An update on amino acid requirements for laying hen

Roshan Adhikari
CJ America

Abstract

With the rise in the global population and demand for protein, egg and meat production will continuously grow in the future. This increase in demand for animal protein will increase in the cost of feed ingredients, constrain land and resources for production and increase overall production cost. The inclusion of available feed-grade amino acids in non-bound form can help to reduce the cost of formulation, reduce crude protein (CP) in the diets, maintain performance, reduce nitrogen excretion and help achieve sustainability goals. Reduction in CP in the diets will also help to improve the health and welfare of laying hens in changing housing systems and cope with environmental and management stress. However, to successfully implement the inclusion of 4th, 5th ,6th limiting amino acids in non bound form in the diets, the optimal requirement of those limiting amino acids for efficient production/growth has to be known to prevent marginal deficiency in birds. This short communication reviews the published literature in requirements of the first 6 limiting amino acids in laying hens diets.

Background

With the rise in global demand for protein, the volume of egg production has increased by 119% from 1990 to 2020 (FAOSTAT). As the global population is expected to surpass 10 billion in 2050, the demand for animal protein sources for consumption will constantly be on the rise. This market requirement will demand more laying hen production and more demand for poultry feed ingredients such as corn, wheat, SBM, DDGS, canola meal, sunflower meal and other cereal energy and protein sources (http://statistics.amis-outlook.org/data/index.html). With increasing feed cost, sustainability development goals, changing housing/welfare system, it is essential to develop reduced CP laying hen diets for cost-effective production. As more non-bound essential amino acids are becoming more economically feasible, the inclusion of such amino acids in the diets will reduce CP in the diets which can maintain/improve production, increase profitability and make cost-effective diets. Such reduced CP diets also reduce water intake, improve litter condition, reduce nitrogen excretion, less ammonia emission in laying hen houses. Furthermore, low CP diets will reduce the amount of excess nitrogen in the hind gut thus reducing the incidence of cocci and NE in laying hens. This is even more important as the housing system has changed from caged to the aviary and free-range production. All these benefits lead to improvement in the welfare and health of laying hens.

The majority of practical laying hen diets are formulated to digestible amino acid basis and minimum CP in the diet using 1st, 2nd, and/or 3rd limiting amino acids in non-bound form (Met, Lys, Thr). Other essential amino acids down the order are met by a bound amino acid source such as SBM, MBM, sunflower meal, canola meal. Such inclusion of SBM will balance 4th and over formulate the lower limiting amino acids in the diet. As we move towards including more non-bound amino acids to formulate better and cost-effective reduced CP diets, the nutritionist must be aware of the limiting amino acid requirement/ratios in their diet formulation so they do not under formulate an essential amino acid down the order that can affect performance.

The amino acid requirement will vary depending on the age, breed, feeding management, housing and environmental conditions. With a long lay cycle and long life span of layers, a proper balance of amino acids becomes essential for optimal production and profitability. However, there is a paucity of available data on the amino acids requirement in laying hens since NRC 1994. This short communication presents the first six limiting amino acid recommendations for laying hens.

Lysine

Lysine is the second limiting amino acid for laying hens in a corn-SBM-based diet. When formulating diets using the concept of ideal protein ratios, it is extremely important to accurately estimate the lysine requirement because the rest of the amino acids are balanced based on the ratio to Lys. An appropriate balance of Lys is essential to improve egg production, egg weight, egg mass, and feed conversion in laying hens (Faria et al., 2003; Spangler et al., 2019). Lys is also important to improve breast muscles development in pullets, maintain immune competency and digestive tract functionality in poultry (Kidd, 2004; Konieczka et al., 2021; Vaezi et al., 2011). Underestimation of Lys can have a negative impact on feed intake and performance. The latest study of Lys requirement of modern Hy-Line W36 fed from 32-44wks had a higher requirement for egg production than egg weight. The Lys requirement was 848mg Lys/hen/day for egg production and 828mg Lys/hen/day for egg weight using Quadratic broken line (Akbari Moghaddam Kakhki et al., 2016). On Lohmann LSL from 22-47wks of age, Lys needs for egg mass ranged from 655 to 817 mg Lys/hen/day, with 706 mg representing the intercept of the broken line and quadratic polynomial regression (Spangler et al., 2019). For the FCR, digestible Lys responses ranged from 690 to 866 mg Lys/hen/d, with 778 representing the digestible Lys estimate when using the broken line and the quadratic polynomial regression (Spangler et al., 2019). The dLys requirements were consistently lower for egg production than for egg mass and feed efficiency. Looking at the Lys requirement across the breeds and types of birds, there is a significant difference in the amount of Lys/bird/day recommendation among breeding companies for their birds. It will also dictate the amount of other amino acids in the diets as the ratios of other essential amino acids:dLys seems to be consistent among the breeds and types of birds (Table 1).

