

## Effect of dietary levels of calcium on performance of pullets and layers, physical characteristics of the egg, and calcium and phosphorus in plasma and bone

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### Summary

Medium-heavy pullets were housed in individual cages with a constant environment, and 432 were selected by weight from a total of 600, at 16 weeks of age. Each of 9 treatments was composed of 12 pullets in 4 replicates. Diets contained 1.5, 3.7 and 5.0 % calcium in several combinations.

1. After a laying phase (at 23 weeks of age when the pullets reached 50 % production), pullets given 5.0 % calcium had significantly lower body weight and an insignificantly higher feed consumption than those given 1.5 or 3.7 % calcium.
2. Calcium in plasma did not significantly differ with level of calcium, whereas inorganic phosphorus in plasma was significantly higher with the lowest level of calcium. As the ratio of calcium to phosphorus decreased, inorganic phosphorus in plasma significantly increased.
3. During the laying phase lasting 22 weeks, egg production was significantly affected by level of calcium. The highest production was with 3.7 % calcium; low levels or high levels of calcium depressed egg production.
4. Egg weight did not differ significantly with level of calcium.
5. Feed consumption increased significantly with level of calcium, because of the low energy level of the high-calcium diet.
6. Egg-shell quality determined as shell thickness, specific gravity (relative density) and proportion of shell was highly significantly affected by each higher level of dietary calcium.
7. Albumen index and yolk index did not differ significantly with calcium level.
8. Tibial ash and breaking strength increased significantly with dietary calcium levels, with clear interrelation.
9. Calcium in plasma was significantly affected by calcium in diet but inorganic phosphorus in plasma was not.

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10. There was a clear relation of calcium in plasma with shell quality and with tibia characteristics.

### Introduction

In recent years, there have been many reports on dietary levels of calcium for laying diets. However few have studied the interaction of calcium levels before and during laying, with the performance of laying hens.

Several research workers (Anderson, 1967; Hurwitz & Bar, 1971; Meyer et al., 1971; Mraz, 1972) have studied the carry-over of prelaying calcium levels on laying performance. Other workers (Berg et al., 1964; Miller & Sunde, 1975) have reported the relation between prelaying diets and laying diets with a range of calcium levels.

There is considerable controversy about the effects of low and high levels on several production traits. Some workers reported depression of egg production (Reddy & Sanford, 1963; Reddy et al., 1968; Ademosun & Kalango, 1973; Tortuero & Centeno, 1973) and of egg weight (Reddy & Sanford, 1963; Reddy et al., 1968; Reichmann & Connor, 1977) with high levels, while others found improved egg production and egg weight with high levels (Mehring, 1965; Miller & Sunde, 1975). No beneficial effect on these parameters (Hurwitz & Bornstein, 1966; Harms & Waldroup, 1971) was found with diets low and high in calcium.

Egg-shell quality as measured by shell thickness, specific gravity (relative density) and shell percentage was improved with the high-calcium diets over low-calcium diets (Reddy & Sanford, 1963; Mehring, 1965; Reddy et al., 1968; Ademosun & Kalango, 1973; Roland et al., 1974; Miller & Sunde, 1975; Reichmann & Connor, 1977).

Internal quality of eggs (Haugh units) was not affected by dietary calcium (Reddy & Sanford, 1963; Reddy et al., 1968).

Hurwitz & Bornstein (1966), Harms & Waldroup (1971), Reichmann & Connor (1977) found that dietary calcium did not affect feed consumption, while Ademosun & Kalango (1973), Tortuero & Centeno (1973) found that high levels depressed feed consumption.

Body weight was decreased with low or high levels according to some workers (Mehring, 1965; Reddy et al., 1968) but not according to others (Hurwitz & Bornstein, 1966; Pepper et al., 1967; Reichmann & Connor, 1977).

Rowland et al. (1967, 1968, 1973), Kalango & Ademosun (1973), and Reichmann & Connor (1977) found that content of ash in tibia and of calcium in tibia ash increased significantly with calcium level.

