Influence of feed form and energy concentration of the rearing phase diets on productivity, digestive tract development and body measurements of brown-egg laying hens fed diets varying in energy concentration from 17 to 46 wk of age

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ABSTRACT

The influence of feed form and energy content of the rearing phase diets on hen productivity, gastrointestinal tract (GIT) traits and body measurements was studied in brown-egg laying hens fed diets differing in energy concentration from 17 to 46 wk of age. The experiment was completely randomized with 12 treatments arranged as a $(2 \times 3) \times 2$ factorial with two feed forms (mash vs. crumbles) and three AME_n concentrations of the rearing phase diets (low, medium and high) and two AME_n concentrations (11.10 vs. 11.52 MJ/kg) of the laying phase diets. The AME_n of the rearing phase diets differed in 0.42 MJ/kg in all three periods (1–5 wk, 5–10 wk and 10–17 wk of age). Hen productivity was determined from 17 to 46 wk and GIT and body traits were measured at 46 wk of age. The characteristics of the rearing phase diets did not affect any of the production, GIT traits or body measurements studied during the laying phase, except body weight gain (BWG) from 17 to 46 wk of age that was higher ($P < 0.01$) in hens that were fed mash during the rearing phase than in hens that were fed crumbles. An increase in the energy content of the laying phase diet from 11.10 to 11.52 MJ AME_n/kg reduced feed intake ($P < 0.001$) and improved feed conversion ratio ($P < 0.001$) from 17 to 46 wk and reduced gizzard contents ($P < 0.01$) at 46 wk of age but did not affect any of the other production trait studied. The characteristics of the rearing and laying hen diets did not affect any of the GIT traits or body measurements at 46 wk of age. Body weight of the hens was positively ($P < 0.001$) related with body length, body mass index, tarsus length and tarsus diameter at this age. The data indicate that neither feed form nor energy concentration of the rearing phase diets affected subsequent hen performance. An increase in the AME_n content of the layer diet from 11.10 to 11.52 MJ/kg decreased feed intake and improved feed conversion ratio but did not affect hen production or egg quality.

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Abbreviations: AME_n, apparent metabolisable energy, corrected for nitrogen; ADFI, average daily feed intake; BMI, body mass index; BW, body weight; BWG, body weight gain; FCR, feed conversion ratio; GIT, gastrointestinal tract; HE, high energy; LE, low energy; LNA, linoleic acid; ME, medium energy.

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1. Introduction

Light hens produce less eggs at the start of the laying period and lay smaller eggs during the laying cycle than heavy hens (Leeson et al., 1997; Pérez-Bonilla et al., 2012a), resulting in a reduction in flock profitability (Pérez-Bonilla et al., 2012b). Two potential nutritional strategies used for increasing the initial body weight (BW) of the hens at the onset of egg production are the feeding of crumbled diets (Frikha et al., 2009a; Gous and Morris, 2001; Saldaña et al., 2015a) and the use of high energy diets (Frikha et al., 2009b; Saldaña et al., 2015b) during the rearing phase. However, Nir et al. (1994) and Serrano et al. (2013) in broilers and Guzmán et al. (2015a) and Saldaña et al. (2015a, b) in pullets, have shown that feeding crumbles or pellets reduced the relative weight (g/kg BW) of the gizzard and the development of the gastrointestinal tract (GIT) as compared with feeding mash. Also, when the energy concentration of the diet increases, fiber content decreases, which also reduces GIT development (González-Alvarado et al., 2007; Jiménez-Moreno et al., 2009a; Sacranie et al., 2012). A poorly developed GIT affects feed intake and might result in pullets not consuming enough energy to meet their requirements for optimal egg production during the first weeks of the laying phase (Pérez-Bonilla et al., 2012b; Guzmán et al., 2016). Consequently, the effects of greater BW but reduced GIT capacity at 17 wk of age, because of feeding a high energy diet in crumble form during the rearing phase, might counteract each other and the final effect on egg production and egg size might depend on other nutritional and management practices.

Laying hens eat to satisfy their energy requirements and therefore, voluntary feed intake decreases as the energy content of the diet increases (Leeson et al., 1997). However, low energy diets might result in hens consuming less feed than required to maximize egg production, especially in light birds at the onset of egg production (Pérez-Bonilla et al., 2012a). Moreover, high energy diets contain usually increased amounts of supplemental fat which may result in improved feed efficiency and an increase in egg size (Mateos and Sell, 1981; Grobas et al., 1999a).

Body measurements, such as body length, body mass index (BMI; g BW/body length²) and tarsus length and diameter, are useful criteria to predict body size and future production of laying hens (Ojedapo et al., 2012; Guzmán et al., 2016). However, the information available on the effects of the characteristics of the rearing and laying hen diets on these variables are limited.

The hypothesis of this research was that pullets fed high energy diets in crumble form during the rearing phase could have greater BW but less developed GIT at the start of the laying phase than pullets fed low energy diets in mash form, which could result in similar productivity during the laying phase. Also, feeding low energy diets during the laying phase could result in reduced egg mass production and egg weight, effects that could be more pronounced in those hens that were lighter or had a poor developed GIT at the onset of the egg production cycle. This research evaluated the effects of feed form and energy concentration of the rearing phase diets on performance, egg quality, digestive tract traits and body measurements of hens fed diets differing in energy concentration from 17 to 46 wk of age.

2. Materials and methods

The procedures described in this research were approved by the Animal Ethics Committee of the Universidad Politécnica de Madrid and were in compliance with the Spanish guidelines for the care and use of animals in research (Boletín Oficial del Estado, 2007).

