

## EFFECTS OF PRE-LAY DIFFERENT PROTEIN AND ENERGY DIETARY LEVELS ON SUBSEQUENT PERFORMANCES IN LOHMANN BROWN

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### Abstract

*The aim of this paper was to find out the effects of diets with different levels of metabolizable energy (ME) and crude protein (CP) administered during the pre-lay period on the subsequent productive performances of the Lohmann Brown hybrid. A low ME diet (2,690 kcal/kg) and a high ME diet (2,820 kcal/kg) were prepared, each one with two variants of CP levels: low (15%) and high (17%). These diets were applied from 15 to 22 weeks of age. From 23 weeks of age, all birds were fed a diet containing 2,700 kcal/kg and 15.6% protein. Egg rate was significantly higher in the high-protein-high-ME fed group. In the same group, the intensity of the yolk colour decreased and the level of total protein concentration in the albumen was higher. Other researched productive parameters (feed intake, egg mass, feed conversion rate) and egg properties (eggshell colour, eggshell breaking strength, Haugh units) were found not significantly modified by the experimental pre-lay feeding.*

**Key words:** Lohmann hybrid, metabolic energy, production performance, egg features.

### INTRODUCTION

The Lohmann Brown hen originates from Germany. Since then, Lohmann Brown hen have become the best hybrid layer due to its large, brown-shelled eggs (Lohmann Management Guide, 2024). The Lohmann Brown hybrid is a cross-breed of the White Rock and Rhode Island Red hen breeds or strains. This Lohmann Brown has been widely used in the egg industry from Germany to other parts of the world. It is also one of the earliest hybrids developed and is still in demand today due to its excellent productivity, egg quality and adaptability (Lohmann Management Guide, 2024). This Lohmann hybrid is the most widespread laying hens in the worldwide. Among the productive performances of this egg-laying hybrid, we specify: a production of 337.5 eggs in a laying period from 19 to 80 weeks of age, egg breaking resistance of over 35 newton, an egg average weight of 65 g, a

feed conversion index of 2.1-2.2 kg feed/kg egg mass, and a daily *ad libitum* consumption of 110-120 g feed (Văcaru Opriş et al., 2002). Sensibility to some parasitic diseases and tuberculosis, predisposition to moulting, nutritional requirements, sensibility to housing conditions and a poor carcass quality are some of the hybrid's problems. As a product of an intense process of selection and amelioration, itself still subject to an intensive selection and amelioration pressure, the hybrid attracted the attention not only of selectors and breeders, but also of other categories of researchers, including those of nutrition physiology (Usturoi et al., 2006; Usturoi et al., 2022). The work of specialists in the nutrition physiology of this hybrid is focused on creating diets that respond as precisely as possible to its nutritional requirements. Digestive absorption and feed conversion capacities (Alagawany et al., 2016), response to different dietary protein sources (Wang et al., 2014), different protein

levels and different energy levels (Kim, 2014; Xin et al., 2022) of the diet are just a few examples of the lines of investigation. The purpose of this work is to investigate the effects of different dietary protein and energy levels during the pre-lay period on subsequent laying performances and egg features in Lohmann layers. The data will be used to establish diets that respond as accurately as possible to the requirements of the species during the egg-laying period.

## MATERIALS AND METHODS

### Experimental design

The experiment was carried out on 360 Lohman brown hens. The birds were grouped into four equal groups of 90 cap. each. The four groups of 90 birds each were housed in four 5.6/4 m cages, totalling 22.4 m<sup>2</sup> each, achieving a density of 4 cap./m<sup>2</sup>. Four experimental diets were prepared: two low metabolic energy diets

and two high metabolic energy diets. Each of the two types of diets with different levels of metabolic energy had in their composition different levels of protein (low and high, Table 1). The four experimental diets were administered *ad libitum* from 15 to 22 weeks of age. From 23 weeks of age all individuals in the four groups were fed the same new diet, formulated according to National Research Center (NRC), Nutrient recommendations for poultry (1994), with a content of 2700 kcal/kg feed and 15.6 g crude protein/100 kg feed (Table 1). The room temperature was maintained at 22 ± 2°C throughout the study. Light period was 8 hour/day (7:00-15:00) at 15 weeks of age, then increased gradually by 1 hour/week till 14 hours/day (5:00-19:00) until 22 weeks of age of the birds.

