 lots were marketed as mechanically extracted soybean meal, and Table 1 shows their proximate composition.

Chemical analyses: Each of the 9 samples was analyzed for:

A — **Proximate analysis** was conducted at Dairyland Labs (Arcadia, Wisconsin).

B — **Solubility of the protein in KOH** was conducted at Dairyland Labs utilizing the standardized procedure by Ruiz and coworkers[9] with only one modification: the sample was ground using a cryomill with a 50mL sample tube and a 1-inch ball bearing. This is necessary because the particle size spec for solvent-extracted soybean meal is not achievable with mechanically extracted soybean meal due to the fat content of the latter.

C — **Trypsin inhibitor activity** was measured at Eurofins Labs (Des Moines, IA) utilizing the official AOCS method[10] published in 2021. Results are expressed as TUI/mg, and mg TId/g per the AOCS method.

Amino acid digestibility bioassay

In vivo amino acid digestibility was conducted at the Animal Science Lab at the University of Illinois by Dr. Carl Parsons. A minimum of 4 cecectomized single-comb, white-leghorn roosters per soybean meal sample were fasted for 24 hours prior to being precision fed (crop intubation) 30g of each sample. Excreta were collected for 48 hours post-feeding. Amino acid analysis is conducted in the intact sample of mechanically extracted soybean meal as well as in the lyophilized sample of the excreta. Therefore, it is possible to estimate by difference how much of each amino acid was absorbed by the birds under the conditions of the experiment, which is expressed as a coefficient of digestibility. A full description of the in vivo amino acid digestibility assay has been described by Corray and coworkers.[11]

Results and discussion

The chemical analyses, including proximate analysis for the 9 samples, are presented in Table 1. Crude protein and fat (ether extract) values are typical of mechanically extracted soybean meal.

Table 2 shows the results of the in vivo (with roosters) amino acid digestibility analysis for the 9 soybean meal samples. All values in Table 2 are below 1.0 because 1.0 would mean 100% digestibility for a given amino acid. In practice, the digestibility of most nutrients in the feed, including amino acids, is lower than 100%. Table 2 shows, for example, that for sample 1 (106), the coefficient of digestibility for the amino acid lysine (dLYS) is 0.8644, which is the same as saying that lysine in that sample is 86.44% digestible.

Table 1. Proximate analysis, KOH protein solubility, and trypsin inhibitor activity in the 9 samples subject of this study. Samples listed in descending order of solubility[1]

Sample number	Sample ID	KOHPS[2] %	TIA[2] TUI/mg	TIA mg TId/g	DM[2] %	CP[2] %	CFb %	ADF %	NDF %	Ether extract, %	Ash %
1	106	83.73	12.85	8.57	89.24	43.67	6.30	6.99	9.80	7.99	5.98
2	1690	82.71	11.50	7.67	92.35	44.03	8.22	7.49	10.33	8.58	5.73
3	1691	77.21	10.50	7.00	94.30	46.51	6.87	6.28	9.73	6.84	6.23
4	1701	76.27	7.59	5.06	96.02	46.60	4.92	6.73	9.13	8.58	5.45
5	121	74.56	16.67	11.11	92.30	47.49	7.25	9.73	12.37	6.13	6.86
6	122	73.44	15.38	10.25	87.38	43.43	6.67	8.70	13.02	6.69	5.62
7	108	66.01	14.40	9.60	89.35	45.05	7.49	10.33	13.08	7.03	6.20
8	1689	63.78	5.03	3.35	92.34	45.81	8.70	6.53	10.02	7.85	5.90
9	1688	38.60	3.50	2.33	91.80	45.06	5.24	7.51	16.25	6.91	6.09

1 KOHPS=KOH protein solubility; TIA=trypsin inhibitor activity; DM=dry matter; CP=crude protein; CFb=crude fiber; ADF=acid detergent fiber; NDF=neutral detergent fiber

2 Average of two determinations

Table 2. In vivo digestible amino acid coefficients for 11 amino acids analyzed in the 9 samples subject of this study[1]