Table 1. Digestible Lys intake and ratio of other essential AA : dLys based on breeder recommendations during peak production

Amino acid requirement	Hy-line W36 -2020	Hy-line brown -2018	Hendrix genetics (Shaver/Bovan /DeKalb)	Lohmann brown lite (2017)	Lohmann LSL lite	Lohman brown classic (2017)	Brazilian table 4th edition
Dig Lysine (mg/bird/d)	800	820	850	680	690	720	
Ratios to dLys							
Met + Cys	91	91	87	90	90	91	98
Threonine	70	70	71	70	70	69	77
Tryptophan	21	21	22	22	22	21	23
Valine	88	88	88	87	88	88	93
Isoleucine	80	80	80	80	80	79	78
Arginine	107	104	105	104	104	104	100

Total Sulphur Amino acids (TSAA)

Methionine and Cystine (TSAA) are the first limiting amino acids in the laying hen diets. In practical poultry nutrition Met receives more attention than Cys as Met can act as a precursor to cysteine. Feed grade non-bound Met was introduced in the 1950s and was rapidly adopted in laying hen diets. TSAA requirements are met by supplementing Met to cover the Cys deficiency. TSAA is essential in poultry for the formation of coenzymes in cellular metabolism, feathering, regulating feed intake, improving egg mass and FCR. From the summary of published literature, the TSAA requirement is generally higher for the FCR than for egg mass. A meta-analysis of the TSAA requirement in laying hens resulted in ratio ranges of 85 to 88% for egg mass and 90 to 93 for the FCR (Spek, 2018; Kidd and Loar, 2021; Table 2).

Table 2. **Estimated optimal SID M+C requirements (% and daily intake) for maximum egg mass (EM) and minimum FCR excluding the values in which estimated SID M+C requirements values were outside the measurement range and where FCR values were lower than 1.8 or higher than 2.3. (Adopted from Spek 2018)**

	Parameter	N*	Mean	Std. Dev.	Min.	Max	%CV
SID M+C (%)	EM	18	0.602	0.1067	0.465	0.783	17.7
	FCR	19	0.628	0.0783	0.514	0.738	12.5
SID M+C intake (mg/d)	EM	18	663	97	521	789	14.6
	FCR	19	687	66.9	576	807	9.7
SID M+C intake per g of EM (mg/g)	EM	18	11.9	1.68	9.2	14.9	14.1
	FCR	19	12.3	1.24	10.3	14	10.1
SID M+C:SID LYS ratio	EM	18	85	15.4	61	115	18.1
	FCR	19	88	11.2	67	109	12.7

*number of titration trials (total number of titration trials is 30 (27 trials + 3 titration trials for which R values were estimated again after excluding the diet containing the lowest dietary SID-M+C level).

