METABOLISM AND NUTRITION

Laying Hen Productivity as Affected by Energy, Supplemental Fat, and Linoleic Acid Concentration of the Diet

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ABSTRACT A trial using 720 Isabrown hens was conducted to determine the influence of energy (AME_n), supplemental fat (SFAT), and linolenic acid (LIN) concentration of the diet on performance and weight of eggs and egg components throughout the laying cycle (22 to 65 wk of age). There were six treatments whose calculated AME_n, SFAT, and LIN content were, respectively: 1) 2,810 kcal/kg, 0%, 1.15%; 2) 2,810 kcal/kg, 4%, 1.15%; 3) 2,810 kcal/kg, 4%, 1.65%; 4) 2,680 kcal/kg, 0%, 1.15%; 5) 2,680 kcal/kg, 4%, 1.15%; and 6) 2,680 kcal/kg, 4%, and 1.65%. All diets were formulated to have the same crude protein, lysine, TSAA, calcium, and nonphytin phosphorus levels per kilocalorie of AME_n. The data were analyzed with SFAT constant (4%) and AME_n and LIN variables (Diets 2, 3, 5, and 6) and with LIN constant (1.15%) and AME_n and SFAT variables (Diets 1, 2, 4, and 5). When LIN was maintained at a constant of 1.15%, an increase in the AME_n of the diets from 2,680 to 2,810 kcal/kg decreased feed intake by 4% ($P < 0.001$). Increasing AME_n also improved feed conversion per dozen eggs and per kilogram of eggs by 4.9 and 4.7% ($P < 0.05$), respectively, and increased BW gain by 55.7% ($P < 0.05$). Egg production rate, egg weight, egg mass output, and energy intake were not modified by treatments. An increase in SFAT within both energy levels from 0 to 4% improved all of the traits studied except feed conversion. Supplemental fat increased both yolk and albumen weight, but the effect was more pronounced on the later. When SFAT was maintained constant at 4%, an increase in AME_n of the diets decreased feed intake and improved feed conversion per dozen and per kilogram of eggs by 5.7, 5.5, and 5.2%, respectively ($P < 0.001$). An increase in LIN content from 1.15 to 1.65% did not modify any of the parameters studied. The results indicate that SFAT consistently improves productivity of hens and egg weight and that the LIN requirement for maximal productivity is 1.15% or less. The beneficial effects of adding SFAT to diets containing more than 1.15% LIN are due to the fat itself rather than to an increase in LIN or AME_n of the diet.

(*Key words*: linoleic acid, supplemental fat, energy, egg weight, yolk weight)

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INTRODUCTION

Age of sexual maturity of pullets can be advanced considerably by breeding programs, light stimulation, nutrition, or by a combination of the three. Decreasing the age of sexual maturity increases the number of eggs laid with the potential disadvantage of the production of a greater proportion of small eggs. Numerous investigations have focused on methods to increase egg weight through diet manipulation at the beginning of the laying period. Increasing the protein (Parsons *et al.*, 1993; Keshavarz and Nakajima, 1995), methionine (Schutte and De Jong, 1994; Keshavarz, 1995), lysine (Zimmerman, 1997), linoleic acid (Jensen and Shutze, 1963; Scragg *et al.*, 1987), AME_n (Harms and Waldroup, 1963; DeGroote, 1972), and the fat content of the diet (Sell *et al.*, 1987) has resulted in improvements in egg weight. The mechanism by which the protein fraction of the diet improves productivity and egg size is well understood, but conflicting results arise when the influence of supplemental fat (SFAT), AME_n, and linoleic acid (LIN) content of the diet on performance of hens and weight of eggs and egg components is studied.

Dietary fat has been reported to increase egg weight, and this effect is primarily attributed to the LIN content of the SFAT. Jensen *et al.* (1958) were the first to demonstrate that corn oil contains a factor necessary to maximize egg weight. Shutze *et al.* (1959, 1962) and Balnave (1970, 1971) concluded that LIN present in vegetable oils was the component responsible for the improvement. However, there are conflicting results on the requirements of LIN for optimal egg size. Shannon and Whitehead (1974) and Whitehead (1984) recommended dietary concentrations of less than 1.0%, whereas Jensen *et al.* (1958), Guenter *et*

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Abbreviation Key: LIN = linoleic acid; SFAT = supplemental fat.

al. (1971), and Scragg *et al.* (1987) obtained advantages in egg weight with LIN levels greater than 2%. Factors such as strain of bird, age at sexual maturity, dietary AME_n concentration, and LIN content of the rearing diets may have contributed to these disparate LIN requirements (Balnave, 1970, 1972; Balnave and Weatherup, 1973, 1974; Scragg *et al.*, 1987; Sell *et al.*, 1987).

The effect of SFAT, independent of its LIN content, on egg size is unclear because LIN is generally supplied to the diet as a component of vegetable oils, and, therefore, both effects are confounded. Some authors (Whitehead, 1981; Sell *et al.*, 1987; Keshavarz and Nakajima, 1995) have observed that the addition of fat to diets containing a concentration of LIN above NRC (1994) requirements (1.0%) increased egg weight. These authors did not find any relationship between egg weight and LIN concentration and concluded that egg weight responds to any readily absorbable fatty acid rather than to LIN alone. Scragg *et al.* (1987), however, reported that increasing the intake of readily absorbable oil without increasing the concentration of LIN did not increase egg weight.