Egg production, daily feed consumption and body weight were monitored through the experiment.

Table 1. Metabolic energy, protein, ingredients and nutrient composition of the diets formulated for experimental groups for pre-laying (15-22 weeks) and laying (23-50 weeks) periods

Energy	Experimental pre-laying feeding period (15-22 weeks)				Laying feeding period (23-50 weeks)
	Low level ME		High level ME		
Protein	Low level CP	High level CP	Low level CP	High level CP	
<i>Ingredients</i>					
Corn	63.1	61.0	61.1	60.2	63.9
Soybean meal	17.1	22.2	16.9	22.4	22.7
Oil	-	-	2.1	1.6	1
Bran	15	12	16	11	-
Limestone	1.74	1.74	1.74	1.74	9.5
Choline chloride	0.08	0.08	0.08	0.08	0.08
Mono-dicalcium phosphate	0.65	0.65	0.65	0.65	0.78
Salt	0.31	0.31	0.31	0.31	0.31
Medical Stone	1.95	1.95	1.95	1.95	0.97
Lysine	0.1	0.1	0.1	0.1	0.12
Methionine	0.07	0.07	0.07	0.07	0.12
Vitamin and mineral premix <sup>1</sup>	0.25	0.25	0.25	0.25	-
Vitamin and mineral premix <sup>2</sup>	-	-	-	-	0.35
<i>Nutrient composition</i>					
Crude protein <sup>3</sup>	15.11	16.92	14.77	16.78	15.60
ME, Kcal/kg <sup>3</sup>	2,690	2,690	2,820	2,820	2,700
Calcium, %	0.85	0.85	0.85	0.85	3.55
Available phosphorus, %	0.56	0.56	0.56	0.56	0.49
Lysine, %	0.79	0.79	0.79	0.79	0.78
Methionine, %	0.30	0.30	0.30	0.30	0.40

<sup>1</sup> Provided per kg of premix: vitamin A, 160,000 IU; vitamin D, 66,000 IU; vitamin E, 550 mg; vitamin K, 40 mg; vitamin B<sub>1</sub>, 60 mg; vitamin B<sub>2</sub>, 130 mg; vitamin B<sub>6</sub>, 50 mg; pantothenate, 220 mg; nicotinamide, 500 mg; folic acid, 10 mg; biotin, 6 mg; Cu, 0.30 g; Fe, 1 g; Mn, 2.50 g; Zn, 2.00 g; I, 13 mg; Se, 10 mg

<sup>2</sup> Provided per kg of premix: vitamin A, 220,000 IU; vitamin D, 72,000 IU; vitamin E, 550 mg; vitamin K, 80 mg; vitamin B<sub>1</sub>, 50 mg; vitamin B<sub>2</sub>, 160 mg; vitamin B<sub>6</sub>, 70 mg; vitamin B<sub>12</sub>, 1 mg; pantothenate, 220 mg; nicotinamide, 700 mg; folic acid, 30 mg; biotin, 6 mg; Cu, 350 g; Fe, 1.5 g; Mn, 3.0 g; Zn, 2.5 g; I, 20 mg; Se, 20 mg

<sup>3</sup> Calculated values

### Egg quality

Egg weight was determined using an ORMA model analytical balance. Shell thickness was measured in the equatorial zone of the egg using an EGG Shell Thickness Gauge tester, Ro.Ma.snc Italia. The breaking strength of the eggs was determined with the Egg Force Reader: the device measures the minimum force required to break the egg shell, expressing the results in mg/cm<sup>2</sup>. Determination of egg white consistency was performed using an EGG Analyser and was expressed in Haugh units. The colour intensity of the eggshell was analysed using a Model Novo-Shade Duo Colour Reflectometer, the comparison being assimilated to the colour shade assessment system provided by the manufacturer of the analysed hybrid. Yolk colour was determined

using a combination of two methods: a method that uses the "Roche Yolk Colour Fan" colour palette with values from 1 to 15. Albumen protein was determined by Kjeldahl method (AOAC, 1995).