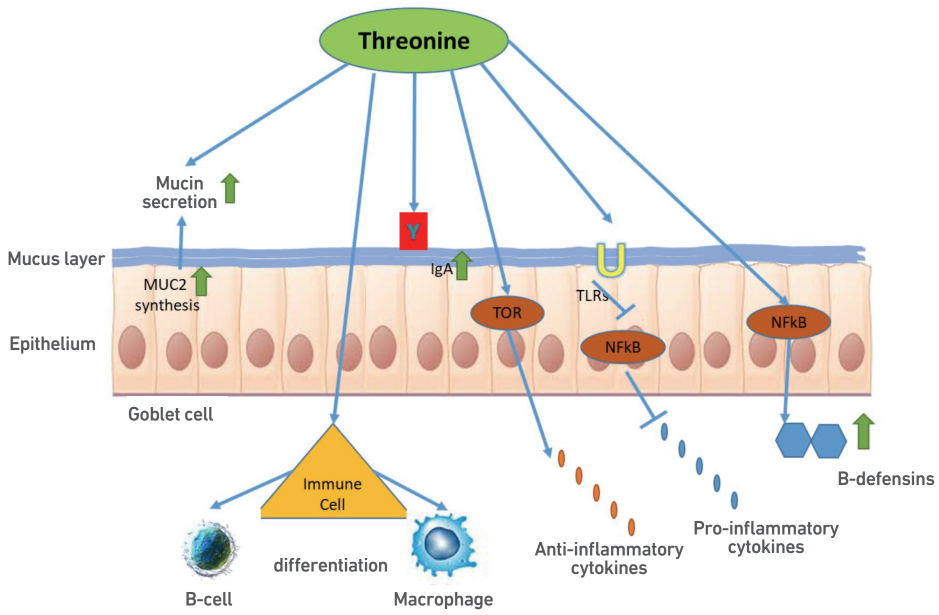
Threonine

Threonine (Thr) is the third limiting amino acid in laying hen diets using corn SBM based diets or wheat or sorghum-based diets as well. Threonine mainly serves as a substrate for protein synthesis, particularly mucin. In addition, Thr can enter the catabolic pathway, where it can be metabolized to a variety of important metabolites (glycine, acetyl CoA, pyruvate) that play a crucial role in host metabolism. Threonine has also been lined to have a benefit on improving intestinal barrier function, intestinal cell turnover, mucin 2 production, overall intestinal health and function (Figure 1), IgA expression as measured mRNA in diets with higher Thr level (Figure 2; Tang et al., 2021).

Comparing across the breeds the dThr:dLys ratio recommendation range from 69 to 71. Brazilian table has indicated the ratio to be 77. The range of Thr requirement (mg/bird/day) for laying hens in published reports from 2003 to 2019 range from 461 (Bregendahl et al., 2008) to 523 (Rocha et al., 2013) for FCR; 414 (Bregendahl et al., 2008) to 505 (Cupertino et al., 2010) for Egg mass; 400 (Bregendahl et al., 2008) – 708 (Azzam et al., 2019) for egg production and 418 (Bregendahl et al., 2008) to 606 (Rocha et al., 2013) for egg weight.

The previous research has indicated that the requirement for Thr can be elevated in chronic diseases, intestinal challenges, or environmental stress. Research has indicated that increasing the dThr:dLys from 67 to 72 improved performance during the NE challenge model. As the housing system has been changing in the USA from cages to the aviary, free-range, organic that included floor system, Thr requirement overall and during a stressful period of cocci and NE outbreak warrants further investigation. Increasing the Thr requirement during such a stressful period could help elevate gut stress and improve performance.

Thr affects goblet cells differentiation and stimulates MUC2 synthesis, regulates immune cell differentiation and immunoglobulins production, modulates release of cytokines.



Abbreviations: IgA: immunoglobulins A; MUC2: mucin-2; TLRs: toll-like receptors; TOR: the target of rapamycin; NF-κB: nuclear factor κ-light-chain-enhancer of activated B cells.

Figure 1. Dietary Threonine modulates intestinal immune function (Adopted from Tang et al., 2021).

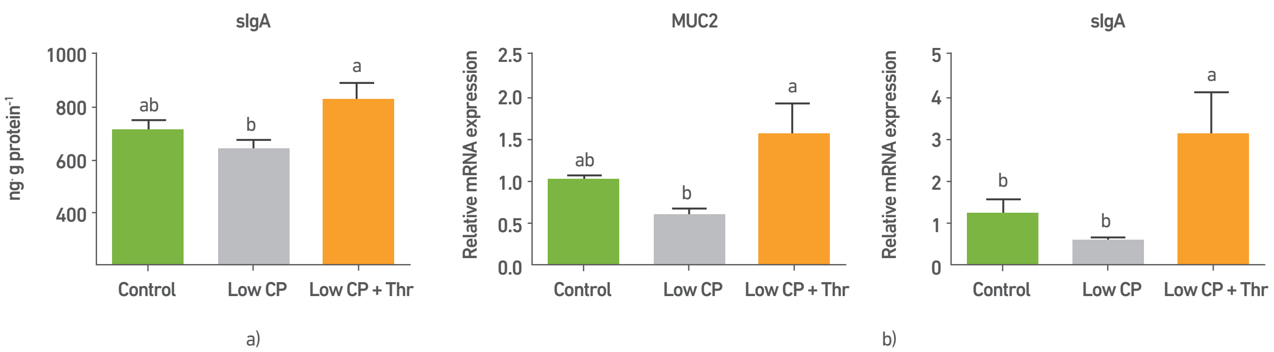


Figure 2. Effects of control (16% CP), low CP (14% CP), or low CP supplemented with 0.3% L-threonine diets on a) the concentration of secretory IgA (sIgA) in the ileum of laying hens; b) the mRNA expression of mucin 2 (MUC2, left) and secretory IgA (sIgA, right) in the ileum of laying hens. Values on each bar with no common letter differ significantly ($P < 0.05$; Adopted from Dong et al., 2017).

