METABOLISM AND NUTRITION

Feeding broiler breeders a reduced balanced protein diet during the rearing and laying period impairs reproductive performance but enhances broiler offspring performance

J. Lesuisse,* C. Li,* S. Schallier,* J. Leblois,† N. Everaert,† and J. Buyse*,[1]

*Laboratory of Livestock Physiology, Department of Biosystems, KU Leuven, Heverlee, Belgium; and †Precision Livestock and Nutrition Unit, TERRA Teaching and Research Centre, University of Liège, Gembloux, Belgium

ABSTRACT Mammalian studies have shown that nutritional constraints during the perinatal period are able to program the progeny (metabolism, performance). The presented research aimed to investigate if broiler breeders and their offspring performance could be influenced by reducing the dietary crude protein (CP) level with 25%. A total of 160 day-old pure line A breeder females were randomly divided over 2 dietary treatments. The control group was fed commercial diets, whereas the reduced balanced protein (RP) breeders received an isoenergetic diet that was decreased with 25% in dietary CP and amino acid during their entire lifespan. The RP birds required an increased feed allowance, varying between 3 and 15%, to meet the same BW goals as their control fed counterparts. The difference in feed allocations and reduction of the dietary CP level resulted in a net protein reduction varying between 14 and 23%. At wk 27 and 40, the body composition of the breeders was changed as a result of the dietary treatment. At both ages, the proportional abdominal fat pad weight of the RP breeders was increased ($P < 0.001$), whereas the proportional breast muscle weight was only higher at wk 27 in the control group compared to the RP group ($P < 0.001$). Egg weight ($P < 0.001$) and egg production ($P < 0.001$) was decreased for the RP fed birds. The lower dietary CP level reduced the proportional albumen weight of the RP eggs ($P = 0.006$). Male offspring from RP breeders were characterized by an increase in BW from 28 d until 35 d of age ($P = 0.015$). Moreover, female progeny of RP breeders showed a reduced FCR ($P = 0.025$), whereas male progeny showed a tendency ($P = 0.052$) towards a lower FCR at 5 wk of age. In conclusion, lowering dietary CP levels in rearing and laying phase of breeders had a negative effect on breeder performance but enhanced live performance of the offspring.

Key words: broiler breeder, offspring, nutrition, low protein, developmental programming

2017 Poultry Science 96:3949–3959
http://dx.doi.org/10.3382/ps/pex211

INTRODUCTION

The importance of crude protein (**CP**) levels in broiler breeder diets is well known and has been an interesting topic of research for the last decades. Both, albumen removal from the eggs prior to incubation (Everaert et al., 2013) and maternal protein reduction (Lopez and Leeson, 1995a; Hocking et al., 2002; van Emous et al., 2013; Moraes et al., 2014), were thoroughly investigated in the past. A decrease in the dietary CP level in poultry production could have a positive impact on the economic and environmental costs of poultry meat production. Broiler production has evolved tremendously over the past 30 years and the selection towards a higher proportional breast muscle weight resulted in an altered body composition of broilers and breeders. The growth curve of female breeders altered from a convex to a concave shaped curve. This resulted in a lower feed allowance in the beginning of the rearing period, but a higher feed allocation towards the start of the laying phase. This concave growth curve assures a more uniform maturation of the flock (Renema et al., 2007). Consequently, breeder production and the feed requirements of both breeder and broiler generations are changed as well. Not only breeding companies, but also the feed industry made tremendous progress in the last decades, resulting in a broiler that is more efficient and grows 4.6 times the rate of a 1957 random-bred strain (Havenstein et al., 2003a,b; Thiruvenkadan et al., 2011; Zuidhof et al., 2014). The interest in low protein and diluted diets is increasing again (Enting et al., 2007; Zhu et al., 2012; Moraes et al., 2014; van Emous et al., 2015a,b). By reducing the CP and/or energy content of the broiler breeder diets, daily feed allocation can be increased, which could

Received March 16, 2017.
Accepted June 21, 2017.
[1]Corresponding author: johan.buyse@kuleuven.be

enhance the welfare of the breeder generation. In addition, maternal dietary treatments have been shown to program the offspring of avian species, with the implication of epigenetic effects (Berghof et al., 2013; Frésard et al., 2013). The study by van Emous et al. (2015a) reduced the dietary CP levels during the rearing period, resulting in an increase of the proportional breast muscle (**BM**) of the male offspring. Moreover, Rao et al. (2009) reduced the dietary CP level during the laying phase and showed an altered gene expression in the yolk-sac membrane, hypothalamus, and muscle of developing chicken embryos. Furthermore, the effect of a reduced albumen amount available for the embryo has been shown to program the chicken early in life with long-lasting effects (Everaert et al., 2013; Willems et al., 2016). Therefore, the presented study investigated the effects of a lower dietary CP level in both rearing and laying phase of breeders. It is hypothesized that reduced dietary CP levels during the entire lifespan of breeder hens could induce maternal programming effects on the broiler offspring towards a better protein efficiency.

MATERIALS AND METHODS

Ethics

The present research was approved by the ethical commission for experimental use of animals of the KU Leuven under accession number P187/2013.

Birds and Management Breeder Trial

A total of 160 1-day-old Pure Line A female breeders were wing tagged and randomly distributed over 10 floor pens (1.5 × 2 m), resulting in 16 birds per pen in the rearing phase and 12 birds per pen in laying phase. Forty 1-day-old male breeders of the same Pure Line A were allotted in 2 pens and kept separate from the females. Pure line A is a pure broiler breeder male line of a commercial breeding company at the A position of a 4-way cross that makes up a commercial broiler. These pens were all located in the same poultry house and the floor was covered with wood shavings. The females were divided into 2 groups: a control group (**C**), fed with standard commercial breeder diets (Aviagen, 2013) and a reduced balanced protein group (**RP**), fed with diets wherein the dietary protein and amino acid level was reduced by 25% during rearing and laying phase (Table 1). This experimental setup was conducted with 5 replicates per treatment. In the first 3 wk, all birds received the same starter feed (Research Diet Service, Wijk bij Duurstede, The Netherlands), which was fed ad libitum from 1 to 14 d. From 15 to 21 d all birds were fed a controlled amount of the starter 1 diet. After 21 d of age the dietary treatment started and the RP group received a feed with a 25% reduction in dietary crude protein and amino acids throughout rearing and laying phase (Table 1). Starter 2 was provided from 22 to 42 d, the grower diet was administered from 43 to 105 d, the pre-breeder diet from 106 to 154 d and breeder diet from 155 d until 40 wk of age. During rearing, the feed allocations were adapted weekly to maintain a similar BW for both groups as all breeders were kept on the same target BW curve recommended in the breeder management guide for a Ross 308 parent stock (Aviagen, 2013). BW and egg production were the directives for calculating the daily feed allocation during the laying phase. For each feeding phase, diets were formulated to be isoenergetically and composed of the same raw materials. Fat percentage was similar in the C and RP diets of each phase, whereas protein was isoenergetically substituted by carbohydrates. The main focus of this research was to evaluate the effects of a RP diet on the progeny and to a lesser extent on the egg production.

The male breeders were not subjected to any dietary treatment and were fed according to the breeder management guide for a Ross 308 parent stock (Aviagen, 2013). The breeders were artificially inseminated from wk 25 onwards. At the age of 19 wk, 12 males were selected and transferred to individual cages (60 × 40 × 60 cm). Their semen was pooled and diluted following the protocol of Sexton (1977), after which 100 μL of the diluted semen was injected once a wk to the vagina of the female breeders. The progeny of these breeders was used for the offspring experiment.

Light and temperature schedules were applied according to the management guidelines for a Ross 308 parent stock (Aviagen, 2013). The photoperiod of the pullets during the first 2 d was 23L:1D and was gradually reduced to 8L:16D at d 10 of age. At 21 wk of age, the breeders were photostimulated and light period was increased to 11L:13D. The photoperiod was further increased by 1 h extra light per wk until 13L:11D was reached at 23 wk of age. During the laying phase, light was on between 0400 h and 1700 h. Feed was supplied daily at 0900 h. Water was available ad libitum during light hours through 1 bell-type drinker per pen. The birds were vaccinated according to the standard commercial vaccination procedures (Galluvet, Lummen, Belgium).

