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The Effect of Feed Restriction Programs and Growth Curves on Reproductive Performance, *in vitro* Lipogenesis and Heterophil to Lymphocyte Ratios in Broiler Breeder Hens

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Abstract: An experiment was conducted to compare Everyday (ED) and Skip-a-day (SK) feeding programs and early Slow growth (SLOW) and Broilerized (BROIL) treatments. Feed restriction programs were implemented from 4 weeks to 5% production. The SLOW group was fed to reach 75% of standard BW by 12 weeks and then to reach standard BW by 21 weeks. The BROIL group was fed *ad libitum* till 7 weeks and then severely restricted to reach standard BW by 21 weeks. Parameters measured included BW, uniformity, age at Sexual Maturity (SM), total and settable egg production, body composition, liver size and composition, *in vitro* Lipogenesis (IVL) and Heterophil-Lymphocyte ratio (H/L). Breeder production performance was evaluated through 45 weeks of age. Birds fed ED grew more efficiently than SK or SLOW. The BROIL treatment resulted in significantly worse feed utilization than all other groups. Frame size was consistently greater in BROIL pullets and consistently smaller in SLOW pullets. Birds fed ED reached SM before SK, who in turn reached SM before SLOW or BROIL birds. Egg production was significantly higher in ED than SK, which in turn was higher than either SLOW or BROIL. The difference of nearly 17 total eggs per hen between ED and BROIL hens could not be explained by differences in BW or body composition. Liver weight and IVL was elevated in SK and SLOW pullets above ED pullets during rearing. Liver weight and IVL were lower in BROIL pullets than other groups during rearing, but after photostimulation dramatic increases in liver weight and IVL resulted in this trend being inverted by 27 weeks. As an indicator of stress, H/L ratios were elevated above ED pullets in SK, SLOW and BROIL pullets at various times during rearing. These times generally coincided with the periods of most severe feed restriction. Feeding regimens and growth curves have a major influence on efficiency and reproductive performance in broiler breeders. These effects were not attributable solely to differences in BW and body composition. The depression of IVL in broilerized pullets even after restricted feeding was implemented was of great interest and warrants further examination.

Key words: Breeder, growth curves, broilerized, feed restriction, performance, metabolism

INTRODUCTION

In order to achieve maximum reproductive performance, broiler breeders must be feed restricted (Robblee *et al*., 1979; McDaniel *et al*., 1981; Bornstein and Lev, 1982; Siegel and Dunnington, 1985; Hocking *et al*., 1987, 1989; Katanbaf *et al*., 1989a, b). Many producers employ restriction programs involving Skip-a-day (SK) rather than Everyday (ED) feeding to ensure maximum uniformity. The effects of the lengthy fasting periods associated with SK feeding are not fully understood.

Males and female pedigree lines are broilerized as part of the process of genetic selection. Primary broiler breeders are initially raised on full feed to allow phenotypic expression of growth rate and yield, revealing their genetic potential for growth. Broilerization helps to provide phenotypic information by which pedigree birds can be selected at early ages. After selection, growth is dramatically reduced by severe feed restriction so that birds reach the standard breeder body weight or close to it around photostimulation. The metabolic and stress related effects of such rearing systems are not well documented.

Growth curves and the timing of *ad libitum* and restricted feeding during critical periods can affect reproductive performance of breeders (Bruggeman *et al*., 1999). Apart from BW, body fat and lean mass are critical determinants of sexual development (Bornstein *et al*., 1984; Soller *et al*., 1984). Different growth curves may result in changes in body composition and differences in performance even though BW does not differ at photostimulation or Sexual Maturity (SM). Rosebrough and Steele (1985) demonstrated that repeated cycles of starvation and refeeding produced lipogenic rates that were higher than those in birds fed *ad libitum*. Skip-a-day feeding in its current state provides for constant cycles of feeding and fasting and possibly results in increased rates of lipogenesis.

Feed restriction can result in elevated levels of stress in broiler breeders (Renema and Robinson, 2004). It has been demonstrated (Hocking *et al*., 1993) that plasma

Heterophil-Lymphocyte (H/L) ratios are elevated in feed restricted birds up to 16 weeks of age but that a certain amount of adaptation occurs to feed restriction. Research has shown that alternate day feeding cycles can affect responses to disease challenges (Boa-Amponsem *et al*., 1997) and impair the immune system (Zulkifli *et al*., 1993).

The objectives of the study was to determine the effects of different types of feed restriction programs on performance, metabolic parameters and stress in broiler breeder females. Furthermore, the effect of differing growth curves on body composition, performance, metabolism and stress was evaluated. While it is common practice to broilerize pedigree stock for selection purposes, the effects of such practices on performance, metabolism and stress are not well defined.

MATERIALS AND METHODS

Stock and management: A total of 840, day old Cobb 500 broiler breeder pullets were randomly assigned to 24 floor pens. The 24 pen experimental units were divided into 4 treatments with 6 replicate pens of 35 pullets each per treatment. The Cobb Breeder Management Guide (Cobb-Vantress, 2005) was used as a reference for all management conditions, including light schedules for dark-out rearing houses. Pullets were weighed weekly in groups from 0-21 weeks of age. Forty pullets per treatment were weighed individually at 4, 7, 14 and 20 weeks of age to obtain estimates of flock uniformity. Shank and keel lengths were measured on 20 birds per treatment at various ages using a vernier caliper as described by Leeson and Summers (1984). Pullets were photostimulated with 13 h of light at 21 weeks, at which time 80 representative birds from each treatment were housed individually in breeder cages. Each cage had an individual feeder and nipple drinker system. Photoperiod was extended by 1 h/week each week until 16 h of light was reached. From 21 weeks of age all hens were weighed individually every week until week 33 and then monthly until the end of the experiment.

Experimental design: A completely randomized design was used to evaluate 4 different treatments. In the first treatment (ED), pullets were full fed for the first two weeks after which time they were fed restricted amounts of feed everyday throughout rearing and production. The Cobb Breeder Management Guide (Cobb-Vantress, 2005) was used as a guide for target BW. Feed allocation was adjusted after weekly weighing in order to keep birds as close to the BW targets as possible. The second treatment (SK) consisted of birds full fed to two weeks and then fed restricted amounts daily till 28 days.

Table 1: Composition (%) of diets and nutrient contents (%)

Ingredient	Pullet starter	Pullet grower	Prebreeder	Breeder I
Corn, Yellow	61.40	61.41	67.78	66.88
Soybean Meal	26.83	15.44	20.37	22.16
Wheat Midds	7.71	19.04	7.09	
CDP[1]	1.83	1.74	1.73	1.80
Limestone	0.69	0.72	1.62	6.36
Termin-8	0.30	0.30	0.30	0.30
Salt 96+%	0.29	0.31	0.31	0.08
Fat, Poultry	0.25	0.50	0.25	1.67
Microsystem Soy	0.25	0.25	0.25	0.25
L-Lysine HCl	0.10			
Alimet-MHAliquid	0.10	0.07	0.08	0.19
Choline Cl-70%	0.09	0.07	0.08	0.09
Trace Mineral[2]	0.06	0.06	0.06	0.06
Copper sulphate	0.05	0.05	0.05	0.05
Vit. Premix[3]	0.04	0.04	0.04	0.05
Ethoxyquin	0.01			
HyD[4]				0.05
Nutrients (%)				
ME, kcal/kg	2870	2820	2920	2920
CP, calculated	18.99	15.16	16.23	15.95
CP, analyzed[5]	18.59	15.28	16.46	15.81
Crude Fat	2.82	3.27	2.96	4.15
Calcium	0.95	0.90	1.25	3.10
Total phosphorous	0.74	0.75	0.69	0.64
Avail. phosphorous	0.45	0.45	0.42	0.41

[1]Calcium diphosphate. [2]Mineral mix provided per kilogram of complete diet: Cu, 55 mg; I, 7.3 mg; Fe, 366 mg; Mn, 310 mg; Zn, 321 mg; K, 2.23 g; Mg, 1.09 g; Se, 0.48 mg. [3]Vitamin mix provided per kilogram of complete diet: vitamin A, 30,800 IU; vitamin D_3, 9,250 IU; vitamin E, 153.9 IU; vitamin B_{12}, 0.154 mg; riboflavin, 46.2 mg; niacin, 185 mg; pantothenic acid, 84 mg; menadione sodium bisulfite, 16.2 mg; folic acid, 12.3 mg; pyridoxine HCl, 46.2 mg; thiamine HCl, 20.5 mg; biotin, 9.3 mg; choline, 2,944 mg; niacin, 185 mg, [4]Hy-D premix supplied 62.5 μg 25-hydroxy D3/kg diet. [5]Corrected to 90% DM

At that time, skip-a-day feeding regimens were employed until 5% production was reached. After 5 % production the birds were fed restricted amounts everyday. Feed allocation was identical to birds from the first treatment. For example, if the ED birds were being fed 50 g everyday, the SK birds would get 100 g every other day. The third treatment (SLOW) consisted of birds fed the same as SK for the first four weeks, but from four to 12 weeks of age restriction was more severe in an attempt to achieve a BW of 75% of the 12 week target set out in the breeder guide. After 12 weeks, feed allocation was increased in order to achieve the recommended target BW at 21 weeks. The SLOW groups was fed skip-a-day from 4 weeks to 5% production. The compositions of the diets utilized for the first three treatments throughout the experiment are shown in Table 1. The starter diet was fed from 0-4 weeks of age, the grower from 4-18 weeks of age, the prebreeder from 18-22 weeks of age and the breeder I diet from 22-45 weeks of age.