### Statistics

The data were statistically processed by determining the mean and the standard error of the mean, using a dedicated software. The differences between the groups were analysed based on the Student's t test, being considered significant at  $P < 0.05$ .

## RESULTS AND DISCUSSIONS

The analysis of the productive parameters presented in Table 2 shows the following particularities.

Table 2. Effect of dietary metabolizable energy and crude protein levels during the pre-lay period (15-22 weeks of age) on the productive performance of Lohmann brown from 23 to 50 weeks of age (mean values)

Item	Low protein diets		High protein diets		SEM	P value
	Low ME diet	High ME diet	Low ME diet	High ME diet		
Feed intake (g/d) <sup>1</sup>	118.1	121.1	118.8	117.3		
Body weight (g) <sup>2</sup>	1,921±32	1,956±44	1,911±33	1,964±51	63	0.611
Laying rate (%) <sup>1</sup>	82.6±2.2 <sup>#</sup>	83.9±0.8	83.3±3.0	86.3±3.0 <sup>#</sup>	5.33	0.011
Egg mass (g/d/cap.) <sup>3</sup>	55.3±5.2	52.5±3.2	52.1±5.3	54.3±4.8	6.33	0.329
Egg weight (g) <sup>1</sup>	62.7±6.5	62.6±5.3	62.6±4.3	63.0±4.0	4.79	0.049
Feed conversion rate (kg food / kg egg) <sup>1</sup>	2.28±0.34	2.29±0.66	2.28±0.21	2.16±0.11	0.43	0.096

<sup>1</sup>Calculated as mean from 23 to 50 weeks of laying

<sup>2</sup>Mean values at the end of experimental monitoring (50-wk-old hens)

<sup>3</sup>Calculated by multiplying the egg production by the egg weigh expressed as mean of weekly values

Values with the same superscript are statistically different

SEM = standard error of mean

Values as mean± standard error of mean

The average feed consumption during the period of 23 to 50 weeks of age of the birds did not show significant differences, being between 117.3 and 121 g/day/cap. The body weight at week 50 of age of the birds was higher in hens fed high ME diets but the difference remained no significant ( $P > 0.05$ ). The average egg laying rate during the period 23-50 weeks of age was the only productive parameter with significant differences between the groups ( $P < 0.05$ ), the higher values being of the group fed high protein high ME diet. The same group presented the lowest feed conversion rate: 2.16 kg food/kg egg mass. Average egg weight and average egg mass were not significantly influenced during the monitored period (23-50

weeks), after feeding the four experimental diets. From the analysis of the data presented in Figure 1 it can be seen that the high-protein-high-ME diet fed group was the first to reach the laying peak and the egg laying values were above those of the other three groups during the following monitoring period, from 23 to 50 weeks of age. In the same period, the egg laying curve of the low-protein-low-ME group was placed below those of the other groups during the whole period. The graphic evolution shows that in the last three weeks of monitoring, the curves of the four batches are approaching, revealing the disappearance of the differences between the groups.