Birds and Management Offspring Trial

In wk 30 and 31, a total of 350 fertile eggs of the control and RP groups were collected. All eggs were incubated in a forced draft incubator (PAS Reform incubator, Zeddam, The Netherlands) for 21 d following the standard incubation conditions, i.e., dry bulb temperature of 37.6°C, a wet bulb temperature of 29.0°C and were turned every hour over an angle of 90°. On embryonic d 18, all eggs were candled and those with signs of viable embryos were transferred from the setter trays to individual hatching baskets. At the end of the incubation period (21.5 d), unhatched eggs were opened

Table 1. Composition and calculated contents of the rearing and laying phase diets (g/kg).

Item	Starter 1 (0 to 21 d)	Starter 2 (22 to 42 d)		Grower (43 to 105 d)		Pre-breeder (106 to 154 d)		Breeder (From 155 d)	
		C[1]	RP[2]	C	RP	C	RP	C	RP
Ingredient									
Wheat	151.6	219.1	293.8	185.7	246.1	194.5	262.4	198.9	268.8
Maize	450.0	400.0	400.0	400.0	400.0	450.0	450.0	450.0	450.0
Soybean meal	215.0	156.0	44.5	107.0	21.0	105.0	7.0	150.0	47.0
Sunflower meal	50.0	50.0	50.0	30.0	30.0	75.0	75.0	40.0	40.0
Wheat middlings	50.0	70.0	70.0	150.0	150.0	100.0	100.0	30.0	30.0
Reapseed meal	20.0	30.0	30.0	-	-	-	-	-	-
Vegetable fat	15.0	20.0	21.5	14.0	14.0	10.0	10.0	15.2	15.2
Cellulose	-	10.0	43.0	35.0	45.0	5.00	16.5	5.00	17.0
Diamol	-	-	-	35.0	45.0	5.00	16.5	5.00	17.0
Animal lard	-	-	-	-	-	7.50	7.50	10.0	10.0
Chalk	15.5	15.5	16.0	14.0	14.6	25.0	25.0	24.5	24.5
Limestone	-	-	-	-	-	-	-	49.0	49.0
Monocalcium phosphate	11.5	12.5	13.0	11.5	11.8	5.50	6.50	6.00	7.00
Sodium carbonate	3.70	2.50	3.20	2.60	3.00	2.80	3.40	2.20	2.80
Potassium carbonate	-	-	-	1.00	5.00	-	4.80	-	5.00
Salt	1.80	2.40	2.00	2.40	2.10	2.20	1.80	2.50	2.10
L-Lysine	2.70	0.60	1.80	0.70	1.60	0.95	2.15	0.35	1.65
DL-Methionine	2.20	1.00	0.60	0.75	0.30	1.15	0.80	1.10	0.70
L-Threonine	0.90	0.30	0.50	0.20	0.40	0.25	0.45	0.20	0.50
L-Tryptophan	-	-	-	-	-	-	-	-	0.15
L-Isoleucine	-	-	-	-	-	-	-	-	1.15
L-Arginine	-	-	-	-	-	-	-	-	0.35
Natuphos 5000G	0.10	0.10	0.10	0.10	0.10	0.10	0.10	0.10	0.10
Premix	10.0	10.0	10.0	10.0	10.0	10.0	10.0	10.0	10.0
Analyzed content[4]									
CP	218.6	195.0	143.0	156.5	117.6	162.0	126.5	161.6	115.5
Calculated content[5]									
AME_n (kcal/kg)	2801	2798	2800	2605	2604	2787	2785	2791	2792
DM	878.4	879	882.0	883.9	886.5	879.5	882.5	886.0	889.0
Crude ash	58.6	57.7	52.7	87.8	95.6	63.0	72.1	108.9	118.3
Crude fat	41.8	45.6	45.8	39.7	38.8	44.3	43.3	49.9	48.7
Crude fibre	41.8	52.7	82.0	72.0	79.9	52.2	61.3	38.6	48.0
Energy: protein ratio	12.8	14.3	19.6	16.6	22.1	17.2	22.0	17.3	24.2
Starch	399.6	409.4	446.8	400.5	430.9	428.1	462.2	418.0	453.1
Sugars	36.7	34.5	26.2	30.3	23.9	30.0	22.7	28.5	20.8
Calcium	10.0	10.0	10.0	9.00	9.00	12.1	12.0	30.1	30.0
Total phosphorus	6.60	6.90	6.50	6.60	6.30	5.30	5.10	4.60	4.4
Available phosphorus	4.30	4.50	4.50	4.20	4.20	2.90	3.00	2.90	3.0
Digestible lysine	0.95	0.66	0.48	0.51	0.38	0.55	0.41	0.56	0.41
Digestible leucine	1.28	1.13	0.83	0.93	0.70	1.00	0.74	1.04	0.77
Digestible M + C	0.73	0.57	0.43	0.46	0.34	0.53	0.41	0.52	0.38
Digestible arginine	1.08	0.94	0.61	0.74	0.49	0.80	0.52	0.81	0.55
Digestible threonine	0.60	0.47	0.34	0.36	0.26	0.40	0.28	0.41	0.30
Digestible tryptophan	0.17	0.15	0.10	0.12	0.08	0.12	0.09	0.13	0.10

[1]Control group: standard breeder diets.

[2]RP group: Reduced balanced protein diet, 25% reduction in dietary crude protein and amino acids.

[3]Provided per kg diet: Vitamin A 10,000 IU; Vitamin D_3 3,500 IU; Vitamin E 100 mg; Vitamin K_3 3.0 mg; Vitamin B_1 3.0 mg; Vitamin B_2 6.0 mg; Vitamin B_6 3.0 mg; Vitamin B_{12} 20 μg; Niacinamide 35 mg; D-pantothenic acid 15 mg; Choline chloride 600 mg; Folic acid 1.5 mg; Biotin 0.15 mg; Iron 40 mg; Copper 16 mg; Manganese 120 mg; Zinc 110 mg; Iodine 1.25 mg; Selenium 0.3 mg.

[4]Based on 2 analyses in duplicate per diet.

[5]Calculated according to CVB, 2012.

to determine the stage of embryonic mortality (early, mid and late death). After cloacal sexing, male and female progeny were randomly allocated to 16 floor pens (1.0 × 0.75 m), with either 10 male or female chicks per pen, resulting in 4 replicates per group per gender. The floor was covered with wood shavings as bedding material. The light and temperature schedules were followed according to the management guide of Ross 308 broilers (Aviagen, 2014). During the first wk, the photoperiod was 23L:1D, from 1 wk of age onwards broilers were kept on a 14L:4D:2L:1D:2L:1D lighting program. Both the progeny from the control and RP group were fed a standard 3 phased broiler feed optimized following the recommendation of CVB (2012) (Table 2) (Research diet service, Wijk bij Duurstede, The Netherlands). The starter diet was supplied from 1 to 10 d, a grower diet from 11 to 25 d and a finisher diet from 26 to 35 d. All birds were wing tagged for individual weekly weighing.

Data Collection Breeder Trial

Body Weight. During both rearing and laying phase, individual BW was monitored weekly for all birds. The weighing was performed before feeding time and daily feed allocations for C and RP group were determined based on this weekly weighing data.

Table 2. Composition and calculated contents of the starter, grower and finisher broiler diets (g/kg).