The fourth treatment (BROIL) was fed *ad libitum* for the first seven weeks. This broilerized group was an attempt to mimic the curve followed in selection of pedigree stock. After seven weeks feed was severely restricted using skip-a-day feeding in order to reach the standard

BW recommended at 21 weeks. The diets used during the broiler period (seven weeks) are shown in Table 2. The starter was fed from day old to three weeks and the grower was fed from three to seven weeks. After seven weeks the BROIL birds were given the same diets (Table 1) as the other treatments. Feed allocation was adjusted weekly after 7 weeks in order to ensure that target BW were reached at 21 weeks. Feed allocations for all treatments during the rearing and production periods are shown in Fig. 1. Figure 2 shows the actual BW curves that were achieved in each of the treatments.

Reproductive performance: Egg production was recorded daily and egg weights were measured by weighing two eggs per hen every week, throughout the production period. Each of the first three eggs from every hen was individually weighed to determine early egg weight. Relative egg weight was calculated as the mean egg weight divided by the BW at housing. All soft shelled, double yolk and cracked eggs were recorded. Settable eggs were defined as eggs weighing >50 g with a hard intact shell and one yolk. Age at first egg (sexual maturity) and peak were recorded. Peak was determined as a five day rolling average. The first three eggs produced by each hen were weighed to gauge early Egg Weight (EW). Subsequently, two eggs from each hen were weighed each week to determine overall mean EW for the entire production period. Relative EW was calculated as the overall mean EW divided by the BW at housing. Feed conversion to Body Weight (BW) at 21 weeks of age was calculated in terms of total feed (g) per kg BW, g protein per kg BW and kcal per kg BW. Efficiency of feed conversion to eggs at 45 weeks of age was calculated as total feed intake (g) per egg, total protein intake (g) per egg and total energy intake (kcal) per egg. Mortality was recorded on a daily basis throughout the experiment.

All hens were artificially inseminated at week 32, 36, 40 and 44. One week's worth of eggs was collected from each hen to determine fertility and hatchability at each interval. Semen was collected from same age, separately reared broiler breeder males using the abdominal massage method as described by Burrows and Quinn (1937). Semen was pooled and sperm cell concentration determined using an IMV Micro-Reader I[1], using an optical density of 381 nm (King and Donoghue, 2000). Semen was diluted to $5x10^7$ sperm/50 µL using Beltsville Poultry Semen Extender to ensure all hens were inseminated with the same number and volume of sperm cells. Each hen was inseminated with 50 µL of diluted semen. Semen was diluted to $5x10^7$ sperm/50 µL prior to insemination to allow detection of variation in fertility levels. While, this low number of sperm cells does not produce exceptionally high fertility levels, by not filling the sperm host glands in the hen it allows for differences among treatments to be determined. All eggs were collected for one week after each insemination and set in Jamesway[2] machines for incubation and hatching. All un-hatched eggs were broken out to determine fertility status. Fertility was calculated as the number of fertile eggs per 100 eggs set. Hatchability was calculated as the number of chicks hatched per 100 eggs set and hatchability of fertile eggs was calculated as the number of chicks hatched per 100 fertile eggs set.

Table 2: Diets used for broiler period

Ingredient	Broiler starter	Broiler grower
Corn, yellow	57.13	57.50
Soybean Meal	36.83	34.00
Fat, Poultry	1.37	4.60
Di-Calcium Phosphate	1.64	1.15
Choline Cl-70%	0.20	0.20
L-Lysine HCl	0.07	0.05
DL-Methionine	0.25	0.16
Ethoxyquin	0.02	0.02
Threonine	0.01	0.06
Limestone	1.37	1.40
Salt 96+%	0.50	0.43
Trace Mineral premix[1]	0.10	0.10
Vitamin premix[2]	0.20	0.20
Propionic acid	0.05	0.05
SACOX 60 (coccidiostat)	0.05	0.05
Selenium premix[3]	0.02	0.02
BMD 50[4]	0.05	0.05
Nutrients (%)		
ME, kcal/kg	2980	3190
CP (%) calculated	21.50	20.25
CP (%) analyzed	20.22	19.55
Lysine	1.25	1.15
Methionine	0.58	0.47
Threonine	0.83	0.83
Crude Fat	3.74	6.95
Calcium	1.00	0.90
Total phosphorous	0.71	0.60
Available phosphorous	0.45	0.35

[1]Manganese, 55 mg as manganous oxide 60%; zinc, 50 mg as zinc oxide 72%; copper, 5 mg as copper sulfate 25%; iron, 30 mg as ferrous sulfate 30%. [2]Supplied per kilogram of diet: vitamin A, 8,800 IU (retinyl palmitate); vitamin D3, 3,300 IU; vitamin E, 11.0 IU (dl-á-tocopheryl acetate); riboflavin, 9.0 mg; biotin, 0.25 mg; thiamin, 4 mg; pantothenic acid, 11.0 mg; vitamin B12, 13 µg; niacin, 26 mg; choline, 900 mg; vitamin K, 1.5 mg; folic acid 125 mg. [3]Se premix supplied 0.2 mg Se from sodium selenite per kg of complete feed. [4]Provided 1 g bacitracin activity/ kg diet

Carcass composition: In order to determine the effect of different feed restriction programs on carcass composition, ten pullets per treatment were sacrificed by CO_2 asphyxiation at 4 , 7, 14, 20, 22 27 and 40 weeks of age. Each breeder carcass was frozen at-20°C before autoclaving. The carcasses were placed in trays, covered with foil and autoclaved at 120°C for 15 h in an AMSCO 3053 sterilizer[3]. The carcasses were homogenized after autoclaving using a Waring 4L blender[4]. Sub-samples were collected after grinding and

Table 3: Coeffcients of variation[1] (%) of broiler hens fed using either Everyday (ED) or Skip-a-day (SK) feeding regimens or with slow early (SLOW) or fast early (BROIL) growth curves

	Feeding Regimen			
Age (weeks)	ED	SK	SLOW	BROIL
	CV (%)			
4	12.7			7.2
7	12.6	10.8	13.4	7.6
14	14.0	12.2	14.9	7.4
20	13.2	10.7	16.4	9.2
22	9.8	8.2	12.1	11.9
27	6.7	6.3	6.5	9.8
40	8.9	8.2	9.5	7.6

[1]CV determined by individually weighing 40 birds per treatment at each interval

Table 4: Keel and shank lengths[1] of broiler breeder hens fed using either everyday (ED) or skip-a-day (SK) feeding regimens or with slow early (SLOW) or fast early (BROIL) growth curves

	Feeding Regimen				
Age (weeks)	ED	SK	SLOW	BROIL	SEM
	Shank (cm)				
4	5.23^{b}			6.00^{a}	0.04
12	7.36^{b}	7.10^{c}	6.84^{d}	8.04^{a}	0.06
20	8.97^{b}	8.70^{c}	8.73^{c}	9.36^{a}	0.08
28	9.15^{b}	8.99^{b}	9.02^{b}	9.59^{a}	0.06
	Keel (cm)				
4	10.13^{b}			11.97^{a}	0.10
12	14.54^{b}	14.01^{c}	13.67^{c}	17.69^{a}	0.15
20	17.86^{b}	17.00^{c}	16.84^{c}	19.49^{a}	0.18
28	18.68^{b}	18.54^{b}	18.04^{b}	19.99^{a}	0.22

[a,b]Means within a row without a common superscript differ significantly ($p \leq 0.05$); [1] Values are means of 20 measurements per treatment

lyophilized in a Genesis SQ 12 EL Freeze drier[5]. After freeze drying, samples were finely ground and carcass protein, ash and fat were analyzed according to AOAC (1990). DM was determined as a % of total wet carcass weight. The percent carcass protein, ash and fat were reported on a dry matter basis. Dry BW was calculated by multiplying the proportion of DM by the total wet carcass weight. Total carcass protein, ash and fat were calculated by multiplying the proportion of each component by the dry BW.