Much of the controversy on the relationship between egg weight and SFAT relates to the confounding effects of the energy contribution of fat with some physiological effects that fat might have *per se*. When fats are included in diets replacing starch, the net energy available to the bird is enhanced, which may result in an increase in BW of the hen and indirectly increase egg size. This effect of fat might have a special significance at the onset of lay when an adequate BW substantially enhances egg weight (Summers and Leeson, 1993). However, Combs (1961) and Keshavarz and Nakajima (1995) found that the beneficial effect of SFAT on egg weight was independent of its energy content.

The present experiment was conducted to determine the influence of LIN, AME_n, and SFAT content of the diet on performance and weight of eggs and egg components of brown laying hens from 22 to 65 wk of age.

MATERIALS AND METHODS

A total of 720 Isabrown hens, 22 wk of age, was used. At 19 wk, the pullets were transferred from the rearing farm to a two-deck cage (41 × 42 cm) facility and housed at random in groups of three birds per cage. During rearing, pullets were housed in cages under light-tight conditions with a constant photoperiod program (10 h light). Pullets were fed following recommendations for commercial layers (Isabrown, 1996).

There were six dietary treatments and four replicates, 10 adjacent cages each, per diet. The experimental diets differed in AME_n, SFAT, and LIN content, and the calculated nutritive values were 2,810 kcal/kg, 0%, and 1.15% for Diet 1; 2,810 kcal/kg, 4% and 1.15% for Diet 2; 2,810 kcal/kg, 4%, and 1.65% for Diet 3; 2,680 kcal/kg, 0%, and 1.15% for Diet 4; 2,680 kcal/kg, 4%, and 1.15% for Diet 5, and 2,680 kcal/kg, 4%, and 1.65% for Diet 6, respectively. The experiment lasted for 11 periods of 28 d (22 to 65 wk of age), and the birds had *ad libitum* access to feed and water throughout the experiment. Animals were placed in an open house with natural ventilation and exposed to a minimum of 16 h light/d. Room temperature ranged from 16 to 34 C.

Crude protein, lysine, methionine, TSAA, calcium, and phosphorus contents of all the ingredients, as determined by laboratory analysis, were used to formulate the experimental diets. Composition of diets is given in Table 1. All diets had a similar protein, lysine, methionine, TSAA, calcium, and nonphytin phosphorus content per unit of energy (Table 2) and met or exceeded the NRC (1994) requirements for layers. Accordingly, the increase in AME_n concentration in our study was parallel to an increase in nutrient density of the diet. All of the feeds were evaluated for AME_n at the beginning of the second experimental period (26 wk of age) by using the total collection procedure described by Mateos and Sell (1980). The diets were also analyzed (AOAC, 1990) to determine moisture by the oven-drying method (930.15), protein by the Kjeldahl method (984.13), ether extract by Fosslet fat analysis (920.39), ash by muffle furnace (942.05), and calcium and phosphorus by spectrophotometry (935.13 and 964.06, respectively). Amino acids were determined by using HPLC (Cohen *et al.*, 1989). All of the samples were analyzed in duplicate.

Hen-day egg production and feed consumption records were kept throughout the trial. All eggs produced during the last 3 d of every week were saved, and weight per replicate was recorded. The data were summarized every 4 wk. The hens were weighed at the start of the experiment and at the end of each 4-wk period except for the 4th and 8th periods. At 24, 26, 30, 34, 38, 42, and 65 wk of age, 12 eggs per replicate were collected at random to determine the weights of yolk, albumen, and shell plus membranes according to the procedure described by Hussein *et al.* (1992). The eggs were individually weighed and broken, and the weights of the shells and yolks were determined; the latter was carefully cleaned of any adhering albumen with a damp paper towel. Albumen weight was calculated by difference between total egg weight and the weight of shell plus yolk. Haugh units, shell thickness, and yolk color as measured by the Roche color fan (Vuilleumier, 1969) were also determined in these eggs.

The results by period and at the end of the experiment were compared by using the ANOVA procedure of the SAS Institute (1990); type of diet was main effect. For the first analysis, the data obtained with the four diets that had 1.15% of LIN (Diets 1, 2, 4, and 5) were used to determine the effects of AME_n and SFAT and their interaction in a factorial arrangement. For the second analysis, the data belonging to the four diets that had 4% SFAT (Diets 2, 3, 5, and 6) were used to determine the effects of AME_n and LIN and their interaction in a factorial arrangement. The effects of experimental period and its interaction with type of diet were determined using a repeated measurement analysis (SAS, 1990).