Reddy et al. (1968), Paul & Snetsinger (1969), Reichmann & Connor (1977) suggested that there is a significant positive correlation between dietary calcium and plasma calcium.

The purpose of this study was to examine the effects of low and high levels of dietary calcium before and during laying on the productive performance of laying hens.

Table 1. Experimental allocation of the rations.

Prelaying phase	Laying phase		
	L	M	H
l	lL	lM	lH
m	mL	mM	mH
h	hL	hM	hH

L = 1.5 %; M = 3.7 %; H = 5.0 % Ca.

## Materials and methods

### *Prelaying phase*

From 600 pullets 16 weeks of age medium-heavy breed, a cross of three strains developed at the Department, 432 were selected at 16 weeks of age by weight and randomly distributed in individual cages in a house of a constant environment (temp. 10 °C, rel.hum. 80 %, and light phase 14 h). Each of the 9 treatments was composed of 12 pullets in 4 replicates. The dietary treatments were 1.5, 3.7 and 5.0 % calcium in several combinations (Table 1). Details of the three diets are given in Table 2. Body weight and feed consumption were recorded for all pullets from 16 weeks of age till 23 weeks (Table 3).

At 23 weeks of age, calcium (method of Fales, 1953) and inorganic phosphorus (Gardiner, 1962) were estimated in plasma from duplicate lots of 4 pullets.

### *Laying phase*

When each pullet reached 50 % production, the dietary treatment was changed as in Table 1. The trial continued for 22 weeks during production. Data were compiled on egg production on hen-day basis, egg weight, feed consumption for each hen (g/day), external and internal quality of eggs (all eggs on one day each month). Ash and breaking strength (Rowland et al., 1967; AOAC, 1970) of the right tibia were estimated for duplicate samples of 4 hens from each group at the end of the trial. Blood plasma of the layers was analysed for calcium and inorganic phosphorus once a month from duplicate samples of 4 hens for each group. The blood samples were drawn immediately after laying.

All data were analysed factorially for variance and, when applicable, treatment means were separated by Duncan's multiple range test (1965).

## Results and discussion

### *Performance before laying*

The results are summarized in Table 3, which shows that the pullets given 5.0 % calcium from 16 to 23 weeks of age were significantly lighter with a lower gain in body weight. The higher feed consumption of pullets fed on diets rich in calcium than of those on diets low in calcium was probably due to the lower energy level

Table 2. Composition of experimental diets and their calculated analysis. Values other than energy and ratios are in per cent.

Ingredients	L	M	H
	Ca 1.5 %	Ca 3.7 %	Ca 5.0 %
Yellow maize (ground)	58.7	49.5	43.7
Barley	10.0	10.0	10.0
Soya bean oil meal (44 %)	20.1	21.9	23.0
Lucerne meal (20 %)	2.5	2.5	2.5
Dried whey	1.5	1.5	1.5
Animal fat hydrolysed	1.2	2.3	3.3
Iodized salt	0.5	0.5	0.5
Vitamin and trace elements <sup>1</sup>	1.0	1.0	1.0
Calcium hydrogen phosphate dihydrate <sup>2</sup> (23.3 % Ca, 19 % P)	2.2	2.2	2.2
Limestone (35 % Ca)	2.3	8.6	12.3
Total	100.0	100.0	100.0
<i>Calculated analysis</i>			
Crude protein	15.9	15.9	15.0
M.E.MJ/kg	12.12	11.35	10.96
Fat	3.90	4.41	4.97
Crude fiber	3.83	3.74	3.67
Ca	1.5	3.7	5.0
P(total)	0.80	0.79	0.77
P(available)	0.59	0.58	0.58
Ca:P(available)	2.54	6.38	8.62
Methionine	0.35	0.34	0.33
Cystine	0.29	0.29	0.28
Lysine	0.79	0.81	0.83

<sup>1</sup> Vitamin and mineral mixture (Farmix 10) contains in 1 kg: vitamin A 750 000 I.U.; vitamin D<sub>3</sub> 150 000 I.U.; vitamin E 500 I.U.; vitamin K<sub>3</sub> 0.1 g; vitamin B<sub>2</sub> 0.4 g; vitamin B<sub>6</sub> 50 mg; nicotinic acid 1.4 g; D. pantho. acid 0.5 g; vitamin B<sub>12</sub> 1 mg; choline chloride 17.5 g; Fe 2 g; Mn 7 g; Cu 1 g; Zn 3 g; I 0.1 g; Co 0.3 g.