***In vitro* lipogenesis:** *In vitro* Lipogenesis (IVL) was measured based on the method described by Rosebrough and Steele (1987). *In vitro* lipogenesis was determined at 4, 7, 14, 20, 22 and 27 weeks of age. Prior to feeding, all birds that were involved in that day's IVL determinations were individually weighed and marked before being returned to their pens. One hour after feeding, marked birds were killed by CO_2 asphyxiation and livers were excised, weighed and washed in phosphate buffered saline to remove blood and debris and then sliced[6] into sections of between 35-70 mg. Slice thickness was set for 3 mm. Six birds were used per treatment and three slices were used per bird. The slices were incubated in 25 mL Erlenmeyer flasks at 37°C for 2 h in 3 mL Hanks balanced salts (Hanks and Wallace, 1949) containing 10 mM HEPES and 10 mM sodium [2-^{14}C] acetate (166 MBq/ mol). Incubations were conducted under a 95 0_2: 5% CO_2 atmosphere, which was obtained by gassing the vials for 15 sec. At the end of the incubation period explants were extracted in 10 mL of 2:1 chloroform: methanol for 18 h. The extract was fractionated with 2 mL 0.88% KCl and then washed according to Folch *et al.* (1957). After the washing process the bottom phase was evaporated to dryness, dispersed in scintillation fluid and counted by liquid scintillation spectroscopy. *In vitro* lipogenesis is expressed as µmoles of acetate incorporated into hepatic lipids per kilogram of body weight.

Liver composition: After slicing, the remaining portions of all excised livers were frozen at -20°C until further processing. Two additional birds were sacrificed and livers were excised and frozen at -20°C for determination of liver composition. These remaining portions from the livers used for IVL determination and the two extra livers were subsequently lyophilized in a Genesis SQ 12 EL Freeze drier[7] to determine the Dry Matter (DM) content. Lyophilized samples were ground before further analysis. Liver protein (N x6.25) and ether extract were then analyzed according to AOAC (1990). Dry liver weight was obtained by multiplying the DM % by the wet liver weight. Both protein and fat % were determined on a DM basis. Total protein and total fat weight was obtained by multiplying the dry liver weight by the % protein and fat in the dry liver sample, respectively.

Heterophil: lymphocyte ratio: The effect of the restriction programs on the health and well being of the pullets was determined by measuring the blood Heterophil-Lymphocyte ratio (H/L). Gross and Siegel (1983) evaluated the use of H/L ratio as a measure of stress in chickens. They concluded that H/L was a good measure of the chicken's perception of stress and could be used to compare groups exposed to various types of stress. Blood H/L ratio was measured at 4, 7, 14, 20, 22, 27 and 40 weeks of age. Blood samples (2 mL), taken one hour after feeding, by cardiac puncture, were collected into EDTA treated vials to prevent clotting. Samples were analyzed at various ages during rearing and production to determine H/L. An Abbott Cell-Dyn 3500[8] automated hematology analyzer was used to determine H/L ratio in 500 µL of the blood sample. Total White Blood Cell (WBC) counts and basophil counts were also obtained in this way. Recommended settings and calibrations for avian hematology were employed according to the manufacturer's operation manual.

Statistical analysis: Data analysis was performed using JMP IN 5.1[9] statistical analysis software. Chicks were assigned to treatments on day one in a completely random manner. All data were analyzed based on a completely randomized design using one way ANOVA. From the beginning of the trial until 21 weeks of age the pen was treated as the experimental unit. After 21 weeks birds were individually caged and each individual bird served as an experimental unit. Data are presented as mean±SEM. When significant differences were observed means were separated using Tukey's Studentized range test. All statements of significance are based on testing at $p \leq 0.05$.

RESULTS

Body weight, uniformity and frame size: Daily feed allocations are shown in Fig. 1. Pullets in the BROIL group were allowed *ad libitum* access to feed until seven weeks of age. Thereafter, only sufficient feed was allocated to reach the 21 week target BW (2420 g). The intention was to avoid any dramatic weight loss in the BROIL group after seven weeks and rather to have a very gradual weight gain to 21 weeks. Actual growth curves and weekly BW are presented in Fig. 2. The mean BW of the BROIL group at 21 weeks was 2518 g. From four to 12 weeks of age, feed allocation for the SLOW group was lower than for ED or SK in an attempt to reduce BW to 75 % of the target at 12 weeks. The recommended BW at 12 weeks was 1290 g. The SLOW group had a mean BW of 1083 g which represented 84% of the target. After 12 weeks feed allocation to the SLOW group was increased in order to reach 2420 g at 21 weeks. Actual 21 week BW for the SLOW group was 2347 g.

Pullets in the ED and SK groups had the same feed allocations throughout the trial period. In spite of this, ED pullets weighed 2497 g at 21 weeks while SK pullets weighed only 2320 g. This difference was statistically significant ($p \leq 0.05$). This represents a difference in BW of approximately 8%. From 25 weeks of age all groups were given equal feed allocations. All groups were fed everyday after 27 weeks. At 40 weeks of age SK hens (3815 g) had significantly lower BW than BROIL (4059 g), SLOW (4000 g) or ED (3977 g) hens.

Forty birds from each treatment were weighed individually at intervals during rearing to determine CV for each treatment (Table 3). At 4 weeks of age the BROIL treatment resulted in a CV of 7.2% which was lower than all other groups (12.7%). At 7, 14 and 20 weeks of age, SK feeding resulted in lower CV than ED feeding. The SLOW growth curve resulted in the highest CV throughout the rearing period. In spite of being severely restricted after seven weeks, the BROIL group had lower CV than all other groups at 7, 14 and 20 weeks. After 21 weeks of age, all birds were individually caged and weighed weekly. After being individually housed, all birds had access to their own feeder and nipple drinker and therefore, feed consumption did not differ between individuals within a treatment group. This is seen in the consistent reduction in CV of all groups after housing. Individual caging at 21 weeks prevented further comparisons in uniformity between treatment groups.

Frame size measurements were taken at intervals during rearing and production (Table 4). By four weeks of age BROIL pullets (6.00 cm) had longer ($p \leq 0.05$) shanks than all other pullets (5.23 cm). In fact, birds from the BROIL group had consistently longer ($p \leq 0.05$) shanks than all other groups at all ages. At 12 weeks of age ED pullets (7.36 cm) had longer ($p \leq 0.05$) shanks than SK (7.10 cm) which in turn had longer shanks than SLOW (6.84 cm). By 20 weeks of age ED pullets still had longer shanks than SK or SLOW, but SK and SLOW no longer differed. At 28 weeks of age, BROIL hens had longer shanks than any other group. None of the other groups differed.

Keel length showed very similar trends to shank length. Birds from the BROIL group had longer ($p \leq 0.05$) keels at 4, 12, 20 and 28 weeks than any of the other groups. At 12 and 20 weeks of age the ED fed birds had longer ($p \leq 0.05$) keels than either SK or SLOW, which did not differ. At 28 weeks of age the keel length of ED, SK and SLOW groups did not differ.

Reproductive performance: Data describing the reproductive performance of the various treatments is presented in Table 5. Sexual Maturity (SM) was defined as age at first oviposition. Everyday feeding resulted in the earliest SM. Birds fed SK took approximately 3.5 days longer ($p \leq 0.05$) to reach SM than ED fed birds. There was a significant difference of 177 g in BW at housing between ED and SK birds (Fig. 2). By 25 weeks (first egg) the difference was still 164 g. The SLOW and BROIL groups both took close to 8 days longer ($p \leq 0.05$) than ED birds to reach SM.

Total egg production was higher in ED than in all other treatments. Hens fed ED during the rearing period produced 97.6 total eggs by 45 weeks, while hens fed SK produced only 90.4 eggs. The SLOW (84.2) and BROIL (80.7) groups produced significantly fewer eggs than SK. Differences between SLOW and BROIL were not significant. Settable egg production (Table 5) was defined as total eggs weighing 50 g or more minus soft-shelled, double yolked or cracked eggs. Settable egg production followed the same trend as total egg production, with ED feeding resulting in more settable eggs than SK, which in turn resulted in more settable eggs than the SLOW growth group or the BROIL group. Total abnormal egg production (double yolk, soft-shelled and cracked eggs) did not differ between treatments. Hens from the BROIL group did produce more ($p \leq 0.05$) double yolk eggs (not shown) than hens from the other treatments.

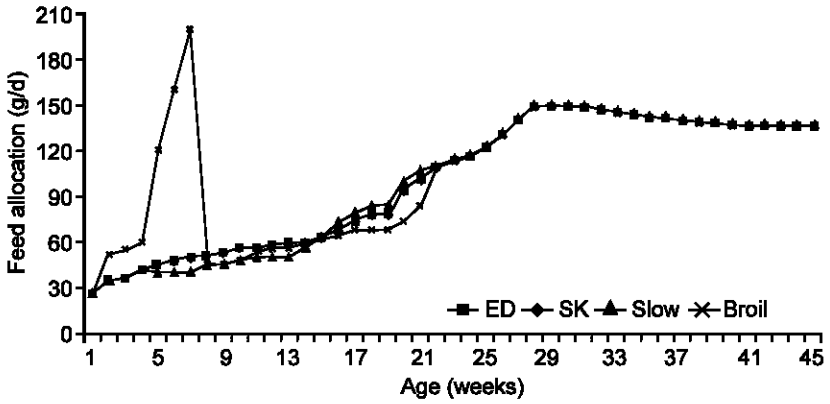

Fig. 1: Feed allocation for broiler breeder hens fed using either everyday (ED) or skip-a-day (SK) feeding regimens or with slow early (SLOW) or fast early (BROIL) growth curves (feed allocation was identical for ED and SK groups)

The first three eggs from each bird were weighed as an indication of early Egg Weight (EW). There were no differences ($p \leq 0.05$) between treatments for early EW. Relative Egg Weight (REW) of the first three eggs was calculated as:

$$\frac{\text{Mean EW of the first three eggs}}{\text{BW at housing}} \times 100$$

Early REW was lower in BROIL hens than in other groups. Other groups had very similar early REW. Overall mean EW was determined by weighing two eggs from each hen every week throughout the production period. Overall EW was higher in BROIL (62.6 g) than in ED (60.9 g) or SK (60.9 g) hens. Hens reared using the SLOW growth curve produced eggs of intermediate size (61. g) but they did not differ significantly from any other treatments. Overall REW was calculated as:

$$\frac{\text{Overall mean EW}}{\text{BW at housing}} \times 100$$

Hens from the SLOW group had higher REW than any of the other treatments.