Tryptophan

Tryptophan comes as possible 4th, 5th, or 6th limiting amino acids in some of the USA-based diets depending on the dTrp:dLys ratio. Tryptophan is primarily used for protein synthesis to maintain egg production but also plays a role as a precursor of serotonin which is synthesized to melatonin to regulate sleep, appetite, and stress responses. Eggshell quality, egg shell thickness and egg composition are also affected by the Trp concentration in the diet (Khattak & Helmbrecht, 2019).

Tryptophan is a serotonin precursor and low serotonin levels in brain tissue lead to aggressive behavior, feather-pecking and cannibalism in layers resulting in substantial production losses (Birkl et al., 2017; Mindus et al., 2021). In addition, Trp can be converted to the B-vitamin niacin through the kynurenine pathway (Rogers & Pesti, 1990).

Large range of dTrp:dLys ratio is currently used in the industry ranging from 17 to 24 with breeder recommendations ranging from 21-22. A study on W-36 hens at 30 wks of age reported the dTrp:dLys ratio ranged from 17.5 to 29 based on the response criteria and regression models (Mousavi et al., 2018). The range of Trp requirement (mg/bird/day) for laying hens in published reports from 2003 to 2019 range from 95 (Bregendahl et al., 2008) to 211(Cardoso et al., 2014) for FCR; 104 (Mousavi et al., 2018) to 212 (Cardoso et al., 2014) for Egg mass; 119 (Bregendahl et al., 2008) to 250 (Khattak & Helmbrecht, 2019) for egg production. A recently published study on laying hens (Hy-Line W 36) from 22 to 34 wks of age reported 173 mg/bird/d for feed efficiency, 183 mg/bird/day for egg mass, and 192 for hen day egg production using the Quadratic polynomial model (Sarsour et al., 2021).

Relation of Tryptophan to serotonin precursor as a stress reducer could play a role in reducing piling mortality in pullets, feather picking, cannibalism which could play an important role in improving the welfare and productivity of birds in alternative housing systems (aviary, cage-free).

Branched Chain Amino acids (BCAA)

Valine (Val) and isoleucine (Ile) can be 4th, 5th, or 6th limiting amino acids in typical corn, SBM, DDGS based laying hen diets. As we learn more about the interaction and antagonism of Val, Ile, and leucine (Leu) in pigs and broilers, it is extremely important to formulate diets with a balanced profile of BCAA. One laying hen study reported that increased dietary Ile in the diet from 0.57 to 1.15% of diet significantly suppressed the egg production and egg mass but restored in the presence of diets increased in both Val and Leu (Peganova & Eder, 2003). Valine and Ile have been most examined for requirement, while there is very limited data on Leu requirement as leu requirement is met by protein source used in the diet. There are very few studies that are designed to investigate the three-way interaction of Val, Ile and Leu in laying hens diets. If we look at the amount of dLeu:dLys in typical corn, SBM, DDGS diets, it could easily range above 165 up to 200 which is way above the requirement of Leu for laying hens. Such imbalance of Leu in the diet can cause marginal deficiency of Val and Ile creating nutritional, health, feathering, bones and production problems in laying hens. BCAA also regulates fatty acid metabolism in the liver which may be important in hepatic yolk-lipoprotein production and could be a rate-limiting factor for egg formation. Deficiency in Val or Ile can cause depression in feed intake, improper feathering, and skeletal issues.