<sup>2</sup> CaHPO<sub>4</sub> · 2H<sub>2</sub>O; trivial name dicalcium phosphate.

Table 3. Effect of level of calcium in diets from 16 to 23 weeks of the prelaying phase on body weight, weight gain, feed consumption, plasma calcium and inorganic phosphorus.

Factor	Low level (l) (1.5 % Ca in diet)	Medium level (m) (3.7 % Ca in diet)	High level (h) (5.0 % Ca in diet)
<i>Body weight (g)</i>			
16 weeks	1907	1903	1901
18 weeks	2109 <sup>b</sup>	2069 <sup>ab</sup>	2054 <sup>a</sup>
20 weeks	2387 <sup>b</sup>	2299 <sup>a</sup>	2263 <sup>a</sup>
22 weeks	2633 <sup>b</sup>	2524 <sup>a</sup>	2485 <sup>a</sup>
23 weeks	2635 <sup>b</sup>	2561 <sup>a</sup>	2533 <sup>a</sup>
Gain 23-16 weeks	728 <sup>b</sup>	658 <sup>a</sup>	632 <sup>a</sup>
<i>Feed consumption</i>			
(g/day)	124.7	122.0	127.3
Ca in plasma (mg/100 ml)	21.4	20.5	20.9
P in plasma (mg/100 ml)	6.63 <sup>b</sup>	4.78 <sup>a</sup>	4.63 <sup>a</sup>

Means with different superscripts horizontally are significantly different ( $P < 0.05$ ) by Duncan's multiple range test.

of the high-calcium diets. In similar trials, Miller & Sunde (1975) found with calcium level of 4.5 % and phosphorus level of 1.0 % body weight was not significantly less than with lower levels of calcium.

Plasma calcium did not show any significant difference with the several levels of calcium; it was insignificantly higher with the lower level. The calcium level in blood plasma seemed to react in another way for laying hens during shell formation than in the prelaying phase. The medullary bone or the stores can accumulate all the calcium consumed. Berg et al. (1964), working with calcium levels from 0.6 to 2.01 % during the growing period 8-21 weeks, showed the same sort of trend in serum calcium but insignificantly.

Inorganic phosphorus in plasma was significantly higher with the lower level of calcium. Inorganic phosphorus in plasma seemed to increase as the ratio of calcium to phosphorus decreases.

#### *Performance during laying*

Results during laying are presented in Tables 4 and 5. Egg production was significantly affected by some of the dietary treatments. The best production was with 3.7 % calcium during laying; low levels and high levels of calcium during laying depressed egg production. Our results agreed with the work of Reddy & Sanford (1963) and Reddy et al. (1968) who noticed that as calcium levels increased from 2.25 to 3.85 % there was a gradual increase in egg production, which was reversed when the levels reached 5.05 % calcium. Ademosun & Kalango (1973) found that a low level (2.0 %) and a high level (4.25 %) of calcium depressed egg production. Mehring (1965) and Miller & Sunde (1975) found the reverse, that egg production increased as calcium level increased to 6.0 %. Reichmann & Connor (1977) found that treatments from 2.4 to 5.69 % calcium did not affect egg production.

For egg weight, there were no significant differences between the several calcium levels (Table 4). A calcium level of 3.7 % gave the insignificantly heaviest egg weight. Our results agree with those of Harms & Waldroup (1971), who noticed that a dietary level up to 5.0 % did not significantly affect egg weight, and are contrary to the results of Mehring (1965), who found that egg weight was improved significantly with high levels to 6.0 %. Reichmann & Connor (1977) observed the reverse, that a high level, 5.69 % calcium, depressed egg weight significantly.