Fertility (Table 5) was determined using artificial insemination of a known amount of sperm cells ($5x10^7$ cells/hen). Fertility, hatchability and hatch of fertile eggs (not shown) did not differ between treatments. Mortality (not shown) did not differ between treatments during the experimental period.

Employing the BROIL treatment resulted in a significant ($p \leq 0.05$) increase in Feed Conversion Ratio (FCR) and had a negative effect on protein and energy utilization from day old to 21 weeks of age compared to all other groups. For example, BROIL pullets required 4.30 kg of feed for each kg of BW while, ED fed pullets required only 3.64 kg of feed per kg BW. Birds fed ED were more efficient in utilizing feed, protein and energy than those fed SK or reared on the SLOW growth curve. Pullets in the SK and SLOW group did not differ. Hens reared on the BROIL curve required more feed, protein and energy per egg than those from the SK or ED groups. Birds fed ED consumed 90 g less feed per egg than BROIL birds. Birds fed ED were also more efficient than birds from the SLOW treatment. Hens fed ED and SK did not differ significantly in their utilization of feed, protein and energy for egg production to 45 weeks. The numerical trend however, was the same as that for the growth parameters to 21 weeks.

Body composition: Birds were sacrificed at various ages for determination of body composition (Table 7). Body compositions were expressed both as percentages of Dry Matter (DM) and as total mass (g) of fat, protein and ash. Dry carcass mass (g) was obtained by multiplying the % DM by the wet carcass weight. The total mass of fat, protein and ash was obtained by multiplying the proportion of each component in the dry carcass by the total dry carcass mass (g). At four weeks of age BROIL pullets had higher ($p \leq 0.05$) DM % and higher protein % than the other treatments. Fat and ash % did not differ at four weeks. Due to differences in BW, BROIL pullets had more ($p \leq 0.05$) total DM, fat, protein and ash than other pullets.

Dry matter % did not differ between treatments at seven weeks of age. Total DM however, was significantly higher in BROIL pullets than other pullets due to large differences in BW. Pullets from ED, SK and SLOW groups had similar total DM at seven weeks. Fat % and total fat was higher in BROIL (38.4%) than in ED (26.9%), SK (26.4%) and SLOW (24.2%) pullets. Fat content was not different in ED, SK or SLOW pullets. Protein % was higher in ED (54.9%), SK (54.8%) and SLOW (53.7 %) pullets than in BROIL (36.4 %) pullets. Total protein was still highest in BROIL due to their greater BW. Total protein was lower ($p \leq 0.05$) in SLOW

Table 5: Age at first egg, peak production, total egg production, settable egg production, abnormal egg production and mean egg weight of broiler breeder hens fed using either everyday (ED) or skip-a-day (SK) feeding regimens or with slow early (SLOW) or fast early (BROIL) growth curves[1,2]

	Feeding Regimen			
Variable	ED	SK	SLOW	BROIL
Age at SM[3] (days)	181.4±0.7[c]	184.9 ± 0.7[b]	189.4 ± 0.8[a]	189.3 ± 0.8[a]
Total eggs/hen	97.6 ± 2.3[a]	90.4±2.3[b]	84.2±2.4[c]	80.7±2.4[c]
Settable eggs/hen[4]	93.7±2.3[a]	86.9±2.3[b]	80.8±2.3[c]	76.7±2.3[c]
Abnormal eggs/hen[5]	1.47±0.28[a]	1.56±0.28[a]	1.65±0.28[a]	2.31±0.28[a]
First three EW[6] (g)	49.2±0.4[a]	49.8±0.4[a]	50.1±0.5[a]	50.2±0.5[a]
Relative EW[7] (first three eggs)	1.82±0.03[a]	1.86±0.03[a]	1.85±0.03[a]	1.71±0.03[b]
Overall EW (g)	60.9±0.4[b]	60.9±0.4[b]	61.3±0.4[ab]	62.6±0.4[a]
Overall relative EW[8]	2.24±0.03[b]	2.27±0.03[b]	2.38± 0.03[a]	2.22±0.04[b]
Fertility (%)[9]	78.3±3.9[a]	75.5±4.0[a]	74.7±3.9[a]	70.7±4.0[a]

[a,b] means within a row without a common superscript differ significantly ($p \leq 0.05$); [1]Mean±SEM; [2]All parameters measured to 45 weeks of age; [3]Sexual maturity, defined as age at first oviposition; [4]Number of eggs weighing > 50g, not including soft shells, cracks or double yolks; [5]Includes soft shell and double yolk eggs; [6] Egg weight; [7] (Mean weight of first three eggs/ BW at housing)*100; [8](Overall mean EW/ BW at housing)*100; [9] Hens were artificially inseminated at 32, 36, 40 and 44 weeks with 7.5 x 10^7 sperm at each insemination

Table 6: Feed conversion ratio expressed in terms of total feed intake, protein intake and energy intake for broiler breeders fed using either Everyday (ED) or Skip-a-day (SK) feeding regimens or with slow early (SLOW) or fast early (BROIL) growth curves[1]

	Feeding Regimen			
Variable	ED	SK	SLOW	BROIL
FCR at 21 weeks (kg feed/ kg BW)	3.64±0.05[c]	3.76±0.05[b]	3.73±0.05[b]	4.30±0.05[a]
Protein intake (g)/ kg BW at 21 weeks	576.3±8.0[c]	594.4±8.0[b]	591.5±8.1[b]	678.7±8.0[a]
Energy intake (kcal)/ kg BW at 21 weeks	10374.0±144.6[c]	10700.9±143.8[b]	10631.4±145.6[b]	12209.2±143.8[a]
Total feed intake (g)/ egg at 45 weeks	342.1±14.5[c]	368.0±14.5[bc]	404.9±14.7[ab]	432.0±14.9[a]
Total protein intake(g)/ egg at 45 weeks	54.5±2.3[c]	58.6±2.3[bc]	64.5±2.3[ab]	68.7±2.4[a]
Total energy intake(kcal)/ egg at 45 weeks	992.5±42.0[c]	1067.7±42.0[bc]	1175.2±42.6[ab]	1251.0±43.3[a]

[a,b] means within a row without a common superscript differ significantly ($p \leq 0.05$); [1]Mean±SEM

(122 g) than in ED (148 g) pullets. Pullets fed SK had intermediate (136 g) total protein and did not differ from either ED or SLOW. Ash % did not differ between groups but BROIL birds had more total ash.

At 14 weeks, DM % was lower in SK and SLOW than in ED and BROIL. Total DM was still highest in BROIL. Pullets fed ED had more total DM than SK or SLOW pullets. Fat % was higher in SLOW pullets than BROIL at 14 weeks. Total fat was still highest in BROIL pullets however. Protein % was higher in BROIL than in SK or SLOW birds. Birds fed ED also had higher protein % than those from the SLOW group. Total protein was still highest in BROIL, while ED birds had more total protein than SK or SLOW birds. Ash % did not differ between groups but total ash was different ($p \leq 0.05$) in all groups. BROIL had the most ash followed by ED, SK and SLOW.

By 22 weeks DM % did not differ between groups but ED and BROIL birds had more ($p \leq 0.05$) total DM than SK or SLOW birds. Fat % and total fat did not differ between any treatments. Protein % also did not differ between treatments but total protein was lower in SK (477 g) and SLOW (489 g) birds than in ED (533 g) or BROIL (549 g). Ash % and total ash was higher ($p \leq 0.05$) in BROIL than in other treatments.

By 27 weeks of age, SM had been reached in all groups. Dry matter % did not differ between groups but total DM was still higher in ED and BROIL birds than in SK or SLOW. Fat % was higher ($p \leq 0.05$) in ED than in SK birds. BROIL and ED hens had more total fat than SK hens. Protein % did not differ between treatments but total protein was higher ($p \leq 0.05$) in ED than in SK or SLOW hens. Hens from the BROIL group no longer had significantly more total protein than SK and SLOW hens. Ash% did not differ at 27 weeks but BROIL hens had more total ash than SK and SLOW hens. Hens fed ED no longer had significantly less total ash than BROIL hens.