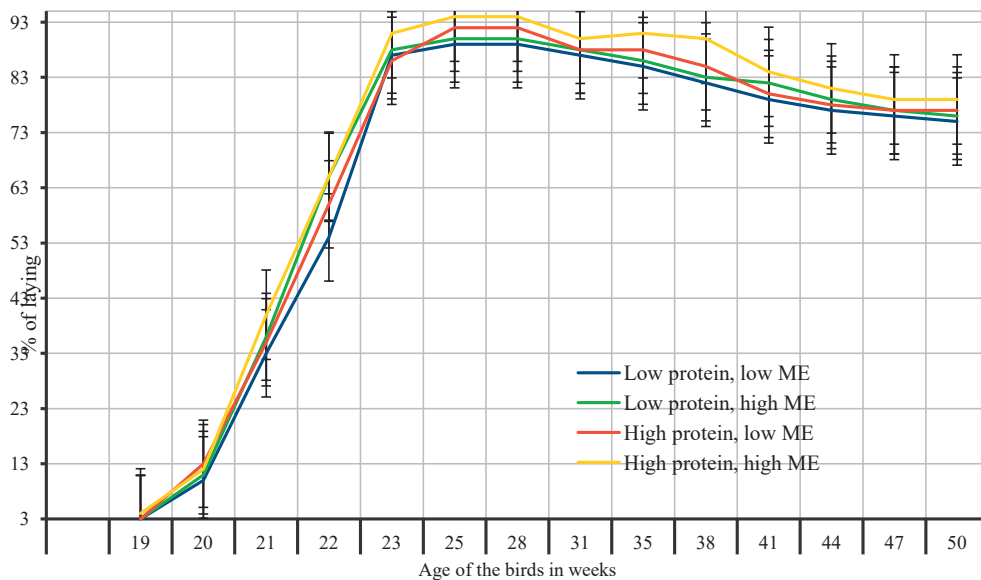


Figure 1. Evolution of laying (as % /housed hens) during experimental feeding and thereafter (23-50 weeks of age)

The results obtained by us on the Lohmann brown hybrid are partially in agreement with the data published by other authors (Xin et al., 2022) in their study on aged He Line brown hens. These authors found that diets with 15 or 16% crude protein and energy levels of 2.700 or 2.800 kcal did not significantly influence the egg production, egg weight, egg mass, and feed intake. Other researchers (Summers et al., 1994) tested diets with crude protein levels of 20, 17, 14, and 11% and found that decreased egg weight proportionally to decreasing protein content of the diet in White Leghorn hens. According to our results, we noted a decrease in the use of feed protein as Lohmann brown birds get older, which is in agreement with the data published by other authors (Schwartz & Allen, 1981) in Japanese Quail females, the aspect being a subject of further investigation.

Our results regarding the increase in laying rate and decrease in feed consumption rate in hens that were fed diets enriched in protein are also confirmed by other authors (Reid, 1976): this author reports a feed consumption rate of 1.89 kg/dozen eggs on a diet with 19% protein, values that increases progressively with the decrease in the percentage of crude protein in the diet. On a range of values from 19 to 13%

crude protein, other authors (Heo et al., 2023) studied the effects of feed protein levels on egg properties finding no significant effects on egg physical properties (breaking strength, shell colour) but a significant effect on egg weight. The same authors also found no significant effects on daily feed consumption.

In our study, the analysis of egg properties (Table 3) reveals some significant influences, which maintained until the end of experimental feeding. A higher protein concentration of albumen ( $P < 0.05$ ) was found in hens fed on protein enriched diets. Also, the shell thickness and egg shape index (not the breaking strength of the eggs) are significantly higher ( $P < 0.05$ ) in the groups fed on protein-enriched diets. Significant influences of the level of metabolizable energy of the diets were not identified at this age of the birds.

In their study regarding the effects of different levels of crude protein and metabolizable energy in the diet in the pre-lay period on egg characteristics, some authors (Xin et al., 2023) found that at the age of 72 weeks of the birds, only the egg shape index, eggshell thickness and yolk colour still retain significant influences.