TABLE 1. Ingredient composition of laying hen diets

	Diet					
	1	2	3	4	5	6
AME_n (kcal/kg)	2,810	2,810	2,810	2,680	2,680	2,680
	(%)					
SFAT[1]	0	4	4	0	4	4
LIN[2]	1.15	1.15	1.65	1.15	1.15	1.65
Wheat	19.3	34.8	36.4	16.4	36.8	32.8
Yellow corn	47.1	22.6	16.1	46.6	11.6	14.6
Manioc	1.5	1.9	7.2	3.2	9.0	9.0
Animal fat	—	4.0	1.7	—	3.3	1.3
Acid oil soapstocks[3]	—	—	2.3	—	0.7	2.7
Soybean meal, 44% CP	4.2	6.8	4.9	15.7	17.1	16.5
Sunflower meal, 36% CP	3.9	12.0	12.0	3.5	10.5	9.1
Meat and bone meal, 51% CP	5.0	3.7	5.0	4.6	—	—
Fish meal, 60% CP	7.0	4.7	5.4	—	—	0.6
Gluten meal, 58% CP	3.2	—	—	0.4	—	2.7
Calcium carbonate	8.1	8.5	8.3	8.0	8.7	8.5
Dicalcium phosphate	0.17	0.45	0.15	1.02	1.73	1.65
Sodium chloride	0.25	0.25	0.25	0.25	0.26	0.25
DL-methionine	0.03	0.05	0.05	0.08	0.06	0.05
Microingredients[4]	0.25	0.25	0.25	0.25	0.25	0.25

[1]SFAT = Supplemental fat.

[2]LIN = Linoleic acid.

[3]Mixture of soy and olive oil containing 55% free fatty acids. Composition in major fatty acids was C16:0, 10.5%; C16:1, 0.18%; C18:0, 3.7%; C18:1, 43.2%; C18:2, 35.0%; C18:3, 5.1%; C20:0, 0.20%; and C20:1, 0.11%.

[4]Provided the following per kilogram of diet: vitamin A (trans-retinyl acetate), 10,000 IU; cholecalciferol, 2,200 IU; Vitamin E (DL-α-tocopheryl acetate), 13 IU; menadione sodium bisulfite, 2.0 mg; riboflavin, 4.0 mg; D-calcium pantothenate, 8.0 mg; nicotinic acid, 28 mg; pyridoxine hydrochloride, 0.8 mg; folic acid, 0.25 mg; d-biotin, 0.05 mg; thiamine hydrochloride, 1.0 mg; vitamin B_{12}, 10 μg; choline cloride, 250 mg; Mn, 80 mg; Zn, 65 mg; Fe, 40 mg; Cu, 8 mg; I, 1.9 mg; Se, 0.25 mg; and ethoxyquin, 125 mg.

Table 2. Analyzed nutrient content of laying hen diets[1]

	Diet					
	1	2	3	4	5	6
AME_n (kcal/kg)	2,810	2,810	2,810	2,680	2,680	2,680
	(%)					
SFAT[2]	0	4	4	0	4	4
LIN[3]	1.15	1.15	1.65	1.15	1.15	1.65
Crude protein	17.5	17.8	17.6	16.8	17.0	17.1
Lysine	0.85	0.82	0.81	0.77	0.80	0.81
TSAA	0.68	0.69	0.69	0.67	0.66	0.67
Calcium	3.94	4.04	4.05	3.61	3.77	3.78
Total phosphorus	0.68	0.71	0.70	0.70	0.68	0.66
Ether extract	3.5	6.6	6.7	3.4	5.9	6.0
LIN[4]	1.08	1.04	1.57	1.13	1.19	1.76
AME_n, kcal/kg[5]	2,780	2,820	2,840	2,650	2,710	2,700
Fatty acids, %						
ΣSAT[6]	0.80	2.05	1.76	0.60	1.66	1.10
ΣMONOUNSAT[7]	0.91	2.46	2.47	0.86	2.07	2.08
ΣPOLYUNSAT[8]	1.16	1.19	1.76	1.19	1.30	1.95

[1]Mean of duplicate analyses.

[2]SFAT = Supplemental fat.

[3]LIN = Linoleic acid.

[4]Calculated values of 1.15% for Diets 1, 2, 4, and 5 and of 1.65% for Diets 3 and 6 (FEDNA, 1994).

[5]Calculated values of 2,810 kcal/kg for Diets 1, 2, and 3 and of 2,680 kcal/kg for Diets 4, 5, and 6 (FEDNA, 1994).

[6]ΣSAT = Σ (C14:0 + C16:0 + C18:0 + C20:0).

[7]ΣMONOUNSAT = Σ (C16:1 + C18:1 + C20:1).

[8]ΣPOLYUNSAT = Σ (C18:2 + C18:3).

TABLE 3. The effects of dietary treatment on performance of hens. Linoleic acid was constant at 1.15% of the diet (22 to 65 wk of age)

	AME_n			Supplemental fat			
	2,680 kcal/kg	2,810 kcal/kg	P[1]	0%	4%	P	SEM[2]
Egg production, %	88.8	88.9	NS	88.0	89.8	0.001	0.22
Egg weight, g	64.9	64.5	NS	64.1	65.3	0.004	0.25
Egg mass output, g/d[3]	57.7	57.3	NS	56.4	58.6	0.001	0.20
Feed consumption, g/d	122	116	0.001	117	121	0.04	1.0
Energy intake, kcal/d	327	326	NS	322	331	0.04	2.75
Feed efficiency							
kg feed/kg eggs	2.12	2.02	0.001	2.08	2.06	NS	0.02
kg feed/dozen eggs	1.65	1.57	0.001	1.60	1.61	NS	0.02
BW change, g	140	218	0.04	88	270	0.03	23.6

[1]NS = $P > 0.05$.