The last body composition analysis occurred at 40 weeks. Hens from the SLOW group had higher DM% than ED hens. Total DM was higher in BROIL and SLOW hens than in ED and SK hens. Fat % did not differ between groups but BROIL (725 g) and SLOW (756 g) hens had more ($p \leq 0.05$) total fat than ED (670 g) or SK (619 g). Protein% was higher in SK than in SLOW hens. Total protein did not differ between groups. Ash % was lower in SLOW hens than in all other hens. Total ash was also lower in SLOW hens than in ED and SK but not BROIL hens.

Liver weight: The weights of excised Livers (LW) at various age intervals are shown in Table 8. The Relative Liver Weight (RLW) was calculated as the LW/BW

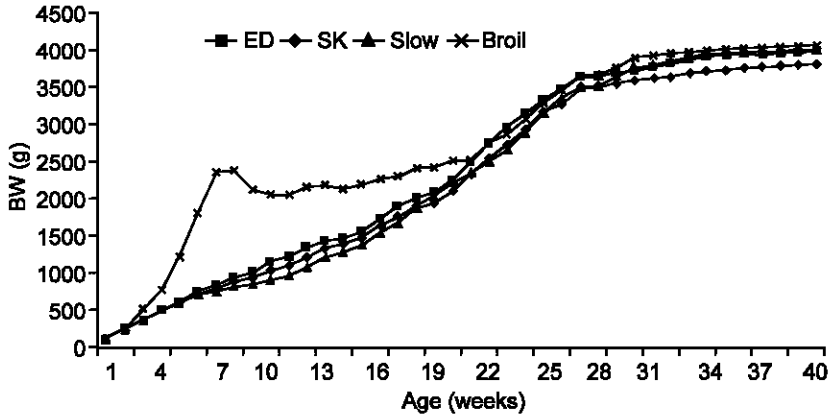

Fig. 2: Growth curves of broiler breeder hens fed using either everyday (ED) or skip-a-day (SK) feeding regimens or with slow early (SLOW) or fast early (BROIL) growth curves

Table 7: Body composition of broiler breeders at 4, 7, 14, 22, 27 and 40 weeks of age fed using either Everyday (ED) or Skip-a-Day (SK) feeding regimens or with slow early (SLOW) or fast early (BROIL) growth curves

Variable	ED		SK		SLOW		BROIL	
	%	g	%	g	%	g	%	g
4 Weeks								
DM[1]	33.9^{b}	170^{y}					38.9^{a}	300^{x}
Fat[2]	31.9	55^{y}					27.7	83^{x}
Protein[2]	47.5^{b}	81^{y}					51.4^{a}	154^{x}
Ash[2]	9.2	16^{y}					8.7	26^{x}
7 Weeks								
DM[1]	31.9	269^{y}	32.1	249^{y}	31.9	229^{y}	32.4	760^{x}
Fat[2]	26.9^{b}	73^{y}	26.4^{b}	66^{y}	24.2^{b}	55^{y}	38.4^{a}	294^{x}
Protein[2]	54.9^{a}	148^{y}	54.8^{a}	136^{yz}	53.7^{a}	122^{z}	36.4^{b}	274^{x}
Ash[2]	8.9	24^{y}	9.3	23^{y}	9.1	21^{y}	9.4	71^{x}
14 Weeks								
DM[1]	34.1^{a}	497^{y}	31.3^{b}	424^{z}	31.2^{b}	397^{z}	33.5^{a}	715^{x}
Fat[2]	25.5^{ab}	127^{y}	25.6^{ab}	109^{y}	27.9^{a}	111^{y}	23.4^{b}	166^{x}
Protein[2]	57.7^{ab}	287^{y}	54.6^{bc}	232^{z}	51.6^{c}	205^{z}	59.1^{a}	424^{x}
Ash[2]	10.7	53^{x}	10.8	46^{y}	10.4	41^{z}	10.5	75^{w}
22 Weeks								
DM[1]	34.8	954^{x}	35.3	893^{y}	34.2	889^{y}	35.1	962^{x}
Fat[2]	29.4	282	30.0	269	28.0	259	28.7	276
Protein[2]	56.0	533^{x}	53.5	477^{y}	55.2	489^{y}	57.2	549^{x}
Ash[2]	9.7^{b}	88^{y}	9.8^{b}	80^{y}	9.6^{b}	87^{y}	10.7^{a}	102^{x}
27 Weeks								
DM[1]	37.2	1348^{x}	35.3	1227^{y}	35.5	1243^{y}	37.0	1347^{x}
Fat[2]	34.2^{a}	464^{x}	29.9^{b}	367^{y}	33.0^{ab}	409^{xy}	34.0^{ab}	458^{x}
Protein[2]	51.0	687^{x}	52.7	645^{y}	51.7	643^{y}	49.5	666^{xy}
Ash[2]	7.9	106^{xy}	8.2	101^{y}	7.9	99^{y}	8.4	114^{x}
40 Weeks								
DM[1]	38.3^{b}	1523^{y}	39.3^{ab}	1501^{y}	42.4^{a}	1699^{x}	39.8^{ab}	1692^{x}
Fat[2]	44.4	670^{y}	41.2	619^{y}	44.4	756^{x}	42.5	725^{x}
Protein[2]	43.4^{ab}	661	46.3^{a}	694	42.4^{b}	720	44.6^{ab}	750
Ash[2]	8.9^{a}	136^{x}	9.2^{a}	138^{x}	6.1^{b}	103^{y}	7.5^{a}	126^{xy}

^{a,b}Within a row, means for % not having the same superscript are significantly different ($p \leq 0.05$); ^{x,y}Within a row means for g not having the same superscript are significantly different ($p \leq 0.05$); [1] Dry matter determined as a percentage of wet carcass weight; [2] Fat, protein and ash determined as a percentage of dry carcass weight

multiplied by 100. The BROIL treatment resulted in greater LW at 4 and 7 weeks than all other treatments. The RLW however, was significantly lower in BROIL than in all other treatments at 4 and 7 weeks. At seven weeks ED, SK and SLOW pullets had no difference in LW. Relative LW however, was higher in SK than in ED or SLOW pullets. At 14 weeks of age BROIL and SK pullets had higher LW than SLOW pullets. Birds from the BROIL group still had significantly lower RLW than all other groups.

Table 8: Absolute and relative liver weights of broiler breeders at various ages fed using either everyday (ED) or skip-a-day (SK) feeding regimens or with slow early (SLOW) or fast early (BROIL) growth curves

Age (weeks)	Feeding Regimen				
	ED	SK	SLOW	BROIL	SEM
	LW[1] (g)				
4	21.1[b]			27.1[a]	1.5
7	22.1[b]	24.6[b]	21.0[b]	42.0[a]	1.0
14	34.7[ab]	38.9[a]	32.2[b]	37.2[a]	1.5
20	44.9[b]	51.9[a]	56.1[a]	45.6[b]	2.1
22	47.6[b]	57.8[a]	54.8[a]	55.0[a]	2.0
27	62.7[b]	75.4[a]	72.8[a]	79.6[a]	3.1
40	83.4[a]	88.6[a]	82.7[a]	84.4[a]	4.2
	RLW[2] (%)				
4	3.33[a]			3.11[b]	0.15
7	1.97[b]	2.12[a]	1.96[b]	1.72[c]	0.04
14	1.97[a]	2.09[a]	1.99[a]	1.59[b]	0.08
20	1.80[b]	2.23[a]	2.23[a]	1.73[b]	0.05
22	1.66[b]	1.92[a]	2.04[a]	1.85[ab]	0.07
27	1.72[b]	2.15[a]	2.03[a]	2.01[a]	0.09
40	2.00[a]	2.11[a]	1.84[a]	1.96[a]	0.11

[a,b]Means within a row without a common superscript differ significantly ($p \leq 0.05$); [1]Absolute liver weight; [2]Relative liver weight calculated as (LW/BW)*100

Table 9: *In vitro* lipogenesis of broiler breeders at various ages fed using either everyday (ED) or skip-a-day (SK) feeding regimens or with slow early (SLOW) or fast early (BROIL) growth curves

Age (weeks)	Feeding Regimen				
	ED	SK	SLOW	BROIL	SEM
	IVL[1] (μmoles/kg BW)				
4	437.9[a]			320.7[b]	32.1
7	227.7[b]	327.4[a]	357.9[a]	41.8[c]	19.9
14	78.8[bc]	97.9[ab]	124.1[a]	56.4[c]	10.5
20	127.3[b]	168.8[a]	154.8[a]	50.1[c]	9.9
22	125.2[b]	160.5[ab]	199.2[a]	135.2[b]	19.7
27	116.1[b]	220.8[a]	150.8[b]	263.7[a]	16.4

[1]*In vitro* lipogenesis, values are presented as μmoles sodium [2-^{14}C] acetate incorporated into hepatic lipids/kg body weight· [a,b]Means within a row without a common superscript differ significantly ($p \leq 0.05$)

By 20 weeks LW and RLW was higher ($p \leq 0.05$) in SK and SLOW groups than in ED or BROIL. Pullets fed ED had lower RLW than SK or SLOW pullets. At 27 weeks of age, ED birds had lower LW than all other groups. The same trend occurred for RLW. By 40 weeks of age, all hens had similar LW and RLW.