2.1. Husbandry, diets and experiment design

Details on bird management and feeding program from hatching to 17 wk of age have been reported by Saldaña et al. (2015b). Briefly, one-day-old Lohmann Brown Classic pullets were placed in an environmentally controlled barn and allotted in groups of 50 into 36 cages with similar initial average BW. The feeding program consisted of three periods (0–5 wk, 5–10 wk and 10–17 wk of age) in which the energy concentrations of the diets varied in 0.42 MJ/kg (from 11.93 to 12.77, 11.30 to 12.14 and 11.10 to 11.94 MJ AME_n/kg for the three feeding periods, respectively). During the rearing phase, the treatments consisted in six diets arranged as a 2 × 3 factorial with two feed forms (crumbles vs. mash) and three energy concentrations (LE, low energy; ME, medium energy; HE, high energy). Each treatment was replicated six times. All diets had similar amino acids and nutrient content per unit of energy. The ingredient composition and the calculated and determined nutritive value of the rearing phase diets are shown in Table 1.

At 17 wk of age, pullets from each of the six previous rearing treatments (LE, ME and HE in mash or crumble form) were pooled and then, distributed in groups of nine birds into ten enriched cages (40 cm × 80 cm × 68 cm; Faccio S.A., Padova, Italy). Half of the cages of each of the six groups received a low energy layer diet (11.10 MJ AME_n/kg) and the other half a high energy layer diet (11.52 MJ AME_n/kg). The two layer diets had the same nutritive value per unit of energy (FEDNA, 2010). Feed in mash form and water were provided for ad libitum consumption. The light program consisted of 15 h for the first week of the experiment and 16 h thereafter. Room temperature was recorded daily throughout the experiment with a maximum of 27 ± 3 °C (July, second period of the experiment) and a minimum of 23 ± 3 °C (January, last period of the experiment). The experiment lasted for seven periods of four week each, with the exception of the first period that lasted five wk (17–22 wk). The ingredient composition and the calculated and determined analyses of the laying phase diets are shown in Table 2.

Table 1

Ingredient composition and calculated and determined analyses (g/kg as fed basis) of the rearing phase diets.

Ingredient	0 to 5 week			5 to 10 week			10 to 17 week		
	LE ^a	ME ^b	HE ^c	LE	ME	HE	LE	ME	HE
Maize	350	350	350	350	350	350	200	200	200
Wheat	192	174	157	–	50	100	315	357	400
Barley	–	–	–	280	233	185	100	103	105
Soybean meal, 455 g CP/kg	333	367	401	220	265	310	90	130	170
Sunflower meal, 320 g CP/kg	–	–	–	100	50	–	–	–	–
Sunflower meal, 280 g CP/kg	60	30	–	–	–	–	150	115	80
Bran	–	–	–	–	–	–	100	50	–
Soy oil	27.1	40.8	53.7	10	15.0	20	8.5	10.3	12
Dicalcium phosphate	20.7	20.7	20.6	15.3	15.7	16	15	15.1	15.3
Calcium carbonate	10.4	10.5	10.6	16.4	14.0	11.6	14.4	12.9	11.6
Sodium chloride	3.5	3.5	3.5	3.5	3.4	3.3	3.5	3.5	3.5
DL-methionine, 990 g/kg	1.3	1.5	1.6	1.1	1.3	1.5	0.5	0.6	0.6
L-threonine, 980 g/kg	–	–	–	0.8	0.7	0.6	–	–	–
L-lysine-HCl, 780 g/kg	–	–	–	0.9	0.5	–	1.1	0.6	–
Premix ^d	2.0	2.0	2.0	2.0	2.0	2.0	2.0	2.0	2.0
Calculated analyses ^e									
AME _n (MJ/kg)	11.93	12.35	12.77	11.30	11.72	12.14	11.10	11.52	11.94
Neutral detergent fiber	111	100	89	136	120	104	180	155	130
Crude protein	217	222	227	190	194	199	161	167	172
Digestible amino acid									
Lys	9.7	10.2	10.8	8.4	8.7	9.0	6.1	6.3	6.5
Met + Cys	7.2	7.4	7.6	6.5	6.7	6.9	5.3	5.5	5.6
Thr	6.8	7.1	7.3	6.7	6.8	6.8	4.5	4.8	5.0
Calcium	11.0	11.0	11.0	10.3	9.4	8.5	10.7	10.2	9.7
Digestible phosphorus	4.4	4.3	4.3	4.3	4.4	4.4	3.7	3.6	3.5
Determined analyses ^f									
Dry matter	911	907	909	904	911	919	914	917	913
GE (MJ/kg)	17.42	17.51	17.94	16.59	16.73	16.93	16.81	16.57	16.75
Ether extract	56	64	75	35	42	46	30	30	31
Crude fiber	41	38	32	62	59	54	66	58	52
Crude protein ^g	213	225	235	183	182	186	161	164	164
Lys	11.3	–	12.1	9.9	–	10.5	6.9	–	7.6
Met + Cys	8.4	–	9.0	7.3	–	7.9	6.4	–	6.5
Thr	8.3	–	8.7	7.7	–	8.1	5.2	–	6.0
Total ash	59	–	59	50	–	62	57	–	53

^a 11.93, 11.30 and 11.10 MJ AME_n/kg from 0 to 5 wk, 5 to 10 wk and 10 to 17 wk of age, respectively.^b 12.35, 11.72 and 11.52 MJ AME_n/kg from 0 to 5 wk, 5 to 10 wk and 10 to 17 wk of age, respectively.^c 12.77, 12.14 and 11.94 MJ AME_n/kg from 0 to 5 wk, 5 to 10 wk and 10 to 17 wk of age, respectively.^d Supplied per kilogram of diet: vitamin A (trans-retinyl acetate), 10,000 IU; vitamin D₃ (cholecalciferol), 3500 IU; vitamin E (all-*rac*-tocopherol-acetate), 35 mg; vitamin B₁, 2 mg; vitamin B₂, 8 mg; vitamin B₆, 4 mg; vitamin B₁₂ (cyanocobalamin), 0.025 mg; vitamin K₃ (bisulphatemenadione complex), 3 mg; choline (choline chloride), 270 mg; nicotinic acid, 60 mg; pantothenic acid (D-calcium pantothenate), 15 mg; folic acid, 1.5 mg; D-biotin, 0.15 mg; zinc (ZnO), 90 mg; manganese (MnO), 75 mg; iron (FeCO₃), 60 mg; copper (CuSO₄•5H₂O), 8 mg; iodine (KI), 2 mg; selenium (Na₂SeO₃), 0.3 mg; Roxazyme, 200 mg [1600 U of *endo*-1,4-β-glucanase (EC 3.2.1.4), 3600 U of *endo*-1,3 (4)-β-glucanase (EC 3.2.1.6) and 5200 U of *endo*-1,4-β-xylanase (EC 3.2.1.8)] supplied by DSM S.A., Madrid, Spain; and Natuphos 5000 [300 FTU/kg 6-phytase (EC 3.1.3.26), 60 mg, supplied by Basf Española S.A, Tarragona, Spain].^e According to FEDNA (2010).^f Data correspond to the average of the mash and crumble diets. The differences in values between the mash and crumble diets were within acceptable ranges.^g Total amino acids was not determined for the ME diets.