Item	Starter (0 to 10 d)	Grower (11 to 25 d)	Finisher (26 to 35 d)
Ingredient			
Wheat	192.4	340.6	500.0
Maize	400.0	250.0	95.1
Soybean meal	290.0	270.0	265.0
Reapseed meal	20.0	30.0	30.0
Potato protein	20.0	10.0	-
Vegetable fat	33.0	29.0	31.0
Animal lard	-	30.0	40.0
Chalk	14.5	13.0	12.6
Monocalcium phosphate	11.0	8.50	6.50
Sodium carbonate	3.00	2.20	2.20
Salt	2.10	2.10	2.00
L-Lysine	1.50	1.80	2.20
DL- Methionine	2.15	2.05	2.10
L-threonine	0.25	0.50	0.80
L-valine	-	0.15	0.45
Natuphos 5000G	0.10	0.10	0.10
Premix[1]	10.0	10.0	10.0
Analyzed content[2]			
CP	222.6	212.6	201.8
Calculated content[3]			
AME_n (kcal/kg)	2843	2954	3000
DM	880.3	881.9	881.9
Crude ash	56.5	52.1	49.9
Crude fat	57.9	79.4	87.1
Crude fiber	26.9	27.5	27.7
Starch	385.2	378.6	375.8
Sugars	38.9	40.1	42.1
Calcium	9.5	8.50	8.00
Total phosphorus	5.9	5.50	5.10
Available phosphorus	4.2	3.70	3.40
Digestible lysine	1.10	1.04	1.01
Digestible leucine	1.54	1.34	1.27
Digestible M + C	0.80	0.76	0.75
Digestible arginine	1.25	1.18	1.15
Digestible threonine	0.68	0.65	0.64
Digestible tryptophan	0.21	0.20	0.20

[1]Provided per kg diet: Vitamin A 10,000 IU; Vitamin D_3 2,500 IU; Vitamin E 50 mg; Vitamin K_3 1.5 mg; Vitamin B_1 2.0 mg; Vitamin B_2 7.5 mg; Vitamin B_6 3.5 mg; Vitamin B_{12} 20 μg; Niacinamide 35 mg; D-pantothenic acid 12 mg; Choline chloride 460 mg; Folic acid 1.0 mg; Biotin 0.2 mg; Iron 80 mg; Copper 12 mg; Manganese 85 mg; Zinc 60 mg; Iodine 0.8 mg; Selenium 0.15 mg.

[2]Based on 2 analyses in duplicate per diet.

[3]Calculated acoording to CVB, 2012.

Body Composition. At 27 and 40 wk of age, 10 birds representing the average weight per treatment were sacrificed for sampling. The birds were sacrificed by electrical stunning, following to the EU regulations, prior to decapitation. The BM (both the pectoralis major and minor of the left side), abdominal fat pad (**AF**), liver, pancreas, heart, oviduct, ovaria, stroma and the largest follicle were dissected and weighed. Additionally, the number of small (between 5 and 10 mm) and large (>10 mm) yellow follicles was recorded. The proportional tissue weights were calculated by dividing the absolute tissue weight by the live weight of the bird.

Egg Production. During the laying period, from 24 to 39 wk of age, eggs were collected daily and weighed individually. The eggs were classified into first and second grade eggs, second grade eggs contained double-yolked eggs or shell abnormalities and were not used for calculation of laying rate. Laying rate in the first 2 wk of production was calculated from 24 to 25 wk of age. Breeder FCR was calculated as the total feed intake (rearing and production phase) divided by the total egg mass from 24 to 39 wk of age (kg feed/kg egg mass).

Egg Composition and Quality. At wk 26, 32, and 39, egg composition and egg quality were determined on 20 eggs per treatment the day after collection. The yolk, albumen, and egg shell of the eggs were separated and weighed. Proportional weights were calculated relative to egg weight. The shell thickness was measured by using a micrometer at 3 places around the equator of the egg. The albumen height measured in Haugh units (**HU**) (Eisen et al., 1962) was determined at the center of the thick albumen using a standard tripod micrometer (Futura-Werner Fürste, Löhne, Germany). Haugh units were calculated using the following equation: HU = 100 × log {albumen height − 5.67 × [(30 × egg weight 0.37 − 100)/100] + 1.9}. The color of the yolk was determined by using a Roche Yolk Color Fan scale (Hoffmann-La Roche, Basel, Switzerland), which ranges from 1 (very light yellow) to 15 (dark orange).

Data Collection Offspring Trial

Body Composition. At hatch and d 35, 10 chicks per group (offspring control and RP group) were sacrificed and dissected. The hatchlings were killed by decapitation, whereas 35-day-old broilers were euthanized by electrical stunning, following the EU regulation, prior to decapitation. The 35-day-old broilers used for sampling were selected based on their BW being near the average BW of the representing pen. At hatch, the weight of residual yolk, breast muscle (pectoralis major and minor, both sides), pancreas, liver and heart was recorded. At d 35, the weight of breast muscle (pectoralis major and minor, left side), abdominal fat pad, pancreas, liver and heart were recorded. Proportional weights were calculated by dividing the organ weight by the live weight.

Body Weight, Feed Intake, and Feed Conversion Ratio. All birds were weighed individually at weekly intervals. The feed intake was calculated weekly by weighing the remaining feed and subtracting this from the amount of feed administered. The feed conversion ratio (**FCR**) was calculated weekly and corrected for mortality and difference in BW. The used empirical equation is based the assumption that 1 point of FCR, i.e., 0.01 is equivalent to a 45 g difference in BW. Heavier birds have higher maintenance requirements and therefore have a higher feed intake. This empirical equation thus corrects for differences in BW between groups.

$FCR_{wmc} = FCR_{actual} + [(\text{Target BW—Actual BW})/45*0.01]$

FCR_{wmc} = FCR corrected for BW differences and mortality

FCR_{actual} = BW gain divided by feed intake, corrected for mortality

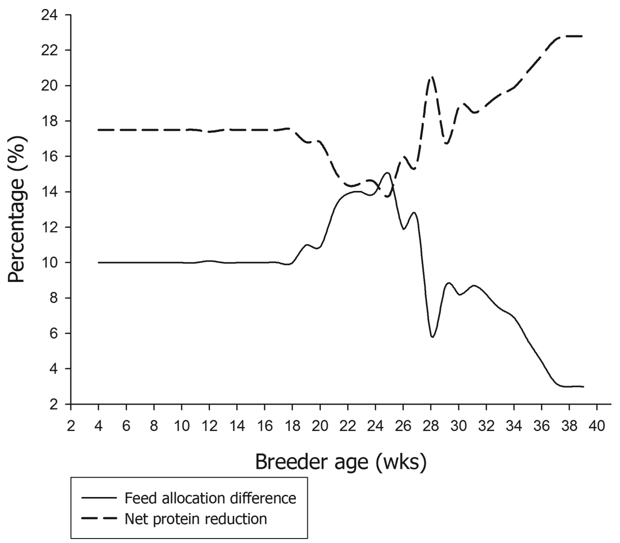

Figure 1. Feed allocation difference and protein reduction between the control and RP group. To maintain the same BW curve different feed allocations between the control and reduced balanced protein group were needed. The protein reduction is the net protein reduction that takes the feed allocation and dietary crude protein level into account.

Statistics

All data regarding the breeder trial were analyzed with the statistical software package SAS University (SAS Institute Inc., Cary, NC). Egg weight, egg production, body and egg composition were analyzed using GLM with treatment, age, and their interaction as classification variables. When there was a significant effect of treatment, or interaction with age, means were further compared by a post-hoc Tukey's test. The zootechnical laying performance of the breeder trial and body composition of the offspring trial were analyzed using one-way analysis of variance (**ANOVA**) with treatment as a classification variable. Average weekly BW, feed intake, and FCR data of the offspring experiment were analyzed using a GLM repeated measurements procedure of JMP pro 12 (SAS Institute Inc., Cary, NC) with age, treatment, and their interactions as classification variables. Data are presented as mean ± SEM. For each result the significance level was set at $P < 0.05$.