***In vitro* lipogenesis:** Table 9 shows the rate of IVL for each of the treatment groups. At four weeks of age, birds fed restricted amounts of feed everyday, had a higher level of IVL than BROIL birds that were being fed *ad libitum* at the time. At seven weeks of age, SK and SLOW pullets showed higher ($p \leq 0.05$) rates of IVL than ED pullets. The BROIL pullets had significantly lower rates of IVL than any of the other birds. At 14 weeks of age the SLOW pullets had higher rates of IVL than the ED or BROIL pullets. Pullets fed SK also had higher rates of IVL than BROIL pullets. At 20 weeks of age SK pullets (168.8 μmoles/kg BW) SLOW (154.8 μmoles/kg BW) and had higher levels of IVL than ED (127.3 μmoles/kg BW), or BROIL (50.1 μmoles/kg BW) pullets. Photostimulation occurred at 21 weeks of age. At 22 weeks, the differences in IVL had narrowed somewhat. Pullets from the SLOW group had significantly higher IVL than ED or BROIL birds. At 27 weeks of age, SK and BROIL hens had higher rates of IVL than either ED or SLOW hens.

Liver composition: Liver protein and fat (Table 10) is presented as% of DM and as total mass (g). All livers were collected approximately 1 h after feeding. Liver fat % and total fat did not differ between ED and BROIL at four weeks of age. Protein% and total protein was higher ($p \leq 0.05$) in BROIL than in ED pullets. At seven weeks of age fat % was higher in ED birds than in was in SLOW birds. SLOW birds, in turn, had higher fat % than BROIL birds. Total fat was lower in SLOW birds (0.33 g) than in ED (0.47 g), SK (0.47 g) or BROIL (0.47 g) birds. Protein% in BROIL (67.2%) was higher than in ED (60.7%), SK (63.4%) or SLOW (62.4%) birds. BROIL pullets had more total protein than ED or SK pullets, which in turn had more than SLOW.

At 14 weeks of age, neither fat nor protein % differed between groups. Total fat was higher in SK than in ED or BROIL birds. Total protein was higher in SK and BROIL birds than in ED or SLOW birds. At 22 weeks of age, SLOW pullets had higher fat% than ED pullets. Total fat was higher in SLOW than in ED or BROIL pullets at 22 weeks. Total fat was also higher in SK than in ED birds. Protein % was higher in ED pullets than in all other treatments. Total protein was higher in SLOW pullets than in ED or BROIL pullets. Pullets fed ED and SKIP also had more total liver protein than BROIL at 22 weeks.

By 27 weeks of age there were no longer any differences in fat% or total fat in any groups. Total fat increased close to 4 fold from 22-27 weeks in each treatment. Protein % did not differ at 27 weeks but total protein was higher in BROIL hens than in SLOW or ED hens. Hens fed ED during rearing, in fact, had less total liver protein than all other groups. Fat% was substantially lower in BROIL hens at 40 weeks of age than it was in all other groups. In spite of there being no differences in LW at 40 weeks (Table 8), total liver fat was lower in BROIL than in ED or SK. Liver protein % did not differ between treatments at 40 weeks. Livers from BROIL hens contained more total protein than livers from SLOW hens.

Heterophil to lymphocyte ratio: H/L ratios for each treatment at various ages are presented in Table 11.

Table 10: Liver composition of broiler breeders at 4, 7, 14, 22, 27 and 40 weeks of age fed using either everyday (ED) or skip-a-day (SK) feeding regimens or with slow early (SLOW) or fast early (BROIL) growth curves

Variable	ED		SK		SLOW		BROIL	
	%[1]	g[2]	%[1]	g[2]	%[1]	g[2]	%[1]	g[2]
4 Weeks								
Fat	11.8	0.71					11.8	0.86
Protein	47.0^{b}	2.81^{y}					52.3^{a}	3.84^{x}
7 Weeks								
Fat	8.0^{a}	0.47^{x}	7.7^{ab}	0.47^{x}	6.3^{b}	0.33^{y}	4.3^{c}	0.47^{x}
Protein	60.7^{b}	3.76^{y}	63.4^{b}	3.90^{y}	62.4^{b}	3.26^{z}	67.2^{a}	7.35^{x}
14 Weeks								
Fat	5.7	0.48^{y}	6.8	0.72^{x}	6.3	0.56^{xy}	4.3	0.45^{y}
Protein	61.1	5.18^{y}	60.8	6.42^{x}	60.2	5.30^{y}	63.7	6.65^{x}
22 Weeks								
Fat	7.17^{b}	0.83^{z}	9.39^{ab}	1.29^{xy}	9.92^{a}	1.47^{x}	8.77^{ab}	1.02^{yz}
Protein	65.6^{a}	7.65^{y}	59.2^{b}	8.13^{xy}	58.4^{b}	8.67^{x}	59.7^{b}	6.94^{z}
27 Weeks								
Fat	21.5	4.02	20.9	4.61	20.7	4.36	18.2	4.14
Protein	54.0	9.78^{z}	52.7	11.40^{xy}	52.7	10.98^{y}	54.1	12.08^{x}
40 Weeks								
Fat	33.3^{a}	9.12^{x}	34.5^{a}	9.85^{x}	29.7^{a}	7.84^{xy}	23.4^{b}	5.93^{y}
Protein	50.2	13.30^{xy}	48.1	12.73^{xy}	51.4	12.57^{y}	54.5	13.57^{x}

^{a,b}Within a row, means for % not having the same superscript are significantly different ($p \leq 0.05$); ^{x,y}Within a row means for g not having the same superscript are significantly different ($p \leq 0.05$) · [1]Liver fat and protein expressed as a percentage of dry liver weight; [2]Total liver fat and protein in grams

Heterophil numbers did not differ between treatments at any age. Lymphocyte count and total white Blood Cell Count (WBC) only showed differences at 14 weeks of age when SLOW pullets had less lymphocytes and total WBC than other groups. H/L ratio did not differ at four or seven weeks between treatments. At 14 weeks however, both SLOW (0.73) and BROIL (0.68) pullets had higher ($p \leq 0.05$) H/L ratios than ED (0.20) and SK (0.30) pullets. At 20 weeks of age SK fed pullets had higher H/L ratio than ED pullets while SLOW and BROIL no longer differed from ED. At 22 weeks the trend is similar but SLOW pullets have higher ($p \leq 0.05$) H/L than ED. There were no differences in H/L at 27 or 40 weeks of age. Basophil count (not shown) did not differ between treatments at any age.

DISCUSSION

The management guides provided by primary breeder companies provide estimates of ideal BW targets through rearing and production. It is not completely clear whether the timing of BW gain is important. The early part of the rearing period is critical for establishing frame size (Hudson *et al*., 2000) while, the importance of the prebreeder phase has been emphasized by several authors (Cave, 1984; Brake *et al*., 1985; Lilburn and Myers-Miller, 1990; Joseph *et al*., 2000). A typical growth curve recommendation is presented in Fig. 3. It is clear that severe feed restriction is employed from 3-16 weeks to maintain low weekly gains. After 16 weeks higher gains are encouraged in the lead up to photostimulation and production. After the onset of production, weekly gains are reduced and minimal gains are recommended after peak.

Birds fed ED grew more efficiently than SK birds in our study. In spite of equal feed intakes, BW was 8% higher in ED pullets by 21 weeks of age. These results are very similar to those reported by other authors. Katanbaf *et al*. (1989a) found that pullets fed ED were 8% heavier than their SK counterparts at 21 weeks even though total feed intake was identical. Leeson and Summers (1985) reported that ED fed breeders weighed about 7% more than SK fed breeders by 20 weeks of age after feeding identical amounts. Lilburn and Myers-Miller (1990) fed 2 groups of pullets 12 kg of feed from 2-24 weeks of age. The first group received a greater allowance of feed early in rearing while the second group received more feed late in rearing. They found that birds receiving high early feed allocations were heavier at 21 weeks in spite of total intake being almost identical, indicating that feed utilization can be altered by manipulation of feed allocation. The BROIL treatment resulted in the poorest FCR. The high early BW of this group meant that the maintenance requirement throughout rearing was higher than for the other groups. More feed was used for maintenance functions and less was allocated to growth.