The experiment was conducted as a completely randomized design with 12 treatments in a factorial arrangement with two feed forms (mash vs. crumbles) and three energy concentrations (LE, ME and HE) during the rearing phase and two diets that differed in AME_n content (low and high) during the laying phase. Each of the 12 treatments was replicated five times and the experimental unit was an enriched cage with nine hens.

2.2. Measurements

Feed disappearance and egg production by replicate were recorded by period as well as for the entire experiment. Hens were weighed by replicate at the beginning of the experiment and at the end of each of the seven experimental periods. Any mortality was recorded and weighed as produced. Egg production data are presented as number of eggs per hen per day. Egg size was estimated by weighing all eggs produced the last two days of each week. The average value of the four weeks (or five wk for the first period) was used to estimate egg weight by period and used for further analysis. From these data, average daily feed intake (ADFI), egg production, egg weight, egg mass, feed conversion ratio (FCR) per kilogram of eggs and per dozen of eggs and BW gain (BWG) were calculated by period and for the entire experiment.

Table 2

Ingredient composition and calculated and determined analyses (g/kg as fed basis) of the laying phase diets.

	11.10 MJ/kg	11.52 MJ/kg
Ingredient		
Wheat	400	400
Maize	200	170
Soybean meal, 470 g CP/kg	171	180
Sunflower meal, 340 g CP/kg	103	110
Sunflower oil soapstocks	14.5	33.9
Dicalcium phosphate	10.8	11.8
Calcium carbonate	82.4	85.0
Sodium chloride	2.8	3.0
DL-methionine, 990 g/kg	1.1	1.3
Sepiolite ^a	9.4	–
Vitamin and mineral premix ^b	5.0	5.0
Calculated analysis ^c		
DM	894	898
AME _n , MJ/kg	11.10	11.52
CP	174	180
Crude fiber	44	46
Neutral detergent fiber	111	114
Digestible amino acid		
Lys	6.8	7.1
Met	3.7	4.0
Met + Cys	6.3	6.6
Thr	5.3	5.5
Trp	1.8	1.9
Calcium	37.5	38.5
Total phosphorus	6.1	6.4
Digestible phosphorus	3.5	3.7
Determined analyses ^d		
Gross energy MJ/kg	16.11	16.21
DM	900	895
CP	182	185
Total amino acid		
Lys	8.2	8.4
Met	4.1	4.3
Met + Cys	7.4	7.5
Thr	6.6	6.7
Trp	2.1	2.2
Ether extract	54	68
Linoleic acid	19	27
Ash	130	133
GMD ± GSD ^e	1112 ± 2.06	1132 ± 2.10

^a A complex magnesium silicate clay incorporated as an inert material.^b Provided the following (per kilogram of diet): vitamin A (*trans*-retinyl acetate), 10,000 IU; vitamin D3 (cholecalciferol), 3750 IU; vitamin E (dl- α -tocopheryl acetate), 10 mg; vitamin B1, 1.3 mg; vitamin B2, 5 mg; vitamin B6, 2 mg; vitamin B12 (cyanocobalamin), 13 mg; niacin, 25 mg; pantothenic acid (d-calcium pantothenate), 10 mg; folic acid, 1 mg; biotin, 13 mcg; choline (choline chloride), 250 mg; manganese (MnO), 88 mg; zinc (ZnO), 63 mg; iron (FeSO₄·H₂O), 38 mg; copper (CuSO₄·5H₂O), 8 mg; iodine [Ca(IO₃)₂], 0.7 mg; selenium (Na₂SeO₃), 0.3 mg; Roxazyme, 200 mg [1600 U of *endo*-1,4- β -glucanase (EC 3.2.1.4), 3600 U of *endo*-1,3 (4)- β -glucanase (EC 3.2.1.6) and 5200 U of *endo*-1,4- β -xylanase (EC 3.2.1.8)] supplied by DSM S.A., Madrid, Spain; Natuphos 5000 (300 FTU/kg supplied by BASF Española, S.A., Tarragona, Spain), 60 mg.^c According to FEDNA (2010).^d Determined analyses were analyzed in duplicate except geometric mean diameter that was determined in triplicate.^e Geometric mean diameter ± Log normal SD.

All eggs produced throughout the experiment were classified and the number of dirty, broken and shell-less eggs, was recorded by replicate. An egg was considered as dirty when a spot of any kind or size was detected on the shell. Other egg quality traits, including yolk and shell color, Haugh units, shell strength and shell thickness, were measured in 12 eggs collected randomly from each replicate the last two days of each of the seven experimental periods. Haugh units and yolk color (Roche color fan) were measured in fresh eggs using a multitester equipment (QCMSys, Technical Services and Supplies, Dunnington, York, UK) as indicated by Pérez-Bonilla et al. (2012a). Shell color was evaluated using a Minolta colorimeter (Chroma Meter Model CR-200, Minolta Corp., Ramsey, NJ) and the Hunter color values [L* (lightness), a* (green to red) and b* (blue to yellow)] were recorded. Egg shell strength, expressed in g/cm², was evaluated using a press meter (Egg Force Reader, SANVO Technology A/S, Odense, Denmark) as indicated by Safaa et al. (2008). Shell thickness was measured at the two pole ends and at the middle section of the egg shell with a digital micrometer (model IT-014UT, Mitotuyo, Kawasaki, Japan) and the average of the three measurements of each of the 12 eggs was used for further analyses.