[2]n = 8 for main effects and n = 4 for interactions.

[3]Interaction ($P < 0.05$) between AME_n and supplemental fat of the diets. Egg mass increased with supplemental fat, but the effect was more pronounced (+3.0 g) with the lowest than with the highest (+1.5 g) AME_n ($P < 0.05$).

RESULTS

Laying Performance

When the LIN content of the diets was maintained constant at 1.15% (Diets 1, 2, 4, and 5), an increase in AME_n of the diets from 2,680 to 2,810 kcal/kg did not influence energy intake, egg production, egg weight, or egg mass output (Table 3). However, hens fed the more concentrated diets ate less feed (116 *vs* 122 g/d; $P < 0.001$) and had better feed conversion per kilogram of egg (2.02 *vs* 2.12 kg; $P < 0.001$) and per dozen eggs (1.57 *vs* 1.65 kg; $P < 0.001$) than hens fed the less concentrated diets. Body weight gain throughout the test was greater for hens fed the high energy diets (218 *vs* 140 g; $P < 0.04$).

An increase of SFAT from 0 to 4% increased feed and energy intake by 3% ($P < 0.05$) (Table 3). Hens fed the 4% added fat diets laid more eggs (89.8 *vs* 88.0%; $P < 0.001$) and produced heavier eggs (65.3 *vs* 64.1 g; $P < 0.01$) than hens fed the unsupplemented diets. As a result, daily egg mass was increased by SFAT (58.6 *vs* 56.4 g; $P < 0.001$). Some of the extra energy ingested was utilized for weight gain, which was greater (270 *vs* 88 g) for hens fed fat-supplemented diets compared with hens not fed supplemental fat. As a result, feed efficiency (kilograms of feed per kilogram or per dozen eggs) was not affected by dietary fat treatments. Most of the improvement in rate of egg production caused by SFAT occurred from 41 to 65 wk of age, after peak egg production (Table 4). However, the beneficial effects of SFAT on egg weight were most noticeable ($P < 0.05$) from 25 to 49 wk of age and decreased thereafter (Table 5). A significant interaction ($P < 0.05$) between SFAT and AME_n of the diet was found with daily egg mass output from 22 to 65 wk of age. Daily egg mass increased more (3 *vs* 1.5 g) when fat was supplemented to low AME_n diets than to high AME_n diets. No other significant effects of this interaction were observed with any other traits studied.

When SFAT was maintained constant at 4% (Diets 2, 3, 5, and 6), an increase in LIN content from 1.15 to 1.65% did not affect any of the parameters studied (Table 6). When the AME_n of the diets increased from 2,680 to 2,810 kcal/kg, feed consumption decreased (123 *vs* 116; $P < 0.001$), whereas feed efficiency [either per kilogram of egg (2.11 *vs* 2.00 kg) or per dozen eggs (1.65 *vs* 1.56 kg)] was improved ($P < 0.001$). None of the other traits (energy

TABLE 4. The influence of age and dietary treatment on rate of egg production (percentage hen-day). Linoleic acid was constant at 1.15% of the diet

				Age of hens									
	Diet	AME_n	Supplemental fat	22–25 wk	26–29 wk	30–33 wk	34–37 wk	38–41 wk	42–45 wk	46–49 wk	50–53 wk	54–57 wk	58–61 wk	62–65 wk
		(kcal/kg)	(%)											
	4	2,680	0	90.0	92.7	93.8	92.7	90.9	87.8	84.6	86.1	83.0	81.1	81.6
	5	2,680	4	90.9	94.4	94.8	93.2	93.2	91.4	89.1	89.9	86.3	84.9	82.0
	1	2,810	0	90.8	92.7	94.1	92.4	90.3	87.7	86.3	85.4	85.2	83.6	82.9
	2	2,810	4	89.5	92.1	94.3	94.4	92.6	90.7	88.3	87.4	86.5	85.4	83.5
SEM[1]				1.41	0.71	1.00	0.88	1.03	0.90	0.59	1.31	1.10	0.98	0.79
				P										
AME_n				0.81	0.13	0.93	0.62	0.55	0.66	0.48	0.25	0.31	0.14	0.09
Supplemental fat				0.89	0.49	0.57	0.16	0.04	0.01	0.01	0.05	0.06	0.01	0.52
AME_n × supplemental fat				0.45	0.13	0.69	0.38	0.99	0.75	0.06	0.53	0.36	0.33	0.91

[1]n = 4.

TABLE 5. The influence of dietary treatment on egg weight. Linoleic acid was constant at 1.15% of the diet

				Age of hens									
	Diet	AME_n	Supplemental fat	22–25 wk	26–29 wk	30–33 wk	34–37 wk	38–41 wk	42–45 wk	46–49 wk	50–53 wk	54–57 wk	58–61 wk	62–65 wk
		(kcal/kg)	(%)											
	4	2,680	0	57.1	60.2	61.9	63.3	63.3	63.0	63.5	65.8	68.5	68.9	70.0
	5	2,680	4	58.5	62.6	63.9	65.3	65.4	65.3	65.1	66.6	69.9	70.2	70.7
	1	2,810	0	57.0	60.6	62.1	63.5	63.2	63.1	63.3	65.3	67.9	69.1	69.6
	2	2,810	4	57.6	62.0	63.1	64.2	63.8	64.0	64.6	66.4	69.0	69.1	70.2
SEM[1]				0.31	0.35	0.42	0.44	0.48	0.45	0.33	0.48	0.44	0.45	0.42
				P										
AME_n				0.14	0.76	0.47	0.34	0.11	0.24	0.31	0.49	0.11	0.33	0.29
Supplemental fat				0.01	0.01	0.01	0.01	0.01	0.01	0.01	0.10	0.02	0.18	0.13
$AME_n \times$ supplemental fat				0.21	0.19	0.28	0.20	0.15	0.14	0.79	0.77	0.77	0.16	0.90