RESULTS

Feed, Protein and Energy Intake

The RP birds had on average 10% more feed allocated to maintain a similar BW throughout the entire experiment (Figure 1), with an increased energy intake as a consequence. In all feeding phases, the diets were formulated isoenergetically and therefore the differences in feed allocation amounts corresponds with an extra energy intake of the RP fed breeders. The net protein reduction takes into account the protein reduction of the RP feed and the difference in feed allocation. An overview of the differences in feed allocation and consequently the net protein reduction and extra energy are shown in Figure 1. From wk 3 to 18, the RP birds were fed approximately 10% more to maintain a similar BW as the control birds. This daily feed allocation corresponded with a 17.5% reduction in dietary protein compared to the control group. In the following weeks, RP birds required a higher feed allowance to reach the same BW. The total feed and CP intake of the control and RP breeders at 10, 15, and 22 wk of age is shown in Table 3. During the start of the laying phase, both groups were fed according to their egg production. After peak production, the birds were fed to realize a growth of approximately 30 g per wk. At 37 wk, the feed allocation difference was only 3.0% which resulted in a 22.8% reduced dietary protein level for the reduced balanced protein group.

Table 3. Effects of dietary protein (CP) level on BW, CV, total feed intake[1] and CP intake[1] of breeders fed control or reduced balanced protein (RP) diets at 10, 15, and 22 wk of age.

Group	BW (g)	CV (%)	Feed intake[1] (g)	CP intake[1] (g)
	10 wks of age			
Control	966 ± 20	16.3 ± 1.6	2653	507
RP	976 ± 8	14.2 ± 1.4	2827	417
P-value CP	NS	NS	/	/
	15 wks of age			
Control	1,460 ± 28	18.8 ± 1.8	5131	833
RP	1,446 ± 13	16.4 ± 1.2	5553	698
P-value CP	NS	NS	/	/
	22 wks of age			
Control	2,308 ± 35	15.8 ± 0.8	9240	1497
RP	2,274 ± 16	15.8 ± 0.7	10,123	1226
P-value CP	NS	NS	/	/

[1]Total feed intake and CP intake is calculated from 2 wk of age. Daily feed allocations within a treatment were the same for all pens.

Body Weight and Body Composition

At wk 27 and 40 BW was not affected by the dietary treatment (Table 4). Absolute BM, AF, liver and pancreas weight increased with age. At 27 wk of age the RP breeders had a lower proportional BM (on average 17.0 ± 0.5%) weight compared to the control breeders (on average 21.4 ± 0.7%), whereas at 40 wk the difference was largely diminished ($P < 0.001$). The RP birds had a higher absolute ($P < 0.001$) and proportional ($P < 0.001$) AF weight at wk 27 and 40. Also the proportional liver ($P = 0.002$) and pancreas ($P = 0.042$) weights were affected by the dietary treatment whereas absolute and proportional weights of the heart, reproductive organs, and brain were not altered (data not shown).

Egg Weight and Egg Production

The control birds came into production at 169 ± 3 d of age, whereas RP breeders produced their first egg 5 d later (Table 5). The laying rate in the first 2 wk of the

Table 4. BW, body composition (breast muscle (BM) and abdominal fat (AF) pad) and organ weight (liver and pancreas) of breeders fed control or reduced balanced protein (RP) diets at 27 and 40 wk of age.

		Treatment				
Variable	Age (wk)	Control	RP	$P_{CP\ level}$	P_{age}	$P_{interaction}$
BW (g)	27	3,600 ± 107	3,440 ± 73	NS	<0.001	0.092
	40	4,301 ± 40	4,484 ± 54			
BM weight (g)	27	762[a] ± 40	587[b] ± 26	0.003	0.001	0.013
	40	796[a] ± 32	779[a] ± 17			
Proportional BM weight (%)	27	21.4[a] ± 0.7	17.0[b] ± 0.5	<0.001	NS	0.02
	40	18.7[ab] ± 0.8	17.4[b] ± 0.3			
AF weight (g)	27	78.0[b] ± 7.7	121.0[a] ± 10.4	<0.001	<0.001	NS
	40	149.2[b] ± 9.9	228.3[a] ± 18.0			
Proportional AF weight (%)	27	2.15[b] ± 0.22	3.50[a] ± 0.27	<0.001	<0.001	NS
	40	3.49[b] ± 0.22	5.06[a] ± 0.39			
Liver weight (g)	27	58.8[b] ± 3.2	67.2[a] ± 2.8	0.002	<0.001	NS
	40	77.6[b] ± 5.9	97.9[a] ± 4.5			
Proportional liver weight (%)	27	1.67[b] ± 0.08	1.96[a] ± 0.09	0.002	NS	NS
	40	1.81[b] ± 0.12	2.19[a] ± 0.11			
Pancreas weight (g)	27	5.46 ± 0.22	4.72 ± 0.21	NS	0.032	NS
	40	5.88 ± 0.32	5.58 ± 0.37			
Proportional pancreas weight (%)	27	0.15[a] ± 0.01	0.14[b] ± 0.01	0.042	NS	NS
	40	0.14[a] ± 0.01	0.12[b] ± 0.01			

[a,b]Means within a tissue, treatment mean with different superscripts have significantly treatment effects ($P < 0.05$).

Table 5. Effect of dietary CP level of breeders fed control or reduced balanced protein (RP) diets during rearing and laying phase on age of first egg, laying rate in the first 2 wks of production (24 to 25 wks of age), total number of eggs per hen (from 24 to 39 wk of age), egg mass (from 24 to 39 wk of age), breeder FCR (from 24 to 39 wk of age), percentage of second grade eggs (from 24 to 39 wk of age) and on fertility and hatchability of eggs collected at 30 and 31 wk of age.

	Treatment		
Zootechnical parameter	Control	RP	$P_{CP\ level}$
Age first egg (d)	169 ± 3	174 ± 2	NS
Laying rate first 2 wks (%)	13.6[a] ± 2.7	5.1[b] ± 1.4	0.022
Total number of eggs per hen	69.3[a] ± 3.5	55.2[b] ± 2.5	0.011
Egg mass (g)	34.2[a] ± 1.5	25.1[b] ± 1.3	0.002
FCR (kg feed/kg egg mass)	52.0[a] ± 2.3	75.3[b] ± 4.1	0.001
Second grade eggs (%)	1.34 ± 0.20	1.93 ± 0.54	NS
Fertility (%)	84.9 ± 1.6	70.4 ± 4.3	0.004
HOF[1] (%)	78.4 ± 1.4	77.2 ± 2.2	NS

[a,b]Means within a zootechnical parameter, treatment mean with different superscripts have significantly treatment effects ($P < 0.05$).
[1]Hatchability calculated on amount of fertile eggs (HOF).

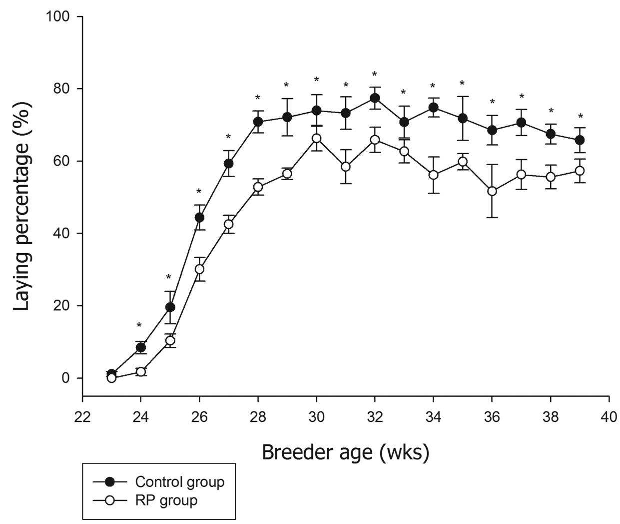

Figure 2. The daily egg production of the control and reduced balanced protein breeders summarized in a weekly overview from wk 23 to 39 (N = 5). Asterisks indicate significant differences ($P < 0.05$) between the treatments at particular age.

control breeders was higher in comparison with their RP fed counterparts ($P = 0.022$). Although they differed at the beginning of the laying phase, both groups reached their peak production in wk 32. The RP breeders were characterized by a decreased laying performance from 24 to 39 wk of age (Figure 2). The average peak production was 77 ± 3% for the control breeders, whereas the RP group reached a laying rate of 66 ± 3%. At the end of the experiment (wk 39) the control and RP birds had an egg production of respectively 66 ± 3% and 58 ± 3%. No differences between the control and RP breeders in the percentage of second-grade eggs was observed. The RP fed breeders were characterized by a decreased fertility, but no difference was found on hatchability (Table 5). The egg weight of birds receiving RP diets was lower ($P < 0.001$) than the control eggs during the whole laying phase (Figure 3). The combined effect of a reduced egg mass output and a higher daily feed intake in both rearing and laying phase resulted in an increased breeder FCR (kg feed/kg egg mass) for the RP fed breeders (Table 5).