At 46 wk of age, after the corresponding hen production control, two hens per replicate were randomly selected, weighed individually and euthanized by CO₂ inhalation. The full digestive tract, from the post-crop esophagus to the cloaca, including digesta content, liver, pancreas and spleen, was removed and weighed. Then, the full proventriculus and gizzard, and the liver were excised and weighed. In addition, the gizzard was emptied from any digest content, cleaned, dried with desiccant paper and weighed again. The weight of the organs was expressed relative (g/kg) to live BW and the weight of the gizzard contents was expressed as a percentage of the full organ weight. Gizzard pH was measured in situ in duplicate in all these hens using a digital pH meter fitted with a fine tip glass electrode (model 507, Crison Instruments S.A., Barcelona, Spain) as indicated by Jiménez-Moreno et al. (2009b). The length of the duodenum (from the gizzard to the pancreo-biliary ducts), jejunum (from the pancreo-biliary ducts to the Meckel's diverticulum), ileum (from the Meckel's diverticulum to the ileo-cecal valve) and of the two ceca (from the ostium to the tip of the right and left ceca) was also measured. The length of the small intestine was determined by adding the length of the duodenum, jejunum and ileum. In addition, the length of the hens from the tip of the beak to the end of the longest phalanx was measured on a glass surface at 17 and 46 wk of age in extended birds, using a flexible tape with a precision of one mm. From these measurements, BMI was estimated by dividing the BW (g) of the hen by the corporal length² (cm) as indicated by Mendes et al. (2007). Also, the length and diameter of the tarsus of these hens were measured with the aid of a digital caliper and expressed as cm per kg of BW. The average value of the traits measured in the two hens was used for further statistical analysis.

2.3. Laboratory analysis

Representative samples of the laying hen diets were ground in a laboratory mill (Retsch Model Z-I, Stuttgart, Germany) equipped with a one-mm screen and analyzed for moisture by the oven-drying (method 930.15), total ash in a muffle furnace (method 942.05) and nitrogen by Dumas (method 968.06) using a Leco analyzer (Model FP-528, Leco Corp., St. Joseph, MI) as indicated by AOAC International (2005). Ether extract was determined after 3N HCl acid hydrolysis (Boletín Oficial del Estado, 1995) and gross energy was determined using an adiabatic bomb calorimeter (model 1356, Parr Instrument Company, Moline, IL, USA). The amino acid content was analyzed by chromatography (Hewlett-Packard 1100, Waldbronn, Germany) in samples ground at 0.5 mm as described by De Coca-Sinova et al. (2008) and the linoleic acid (LNA) content as indicated by Grobas et al. (1999a). Particle size distribution and mean particle size of the diets, expressed as geometric mean diameter, were determined in a 100 g sample using a shaker (Retsch, Stuttgart, Germany) provided with eight sieves ranging in mesh from 5000 to 40 µm as indicated by the ASAE (1995).

2.4. Statistical analysis

The normal distribution of residuals and variance homogeneity of the data were tested by the UNIVARIATE procedure and the Levene's Test, respectively (SAS Institute, 2004). The effects of dietary treatment (feed form and AMEn content of the rearing phase diet and AMEn content of the laying phase diet) and their interactions on hen production were analyzed as repeated measures using the MIXED. Data on GIT development and body measurements were analyzed using the GLM procedure of SAS (SAS Institute, 2004). When significant differences among treatments were detected, means were separated using the Tukey test. In addition, the Pearson correlation analyses (SAS Institute, 2004) was used to study the relation between BW of the hens and body length, BMI and tarsus length and diameter at 46 wk of age. Differences were considered significant at $P < 0.05$.

3. Results

3.1. Hen performance and egg quality

Details on pullet performance from hatching to 17 wk of age have been reported by Saldaña et al. (2015b). Briefly, feeding crumbles increased ADFI (52.9 vs. 49.7 g; $P < 0.001$) and BWG (12.7 vs. 11.6 g; $P < 0.001$) and improved FCR (4.18 vs. 4.27; $P < 0.001$) of the pullets. Consequently, BW at 17 wk of age was higher in pullets that were fed crumbles than in pullets that were fed mash (1554 vs. 1432 g; $P < 0.001$). An increase in the energy content of the rearing diets decreased ADFI linearly ($P < 0.001$) and improved FCR quadratically ($P < 0.01$) but energy intake (KJ AME_n/d) was not affected.

Mortality during the laying phase was 1.8% and was not related to treatment (data not shown). No interactions among main effects on hen productivity were detected and therefore, only main effects are presented (Table 3). From 17–46 wk of age, type of diet fed during the rearing phase did not affect any of the productive traits studied, except BWG that was higher (386 vs. 282 g; $P < 0.001$) for hens that were fed mash during the rearing phase than for hens that were fed crumbles (Figs. 1 and 2). Most of the differences in BWG between the two groups of hens occurred from 17 to 22 wk of age and in fact, no significant differences were detected after this age. As a result, BW of the hens at 46 wk of age was similar for all hens, irrespective of the feed form or energy concentration of the rearing phase diets.

An increase in the energy content of the laying phase diets reduced ADFI (113 vs. 109 g; $P < 0.001$) and improved FCR per kg (1.98 vs. 1.92; $P < 0.001$) and per dozen (1.44 vs. 1.41; $P < 0.005$) of eggs but did not affect any of the other traits studied. The effects of energy concentration of the diet on hen productivity by period are shown in Fig. 3. Egg quality (percentage of

Table 3

Effects of feed form and energy concentration of the rearing diets and energy concentration (AME_n/kg) of the laying hen diet on hen production from 17 to 46 wk of age.