Sample number	Sample ID	dLYS %	dARG %	dMET %	dCYS %	dTHR %	dVAL %	dILE %	dLEU %	dTRP %	dASP %	dGLU %
1	106	0.8644	0.9230	0.8593	0.7868	0.8545	0.8616	0.8798	0.8778	0.9144	0.8965	0.9253
2	1690	0.8840	0.9334	0.8868	0.7976	0.8442	0.8721	0.8904	0.8863	0.9167	0.8917	0.9194
3	1691	0.8549	0.9298	0.8939	0.8028	0.8643	0.8806	0.9004	0.8970	0.9177	0.8927	0.9213
4	1701	0.8035	0.9087	0.8642	0.8946	0.8031	0.8509	0.8699	0.8692	0.9004	0.8349	0.8858
5	121	0.8476	0.9157	0.8275	0.7986	0.8283	0.8360	0.8561	0.8581	0.9121	0.8710	0.9058
6	122	0.8033	0.8953	0.8100	0.7164	0.7824	0.8011	0.8075	0.8178	0.8921	0.8258	0.8754
7	108	0.7990	0.8907	0.7919	0.6900	0.7679	0.7927	0.8120	0.8124	0.8754	0.8201	0.8704
8	1689	0.8051	0.9228	0.8826	0.7467	0.8360	0.8677	0.8864	0.8855	0.9057	0.8512	0.8955
9	1688	0.7677	0.9242	0.8802	0.7104	0.8172	0.8708	0.8842	0.8920	0.8941	0.8119	0.8732

1 dLYS=digestible lysine; dARG=digestible arginine; dMET=digestible methionine; dCYS=digestible cystine; dTHR=digestible threonine; dVAL=digestible valine; dILE=digestible isoleucine; dLEU=digestible leucine; dTRP=digestible tryptophan; dASP=digestible aspartic acid; dGLU=digestible glutamic acid.

Table 3 shows the statistical analysis of the correlations between the KOH protein solubility values and the digestible amino acid coefficients of the 9 samples of mechanically extracted soybean meal. The correlation between the KOH protein solubility values and lysine digestibility is strong, as indicated by the R^2 value of 0.66, which means that 66% of the variation of the KOH protein solubility value is explained by digestibility. In other words, **the lower the value of KOH protein solubility, the lower the digestibility of lysine is.** Or expressed by the r value (the correlation coefficient) of 0.81, there is 81% correlation between solubility and digestibility of lysine. The P value (the last row in Table 3) indicates the probability that this correlation between KOH protein solubility and lysine digestibility is repeatable. P values lower than 0.05 indicate that the probability that the correlation occurs again in similar experiments is very high.

In contrast, in Table 3, the correlation between KOH protein solubility and the digestibility of the amino acid methionine (dMET) doesn't exist because not only is the R^2 value for practical purposes zero, but also the r value indicates that there is only 4.73% correlation, which is negligible. Like lysine, correlations exist between KOH protein solubility and digestible aspartic acid (dASP) and digestible glutamic acid (dGLU). However, out of these three amino acids, only lysine is essential in the diet for monogastric animals.

Table 3. Statistics of the correlations between KOH protein solubility and digestible amino acid coefficients in 9 samples of mechanically extracted soybean meal

Line number	Sample code	KOHPS %	dLYS %	dASP %	dGLU %	dMET %
1	106	83.73	0.8644	0.8965	0.9253	0.8593
2	1690	82.71	0.8840	0.8917	0.9194	0.8868
3	1691	77.21	0.8549	0.8927	0.9213	0.8939
4	1701	76.27	0.8035	0.8349	0.8858	0.8642
5	121	74.56	0.8476	0.8710	0.9058	0.8275
6	122	73.44	0.8033	0.8258	0.8754	0.8100
7	108	66.01	0.7990	0.8201	0.8704	0.7919
8	1689	63.78	0.8051	0.8512	0.8955	0.8826
9	1688	38.60	0.7677	0.8119	0.8732	0.8802
R^2			0.6620	0.5192	0.4457	0.0022
r			0.8136	0.7205	0.6676	0.0473
P<0.05			0.0076	0.0285	0.0494	0.9038[1]

1 For dMET P>0.05

The significance of the data in Table 3 indicates that mechanically extracted soybean meal manufacturers must consider that excess heat during dry extrusion may result in damage to the essential amino acid lysine. Additionally, manufacturers may use the standardized KOH protein solubility method to determine the adequacy of the heat treatment in their process.

Regarding the second parameter measured in the 9 soybean meal samples, trypsin inhibitor activity, Table 4 presents the correlations between trypsin inhibitor activity and in vivo amino acid coefficients. Actually, the trypsin inhibitor activity in this study is correlated to only one amino acid, out of the 11 amino acids reported in Table 2. Trypsin inhibitor activity is inversely correlated to digestible methionine (dMET). This correlation is meaningful in the context of this experiment because it is well known, dating back to the 1960s, that trypsin inhibitors and methionine interact in a particular way.[12] As already shown in Table 3, KOH protein solubility is not correlated to dMET in this study; and precisely because of this, no correlation, the inverse correlation of trypsin inhibitor activity versus dMET is clearly expressed in Table 4.