Egg Composition

The effects of a reduced dietary CP level on egg composition and egg quality traits at 26, 32, and 39 wk of age is shown in Table 6. The egg weight of the RP breeders was lower ($P < 0.001$) at wk 26, 32, and 39, an

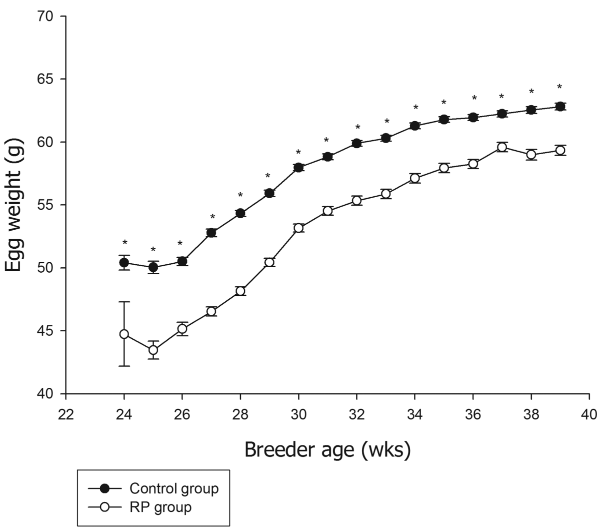

Figure 3. The average egg weight of the control and reduced balanced protein (RP) breeders summarized in a weekly overview from wk 24 to 39 of age. Asterisks indicate significant differences ($P < 0.05$) between the treatments at particular age.

effect that was consistently observed during the whole laying period (Figure 3). Both the absolute ($P < 0.001$) and proportional ($P = 0.006$) albumen weight of eggs were affected by age in an opposite way but were both decreased as a result of reducing the dietary protein level. The proportional albumen weights at peak production, namely at 32 wk of age, was 57.9 ± 0.4% and 56.3 ± 0.5% for respectively the control and RP group. The eggs from the control group had a higher absolute yolk weight ($P < 0.001$), while no effects were observed in the proportional yolk weight. No effects of the dietary treatment on the yolk color (data not shown) and albumen height were observed. The proportional egg shell weight decreased with breeder age and was higher ($P = 0.046$) for breeders receiving the reduced balanced protein diet.

Offspring Performance

Body Weight. The male and female BW trajectories from hatch until wk 5 are presented in Table 7. Male offspring results showed an interaction effect between the dietary CP level and age ($P = 0.002$).

Table 6. Egg composition and egg quality traits of breeders fed control or reduced balanced protein (RP) diets at wk 26, 32 and 39 of age.

		Treatment				
Variable	Age (wks)	Controls	RP	$P_{CP\ level}$	P_{age}	$P_{interaction}$
Egg weight (g)[1]	26	50.1[a] ± 0.3	45.1[b] ± 0.5	<0.001	<0.001	NS
	32	59.9[a] ± 0.2	55.3[b] ± 0.4			
	39	62.8[a] ± 0.3	59.3[b] ± 0.4			
Albumen weight (g)	26	29.8[a] ± 0.5	25.0[b] ± 0.6	<0.001	<0.001	NS
	32	33.7[a] ± 0.7	30.1[b] ± 0.6			
	39	35.3[a] ± 0.6	32.2[b] ± 0.6			
Proportional albumen weight (%)	26	58.8[a] ± 0.4	57.9[b] ± 0.5	0.006	<0.001	NS
	32	57.9[a] ± 0.4	56.3[b] ± 0.4			
	39	55.7[a] ± 0.3	55.3[b] ± 0.5			
Yolk weight (g)	26	13.6[a] ± 0.2	11.5[b] ± 0.4	<0.001	<0.001	NS
	32	17.1[a] ± 0.3	16.3[b] ± 0.3			
	39	20.2[a] ± 0.6	18.6[b] ± 0.4			
Proportional yolk weight (%)	26	26.7 ± 0.3	26.6[z] ± 0.9	NS	<0.001	NS
	32	29.4 ± 0.4	30.5[y] ± 0.3			
	39	31.9 ± 0.3	32.0[x] ± 0.5			
Yolk: albumen (%)	26	45.6 ± 0.8	46.1 ± 1.7	NS	<0.001	NS
	32	51.0 ± 1.0	54.3 ± 0.9			
	39	57.4 ± 0.8	58.1 ± 1.3			
Shell weight (g)	26	7.19[a] ± 0.11	6.79[b] ± 0.13	0.011	0.010	NS
	32	7.37[a] ± 0.13	6.98[b] ± 0.09			
	39	7.86[a] ± 0.12	7.35[b] ± 0.16			
Proportional shell weight (%)	26	14.2[b] ± 0.3	15.8[a] ± 0.3	0.046	<0.001	NS
	32	12.7[b] ± 0.2	13.1[a] ± 0.1			
	39	12.4[b] ± 0.2	12.7[a] ± 0.3			
Shell thickness (mm)	26	0.337 ± 0.011	0.342 ± 0.010	NS	NS	NS
	32	0.353 ± 0.006	0.350 ± 0.006			
	39	0.348 ± 0.005	0.337 ± 0.005			
Albumen height (HU)	26	77.5 ± 1.2	74.2 ± 1.8	NS	<0.001	NS
	32	80.3 ± 1.5	78.1 ± 1.7			
	39	83.5 ± 1.2	81.1 ± 1.8			

[a,b]Means within a row, treatment mean with different superscripts have significantly treatment effects ($P < 0.05$).
[1]All eggs of these wk were weighed.
[2](Proportional) albumen, (proportional) yolk, yolk: albumen, (proportional) shell weight, shell thickness and albumen height were measured on 20 eggs with an average egg weight within the corresponding group.

Table 7. BW and FCR corrected for weight differences and mortality (FCR_{wmc}) of male and female progeny of control and reduced balanced protein (RP) breeders at hatch, wk 1, wk 2, wk 3, wk 4 and wk 5 of age.

		Broiler age (wks)					
Maternal treatment	*P*-value	Hatch	1	2	3	4	5
Male offspring							
BW (g)							
Control		$43.5^{a} \pm 0.5$	169 ± 2	496 ± 5	$1,008 \pm 13$	$1,579^{b} \pm 17$	$2,263^{b} \pm 22$
RP		$39.5^{b} \pm 0.7$	161 ± 3	479 ± 8	$1,013 \pm 16$	$1,631^{a} \pm 22$	$2,345^{a} \pm 31$
P-value CP	0.025						
P-value age	<0.001						
P-value CP × age	0.002						
P-value CP per wk		<0.001	NS	NS	NS	0.021	0.015
FCR_{wmc}							
Control			1.118 ± 0.012	1.192 ± 0.013	1.226 ± 0.006	1.337 ± 0.005	1.406 ± 0.007
RP			1.118 ± 0.015	1.185 ± 0.011	1.211 ± 0.004	1.334 ± 0.006	1.364 ± 0.010
P-value CP	NS						
P-value age	<0.001						
P-value CP × age	NS						
Female offspring							
BW (g)							
Control		$42.3^{a} \pm 0.5$	$166^{a} \pm 2$	$465^{a} \pm 6$	$948^{a} \pm 13$	$1458^{a} \pm 16$	$2088^{a} \pm 23$
RP		$38.9^{b} \pm 0.6$	$153^{b} \pm 3$	$445^{b} \pm 7$	$897^{b} \pm 14$	$1430^{b} \pm 19$	$2050^{b} \pm 26$
P-value CP	<0.001						
P-value age	<0.001						
P-value CP × age	NS						
FCR_{wmc}							
Control			$1.151^{b} \pm 0.024$	$1.242^{b} \pm 0.031$	$1.267^{b} \pm 0.013$	$1.373^{b} \pm 0.017$	$1.431^{b} \pm 0.009$
RP			$1.145^{a} \pm 0.014$	$1.211^{a} \pm 0.006$	$1.246^{a} \pm 0.015$	$1.336^{a} \pm 0.018$	$1.402^{a} \pm 0.010$
P-value CP	0.025						
P-value age	<0.001						
P-value CP × age	NS						

[a,b]Means within a zootechnical parameter, treatment mean with different superscripts have significantly effects ($P < 0.05$).