Feed consumption increased significantly with increase in dietary calcium during the laying period, perhaps because of the low energy level of the high-calcium diet. Our results agree with those of Miller & Sunde (1975) and conflict with those of Mehring (1965), and Hurwitz & Bornstein (1966). In our work, the lower egg production obtained with the lower level of calcium may be due to the low feed consumption. We could not draw any clear conclusions on the relation prelaying and laying.

The level of calcium had its greatest effect on shell quality as measured by shell thickness, specific gravity (relative density) and proportion of shell (Table 4). If the prelaying treatments are aggregated, shell quality improved significantly with each calcium level. Our results closely agree with those of Reddy & Sanford (1963),

Table 4. Effect of level of calcium in the pre-laying and laying diets on various performance characteristics of the laying hens. For codes of dietary treatments see Table 1.

	L			M			H			Production phase			
	1.5 %	m 3.7 %	h 5.0 %	1.5 %	m 3.7 %	h 5.0 %	1.5 %	m 3.7 %	h 5.0 %	1+m+h	L	M	H
Egg production (%)	72.8 <sup>bc</sup>	73.9 <sup>c</sup>	74.6 <sup>c</sup>	69.9 <sup>ab</sup>	77.0 <sup>d</sup>	69.3 <sup>a</sup>	69.0 <sup>a</sup>	72.9 <sup>bc</sup>	72.8 <sup>bc</sup>	70.6 <sup>a</sup>	74.6 <sup>c</sup>	74.6 <sup>c</sup>	72.3 <sup>b</sup>
Egg weight (g)	59.5	60.9	60.2	60.8	60.5	61.0	59.7	61.6	60.7	60.0	61.0	61.0	60.6
Feed consumption (g/day)	123.4 <sup>a</sup>	130.7 <sup>e</sup>	133.1 <sup>de</sup>	124.7 <sup>ab</sup>	130.5 <sup>e</sup>	131.8 <sup>cd</sup>	126.3 <sup>b</sup>	129.6 <sup>c</sup>	134.6 <sup>e</sup>	124.8 <sup>a</sup>	130.3 <sup>b</sup>	130.3 <sup>b</sup>	133.2 <sup>c</sup>
Shell thickness (mm)	0.326 <sup>a</sup>	0.349 <sup>bc</sup>	0.363 <sup>d</sup>	0.328 <sup>a</sup>	0.352 <sup>cd</sup>	0.364 <sup>d</sup>	0.335 <sup>ab</sup>	0.358 <sup>cd</sup>	0.365 <sup>d</sup>	0.330 <sup>a</sup>	0.353 <sup>b</sup>	0.353 <sup>b</sup>	0.364 <sup>c</sup>
Specific gravity*	796 <sup>a</sup>	833 <sup>bc</sup>	858 <sup>c</sup>	788 <sup>a</sup>	840 <sup>e</sup>	852 <sup>c</sup>	806 <sup>ab</sup>	842 <sup>c</sup>	856 <sup>c</sup>	797 <sup>a</sup>	838 <sup>b</sup>	838 <sup>b</sup>	855 <sup>c</sup>
Shell percentage	7.34 <sup>a</sup>	8.16 <sup>bc</sup>	8.77 <sup>d</sup>	7.24 <sup>a</sup>	8.31 <sup>cd</sup>	8.71 <sup>cd</sup>	7.56 <sup>ab</sup>	8.42 <sup>cd</sup>	8.74 <sup>d</sup>	7.38 <sup>a</sup>	8.30 <sup>b</sup>	8.30 <sup>b</sup>	8.74 <sup>c</sup>
Yolk index	0.428	0.427	0.429	0.422	0.426	0.424	0.425	0.427	0.427	0.425	0.426	0.426	0.427
Albumen index	0.089	0.085	0.080	0.085	0.083	0.081	0.085	0.084	0.086	0.084	0.083	0.083	0.085

Means with different superscripts horizontally are significantly different ( $P < 0.05$ ) by Duncan's multiple range test in the two combinations.