[1]n = 4.

intake, rate of egg production, egg weight, egg mass output, and BW change) were affected by the AME_n of the diets. There were no significant interactions between SFAT and AME_n in any variables examined.

Weight of Egg Components

Yolk (Tables 7 and 8) and albumen (Table 9) weights increased with the age of the hens. Proportionally, most of the increase was due to an increase in yolk weight, and, therefore, the yolk-to-albumen ratio increased with age (Table 10). Shell weight also increased with age, but at a lower rate than egg weight. From 26 to 65 wk of age, egg weight increased by 21.8%, whereas egg shell weight increased only by 14.5% (data not shown). Accordingly, the percentage of shell of the total weight of the egg was 9.6% at 26 wk of age but only 8.9% at 65 wk ($P < 0.05$) (data not shown).

An increase in the AME_n of the diets from 2,680 to 2,810 kcal/kg increased the weight of the yolks only when hens were 30 wk old when dietary LIN was at 1.15% of the diet (Table 8). Similarly, when SFAT was constant at 4% of the diet, AME_n increased yolk weight during the 26- and 30-wk age periods (Tables 7 and 8, respectively). Albumen weight was not modified by the AME_n of the diets ($P > 0.10$) during any part of the laying period (Table 9). As a consequence, yolk-to-albumen ratio [(yolk weight/albumen weight) × 100] increased at weeks 24, 26, and 30 for diets high in energy in the series of diets with the SFAT held constant at 4% (Table 10).

The influence of SFAT on yolk weight was positive but inconsistent. The increase in yolk weight was only significant ($P \leq 0.06$) at 30, 42, and 65 wk of age (Table 7). A significant interaction ($P < 0.01$) between AME_n and SFAT was observed at 42 wk, so that the improvement of yolk weight only occurred with low AME_n diets. Supplemental fat, however, consistently increased albumen weight ($P < 0.05$) during the first part of the laying cycle, but the improvement was inconsistent beyond 34 wk of age (Table 9).

Dietary LIN concentration had little effect on the weight of egg components throughout the laying cycle. Only yolk weight at 65 wk increased by 4.0%, and the yolk-to-albumen ratio increased by 2.8 and 3.7% at 24 and 26 wk of age when LIN increased from 1.15 to 1.65% (Tables 8 and 10, respectively).

DISCUSSION

The information provided by this experiment indicates that the LIN requirements of brown layers for maximum productivity is no greater than 1.15% of the diet. An in-

TABLE 6. The effects of dietary treatment on performance of hens. Supplemental fat was constant at 4% of the diet (22 to 65 wk of age)

	AME_n			Linoleic acid			
	2,680 kcal/kg	2,810 kcal/kg	P^1	1.15%	1.65%	*P*	SEM^2
Hen-day production, %	89.4	89.2	NS	89.7	88.9	NS	0.29
Egg weight, g	65.4	65.0	NS	65.3	65.1	NS	0.32
Egg mass, g/d	58.5	58.0	NS	58.6	57.8	NS	0.29
Feed consumption, g/d	123	116	0.001	120	118	NS	0.94
Energy intake, kcal/d	330	325	NS	331	324	NS	2.58
Feed efficiency							
kg feed/kg eggs	2.11	2.00	0.001	2.06	2.05	NS	0.02
kg feed/dozen eggs	1.65	1.56	0.001	1.61	1.60	NS	0.02
BW change, g	251	245	NS	270	226	NS	24.3

[1]NS = $P > 0.05$.

[2]n = 8 for main effects and n = 4 for interactions.

TABLE 7. The influence of dietary treatment on yolk weight. Linoleic acid was constant at 1.15% of the diet

	Diet	AME_n	Supplemental fat	Age of hens						
				24 wk	26 wk	30 wk	34 wk	38 wk	42 wk	65 wk
		(kcal/kg)	(%)							
	4	2,680	0	12.0	13.8	14.5	15.3	17.1	16.8[z]	18.0
	5	2,680	4	12.1	13.9	15.2	16.0	17.3	18.0[x]	18.8
	1	2,810	0	12.1	13.8	15.1	16.1	16.8	17.4[y]	18.2
	2	2,810	4	12.3	14.2	15.4	16.2	17.2	17.3[y]	18.5
SEM[1]				0.13	0.15	0.17	0.28	0.15	0.21	0.26
				P						
AME_n				0.44	0.22	0.03	0.09	0.21	0.77	0.74
Supplemental fat				0.14	0.10	0.02	0.18	0.12	0.02	0.06
AME_n × supplemental fat				0.84	0.43	0.47	0.31	0.63	0.01	0.33

[x–z]Means with different superscript within the same column differ ($P < 0.05$).