	Egg production ^c	ADFI (g/hen)	Egg weight (g)	Egg mass (g/d)	FCR		BW (g)		BW gain (g)
					(kg:kg)	(kg:dozen)	17 wk	46 wk	
Feed form, rearing phase									
Mash	0.936	111	61.0	57.0	1.95	1.43	1432 ^b	1816	386 ^a
Crumbles	0.930	110	60.9	56.6	1.95	1.42	1554 ^a	1836	282 ^b
Energy level, rearing phase									
LE ^d	0.942	111	61.1	57.2	1.94	1.42	1486	1833	347
ME ^e	0.927	111	60.8	56.3	1.96	1.43	1487	1802	316
HE ^f	0.930	110	61.0	56.7	1.96	1.43	1506	1842	340
Energy level, laying phase									
Low ^g	0.938	113 ^a	60.9	57.0	1.98 ^a	1.44 ^a	1494	1815	321
High ^h	0.928	109 ^b	61.0	56.6	1.92 ^b	1.41 ^b	1492	1837	347
SD ⁱ	0.0298	3.07	0.95	1.55	0.047	0.047	37.6	60.4	58.5
P-value ^j									
Rearing phase									
Feed form	0.380	0.423	0.738	0.325	0.721	0.840	0.001	0.210	0.002
Energy level	0.296	0.613	0.743	0.109	0.199	0.532	0.174	0.104	0.338
Laying phase									
Energy level	0.198	0.001	0.797	0.476	0.001	0.005	0.848	0.174	0.102

^a Means with different superscript within each main effect are significantly different ($P < 0.05$).

^b Means with different superscript within each main effect are significantly different ($P < 0.05$).

^c Eggs/hen per day.

^d 11.93, 11.30 and 11.10 MJ AME_n/kg from 0 to 5 wk, 5 to 10 wk and 10 to 17 wk of age, respectively.

^e 12.35, 11.72 and 11.52 MJ AME_n/kg from 0 to 5 wk, 5 to 10 wk and 10 to 17 wk of age, respectively.

^f 12.77, 12.14 and 11.94 MJ AME_n/kg from 0 to 5 wk, 5 to 10 wk and 10 to 17 wk of age, respectively.

^g 11.10 MJ AME_n/kg.

^h 11.52 MJ AME_n/kg.

ⁱ 30 replicates for feed form in the rearing phase, 20 replicates for energy level in the rearing phase and 30 replicates for energy level in the laying phase.

^j The interactions among main effects were not significant ($P > 0.05$).

dirty eggs, Haugh units, yolk color, shell color, shell strength and shell thickness) was not affected by the type of diet used during the rearing or the laying phases (Table 4).

3.2. Digestive traits and body measurements

Details on the effects of the characteristics of the rearing phase diet on GIT traits and body measurements of the pullets have been reported elsewhere (Saldaña et al., 2015b). Briefly, the relative weight of the GIT and the gizzard and gizzard digesta content was lower ($P < 0.05$ to $P < 0.001$) and gizzard pH was higher ($P < 0.05$ to $P < 0.001$) in pullets fed crumbles than in pullets fed mash at all ages (5, 10 and 17 wk of age). Body and tarsus length was shorter ($P < 0.01$) at all ages in pullets fed crumbles than in pullets fed mash. Energy concentration of the diet did not affect any of the GIT traits or body measurements evaluated.

During the laying period, no interactions between the characteristics of the rearing phase diets and the energy content of the laying phase diets were detected for any of the traits studied and therefore, only main effects are presented (Tables 5 and 6). At 46 wk of age, gizzard content was higher in hens fed the low energy layer diet than in hens fed the high energy layer diet (281 vs. 261 g/kg; $P < 0.01$). The RW of the full proventriculus, full and empty gizzard and the liver, as well as the pH of the gizzard contents and the length of the small intestine and cecum, were not affected by the energy content of the layer diet. Similarly, body length, BMI and tarsus length and diameter of the hens were not affected by dietary treatment (Table 7). Hen BW showed a positive correlation ($P < 0.001$) with body length, BMI and tarsus length and diameter (Table 8).

4. Discussion

4.1. Hen performance

4.1.1. Rearing phase diets

In general, pullets that are heavier at 17 wk of age anticipate sexual maturity and egg production and lay larger eggs than pullets that are lighter (Renema et al., 1999a; Pérez-Bonilla et al., 2012b). In this respect, Summers et al. (1987) and Renema et al. (1999b) reported a positive relation between BW of the hens and the maturation of the reproductive traits and the onset of egg production. In the current research, hens that received crumbles during the rearing phase were heavier at 17 wk of age and started egg production sooner (data not shown) and laid more eggs (0.913 vs. 0.898) that were heavier (56.2 vs. 55.4 g) from 17 to 22 wk of age than hens that were fed mash, although the differences did not reach significance. Moreover,

Table 4Effects of feed form and energy concentration of the rearing diets and energy concentration (AME_n/kg) of the laying hen diets on egg quality from 17 to 46 wk of age.

	Dirty eggs(%)	Broken(%)	Shell-less(%)	Haugh units	Shell color ^a			Yolk color ^b	Shell strength (g/cm ²)	Shell thickness (mm)
					L	a	b			
Feed form, rearing phase										
Mash	1.47	0.51	0.22	92.1	61.2	15.5	32.1	10.3	4939	0.38
Crumbles	1.29	0.46	0.18	92.0	61.2	15.5	32.1	10.2	4898	0.38
Energy level, rearing phase										
LE ^c	1.19	0.49	0.18	91.8	61.2	15.6	32.3	10.3	4992	0.38
ME ^d	1.46	0.54	0.17	92.1	61.5	15.3	31.9	10.3	4846	0.38
HE ^e	1.47	0.46	0.19	92.3	61.1	15.6	32.4	10.3	4916	0.38
Energy level, laying phase										
Low ^f	1.39	0.44	0.17	92.1	61.0	15.6	32.1	10.3	4915	0.38
High ^g	1.36	0.52	0.23	91.9	61.4	15.4	32.1	10.3	4925	0.38
SD ^h	0.528	0.109	0.028	0.93	0.86	0.90	0.52	0.41	216.2	0.007
P-value ⁱ										
Rearing phase										
Feed form	0.272	0.414	0.347	0.833	0.896	0.695	0.590	0.322	0.560	0.543
Energy level	0.193	0.291	0.100	0.158	0.249	0.265	0.124	0.793	0.170	0.399
Laying phase										
Energy level	0.876	0.175	0.144	0.629	0.112	0.151	0.688	0.794	0.867	0.320