The periods of fasting and then refeeding in SK regimens require that the bird mobilize nutrients from body reserves during the fast in order to meet its energy demands. This process of deposition of nutrients after a meal and then remobilization is not totally efficient. In our trial, BW of SK fed breeders was still lower than ED at 40 weeks of age. There are obvious benefits in feed utilization when feeding ED rather than SK. Feeding SK however, resulted in improvements in uniformity compared to ED. This improvement in uniformity is of

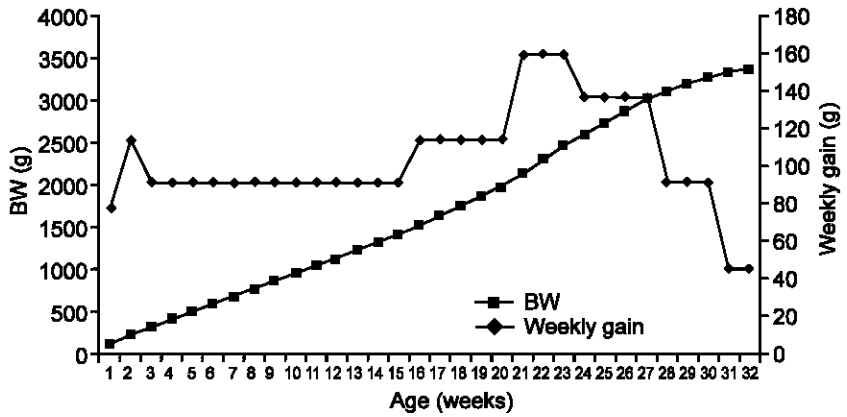

Fig. 3: A typical body weight curve and average weekly gain from 0 to 32 weeks of age as recommended by primary breeders

great importance in commercial breeder operations. Whether the benefits in feed utilization outweigh the losses in uniformity is unclear.

The BROIL treatment consistently had the best uniformity throughout the entire rearing period even though feed restriction in the group after 7 weeks was the most severe. The SLOW treatment resulted in the worst uniformity. This was to be expected as the early feed restriction was most severe in this group. Interestingly, the higher feed allocations after 12 weeks in this group did not improve uniformity to the level of the SK group. These results indicate the importance of the early rearing period to establish good flock uniformity and the difficulty in attempting to correct poor uniformity.

The BROIL pullets consistently had larger frames than other groups. This trend persisted even after the BROIL birds were exposed to severe feed restriction after 7 weeks of age. Up to 20 weeks the SK and SLOW birds generally had smaller frames than ED birds. This most likely reflects the correlation between body weight and frame size. It is clear that the period up to 12 weeks is important in determining frame size as the SLOW group were never quite able to catch up to the frame size of the ED birds, even though their BW was almost identical at 20 weeks. BROIL birds continued to have the largest frames through 28 weeks due to their greater intakes during the early rearing period. A further indicator of the importance of the early rearing period in determining frame size and bone mineral reserve is the fact that SLOW birds had significantly lower carcass ash% and total ash than all other groups at 40 weeks of age. This result was in spite of the SLOW birds producing significantly fewer eggs than ED or SK birds.

Birds fed ED reached SM before any other group. Differences in BW may be responsible for the difference between ED and SK birds. The SLOW and BROIL birds both took around 8 days longer to reach SM than ED birds and this difference could not be attributed to BW. Hens from the BROIL group had very similar BW to ED hens from week 20. Hens from the SLOW group had slightly lower BW than the ED group but their BW did not differ from the SK hens, who still reached SM before they did. Bruggeman *et al.* (1999) found that birds fed *ad libitum* from two to six weeks of age and then restricted after that, had delayed SM. These results show the importance of timing of growth and feed allocation in determining SM rather than BW alone. The delay in SM for SK compared to ED birds could be a result of differences in BW and carcass protein content. Fat content did not differ between these groups at 22 weeks. The delay in SM for SLOW birds compared to SK birds is more difficult to explain however, as they did not differ in BW, carcass fat or carcass protein content at 22 weeks.

Many of the factors determining reproductive fitness occur before SM (Robinson *et al.*, 1995). Several researchers have shown the importance of chronological age, BW, carcass fat content and carcass protein content (Brody *et al.*, 1980; Dunnington *et al.*, 1983; Leeson and Summers, 1983; Bornstein *et al.*, 1984; Soller *et al.*, 1984; Zelenka *et al.*, 1986; Wilson *et al.*, 1995; Sun *et al.*, 2006) as thresholds for development of SM. In this study, BROIL pullets had equal BW, fat content and protein content and more ash than ED fed birds at 22 weeks and yet still took 8 days longer to reach SM than ED birds. Variations in nutrient intake and subsequent energy status are communicated to the hypothalamic-pituitary axis by changes in plasma concentrations of hormones such as glucagon and insulin as well as metabolites such as glucose and free fatty acids. Feed allocation was lower in BROIL pullets just prior to photostimulation which may have caused the subsequent delay in onset of SM.

Wilson *et al.* (1995) demonstrated that breeder pullets reared on a slow early growth curve had less carcass protein at 25 weeks of age despite similar BW to standard birds. These slow early growth birds were last

Table 11: Blood heterophil: lymphocyte ratios of broiler breeders fed using either everyday (ED) or skip-a-day (SK) feeding regimens or with slow early (SLOW) or fast early (BROIL) growth curves[1]

Age (weeks)	Feeding regimen				
	ED	SK	SLOW	BROIL	SEM
	Heterophil/lymphocyte ratio				
4	0.21[a]			0.25[a]	0.07
7	0.13[a]	0.25[a]	0.19[a]	0.15[a]	0.04
14	0.20[b]	0.30[b]	0.73[a]	0.68[a]	0.08
20	0.22[b]	0.38[a]	0.31[ab]	0.32[ab]	0.08
22	0.26[b]	0.40[ab]	0.56[a]	0.41[ab]	0.09
27	0.20[a]	0.37[a]	0.22[a]	0.50[a]	0.12
40	0.41[a]	0.71[a]	0.66[a]	0.64[a]	0.25
	Heterophils (count*10^3/μL)				
4	4.42[a]			3.37[a]	0.50
7	4.35[a]	4.88[a]	4.42[a]	5.00[a]	0.32
14	4.16[a]	5.39[a]	4.73[a]	5.21[a]	0.49
20	4.44[a]	5.09[a]	4.12[a]	4.36[a]	0.41
22	4.16[a]	4.43[a]	5.65[a]	4.43[a]	0.83
27	4.27[a]	4.50[a]	5.03[a]	5.51[a]	0.52
40	4.21[a]	5.21[a]	5.93[a]	4.91[a]	1.18
	Lymphocytes (count*10^3/μL)				
4	27.8[a]			23.4[a]	4.3
7	33.9[a]	28.7[a]	29.7[a]	34.1[a]	3.4
14	21.7[a]	18.3[a]	8.4[b]	17.4[a]	2.1
20	24.3[a]	18.3[a]	20.0[a]	16.7[a]	2.7
22	18.45[a]	18.4[a]	16.6[a]	23.0[a]	3.7
27	24.0[a]	18.4[a]	26.5[a]	20.1[a]	3.3
40	12.4[a]	13.3[a]	15.3[a]	14.9[a]	3.3
	Total WBC[2] (count*10^3/μL)				
4	33.0[a]			27.8[a]	4.6
7	40.3[a]	35.5[a]	35.5[a]	40.8[a]	3.6
14	28.1[a]	21.4[a]	13.7[b]	24.2[a]	2.6
20	30.3[a]	31.4[a]	27.7[a]	24.2[a]	2.9
22	20.1[a]	23.2[a]	24.0[a]	28.9[a]	4.4
27	30.0[a]	23.8[a]	32.6[a]	27.1[a]	3.6
40	16.0[a]	19.5[a]	22.4[a]	21.1[a]	3.4

[a,b]Means within a row without a common superscript differ significantly ($p \leq 0.05$); [1]Differential cell counts were obtained using an Abbott Cell-Dyn 3500 (Diamond Diagnostics Inc. Holliston, Massachusetts) automated hematology analyzer
[2]White blood cells

to reach SM. They concluded neither BW nor carcass fat were limiting in delaying sexual maturation in their experiment. Their results suggested that carcass protein levels were the limiting factor determining age at SM. It is clear that no single factor is absolutely responsible for determining sexual maturation. More likely, combinations of all the aforementioned factors and their levels relative to each other interact to determine the pullet's readiness for sexual maturation.

Egg production was higher in ED compared to SK fed breeders at 45 weeks of age. This may be partly explained by differences in BW. However, Wilson *et al.* (1989) showed that egg production was lower for birds fed using SK programs from eight weeks of age compared to birds fed restricted amounts everyday from two weeks of age. In this study, BW did not differ between the two groups. Wilson *et al.* (1995) found that 20 week BW did not have a consistent effect on performance parameters in breeder hens.

The difference in egg production between ED and BROIL birds was close to 17 eggs per hen by 45 weeks of age. This difference occurred in spite of body weight and body composition at 22 and 27 weeks being almost identical for each of the two groups. The BROIL birds were allocated less feed in the period immediately prior to photostimulation and right after photostimulation. Feed allocation after 22 weeks was very similar for ED and BROIL birds. Lilburn and Myers-Miller (1990) found that higher feed allocations from 16 weeks of age lead to improved performance over lower allocations. They also found that hens with higher BW (1988 g) at 16 weeks of age had higher hen-day production than lower BW (1591 g) hens.