Conversely, while dLYS is strongly correlated with KOH protein solubility (Table 3), it is not correlated with trypsin inhibitor (Table 4) **in this study**, as the correlation of solubility with dLYS predominates. Ruiz and collaborators reported in 2023[13] that trypsin and chymotrypsin inhibitors were inversely correlated to several amino acids, including dLYS, in a set of 12 commercial solvent-extracted soybean meals. In contrast to the present experiment, the KOH protein solubility values of those 12 soybean meal samples were in the 80s. Consequently, the inverse correlations were measured without **interference** from the damage to amino acid digestibility caused by overprocessing.

Table 4. Statistics of the correlations between trypsin inhibitor activity and digestible amino acid coefficients in 9 samples of mechanically extracted soybean meal

Samples listed in descending order of trypsin inhibitor activity

Line number	Sample code	DM %	CP %	TIA TUI/mg	dMET %	dLYS %
5	121	92.30	47.49	16.67	0.8275	0.8476
6	122	87.38	43.43	15.38	0.8100	0.8033
7	108	89.35	45.05	14.40	0.7919	0.7990
1	106	89.24	43.67	12.85	0.8593	0.8644
2	1690	92.35	44.03	11.50	0.8868	0.8840
3	1691	94.30	46.51	10.50	0.8939	0.8549
4	1701	95.07	47.00	7.59	0.8642	0.8035
8	1689	92.34	45.81	5.03	0.8826	0.8051
9	1688	91.80	45.06	3.50	0.8802	0.7677
R^2					0.4900	0.2150
r					0.7000	0.4637
P<0.05					0.0358	0.2086[1]

1 For dLYS P>0.05

Conclusions

1. Even though solvent-extracted soybean meal and mechanically extracted soybean meal are two different ingredients due to process differences, both processes are susceptible to protein quality damage from excess heat treatment. **KOH protein solubility is an effective chemical analysis for detecting overprocessing in both processes**. KOH protein solubility is directly correlated to dLYS in both processes. In fact, this is not entirely new, as Evans and St. John[3] reported that the solubility of soybean meal protein in a dilute solution of KOH was correlated with broiler chick performance. They worked with solvent-extracted soybean meal and Expeller soybean meal, different methods of oil extraction. KOH protein solubility was correlated to chick performance for birds fed soybean meal by either method of extraction. The KOH protein solubility test and its correlation with digestible amino acid coefficients are not exclusive of the solvent extraction process.

2. Trypsin inhibitors are the main anti-nutritional factors in soybeans, and their quantitative reduction is the main reason for the heat treatment during processing. Trypsin inhibitor activity needs to be measured and controlled because insufficient heat treatment leads to underprocessed mechanically extracted soybean meal. Underprocessed soybean meal (either solvent or mechanical extraction) results in trypsin inhibitors and other thermolabile antinutritional factors remaining at levels high enough to cause problems in the field with poultry[8] and swine,[14] such as poor feed conversion, loss of body weight gain, and intestinal integrity problems. Not to mention the negative effects on breeding animals.[15]

3. The practical conclusions of this study are drawn from the somewhat contradictory results presented in Tables 3 and 4 above. It appears that optimum KOH protein solubility lies in the 82-83% range for the digestible lysine coefficient (dLYS) to be in the 0.86-0.88 range (Table 3). However, the KOH protein solubility of 82-83% in this study corresponds to 11-13 TUI/mg of trypsin inhibitors (Table 1). Nevertheless, research with trypsin inhibitors has demonstrated that they need to be below 10 TUI/mg (or below 6.67 mg TId/g) for optimum performance in poultry.[16] Consequently, **the study's data suggests that a compromise must be made between KOH protein solubility and trypsin inhibitor activity to define practical quality parameters for mechanically extracted soybean meal**. The sample coded as 1691 (Tables 3 and 4) indicates that at a KOH protein solubility of ~77%, the trypsin inhibitor activity is reduced to ~10.5 TUI/mg (the same as ~7 mg TId/g). **Therefore, the practical recommendation for mechanically extracted soybean meal manufacturers by dry extrusion is a KOH protein solubility of about 77-78% and simultaneously trypsin inhibitor activity of no more than 10 TUI/mg (6.67 mg TId/g).**

4. The above highlighted recommendations imply that even though KOH protein solubility in the 80-83% range may deliver, in theory, a higher digestible lysine value, **in practice, the adverse effects of the excess trypsin inhibitor activity at that KOH protein solubility level may nullify the advantage of higher dLYS**. It is a paradox!

5. Finally, the KOH protein solubility optimum and the trypsin inhibitor activity optimum move in opposite directions. A very similar situation has been observed with solvent-extracted soybean meal.[16]

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