^a Hunter color values, L* (lightness), a* (green to red) and b* (blue to yellow).^b Measured with roche color fan.^c 11.93, 11.30 and 11.10 MJ AME_n/kg from 0 to 5 wk, 5 to 10 wk and 10 to 17 wk of age, respectively.^d 12.35, 11.72 and 11.52 MJ AME_n/kg from 0 to 5 wk, 5 to 10 wk and 10 to 17 wk of age, respectively.^e 12.77, 12.14 and 11.94 MJ AME_n/kg from 0 to 5 wk, 5 to 10 wk and 10 to 17 wk of age, respectively.^f 11.10 MJ AME_n/kg.^g 11.52 MJ AME_n/kg.^h 30 replicates for feed form in the rearing phase, 20 replicates for energy level in the rearing phase and 30 replicates for energy level in the laying phase.ⁱ The interactions among main effects were not significant (P > 0.05).**Table 5**

Effects of feed form and energy concentration of the rearing diets and energy concentration of the laying hen diet on the relative weight (g/kg BW) of the full gastrointestinal tract (GIT) of organs of the hens at 46 wk of age.

	BW (g)	GIT (g/kg BW)	Liver (g/kg BW)	Proventriculus (g/kg BW)	Gizzard			
					Full (g/kg BW)	Empty (g/kg BW)	Content (g/kg organ W)	pH
Feed form, rearing phase								
Mash	1838	121	25.9	5.0	25.8	18.8	272	3.96
Crumbles	1858	119	25.1	5.3	25.1	18.1	269	3.88
Energy level, rearing phase								
LE ^c	1850	120	25.2	4.9	25.5	18.3	273	3.85
ME ^d	1844	120	25.8	5.4	25.4	18.7	274	3.91
HE ^e	1838	121	25.5	5.1	25.4	18.6	267	4.01
Energy level, laying phase								
Low ^f	1849	120	25.7	5.1	25.6	18.3	281 ^a	3.97
High ^g	1848	120	25.3	5.1	25.3	18.7	261 ^b	3.88
SD ^h	79.4	04.5	2.91	0.67	2.24	1.79	14.3	0.335
P-value ⁱ								
Rearing phase								
Feed form		0.234	0.307	0.157	0.995	0.885	0.780	0.429
Energy level		0.965	0.812	0.180	0.109	0.156	0.825	0.370
Laying phase								
Energy level		0.545	0.636	0.962	0.724	0.313	0.026	0.311

^a Means with different superscript within each main effect are significantly different (P < 0.05).^b Means with different superscript within each main effect are significantly different (P < 0.05).^c 11.93, 11.30 and 11.10 MJ AME_n/kg from 0 to 5 wk, 5 to 10 wk and 10 to 17 wk of age, respectively.^d 12.35, 11.72 and 11.52 MJ AME_n/kg from 0 to 5 wk, 5 to 10 wk and 10 to 17 wk of age, respectively.^e 12.77, 12.14 and 11.94 MJ AME_n/kg from 0 to 5 wk, 5 to 10 wk and 10 to 17 wk of age, respectively.^f 11.10 MJ AME_n/kg.^g 11.52 MJ AME_n/kg.^h 30 replicates for feed form in the rearing phase, 20 replicates for energy level in the rearing phase and 30 replicates for energy level in the laying phase.ⁱ The interactions among main effects were not significant (P > 0.05).

Fig. 1. Effect of feed form of the rearing diets on egg production (A), feed intake (B), egg weight (C), feed conversion ratio (D) and BW gain (BWG) (E) of the hens from 17 to 46 wk of age.

NS $P > 0.05$; *** $P < 0.005$.

The effect of the period was significant ($P < 0.005$) in all the variables studied.

no differences in egg production or egg weight among treatments were observed after this age. On the other hand, hens fed mash during the rearing phase were lighter at 17 wk of age than hens fed crumbles but they gained weight more rapidly and in fact, no differences in BW were detected at 22 wk of age. Consequently, the benefits of feeding crumbles to pullets on subsequent egg production might not be as relevant as expected and in most cases the potential benefits might be limited to the first few weeks of the laying period.

The authors have not found any published research on the effects of energy concentration of the pullet diets on subsequent hen performance. Usually, an increase in dietary energy results in heavier pullets that produce more egg mass and lay heavier eggs during the laying period (Pérez-Bonilla et al., 2012a). In the current research, BW at 17 wk of age was greater in pullets that were fed crumbles during the rearing phase than in pullets that were fed mash (1554 vs. 1432 g) but after 22 wk of age, BW was not affected by the energy content of the rearing phase diets. The results reported herein indicate that a variation in the energy concentration of the rearing diets of ± 0.42 MJ AME_h/kg did not have any effect on subsequent hen performance.

Fig. 2. Effect of energy content of the rearing diets^a on egg production (A), feed intake (B), egg weight (C), feed conversion ratio (D) and BW gain (BWG) (E) of the hens from 17 to 46 wk of age.

^aLow: 11.93, 11.30 and 11.10 MJ AME_n/kg from 0 to 5 wk, 5 to 10 wk and 10 to 17 wk of age, respectively; medium: 12.35, 11.72 and 11.52 MJ AME_n/kg from 0 to 5 wk, 5 to 10 wk and 10 to 17 wk of age, respectively; high: 12.77, 12.14 and 11.94 MJ AME_n/kg from 0 to 5 wk, 5 to 10 wk and 10 to 17 wk of age, respectively.

^{NS} P > 0.05.

The effect of the period was significant (P < 0.005) in all the variables studied.

The data suggest that nutritionists might modify the energy level of the rearing phase diets, within the indicated limits, according to the relative prices of available ingredients.