\* Specific gravity = 1.0.

Table 5. Effect of level of calcium in the pre-laying and laying diets on tibia ash, breaking strength, plasma calcium and inorganic phosphorus of the laying hens.

	L : 1.5 %			M : 3.7 %			H : 5.0 %			Production phase			
	1.5 %	m 3.7 %	h 5.0 %	1.5 %	m 3.7 %	h 5.0 %	1.5 %	m 3.7 %	h 5.0 %	1+m+h	L	M	H
Ash in tibia (%)	58.1 <sup>ab</sup>	59.4 <sup>b</sup>	59.1 <sup>ab</sup>	57.6 <sup>ab</sup>	59.1 <sup>ab</sup>	59.1 <sup>ab</sup>	56.9 <sup>a</sup>	58.9 <sup>ab</sup>	59.6 <sup>b</sup>	57.5 <sup>a</sup>	59.2 <sup>b</sup>	59.2 <sup>b</sup>	59.3 <sup>b</sup>
Breaking strength of tibia (kgf)	9.70 <sup>a</sup>	10.36 <sup>ab</sup>	12.08 <sup>ab</sup>	10.29 <sup>ab</sup>	10.48 <sup>ab</sup>	11.69 <sup>ab</sup>	8.63 <sup>a</sup>	11.49 <sup>ab</sup>	13.66 <sup>b</sup>	9.54 <sup>a</sup>	10.78 <sup>a</sup>	10.78 <sup>a</sup>	12.48 <sup>b</sup>
Ca in plasma (mg/100 ml)	23.7 <sup>a</sup>	26.8 <sup>bc</sup>	27.7 <sup>c</sup>	24.0 <sup>ab</sup>	28.2 <sup>c</sup>	27.5 <sup>c</sup>	26.0 <sup>abc</sup>	27.7 <sup>c</sup>	28.2 <sup>c</sup>	24.6 <sup>a</sup>	27.6 <sup>b</sup>	27.6 <sup>b</sup>	27.8 <sup>b</sup>
Inorganic P in plasma (mg/100 ml)	5.38	5.47	5.03	5.63	5.83	5.69	5.80	5.49	5.61	5.60	5.60	5.60	5.44

Means with different superscripts horizontally are significantly different ( $P < 0.05$ ) by Duncan's multiple range test in the two combinations.

1.1 kgf = 9.8 N.

Mehring (1965), Reddy et al. (1968), Ademosun & Kalango (1973), Roland et al. (1974), Miller & Sunde (1975) and Reichmann & Connor (1977). The highest level of calcium during laying seems to increase the stores of calcium in medullary bone, from which more would be released into the blood as protein-bound calcium, so increasing shell calcification in the uterus. It is not worth-while to increase shell thickness and to decrease egg production with extra high calcium levels in the diet. Our results do not support those of Paul & Snetsinger (1969), Harms & Waldroup (1971), who observed that high levels of 4.25 % and 5.0 % calcium depressed shell quality.

Internal quality of eggs as measured by yolk and albumen indices did not show any significant difference with level of calcium (Table 4). Reddy & Sanford (1963) and Reddy et al. (1968) found that egg quality (Haugh units) was not affected by dietary level. However our treatments are not comparable with theirs.

Increasing calcium levels significantly increased proportion of tibia ash and breaking strength with clear relation between the two (Table 5). Rowland et al. (1967; 1968; 1973), Kalango & Ademosun (1973), Reichmann & Connor (1977) came to a similar conclusion. So bone minerals and breaking strength vary with level of dietary calcium before and during laying.

Plasma calcium levels increased insignificantly with each step of dietary calcium, but only the 1.5 % level of calcium was significantly different from the others (Table 5). Inorganic phosphorus in plasma did not react. Our results agree with the work of Reddy et al. (1968), Paul & Snetsinger (1969), Reichmann & Connor (1977), who pointed out that plasma calcium increased significantly with dietary calcium.

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