Table 3. Effect of dietary metabolizable energy and crude protein levels during the pre-lay period (15-22 weeks of age) on the egg features of Lohmann brown hen (mean values of 30 eggs from five working days in the 50<sup>th</sup> week of life)

\Item	Low protein diets			High protein diets			SEM	P value
	Low ME diet	High ME diet	Mean	Low ME diet	High ME diet	Mean		
Egg shape index	1.36	1.39	1.38 <sup>a</sup>	1.34	1.37	1.36 <sup>a</sup>	0.23	0.011
Eggshell breaking strength (kg/cm <sup>2</sup> )	3.32	3.43		3.44	3.21		0.54	0.321
Eggshell thickness (µm)	330	330	330 <sup>b</sup>	355	360	354 <sup>b</sup>	54	0.110
Eggshell colour (absorbance)*	55	58		60	60		33	0.202
Yolk colour intensity**	6.6	6.8	6.7 <sup>c</sup>	6.3	6.0	6.22 <sup>c</sup>	1.32	0.032
Haugh unit (egg freshness)	78.2	77.7		81.0	80.3		18.5	0.065
Albumen protein conc. (mg/mL)	124.3	131.0	127.6 <sup>d</sup>	126.4	133.0	129.7 <sup>d</sup>	32.3	0.087

\*Values expressed by the degree of light absorption from 0 to 100

\*\*Mean values of a minimum three readings on the colour scale from 1 to 15

Values with the same superscript differ significantly

The other investigated egg properties in the experiment (eggshell breaking strength, yolk colour and Haugh units) no longer retained significant influences from energy and crude protein differentiated diets during the pre-lay period. The analysis of the graphic evolution of the total protein content of the albumen shows that these differences between the groups appeared as early as the age of 23 weeks of the birds, the differences reducing with increasing age (Figure 2). On the other hand, some

researchers (Heo et al., 2023) in experiments on 13-week-old He Line brown found the increase in the intensity of the colour of the yolk proportional to the decrease in the crude protein content of the diet. The explanation given by Moros et al. (2022) and Galobart et al. (2004) was that this effect was secondary to the increase in corn gluten concentration of low crude protein diets. It is well known that corn gluten meal is rich in natural pigments (e.g., xanthophylls) for egg yolk colouration.

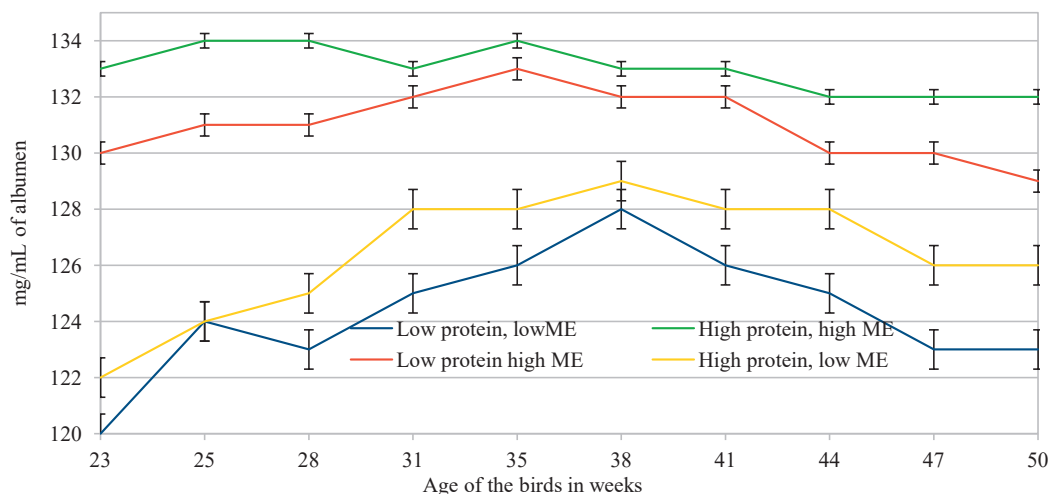


Figure 2. Evolution of the total albumen protein concentration in Lohmann brown hens fed on different protein and EM level diets in prelay period (15-22 weeks)

## CONCLUSIONS

Feeding Lohmann brown chickens on diets containing different protein and ME levels in the pre-lay period (15-22 weeks) allowed the characterization of the effects on the productive performance and egg features during the subsequent laying period. Egg rate was significantly higher in the high protein high ME group. In the same group, the intensity of the yolk colour decreased and the level of total protein concentration in the albumen was higher. Other studied productive parameters (feed intake, egg mass, egg consumption rate) and egg properties (eggshell colour, eggshell breaking strength, Hough units) were not significantly modified by the experimental pre-lay feeding.