4.1.2. Laying phase diets

From 17–46 wk of age, hens fed the high energy diet consumed 3.7% less feed and had 3.1% better FCR than hens fed the low energy diet but egg production was not affected, consistent with data of Grobas et al. (2001) and Safaa et al. (2009). The decrease in ADFI observed in hens fed the high energy diet was expected because poultry eat to satisfy their energy requirements, provided that the capacity of the GIT is not compromised (Hill et al., 1956; Leeson et al., 1997). Egg production and egg size were not affected by the energy content of the diet, consistent with data of Grobas et al. (1999b) in brown hens

Fig. 3. Effect of energy concentration (AME_n/kg) on of the laying hen diet^a on egg production (A), feed intake (B), egg weight (C), feed conversion ratio (D) and BW gain (BWG) (E) from 17 to 46 wk of age.

^aLow 11.10 MJ AME_n/kg ; high: 11.52 MJ AME_n/kg .

NS $P > 0.05$; * $P < 0.05$; ** $P < 0.01$; *** $P < 0.005$.

The effect of the period was significant ($P < 0.005$) in all the variables studied.

fed diets varying in AME_n from 11.22 to 11.76 MJ. In contrast, Mathlouthi et al. (2002) reported higher egg mass production in hens fed diets with 11.52 MJ AME_n/kg than in hens fed diets with 11.10 MJ AME_n/kg . Similarly, Pérez-Bonilla et al. (2012a) reported higher egg production with increases in the AME_n content of the diet from 11.10 to 11.94 MJ/kg. The reason for the discrepancy among researches on the effects of energy concentration of the diet on egg production and egg mass, is not known but might be related to the strain of hens used as well as on the nutritional characteristics of the experimental diets. For example, in the experiment of Mathlouthi et al. (2002) Single Comb White Leghorn hens were used and most of the difference in egg mass reported was caused by the lower weight of the eggs produced by hens fed the low energy diet (53.7 vs. 56.0 g). The authors did not give any reason for the difference in egg size observed but the LNA content of the low and high energy diets used was 10.7 and 12.7 g/kg, respectively. The NRC (1994) recommended a minimum daily intake of 1.20 g LNA

Table 6

Effects of feed form and energy concentration of the rearing diets and energy concentration (AME_n/kg) of the laying hen diet on the relative length (cm/kg BW^a) of the gastrointestinal tract at 46 wk of age.

	Duodenum	Jejunum	Ileum	SI ^b	Cecum
Feed form, rearing phase					
Mash	14.0	41.8	38.4	94.2	24.4
Crumbles	13.8	42.0	37.8	93.6	23.8
Energy level, rearing phase					
LE ^c	13.7	41.1	38.0	92.7	23.5
ME ^d	14.5	42.8	38.5	95.8	24.5
HE ^e	13.7	41.7	37.9	93.3	24.3
Energy level, laying phase					
Low ^f	14.1	41.8	37.8	93.7	23.8
High ^g	13.8	42.0	38.4	94.2	24.4
SD ^h	1.79	3.42	2.68	6.69	2.91
P-value ⁱ					
Rearing phase					
Feed form	0.654	0.822	0.405	0.729	0.408
Energy level	0.201	0.285	0.734	0.290	0.530
Laying phase					
Energy level	0.489	0.810	0.399	0.776	0.455

^a The BW of the hens is shown in Table 5.

^b Small intestine (duodenum, jejunum and ileum).

^c 11.93, 11.30 and 11.10 MJ AME_n/kg from 0 to 5 wk, 5 to 10 wk and 10 to 17 wk of age, respectively.

^d 12.35, 11.72 and 11.52 MJ AME_n/kg from 0 to 5 wk, 5 to 10 wk and 10 to 17 wk of age, respectively.

^e 12.77, 12.14 and 11.94 MJ AME_n/kg from 0 to 5 wk, 5 to 10 wk and 10 to 17 wk of age, respectively.

^f 11.10 MJ AME_n/kg.

^g 11.52 MJ AME_n/kg.

^h 30 replicates for feed form in the rearing phase, 20 replicates for energy level in the rearing phase and 30 replicates for energy level in the laying phase.

ⁱ The interactions among main effects were not significant ($P > 0.05$).

Table 7

Effects of feed form and energy content of the rearing diets and energy concentration (AME_n/kg) of the laying hen diet on body measurements at 46 wk of age.

	Hen length(cm/BW ^a)	BMI ^b (g/cm ²)	Tarsus (cm/BW)	
			Length	Diameter
Feed form, rearing phase				
Mash	36.0	0.43	5.54	0.71
Crumbles	35.6	0.44	5.46	0.72
Energy level, rearing phase				
LE ^c	35.7	0.45	5.46	0.71
ME ^d	35.8	0.43	5.46	0.71
HE ^e	36.0	0.43	5.57	0.72
Energy level, laying phase				
Low ^f	35.8	0.44	5.53	0.71
High ^g	35.8	0.43	5.47	0.72
SD ^h	1.68	0.022	0.291	0.022
P-value ⁱ				
Rearing phase				
Feed form	0.347	0.193	0.353	0.339
Energy level	0.822	0.928	0.361	0.301
Laying phase				
Energy level	0.976	0.384	0.360	0.309

^a The BW of the hens is shown in Table 5.

^b Body mass index.

^c 11.93, 11.30 and 11.10 MJ AME_n/kg from 0 to 5 wk, 5 to 10 wk and 10 to 17 wk of age, respectively.

^d 12.35, 11.72 and 11.52 MJ AME_n/kg from 0 to 5 wk, 5 to 10 wk and 10 to 17 wk of age, respectively.

^e 12.77, 12.14 and 11.94 MJ AME_n/kg from 0 to 5 wk, 5 to 10 wk and 10 to 17 wk of age, respectively.

^f 11.10 MJ AME_n/kg.

^g 11.52 MJ AME_n/kg.

^h 30 replicates for feed form in the rearing phase, 20 replicates for energy level in the rearing phase and 30 replicates for energy level in the laying phase.