Large range of dVal:dLys ratio is currently used in the industry with breeder recommendations ranging from 87-88 and Brazilian tables recommending 93. A study on W-36 hens at 52-58 wks of age reported the dVal:dLys ratio of 93 to optimize egg mass (Bregendahl et al., 2008). Another study on Hy-Line W36 hens from 41 to 60 wks old reported the dVal:dLys ratio of 89% to optimize egg mass, 85% to optimize hen housed egg production and 80% to optimize FCR (Wen et al., 2019). If we compare the requirement for egg mass between the above two studies, there is an 8 percentage points higher requirement of Val in Bregendahl et al., 2008 compared to Wen et al., 2019. The diet from Berendahl et al., 2008 is a corn-SBM-MBM based diet that can have dLeu:dLys as high as 180 whereas the diets from Wen et al.,2019 is a corn-peanut meal-based diet which contains dLeu:dLys ratio of 118, close to the requirement in laying hens. This increase in Leu ratio in the Bregendahl study could warrant a higher level of Val in the diet.

Ile requirement studies in laying hens are scarce. Several studies have linked Ile to egg size and uniformity. Large range of dIle:dLys ratio is currently used in the industry with breeder recommendations ranging from 79-80 and Brazilian tables recommending 78. A study on W-36 hens at 52-58 wks of age reported the dIle:dLys ratio of 79 to optimize egg mass (Bregendahl et al., 2008). Another study on Shaver hens from 20 to 46 wks old reported the dIle:dLys ratio of 82-88% depending on the parameter. Both studies have the requirement ratio close to each other with Leu in the diet ranging from 160-180 in those diets.
The requirement of Val and Ile could depend on the amount of Leu in the diet. Further studies must be conducted in laying hens to understand the Val and Ile requirement for different age and production at different Leu ratios.

Summary

Amino acid requirement of laying hens are altered because of numerous experimental conditions including breed, age, housing system, ingredinets used, basal diet variation. Valuation of amino acids are highly subjected to the production parameters such as weight gain, egg weight, FCR, egg production, and egg mass that use optimization. Proper implementation of amino acid requirements and adoption of available non-bound limiting amino acids can help nutritionists formulate cost-effective, low CP diets meeting all the essential amino acid requirements. Such diets will also maintain the performance, reduce nitrogen excretion, reduce ammonia emission in the house, reduce water intake, improve digestibility coefficient, increase production, reduce health challenges thus improving the profitability, health, and welfare of birds.

REFERENCES

1. Akbari Moghaddam Kakhki, R., Golian, A., & Zarghi, H. (2016). Effect of dietary digestible lysine concentration on performance, egg quality, and blood metabolites in laying hens. Journal of Applied Poultry Research, 25(4), 506–517. https://doi.org/10.3382/japr/pfw032
2. Azzam, M. M., Alhotan, R., Al-Abdullatif, A., Al-Mufarrej, S., Mabkhot, M., Alhidary, I. A., & Zheng, C. (2019). Threonine Requirements in Dietary Low Crude Protein for Laying Hens under High-Temperature Environmental Climate. Animals, 9(9), 586. https://doi.org/10.3390/ani9090586
3. Birkl, P., Franke, L., Bas Rodenburg, T., Ellen, E., & Harlander-Matauschek, A. (2017). A role for plasma aromatic amino acids in injurious pecking behavior in laying hens. Physiology & Behavior, 175, 88–96. https://doi.org/10.1016/j.physbeh.2017.03.041
4. Bregendahl, K., Roberts, S. A., Kerr, B., & Hoehler, D. (2008). Ideal ratios of isoleucine, methionine, methionine plus cystine, threonine, tryptophan, and valine relative to lysine for white leghorn-type laying hens of twenty-eight to thirty-four weeks of age. Poultry Science, 87(4), 744–758. https://doi.org/10.3382/ps.2007-00412