[1]n = 4.

crease in LIN content from 1.15 to 1.65% did not improve any trait at any point of the laying cycle, although an increase ($P < 0.06$) of the yolk proportion was observed in the early laying period (24 and 26 wk of age). These results contrast with those of Scragg *et al.* (1987), who observed an increase in mean egg weight with LIN concentrations up to 2.33% in the diet. However, other authors (Shannon and Whitehead, 1974; Whitehead, 1981) did not find any beneficial effect on this trait with levels of LIN from 0.8 to 1.15%.

An increase in the AME_n of the diet often results in an improved performance and increased egg weight (Bray, 1967; DeGroote, 1972). However, in most studies conducted with variable AME_n, the increase in energy was obtained by adding fat, and, therefore, the effects of increased AME_n and SFAT are confounded (Parsons *et al.*, 1993).

In our trial, an increase in the AME_n of the diet did not improve laying rate, egg weight, or egg mass output, which agrees with the results of Summers and Leeson (1983, 1993), Sell *et al.* (1987), and Keshavarz and Nakajima (1995). Hens fed the higher energy diets had greater BW gains than hens fed the lower energy diets. Rate of egg production and egg weight during the early stages of the laying cycle are positively related to the BW of the pullets. Summers and Lesson (1983) found that heavier pullets laid significantly more and heavier eggs than did lighter pullets from 19 to 25 wk of age. Therefore, an increase in AME_n of the diet might be of value for underweight pullets but not for well-managed pullets (Keshavarz, 1995). The pullets in this trial weighed 1,710 g at 20 wk, which slightly exceeded the recommendations for the Isabrown strain at this age. Therefore, no improvement in egg weight because of an increase in AME_n of the diet should be expected in our case.

Supplemental fat resulted in an improvement in most of the traits studied, including egg weight. The beneficial effects of SFAT on egg weight were more pronounced with the low-energy diets at the beginning of the laying period and were independent of the LIN content of the diets. Previous research showed also that SFAT exerts a favorable effect on egg weight beyond that attributable to an increase in LIN concentration (Shannon and Whitehead, 1974; Sell *et al.*, 1987; Keshavarz, 1995) or in the AME_n of the diets (Sell *et al.*, 1987; Parsons *et al.*, 1993; Keshavarz and Nakajima, 1995). However, Sell *et al.* (1979), Summers and Leeson (1983), and Atteh and Leeson (1985) were not able to detect any effect of SFAT

TABLE 8. The influence of dietary treatment on yolk weight. Supplemental fat was constant at 4% of the diet

	Diet	AME_n	Linoleic acid	Age of hens						
				24 wk	26 wk	30 wk	34 wk	38 wk	42 wk	65 wk
		(kcal/kg)	(%)							
	5	2,680	1.15	12.1	13.9	15.2	16.0	17.3	18.0	18.8
	6	2,680	1.65	12.5	13.9	15.1	16.1	17.6	17.4	19.1
	2	2,810	1.15	12.3	14.3	15.5	16.2	17.2	17.3	18.5
	3	2,810	1.65	12.5	14.5	15.6	16.3	17.3	17.4	19.7
SEM[1]				0.13	0.11	0.16	0.20	0.14	0.23	0.13
				P						
AME_n				0.70	0.01	0.03	0.27	0.19	0.12	0.64
Linoleic acid				0.08	0.52	0.94	0.80	0.19	0.25	0.03
AME_n × linoleic acid				0.57	0.21	0.59	0.99	0.61	0.18	0.13

[1]n = 4.

TABLE 9. The influence of dietary treatment on albumen weight. Linoleic acid was constant at 1.15% of the diet

	Diet	AME_n	Supplemental fat	Age of hens						
				24 wk	26 wk	30 wk	34 wk	38 wk	42 wk	65 wk
		(kcal/kg)	(%)							
	4	2,680	0	36.9^{z}	37.2	39.6	39.1^{z}	41.0	37.9	42.1
	5	2,680	4	39.5^{x}	40.6	41.4	41.8^{y}	41.2	41.1	41.4
	1	2,810	0	38.1^{xy}	37.6	40.5	40.4^{x}	40.1	38.3	40.4
	2	2,810	4	37.9^{y}	39.2	41.3	40.8^{x}	40.8	39.3	42.4
SEM[1]				0.61	0.77	0.56	0.50	0.55	0.68	1.42
				P						
AME_n				0.78	0.57	0.46	0.75	0.25	0.33	0.82
Supplemental fat				0.08	0.01	0.05	0.01	0.44	0.01	0.90
AME_n × supplemental fat				0.05	0.27	0.37	0.04	0.64	0.12	0.36

$^{x-z}$Means with different superscript in the same column differ ($P < 0.05$).

[1]n = 4.

on egg weight. The reasons for these discrepancies are not known, but are probably due to the distinct experimental protocols used. For example, the hens used by these previous authors were from different strains, had different weights and age, and had a different rearing program than the hens used by us. Furthermore, the major dietary ingredients used also differed among studies. Our results clearly show that SFAT improves hen performance and egg weight and that these effects were independent of the LIN and the energy supplied by the SFAT. Therefore, a positive effect of supplemental fat *per se* on performance of hens should be expected under practical conditions.