ⁱ The interactions among main effects were not significant ($P > 0.05$).

per day to maximize egg size in brown-egg laying hens. Probably, the low LNA intake was limiting egg mass production in hens fed the low energy diet, penalizing feed efficiency. In contrast, in the current research LNA intake was 2.94 and 2.15 g/d for the high and low energy diets, respectively, values that were above requirements for both groups of hens (Grobas et al., 2001; Bouvarel et al., 2010). Also, in the research of Pérez-Bonilla et al. (2012a), in which egg production increased with increases in the energy content of the diet, an increase in energy concentration of 7.5% (11.10–11.94 MJ AME_n/kg) decreased

Table 8Correlations between full BW^a and body length, tarsus length and diameter and body mass index (BMI)^b of the hens at 46 wk of age.

	BW			
	Body length (cm)	BMI (g/cm ²)	Tarsus(cm)	
			Length	Diameter
R	0.394	0.637	0.414	0.351
P-value	<0.001	<0.001	<0.001	<0.001

^a The BW of the hens is shown in Table 5.^b Body length, BMI and tarsus length and diameter of the hens are shown in Table 7.

ADFI by 3.5%, which resulted in a 3.9% increase in energy intake and in a subsequent improvement in egg production. In the current experiment, however, an increase of 0.42 MJ AME_n/kg diet (a 3.8% increase) resulted in a similar increase in energy intake and consequently, no changes in hen performance between the two groups of hens were expected.

An increase in dietary energy is generally attained by increasing the amount of supplemental fat. Mateos and Sell (1980, 1981) showed that supplemental fat reduced the rate of passage of the digesta, facilitating the contact between nutrients and enzymes and improving the utilization of the non-fiber components of the diet. Moreover, when the lipid content of the diet increases, the level of LNA content also increases. Both effects, reduced rate of passage and increased LNA content, might result in an increase in egg size and an improvement in FCR (Grobas et al., 1999c). In the current experiment, the low and high energy diets contained 54 and 68 g/kg ether extract and 19 and 27 g/kg LNA, respectively. Probably, the small difference in ether extract content of the 2 diets did not result in any visible effect on egg weight. On the other hand, the LNA content of the low energy diet (19 g/kg) met already the requirement of the hens to maximize egg weight (Grobas et al., 1999a; Safaa et al., 2009). The data suggest that laying hens, in the range of energy concentration studied (11.10–11.52 MJ AME_n/kg), and provided that the requirements in other essential nutrients (indispensable amino acids, LNA and fat content) are satisfied, regulate ADFI precisely according to energy requirements.

4.2. Egg quality

Egg quality was not affected by feed form of the rearing phase diets or by the energy concentration of the rearing and laying phase diets. The authors have not found any published research on the effects of the characteristics of the rearing diets on egg quality to compare with the results reported herein. In regard to dietary energy, Grobas et al. (1999b) and Pérez-Bonilla et al. (2012a) observed similar egg quality (dirty, broken, shell-less, Haugh units, yolk pigmentation and yolk to albumen ratio) for hens fed diets containing 11.10 or 11.52 MJ AME_n/kg, in agreement with the results reported herein. However, Zimmermann and Andrews (1987) and Wu et al. (2005) observed a reduction in albumen quality and Junqueira et al. (2006) a decrease in the percentage of shell-less eggs with increases in the energy concentration of the diet. Factors such as strain and age of the hens, as well as the ingredient composition of the diet, might affect the response of egg quality traits to variations in energy content of the diet. For example, Zimmermann and Andrews (1987) and Junqueira et al. (2006) used Leghorn hens in the second cycle of egg production and Wu et al. (2005) reported that the decrease in AME_n of the diet reduced yolk size, which in turn could affect egg size and other egg quality traits.

4.3. Digestive traits and body measurements

The interest on the effects of the ingredient composition and physico-chemical characteristics of the diets on the anatomy and physiology of the GIT in poultry has increased in the last decade but most available data have been obtained in broilers (Mateos et al., 2002, 2012; Serrano et al., 2012; Jiménez-Moreno et al., 2016) and pullets (Saldaña et al., 2015a), with little information in laying hens. In fact, we have not found any published report on the effects of feed form and energy content of the rearing phase diets on digestive tract traits of the hens at the end of the laying period. In the current research, neither feed form nor energy content of the rearing phase diets affected the development and function of the different organs of the GIT of the hens at 46 wk of age. Moreover, the energy content of the layer diet had little effect on the development of the GIT at this age. In fact, the only difference observed was for gizzard digesta content that was greater in hens fed the low energy diet. In this respect, feed intake was higher in hens fed the low energy diet than in hens fed the high energy diets and consequently, an increase in the digesta content of the gizzard was expected. In addition, fiber content was higher in the low- than in the high energy diet, which may result in an increase in digesta content, as has been demonstrated in broilers by González-Alvarado et al. (2008). The results show that feed form and energy concentration of the rearing phase diets and energy concentration of the laying phase diets, had little effect on subsequent hen performance or on the characteristics of the digestive tract at the end of the laying cycle.

Body length, BMI and tarsus measurements are used to evaluate body composition and size and to predict future performance of broilers (Mendes et al., 2007; Van Roovert-Reijrink, 2013), pullets (Itza et al., 2011; Guzmán et al., 2015b) and laying hens (Ojedapo et al., 2012; Guzmán et al., 2016). Body length, BMI and length and diameter of the tarsus were not affected by dietary treatment, indicating that neither the characteristics of the rearing phase diets nor the energy content

of the laying phase diets affected body size of the mature hens. However, a significant relationship between BW of the hens and body length, BMI and tarsus length and diameter was detected, consistent with the results of Guzmán et al. (2015b).

5. Conclusions

Feed form and energy concentration of the rearing phase diets had little effect on the performance and egg quality of the hens during the entire laying cycle or on GIT traits and morphometric measurements of the hens at 46 wk of age. An increase in the energy content of the laying phase diets from 11.10 to 11.52 MJ/kg decreased ADFI and improved FCR proportionally to the increase in energy concentration but did not affect any of the other traits studied. Consequently, nutritionists may adapt the characteristics of the rearing and laying phase diets, within the range of values studied, to the cost of available ingredients, without any detectable effect on hen productivity or egg quality.

Conflict of interest statement

The authors confirm they are not conflict of interest in this research.

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