## REFERENCES

- Alagawany, M., Abd El-Hack, M.E., Ragab, M., Tiwari, R., Sachan, S., Karthik K. & Dhama, K (2016). Positive and negative impacts of dietary protein levels in laying hens. *Asian J. Anim. Sci.*, 10,165-174.
- AOAC (1995). *International, Eggs and egg products, Official methods of analysis of AOAC International*. Ed. Gaithersburg M.D.
- Kim, J.-H. (2014). Energy metabolism and protein utilization in chicken. A review. *Korean Journal of Poultry Science*. *The Korean Society of Poultry Science*. 4, 313-322. <http://dx.doi.org/10.5536/KJPS.2014.41.4.313>.
- Galobart, J., Sala, R., Rinc on-Carruyo, W., Manzanilla, E.G., Vila, ., & J. Gasa, J. (2004). Egg yolk color as affected by saponification of different natural pigmentation sources. *J. Appl. Poult. Res.* 13:328–334.
- Heo, Y.-J., Park, J., Kim, Y.-B., Kwon, B.-Y., Kim, D.-H., Song, J., & Lee, K.-W. (2023). Effects of dietary protein levels on performance, nitrogen excretion, and odor emission of growing pullets and laying hens. *Poultry Science*. 102, 102798. <https://doi.org/10.1016/j.psj.2023.102798>.
- Lohmann brown classic management guide. Lohman breeders, <https://lohmman-breeders.com/media/strains/cage/management/Lohmann-Brown-Classic-Cage.pdf> (accessed 2024)
- Moros, E. E., Darnoko, D., Cheryan, M., Perkins, E.G., & Jerrell J. (2002). Analysis of xanthophylls in corn by HPLC. *J. Agric. Food Chem.* 50,787–5790.
- NRC. "Nutrient Requirements of Poultry. ed." *National Academy of Sciences - NRC, Washington, DC* (1994).
- Reid, B.L. (1975). Estimated daily protein requirements in laying hens. *Poultry science*. 55, 461-465.
- Schwartz, R.W. Allen, N.K. (1981). Effect of ageing on protein requirements. *Poultry science*. 60, 342-348.
- Usturoi, M.G., I. Văcaru Opreș, I., & Ciocan, I. (2006). Productive parameters achieved by the laying hybrid Lohmann Brown on permanent layer. *Cercetări Agronomice în Moldova* 39 (2), 61-70 .
- Usturoi, M.G., Radu-Rusu, R.M., Usturoi, A. & Rațu, R.N. (2022). Adaptability of Lohmann Brown Hybrid to different production systems. *Animal & Food Sciences Journal. Iași*. 78, 59-65.
- Văcaru Opreș, I., S. Țârlea, S., Plăcintă, P., Van, I., & Bunaciu, P. (2002) – *Treatise of Aviculture*, II, Ceres, Bucharest.
- Wang, X., Peebles, E.D., & Zhai, W. (2014). Effects of protein source and nutrient density in the diets of male broilers from 8 to 21 days of age on their subsequent growth, blood constituents, and carcass compositions. *Poultry Science* 93,1463–1474. <http://dx.doi.org/10.3382/ps.2013-03838>.
- Xin, Q., Ma, N., Jiao, H., Wang, X., Li, H., Zhou, Y., Zhao, J., & Lin, H. (2022). Dietary energy and protein levels during the pre-lay period on production performance, egg quality, expression of genes in hypothalamus-pituitary-ovary axis, and bone parameters in aged laying hens. *Front. Physiol.* 13,1–17. doi: 10.3389/fphys.2022.88738.1.

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