The period between 41 and 57 wk of age corresponded to summer conditions in which temperatures inside the poultry house often reached 32 C and more. Under these circumstances, feed intake was significantly improved by SFAT (Figure 1). When LIN was maintained constant at 1.15%, hens fed diets containing 4% fat ate more feed than hens fed diets with no supplemental fat, and the extra nutrients consumed by SFAT-fed hens were used to produce more eggs that were heavier (Tables 4 and 5) rather than to increase the BW of the hens (Figure 2). These data illustrate that the beneficial effects of SFAT are more pronounced under situations of heat stress, as has been previously reported by other authors (Mateos and Méndez, 1990; Teeter and Belay, 1996; Bonnet *et al.*, 1997; Rand *et al.*, 1997). The influence of increasing the AME_n of the diets under conditions of hot weather was less evident as neither egg rate nor egg weight were significantly improved.

The mechanism by which SFAT influences egg weight remains unclear. Most of the research conducted on the response of egg weight to LIN, AME_n, and SFAT concentrations in the diet have focused on the egg weight, and little attention has been paid to the concurrent changes in yolk and albumen weights that might have taken place. March and McMillan (1990) suggested that LIN content might enhance synthesis of lipoproteins in the liver that eventually would be secreted and taken up by the developing oocytes. Dietary fat (*i.e.*, long-chain fatty acids) entering the portal blood would be in the form of portomicrons, which are broken down by the liver. The lipids are then re-synthesized as components of very low density lipoprotein particles for direct transport and deposition into the yolk (see review by Nimpf and Schneider, 1991). Jensen *et al.* have pointed out that LIN increased egg weight by augmenting yolk weight, and that the beneficial

TABLE 10. The influence of dietary treatment on yolk to albumen ratio. Supplemental fat constant at 4% of the diet

	Diet	AME_n	Linoleic acid	Age of hens						
				24 wk	26 wk	30 wk	34 wk	38 wk	42 wk	65 wk
		(kcal/kg)	(%)							
	5	2,680	1.15	0.31	0.34	0.36	0.38	0.42	0.44	0.47
	6	2,680	1.65	0.32	0.36	0.37	0.39	0.42	0.44	0.46
	2	2,810	1.15	0.32	0.36	0.37	0.40	0.42	0.44	0.45
	3	2,810	1.65	0.32	0.37	0.38	0.39	0.43	0.43	0.47
SEM[1]				0.42	0.58	0.45	0.61	0.58	0.36	1.11
				P						
AME_n				0.04	0.05	0.04	0.16	0.60	0.19	0.58
Linoleic acid				0.04	0.06	0.48	0.44	0.16	0.19	0.80
AME_n × linoleic acid				0.20	0.16	0.87	0.23	0.88	0.08	0.21

[1]n = 4.

	P										
ANOVA											
Source of variation											
AME_n	0.61	0.03	0.07	0.01	0.01	0.01	0.01	0.01	0.01	0.02	0.09
Supplemental fat	0.20	0.10	0.10	0.45	0.02	0.02	0.01	0.05	0.08	0.78	0.72
AME_n × supplemental fat	0.18	0.73	0.89	0.29	0.99	0.19	0.57	0.49	0.30	0.34	0.97

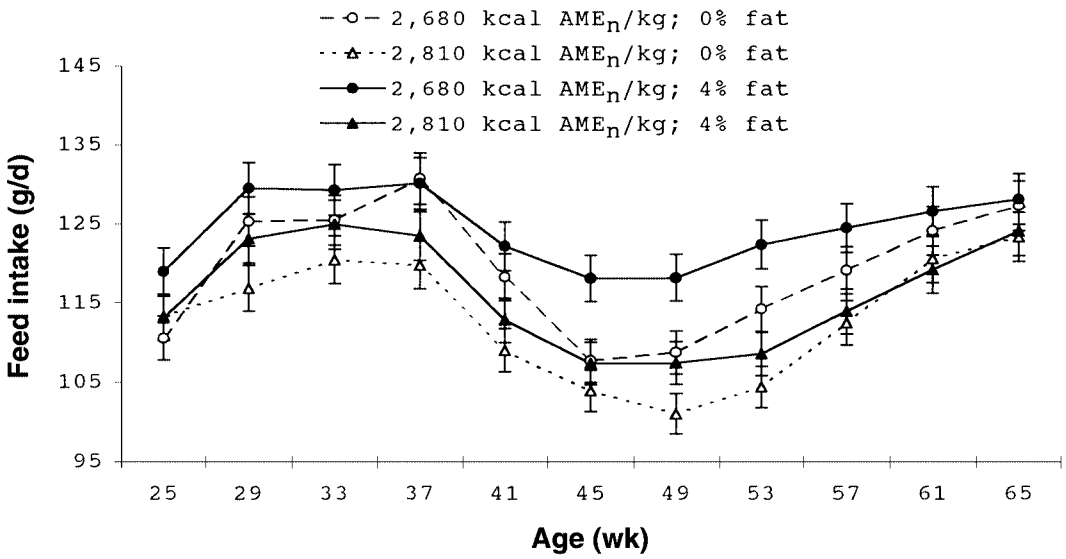

FIGURE 1. The influence of dietary treatment on feed intake. Linoleic acid was constant at 1.15% of the diet (n = 4 observations per each combination of treatments; SD = 6.5).

effects will take place only if LIN-deficient diets are used as controls. In our study, supplementation of the diet with LIN above 1.15% did not increase egg weight, although yolk weight increased at the end of the laying cycle (65 wk) and yolk-to-albumen ratio increased at the early laying period (24 and 26 wk of age).