Average hatch weight was lower in both male and female offspring ($P < 0.001$) for chicks originating from the RP breeders. Proportional chick weight was not affected by the dietary treatment. Male progeny of RP hens was on average 4 g lighter at hatch than hatchlings from the control group. At wk 3, BW of both groups was approximately 1,010 ± 11 g, while at wk 5 BW was higher ($P = 0.025$) for the progeny of the RP breeders. At 5 wk of age, the male offspring of RP fed breeders was on average 82 g heavier compared to offspring of control fed breeders. In contrast, female progeny of the RP breeders were characterized by a lower BW from hatch until 5 wk of age ($P < 0.001$) (Table 7). At 5 wk of age, the female progeny originating from control breeders was on average 38 g heavier compared to the female offspring of RP breeders.

Feed Conversion Ratio. The weight and mortality corrected FCR (**FCR_{wmc}**) for males and females (Table 7) increased with age ($P < 0.001$). A tendency towards a lower FCR_{wmc} was observed for male progeny of RP breeders ($P = 0.052$). The FCR_{wmc} at slaughter age was on average 1.40 ± 0.01 and 1.36 ± 0.01 for broilers originating from respectively the control and RP breeders. The female progeny of RP birds had a decreased FCR_{wmc} ($P = 0.025$) compared to the chicks descending from the control breeders (on average 1.40 ± 0.01 and 1.43 ± 0.01 respectively).

Body Composition. Absolute and proportional organ weights for the male and female offspring are shown in Table 8. At 5 wk of age, the proportional BM weight of the male broiler offspring from the RP birds was higher ($P = 0.019$) compared to their control fed counterparts (on average 10.4 ± 0.1% and 11 ± 0.1%). The absolute and proportional AF and liver weights of both

Table 8. Body composition (breast muscle (BM) and abdominal fat (AF) pad) and organ weight (liver and pancreas) of the male and female broiler progeny of breeders fed control or reduced balanced protein (RP) diets at 5 wk of age.

	Breeder treatment		
Variable	Control	RP	$P_{CP\ level}$
Male progeny			
BM[1] (g)	$238^{b} \pm 7$	$264^{a} \pm 8$	0.019
Proportional BM[1] (%)	$10.4^{b} \pm 0.1$	$11.0^{a} \pm 0.1$	0.005
AF (g)	23.1 ± 0.9	23.9 ± 1.7	NS
Proportional AF (%)	1.00 ± 0.03	0.98 ± 0.07	NS
Liver (g)	46.0 ± 2.2	46.9 ± 1.7	NS
Proportional liver (%)	2.00 ± 0.9	1.93 ± 0.6	NS
Pancreas (g)	3.50 ± 0.19	3.46 ± 0.16	NS
Proportional pancreas (%)	0.15 ± 0.01	0.14 ± 0.01	NS
Female progeny			
BM[1] (g)	228 ± 7	230 ± 9	NS
Proportional BM[1] (%)	10.8 ± 0.2	10.9 ± 0.2	NS
AF (g)	25.5 ± 2.1	23.0 ± 1.9	NS
Proportional AF (%)	1.20 ± 0.03	1.10 ± 0.07	NS
Liver (g)	44.2 ± 2.9	43.9 ± 1.8	NS
Proportional liver (%)	2.08 ± 0.10	2.09 ± 0.06	NS
Pancreas (g)	$3.26^{b} \pm 0.16$	$3.77^{a} \pm 0.15$	0.035
Proportional pancreas (%)	$0.15^{b} \pm 0.01$	$0.18^{a} \pm 0.01$	0.049

[a,b]Within a tissue, treatment means with different superscripts are significantly different ($P < 0.05$).

[1]The breast muscle (both the pectoralis major and minor) of the left side.

male and female progeny were not affected by the dietary treatment of the breeder generation. However, the absolute and proportional pancreas weights of the female RP progeny was increased ($P = 0.049$).

DISCUSSION

The present study investigated the effects of a lower dietary CP level in both rearing and laying phase of breeders. The reduced dietary CP levels during the entire lifespan of the breeder hens induced maternal programming effects of the broiler offspring towards an improved zootechnical performance.

Breeder Performance

At 27 wk of age, the RP breeders were characterized by a respectively higher and lower proportional AF and BM weight compared to their control counterparts. These effects of a reduced dietary CP level on the body composition of breeders are in accordance with previous studies (Spratt and Leeson, 1987; Miles et al., 1997; Hudson et al., 2000; van Emous et al., 2013, 2015b). The higher AF in the RP breeders could be explained by the excess of energy consumed by the RP group (De Beer et al., 2007; Mohiti-Asli et al., 2012). The possibly increased hepatic de novo lipogenesis, mediated by the alteration in the energy:protein ratio of (pre-breeder: control diet 17.2 versus 22.0 in the RP diet), resulted in an increased lipid deposition in the abdominal fat, and an increased (proportional) liver weight, which is in accordance with Adams and Davis (2001) and Swennen et al. (2007). At 40 wk of age, the end of the experiment, the (proportional) AF weight was still higher for the RP fed birds, as the energy:protein ratio was 17.3 versus 24.2 for respectively the control and RP diet. At 40 wk of age, the (proportional) BM weight was similar for the control and RP group, as also observed by van Emous et al. (2015b). In contrast to the latter study, the RP birds were fed also a RP diet in the laying period. Hence, these results suggest that the dietary CP content during the rearing phase had a higher impact on the body composition than the CP level in the breeder diet.

Not only body composition, but also egg weight and composition were influenced by the dietary CP level. A lower egg, yolk, albumen and shell weight was observed for breeders that were fed the RP diets. Lopez and Leeson (1995b) also reported a 2 g lower egg weight for breeders fed low protein diets in the laying phase, whereas Spratt and Leeson (1987) found a minor decrease in egg weight from 32 until 35 wk of age as a consequence of a reduced dietary CP in the laying phase, but reported a 0.5 g decrease in egg weight throughout the entire laying period for breeders fed lower energy diets. Hocking et al. (2002) found no effects of a lower dietary CP level during rearing phase on egg weight. However, in contrast with the work of Spratt and Leeson (1987), Lopez and Leeson (1995b) and Hocking et al. (2002), the BW of our C and RP breeders was kept equal. As BW, a potential predominant factor in egg weight, did not vary between C and RP groups, this factor can be excluded in our study. Hence, it is hypothesized that besides BW, also dietary CP level is a major determining factor for egg weight. In addition to a reduced egg weight, our results also showed a decreased proportional albumen and shell weight. Spratt and Leeson (1987) also reported a decreased proportional albumen weight at 32 wk, but not at 28, 36, and 40 wk of age. In contrast, Lopez and Leeson (1995b) and Naber (1979) observed no effect of dietary CP levels on egg composition, which could be explained by the fact that the reduced dietary CP levels were only applied during the rearing phase, whereas in our study the breeders were fed RP diets throughout their entire lifespan. A lower shell weight was reported by Spratt and Leeson (1987) as consequence of an increased energy intake, which is in agreement with our findings.