Pullets from the SLOW group had slightly lower BW and less total carcass protein than ED fed birds at 22 and 27 weeks of age. Feed allocation prior to photostimulation was higher in SLOW than ED birds and did not differ after photostimulation. These differences in BW and carcass protein do not seem sufficient to explain that these birds produced >13 less total eggs than ED birds by 45 weeks of age. The exact mechanisms by which the SLOW early growth reduced egg production are not immediately obvious but likely relate to body composition, frame size and perhaps to different endocrine profiles in these birds.

Wilson *et al.* (1995) reared three sets of broiler breeders on different growth curves. A standard growth curve, an early slow and an early fast growth curve were compared. Despite having equal BW at 24 weeks, slow early growth curve birds had higher BW by 56 weeks. Slow early growth also resulted in numerically lower shank lengths compared to early fast groups. This was reflected in carcass composition data, in which ash levels were lower in early slow than in early fast programs. This data supports the notion that early growth is important in establishing frame size in breeders. The slow early growth resulted in fewer settable eggs and fewer chicks than either the standard or the early fast program. This was very similar to our findings with regard to egg production in the SLOW versus the ED treatment. They noted that the slow early growth program resulted in birds entering production with lower carcass protein levels which was similar to our findings.

Renema *et al.* (2001a) determined the effects of three different growth curves on egg production. They found that birds reared to standard weight, 150 g below the standard weight, or 150 g above the standard weight at 20 weeks of age did not differ in their egg or chick production. They did find some negative effects on ovarian morphology in the heavier birds (Renema *et al.*, 2001b). In their research, the differences in BW were

achieved over the whole length of the rearing period. The general shape of the growth curve did not differ between treatments. Their data proved that BW alone is not the only factor that determines reproductive efficiency. In combination with the results presented in this study it seems the shape of the growth curve may be of great importance in determining subsequent performance in breeder hens.

Early and overall relative EW was lowest in BROIL hens and overall REW was highest for SLOW hens. Hens from the BROIL treatment produced larger eggs than ED hens even though BW was not different at photostimulation or SM.

Fertility and hatchability were not affected by feeding regimens or growth curves in our studies. Fertility was numerically highest in ED and numerically lowest in BROIL, which mimics the trends seen for total and settable egg production. Other authors (Walsh and Brake, 1997) have shown that a certain minimum protein intake (1180 g CP) is required by 20 weeks in order to maximize fertility. All of our birds exceeded this protein intake by 20 weeks.

Liver weight was affected by feeding regimens and growth curves. Pullets from the SK and SLOW groups generally had higher LW than ED or BROIL pullets during rearing. Wilson *et al.* (1995) also noted that birds reared on a slow early growth curve had larger livers than standard birds or early fast growth curve birds. Muiruri *et al.* (1975) demonstrated in two experiments that RLW was close to 50% higher in meal-fed chicks one hour after feeding than it was in *ad libitum* fed chicks. Rosebrough and Steel (1985) also reported an increase in RLW of fasted and refed birds over *ad libitum* birds. Pullets from the BROIL group had the lowest RLW at 4, 7, 14 and 20 weeks of age. After photostimulation however, BROIL pullets' LW increased far more dramatically than that of other groups. The reason for the different response to photostimulation is unclear. By 40 weeks the RLW was no longer affected by any of the treatments used during the rearing period. Muiruri *et al.* (1975) believed that much of the increased LW after a meal was due glycogen and water accumulation rather than lipid. This notion was supported by the work of Leveille (1966), who found a near two-fold increase in liver glycogen concentration after refeeding of meal-fed chicks. In contrast to the speculation of Muiruri *et al.* (1975), our results and those of other groups (Akiba *et al.*, 1983; Katanbaf *et al.*, 1989c) show that part of the increased LW in birds fed on programs that involve skipped days is a result of higher lipid levels.

The liver is the major site of fatty acid synthesis in the chicken (O'Hea and Leveille, 1969; Hermier, 1997). Nutritional alteration of lipogenesis in birds is generally achieved either by fasting and refeeding (Rosebrough, 2000), or by altering energy-protein ratios in the diet (Donaldson, 1981; Rosebrough, 2000; Yeh and Leveille, 1969). The increase reported here, in IVL at four weeks for birds fed restricted amounts of feed ED compared to *ad libitum* fed BROIL birds are in agreement with previous findings (Yeh and Leveille, 1972; Rosebrough and Steele, 1985; Rosebrough, 2000). The SK and SLOW groups were both fed every other day from 4 weeks to 5% production. These fasting periods followed by larger meals resulted in elevated rates of IVL compared to ED fed birds at each interval tested.

Mechanisms by which repeated cycles of fasting and refeeding influence hepatic lipogenesis include, increased hepatic acetyl-CoA carboxylase (ACC) mRNA expression (Hillgartner *et al.*, 1996; Richards *et al.*, 2003), increased availability of NADPH reducing equivalents from the Malic Enzyme (ME) reaction and increased circulating triiodothyronine (T_3) levels (Rosebrough, 2000). The rate limiting step in lipogenesis occurs at ACC. This enzyme is regulated in the short-term by covalent modification (phosphorylation) and allosteric control by citrate (Hillgartner *et al.*, 1995, 1996). Long term transcriptional regulation of ACC is mediated by insulin, glucagon, T_3 and glucose (Hillgartner *et al.*, 1996). Feed restriction also increases the expression of other genes involved in lipogenesis. Richards *et al.* (2003) demonstrated increased expression of sterol regulatory element binding protein-1, ATP-citrate lyase, fatty acid synthase, ME and stearoyl-CoA (Δ9) desaturase-1 genes in feed restricted (meal-fed) birds compared to *ad libitum* fed birds.

The dramatically lower rates of IVL in BROIL pullets at 14 and 20 weeks of age are of great interest. They were being fed every other day at that time. It would be expected that IVL would be greatly enhanced under those circumstances. The exact opposite occurred however. After lighting, BROIL birds showed a dramatic increase in IVL, to the point that it was highest in BROIL hens by 27 weeks of age. Their LW followed a similar pattern during the pullet to breeder transition. Understanding the reasons for these patterns of IVL may help to explain why these birds produce so many fewer eggs than other birds of similar BW and body composition. It would be of great interest to study the endocrine response to feeding in the BROIL birds before and after photostimulation.

Feed restriction is considered essential for the well being (Katanbaf *et al.*, 1989a) and reproductive performance (Katanbaf *et al.*, 1989b) of broiler breeders, but can cause behaviors indicative of hunger and frustration (Savory *et al.*, 1992; Hocking *et al.*, 1993, 1996). During rearing the H/L ratios of SK, SLOW and BROIL birds were higher than those of ED birds at various times. Interestingly, restricted feeding did not elevate H/L ratios above the *ad libitum* fed BROIL group at 4 or 7 weeks of age. The most dramatic differences were at 14 weeks, when SLOW and BROIL birds had

much higher H/L ratios than either ED or SK. This is indicative of the more severe feed restriction that these 2 groups had been experiencing in the weeks before sampling. After 27 weeks differences were no longer evident. These data once again show the potential for feed restriction to elevate H/L ratios. Hocking *et al.* (1993) found that H/L ratio of restricted pullets was elevated at 8, 12 and 16 weeks of age as compared to full–fed control groups. Maxwell *et al.* (1990) observed elevated basophil numbers when comparing feed restricted to *ad libitum* fed birds. In the studies conducted here, basophil numbers were not affected by different feeding programs or different growth curves.

Present data suggests that growth curve and feed allocation patterns are as important as BW and total feed intake in determining reproductive efficiency. Bruggeman *et al.* (1999) determined that the critical age for feed restriction was from 7-15 weeks and that the level and timing of feed restriction has important effects on growth and the reproductive axis (hypothalamic-pituitary and ovary-oviduct) development. They suggested that longer term feed restriction was unnecessary to ensure maximum production. Yu *et al.* (1992) determined that feed restriction from 4-18 weeks was necessary to maximize reproductive efficiency. Present studies indicate that BW and body composition at photostimulation, while, important are not the only determinants of reproductive efficiency. The shape of the growth curve can have dramatic effects on egg production and other metabolic parameters such as lipogenesis. Further research, perhaps into endocrine related effects, is required to determine why broilerized pedigree stock are unable to sustain acceptable levels of performance.

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Abbreviation key: ACC = acetyl-CoA carboxylase; ED = everyday fed pullets; EW = egg weight; FCR = feed conversion ratio; H/L = heterophil to lymphocyte ratio; IVL = *in vitro* lipogenesis; LW = liver weight; RLW = relative liver weight; REW = relative egg weight; SK = skip-a-day fed pullets; SM = sexual maturity; T_3 = triiodothyronine.

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[6]McIlwain Tissue Chopper, Vibratome Company, St Louis, Missouri
[7]The Virtis Company, Gardiner, New York
[8]Abbott Diagnostics, Abbott Park, Illinois
[9]SAS Inst., Inc., Cary, North Carolina