REFERENCES

5. Cardoso, A. S., Costa, F. G. P., Vilar da Silva, J. H., Saraiva, E. P., Nogueira, E. T., Santos, C. S., Ramalho de Lima, M., & Vieira, D. V. G. (2014). Nutritional requirement of digestible tryptophan for white-egg layers of 60 to 76 weeks of age. Journal of Applied Poultry Research, 23(4), 729–734. https://doi.org/10.3382/japr.2014-01012
6. Dong, X. Y., Azzam, M. M. M., & Zou, X. T. (2017). Effects of dietary threonine supplementation on intestinal barrier function and gut microbiota of laying hens. Poultry Science, 96(10), 3654–3663. https://doi.org/10.3382/ps/pex185
7. FAOSTAT. (n.d.). Retrieved December 28, 2021, from https://www.fao.org/faostat/en/#home
8. Faria, D. E., Harms, R. H., Antar, R. S., & Russell, G. B. (2003). Re-evaluation of the Lysine Requirement of the Commercial Laying Hen in a Corn-Soybean Meal Diet. Journal of Applied Animal Research, 23(2), 161–174. https://-doi.org/10.1080/09712119.2003.9706418
9. Khattak, F., & Helmbrecht, A. (2019). Effect of different levels of tryptophan on productive performance, egg quality, blood biochemistry, and caecal microbiota of hens housed in enriched colony cages under commercial stocking density. Poultry Science, 98(5), 2094–2104. https://doi.org/10.3382/ps/pey562
10. Kidd, M. (2004). Nutritional modulation of immune function in broilers1. Poultry Science, 83(4), 650–657. https://-doi.org/10.1093/ps/83.4.650
11. Konieczka, P., Mikulski, D., Ognik, K., Juśkiewicz, J., Zduńczyk, Z., & Jankowski, J. (2021). Increased Dietary Inclusion Levels of Lysine Are More Effective than Arginine in Supporting the Functional Status of the Gut in Growing Turkeys. Animals : An Open Access Journal from MDPI, 11(8), 2351. https://doi.org/10.3390/ani11082351
12. Mindus, C., van Staaveren, N., Fuchs, D., Gostner, J. M., Kjaer, J. B., Kunze, W., Mian, M. F., Shoveller, A. K., Forsythe, P., & Harlander-Matauschek, A. (2021). L. rhamnosus improves the immune response and tryptophan catabolism in laying hen pullets. Scientific Reports, 11(1), 19538. https://doi.org/10.1038/s41598-021-98459-x
13. Mousavi, S. N., Afsar, A., Khalaji, S., & Abbasi, M. (2018). Estimation of digestible tryptophan:lysine ratios for maximum performance, egg quality and welfare of white-egg-laying hens by fitting the different non-linear models. Journal of Applied Animal Research, 46(1), 411–416. https://doi.org/10.1080/09712119.2017.1316278
14. Peganova, S., & Eder, K. (2003). Interactions of various supplies of isoleucine, valine, leucine and tryptophan on the performance of laying hens. Poultry Science, 82(1), 100–105. https://doi.org/10.1093/ps/82.1.100
15. Rocha, T. C. da, Gomes, P. C., Donzele, J. L., Rostagno, H. S., Mello, H. H. de C., Ribeiro, C. L. N., & Troni, A. R. (2013). Digestible threonine to lysine ratio in diets for laying hens aged 24-40 weeks. Revista Brasileira de Zootecnia, 42, 879–884. https://doi.org/10.1590/S1516-35982013001200007
16. Rogers, S. R., & Pesti, G. M. (1990). The Influence of Dietary Tryptophan on Broiler Chick Growth and Lipid Metabolism as Mediated by Dietary Protein Levels. Poultry Science, 69(5), 746–756. https://doi.org/10.3382/ps.0690746
17. Sarsour, A. H., Lee, J. T., Haydon, K., & Persia, M. E. (2021). Tryptophan requirement of first-cycle commercial laying hens in peak egg production. Poultry Science, 100(3), 100896. https://doi.org/10.1016/j.psj.2020.11.065
18. Spangler, H., Utterback, P., Parsons, C. M., & Tillman, P. (2019). Determining the digestible lysine requirement of 22- to 47-week-old Lohmann laying hens using an increasing protein titration methodology. Poultry Science, 98(4), 1706–1715. https://doi.org/10.3382/ps/pey503
19. Tang, Q., Tan, P., Ma, N., & Ma, X. (2021). Physiological Functions of Threonine in Animals: Beyond Nutrition Metabolism. Nutrients, 13(8), 2592. https://doi.org/10.3390/nu13082592
20. Vaezi, G., Teshfam, M., Bahadoran, S., Farazyan, H., & Hosseini, S. (2011). Effects of Different Levels of Lysine on Small Intestinal Villous Morphology in Starter Diet of Broiler Chickens. 4
21. Wen, J., Helmbrecht, A., Elliot, M. A., Thomson, J., & Persia, M. E. (2019). Evaluation of the Valine requirement of small-framed first cycle laying hens. Poultry Science, 98(3), 1272–1279. https://doi.org/10.3382/ps/pey448