The lower laying performance by RP breeder was in contrast with the findings of van Emous et al. (2015b), whereby an increased egg production was reported for breeders that were fed a lower dietary CP level during the rearing period. Hocking et al. (2002) reported a decreased egg production when pullets were fed a low protein diet (10% CP) during rearing, which is slightly lower compared to our RP grower diet (11.8% CP), while the study of van Emous et al. (2015b) decreased the dietary CP content of the grower diet only to 12.8%. However, in the present study RP breeders were also fed an RP breeder diet (11.6% CP), whereas in the study of van Emous et al. (2015b) and Hocking et al. (2002) all breeders received a control diet during the laying period with respectively 14.5% and 15.0% CP. Joseph et al. (2000) reported a decrease in early and late stage egg production when birds were reared on control diets but fed a low protein (14% CP) compared to a control (16% CP) and high protein (18% CP) diet during the laying period. It is postulated that a deficiency in methionine and lysine levels, which were lower in the present study compared to van Emous et al. (2015b), may be responsible for the decreased laying performance of the RP breeders (Bowmaker and Gous, 1991). Hence, not only the dietary CP in the breeder feed, but also the magnitude of dietary CP reduction relative to normal CP levels in rearing diets may be a determining factor for laying performance afterwards. van Emous et al. (2015b) also observed a better laying persistency from 48 wk onwards when the breeder pullets were reared on diets with a lower dietary CP level. The improved persistency was probably mediated by the increased AF and decreased BM at the onset of lay. Even though our experiment was terminated at wk 40, the extra fat reserve at the onset of lay was demonstrated.

In accordance with Walsh and Brake (1997) and van Emous et al. (2015a), a decreased fertility was found for RP fed breeders. In the present research total dietary CP intake of the RP fed breeders was only 1059 g at

20 wk of age. Walsh and Brake (1997) a reported that a minimal total dietary CP intake of 1,180 g between 0 and 20 wk of age is needed for an adequate level of fertility. Whereas van Emous et al. (2015a) found that a minimal total CP intake of 1,300 g was needed for the modern broiler breeders.

Feed allocations were weekly adapted to maintain a similar BW for both groups throughout the entire experiment, as BW is one of the main factors that could influence breeder and offspring performance. The breeders fed RP diets required approximately 10%, with a maximum of 14%, more feed during the rearing phase to obtain a similar BW at 23 wk. This observation is in agreement with literature, although the absolute amount of extra feed, required to meet the same BW goals, varied between published results (Lilburn and Myers-miller, 1990; Miles et al., 1997; Hudson et al., 2000; Hocking et al., 2002; van Emous et al., 2013, 2015b). The higher feed allocation of the RP breeders must be taken into account for calculating the net protein reduction of RP group. The control and RP diets were formulated isoenergetically, thus a higher feed allocation resulted in a higher energy intake for the RP fed birds. During the rearing phase, the RP diets contained 25% less protein which corresponds in an average net reduction of 17.5% in CP intake and a 10% increase in ME intake. As a result, not only the dietary protein and ME levels, but also energy:protein ratio was altered for the RP fed birds. At the beginning of the laying phase, the difference in feed allocation between the RP and C breeders varied as a result of feeding the breeders according to their egg production. At peak production, the RP fed birds received 8% more feed compared to their control counterparts. The difference gradually decreased to 3% at 38 wk, while the difference in egg production declined from 11 to 8%. At wk 38 of age, the net difference in daily protein and energy intake was respectively 22.8 and 3.0%. These results suggest that the dietary CP content during the rearing phase had a higher impact on BW and body composition than in the laying phase.

Offspring Performance

Despite a lower hatch weight, as a result of a lower egg weight, male offspring of the RP fed breeders had a better growth performance in comparison with the progeny of their control fed counterparts. The RP male progeny had a significantly higher slaughter weight (on average + 82 g) and a tendency towards a lower FCR_{wmc}. The female RP progeny on the other hand had a lower BW at slaughter age, but a significantly decreased FCR_{wmc}. A differential gender effect, as a consequence of the maternal nutrition was also reported by Spratt and Leeson (1987), Zambrano et al. (2005), van der Waaij et al. (2011) and van Emous et al. (2015a). A lower FCR is in accordance with the research of Lopez and Leeson (1995a). Rao et al. (2009) also reported a higher BW and proportional BM weight for the progeny of low protein fed Langshan breeders. These positive effects on the offspring were supported by alterations of the gene expression in yolk-sac membrane, hypothalamus and muscle of developing chicken embryos (Rao et al., 2009). However, Moraes et al. (2014) and van Emous et al. (2015a) did not observe an effect of a lower dietary CP level in the breeder rearing diets on offspring live performance. These apparent discrepancies between published results can be explained by the period when a lower dietary CP level was provided to the breeders. Interestingly, the reduced proportional albumen weight and hence the possibly reduced protein amount available for the broiler embryo, was followed by an improved postnatal performance. The study of Willems et al. (2016) reported that prenatal protein undernutrition, by albumen removal early during incubation, in laying hens influenced gene expression and associated metabolic pathways in the liver such as "embryonic and organismal development", "organ morphology and development", "tissue development" and "reproductive system development and function", which resulted in long-lasting effects on the protein and carbohydrate metabolism of adult laying hens. Additionally, Piestun et al. (2013) reported that increasing temperature during the mid-phase of incubation had a positive influence on the energy metabolism and an improved thermotolerance of broilers in later life. Thus an increased incubation temperature programmed broilers to cope better with higher temperatures. In mammals, it is also known that prenatal nutritional constraints can program the offspring, resulting in an altered metabolic phenotype in adult life (Lillycrop et al., 2009). One of the most interesting and important features of developmental programming is the evidence in mammals that the effects of maternal malnutrition can be passed transgenerationally from mother over daughter to the F2 progeny (Aerts and Assche, 2003; Zambrano et al., 2005; Pinheiro et al., 2008). Prenatal programming by maternal influences is often referred to as epigenetic programming. Indeed, the inheritance of some effects without selection or altering the DNA sequence of the hens implies that epigenetic mechanisms must be involved.

CONCLUSION

In conclusion, reducing the dietary CP levels in both rearing and laying phase of broiler breeders had a negative effect on egg production rate and egg weight and altered breeder body composition. However, the reduced dietary CP levels had a positive effect on zootechnical performance of the offspring.

ACKNOWLEDGMENTS

The diligent technical assistance of Daniel Vermeulen, Astrid Koppenol, Yufeng Wang, Els Willems and André Respen is gratefully acknowledged.

REFERENCES

Adams, K. A., and A. J. Davis. 2001. Nutrient-gene expression dietary protein concentration regulates the mRNA expression of chicken hepatic malic enzyme. J. Nutr. 131:2269–2274.

Aerts, L., and F. A. Van Assche. 2003. Intra-uterine transmission of disease. Placenta 24:905–911.

Aviagen. 2013. Ross 308 parent stock management guide. Aviagen Incorporated, Huntsville, AL.

Aviagen. 2014. Ross 308 broiler management guide. Aviagen Incorporated, Huntsville, AL.

Berghof, T. V. L., H. K. Parmentier, and A. Lammers. 2013. Transgenerational epigenetic effects on innate immunity in broilers: An underestimated field to be explored? Poult. Sci. 92:2904–2913.

Bowmaker, J. E., and R. M. Gous. 1991. The response of broiler breeder hens to dietary lysine and methionine. Br. Poult. Sci. 32:37–41.

CVB. 2012. CVB Tabellenboek Veevoeding (Feedstuff table 2012). Centraal Veevoeder Bureau, Lelystad, the Netherlands.

De Beer, M., R. W. Rosebrough, B. A. Russell, S. M. Poch, M. P. Richards, and C. N. Coon. 2007. An examination of the role of feeding regimens in regulating metabolism during the broiler breeder grower period. 1. hepatic lipid metabolism. Poult. Sci. 86:1726–1738.

Eisen, E. J., B. B. Bohren, and H. E. Mckean. 1962. The Haugh unit as a measure of egg albumen quality. Poult. Sci. 1461–1468.