In this trial, SFAT increased yolk and albumen weight and modified the composition of the eggs. Sell *et al.* (1987)

	P								
ANOVA									
Source of variation									
AME_n	0.39	0.95	0.22	0.45	0.26	0.02	0.06	0.12	0.20
Supplemental fat	0.10	0.34	0.10	0.26	0.14	0.02	0.01	0.03	0.02
AME_n × supplemental fat	0.27	0.51	0.15	0.21	0.23	0.19	0.40	0.26	0.47

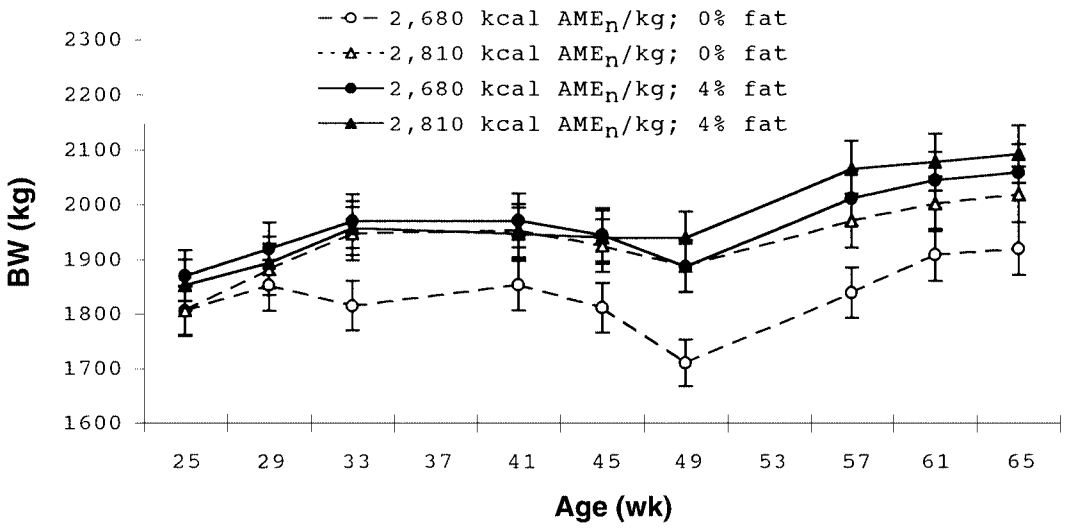

FIGURE 2. The influence of dietary treatment on BW of hens. Linoleic acid was constant at 1.15% of the diet (n = 4 observations per each combination of treatments; SD = 119).

found that fat supplementation to isocaloric diets of White Leghorn hens increased yolk weight, but weight of the albumen was not affected. They hypothesized that the rate of hepatic synthesis of lipoproteins by hens during early egg production was insufficient to supply the amounts of lipids needed to achieve optimum egg yolk development and that the exogenous fat might help to meet these needs. Older hens might have an adequate capacity for hepatic very low density lipoprotein synthesis to fulfill requirements for yolk formation. These authors observed that SFAT had the greatest effects on yolk weight from 24 to 38 wk and that the beneficial effects essentially disappeared by 38 wk of age. In our trial, SFAT increased yolk and albumen weight, but the effect was more consistent for albumen weight. These results agree with those of Keshavarz and Nakajima (1995). Whitehead *et al.* (1991) found that the increase in egg weight with age was associated with a greater increase in the proportion of yolk at the expense of albumen, whereas the increase in egg weight caused by SFAT was mostly due to an increase in egg albumen, although yolk weight was also increased. Whitehead *et al.* (1993) and Whitehead (1995) indicated that dietary fatty acids may increase egg weight by stimulating the synthesis of oviducal proteins, a mechanism that is different from that causing the age-related increase in weight. Because the formation of most of the oviduct proteins is stimulated by estrogens, it is possible that the function of this hormone in the laying hen is influenced by dietary fatty acids. In fact, Whitehead *et al.* (1993) observed that egg albumen was increased in young (< 30 wk) and old (> 46 wk) hens and that the improvement in egg weight in the old hens was primarily due to an increase in albumen. Mean plasma estradiol concentrations were highly correlated with the changes in egg weight. They concluded that estrogens are important in controlling egg weight and that dietary fats influence egg weight by modifying estrogen metabolism. Furthermore, under the influence of estrogen, the avian liver markedly increases very low density lipoprotein synthesis and begins producing vitellogenin (Nimpf and Schneider, 1991).

The results reported here show that once the LIN requirement of hens is fulfilled (1.15% of the diet or less), SFAT increased egg weight independently of LIN and AME_n content of the diet. This observation supports the conclusions of Balnave (1971), Whitehead (1981), and Keshavarz and Nakajima (1995), indicating that SFAT exerts favorable effects on egg weight beyond those attributable to other nutrients of the diet.

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