Enting, H., A. Veldman, M. W. A. Verstegen, and P. J. van der Aar. 2007. The effect of low-density diets on broiler breeder development and nutrient digestibility during the rearing period. Poult. Sci. 86:720–726.

Everaert, N., S. Metayer-Coustard, H. Willemsen, H. Han, Z. Song, N. Everaert, S. Me, Z. Ansari, E. Decuypere, J. Buyse, and S. Tesseraud. 2013. The effect of albumen removal before incubation (embryonic protein under-nutrition) on the post-hatch performance, regulators of protein translation activation and proteolysis in neonatal broilers. Br. J. Nutr. 110:265–274.

Frésard, L., M. Morisson, J. Brun, A. Collin, B. Pain, and F. Minvielle. 2013. Epigenetics and phenotypic variability: some interesting insights from birds. Genet. Sel. Evol. 45:1–12.

Havenstein, G. B., P. R. Ferket, and M. A. Qureshi. 2003a. Growth, livability and feed conversion of 1957 versus 2001 broilers when fed representative 1957 and 2001 broiler diets. Poult. Sci. 82:1500–1508.

Havenstein, G. B., P. R. Ferket, and M. A. Qureshi. 2003b. Carcass composition and yield of 1957 versus 2001 broilers when fed representative 1957 and 2001 broiler diets. Poult. Sci. 82:1509–1518.

Hocking, P. M., R. Bernard, and G. W. Robertson. 2002. Effects of low dietary protein and different allocations of food during rearing and restricted feeding after peak rate of lay on egg production, fertility and hatchability in female broiler breeders. Br. J. Nutr. 43:94–103.

Hudson, B. P., R. J. Lien, and J. B. Hess. 2000. Effects of early protein intake on development and subsequent egg production of broiler breeder hens. J. Appl. Poult. Res. 9:324–333.

Joseph, N. S., F. E. Robinson, D. R. Korver, and R. A. Renema. 2000. Effect of dietary protein intake during the pullet-to-breeder transition period on early egg weight and production in broiler breeders. Poult. Sci. 79:1790–1796.

Lilburn, M. S., and D. J. Myers-miller. 1990. Effect of body weight, feed allowance and dietary protein intake during the prebreeder period on early reproductive performance of broiler breeder hens. Poult. Sci. 69:1118–1125.

Lillycrop, K. A., M. A. Hanson, and G. C. Burdge. 2009. Epigenetics and the influence of maternal diet. Pages 11–20 in Early Life Origins of Human Health and Disease. J. P. Newnham, and M. G. Ross, eds. Karger, Basel, Switzerland.

Lopez, G., and S. Leeson. 1995a. Response of broiler breeders to low-protein diets. 2. offspring performance. Poult. Sci. 74:696–701.

Lopez, G., and S. Leeson. 1995b. Response of broiler breeders to low-protein diets. 1. adult breeder performance. Poult. Sci. 74:685–695.

Miles, R. D., H. R. Wilson, and R. H. Harms. 1997. Protein intake of broiler breeder replacements and its effect on body composition and subsequent performance. J. Appl. Anim. Res. 11:25–36.

Mohiti-Asli, M., M. Shivazad, M. Zaghari, S. Aminzadeh, M. Rezaian, and G. G. Mateos. 2012. Dietary fibers and crude protein content alleviate hepatic fat deposition and obesity in broiler breeder hens. Poult. Sci. 91:3107–3114.

Moraes, T., A. Pishnamazi, and E. Mba. 2014. Effect of maternal dietary energy and protein on live performance and yield dynamics of broiler progeny from young breeders. Poult. Sci. 93:2818–2826.

Naber, E. C. 1979. The effect of nutrition on the composition of eggs. Poult. Sci. 58:518–528.

Piestun, Y., S. Druyan, J. Brake, and S. Yahav. 2013. Thermal manipulations during broiler incubation alter performance of broilers to 70 days of age. Poult. Sci. 92:1155–1163.

Pinheiro, A. R., I. D. M. Salvucci, M. B. Aguila, and C. A. M.-De. 2008. Protein restriction during gestation and / or lactation causes adverse transgenerational effects on biometry and glucose metabolism in F1 and F2 progenies of rats. Clin. Sci. 114:381–392.

Rao, K., J. Xie, X. Yang, L. Chen, R. Grossmann, and R. Zhao. 2009. Maternal low-protein diet programmes offspring growth in association with alterations in yolk leptin deposition and gene expression in yolk-sac membrane, hypothalamus and muscle of developing Langshan chicken embryos. Br. J. Nutr. 102:848–857.

Renema, R. A., M. E. Rustad, and F. E. Robinson. 2007. Implications of changes to commercial broiler and broiler breeder body weight targets over the past 30 years. Worlds Poult. Sci. J. 63:457–472.

Sexton, T. J. 1977. A new poultry semen extender 1. effect of extension on the fertility of chicken semen. Poult. Sci. 56:1443–1446.

Spratt, R. S., and S. Leeson. 1987. Broiler breeder performance in response to diet protein and energy. Poult. Sci. 66:683–693.

Swennen, Q., E. Decuypere, and J. Buyse. 2007. Implications of dietary macronutrients for growth and metabolism in broiler. Worlds Poult. Sci. J. 63:541–556.

Thiruvenkadan, A. K., R. Prabakaran, and S. Panneerselvam. 2011. Broiler breeding strategies over the decades: an overview. Worlds Poult. Sci. J. 67:309–336.

van der Waaij, E. H., H. van den Brand, J. van Arendonk, and B. Kemp. 2011. Effect of match or mismatch of maternal – offspring nutritional environment on the development of offspring in broiler chickens. Animal 5:741–748.

van Emous, R. A, R. P. Kwakkel, M. M. van Krimpen, H. van den Brand, and W. H. Hendriks. 2013. Effects of growth patterns and dietary crude protein levels during rearing on body composition and performance in broiler breeder females during the rearing and laying period. Poult. Sci. 92:2091–2100.

van Emous, R. A., R. P. Kwakkel, M. M. Van Krimpen, H. Van Den Brand, and W. H. Hendriks. 2015a. Effects of growth patterns and dietary protein levels during rearing of broiler breeders on fertility, hatchability, embryonic mortality and offspring performance. Poult. Sci. 94:681–691.

van Emous, R. A., R. P. Kwakkel, M. M. Van Krimpen, and W. H. Hendriks. 2015b. Effects of dietary protein levels during rearing and dietary energy levels during lay on body composition and reproduction in broiler breeder females. Poult. Sci. 94:1030–1042.

Walsh, T. J., and J. Brake. 1997. The effect of nutrient intake during rearing of broiler breeder females on subsequent fertility. Poult. Sci. 297–305.

Willems, E., C. Guerrero-bosagna, E. Decuypere, S. Janssens, J. Buyse, N. Buys, P. Jensen, and N. Everaert. 2016. Differential expression of genes and DNA methylation associated with prenatal Protein undernutrition by albumen removal in an avian model. Sci. Rep. 6:1–12.

Zambrano, E., P. M. Martinez-Samayoa, C. J. Bautista, M. Deas, L. Guillen, C. Rodrıguez-Gonzalez, G. L. Guzman, F. Larrea, and P. W. Nathanielsz. 2005. Sex differences in transgenerational alterations of growth and metabolism in progeny (F2) of female offspring (F1) of rats fed a low protein diet during pregnancy and lactation. J. Physiol. 566:225–236.

Zhu, C., Z. Y. Jiang, S. Q. Jiang, G. L. Zhou, Y. C. Lin, F. Chen, and P. Hong. 2012. Maternal energy and protein affect subsequent growth performance, carcass yield, and meat color in Chinese Yellow broilers. Poult. Sci. 91:1869–1878.

Zuidhof, M. J., B. L. Schneider, V. L. Carney, D. R. Korver, and F. E. Robinson. 2014. Growth, efficiency and yield of commercial broilers from 1957, 1978 and 2005. Poult. Sci. 93:2970–2982.